生物物理

SEIBUTSU BUTSURI

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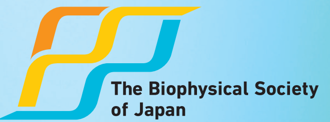
第54回年会予稿集

2016.11.25（金）～27（日）

つくば国際会議場

主催　一般社団法人　日本生物物理学会



一般社団法人　日本生物物理学会　http://www.biophys.jp

第 54 回日本生物物理学会年会

The 54th Annual Meeting of the Biophysical Society of Japan

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The 54th Annual Meeting of the Biophysical Society of Japan（BSJ2016）

第54回日本生物物理学会年会（2016年度）

会 期：
Date：

2016年 11月25日（金）－ 27日（日）
November 25（Fri）－ 27（Sun）, 2016

会 場：
Venue：

つくば国際会議場
（茨城県つくば市竹園2-20-3）
Tsukuba International Congress Center
（2-20-3 Takezono, Tsukuba, Ibaraki 305-0032, Japan）

年会実行委員長：
Chair：

豊島 陽子
（東京大学 大学院総合文化研究科）
Yoko Toyoshima
（The University of Tokyo）

HOMEPAGE　http://www.aeplan.co.jp/bsj2016/

※会期後は日本生物物理学会にデータが移行されますので、学会ホームページよりご覧ください。

抄録本文（Abstract）...

オンライン講演予稿集は、こちらからダウンロードいただけます。

http://www.biophys.jp/dl/pro/54th_proceedings.pdf
ID：ambsj54　Password ： tsukuba2016

※スマートフォン・タブレット端末向けのプログラム検索・要旨閲覧アプリは、
2016年 11月21日（月）に公開予定です。

（筑波山）

編集・発行 ： 第54回日本生物物理学会年会実行委員会

第54回日本生物物理学会年会事務局
東京都目黒区駒場3-8-1
東京大学 大学院総合文化研究科 広域科学専攻 生命環境科学系内
E-mail: bp_nenkai54@bio.c.u-tokyo.ac.jp
発行日：表4（裏表紙）記載

第54回日本生物物理学会年会（2016年度）

開催にあたって

第54回年会 実行委員長

豊島 陽子

（東京大学 大学院総合文化研究科）

一般社団法人日本生物物理学会第54回年会を、つくば国際会議場にて2016年11月25日～27日の日程で開催いたします。つくばでの開催は1996年以来20年ぶりとなりますが、この間、つくば国際会議場が開設され、東京とつくばを結ぶ「つくばエクスプレス」が開通し、利便性が向上しました。秋も深まる中、つくばにおいて熱い議論や研究者間の交流が深まることを期待しています。

つくば年会では、例年通り、すべての発表は英語で行われます。34のシンポジウムには、生物物理分野のフロントをめざす一般会員からの公募によるものと大型プロジェクトを組織している研究者に応募いただいたものに加え、本年会では日中、日韓、日豪、の3つの二国間交流シンポジウムを学会本部に企画していただきました。東アジアにおける国際交流を促進する機会になることが期待されます。

一般発表は、937件の応募がありまして、すべてポスター発表といたします。12回目を迎える若手奨励賞と若手招待講演賞に加えて、今回、新たに学生発表賞が創設されました。これは、大学院生や学部学生を対象に、優秀なポスター発表を表彰するもので、若手研究者を対象とした従来の賞と比べ、より若い年代層の会員をエンカレッジすることと思います。

さらに今年は、男女共同参画・若手支援シンポジウムとキャリア支援説明会の２つの企画に連携をもたせ、若手会員のキャリア構築の一助となるように工夫をいたしました。総会シンポジウムやBPPB論文賞受賞講演会なども年会におけるイベントとして定着してきました。また、科研費説明会では、科研費制度改革に関する最新の情報を提供いたします。協賛企業による展示会やランチョンセミナーにおいては、最新の計測機器や研究を支援するシステムや製品等の情報が紹介されます。

本年会における研究発表や討論、さまざまなアクティビティが、会員の皆様にとって有益なものとなり、生物物理分野の研究の新たな一歩につながり、今後、益々発展することを願っております。

第54回日本生物物理学会年会実行委員会
- Organizers -

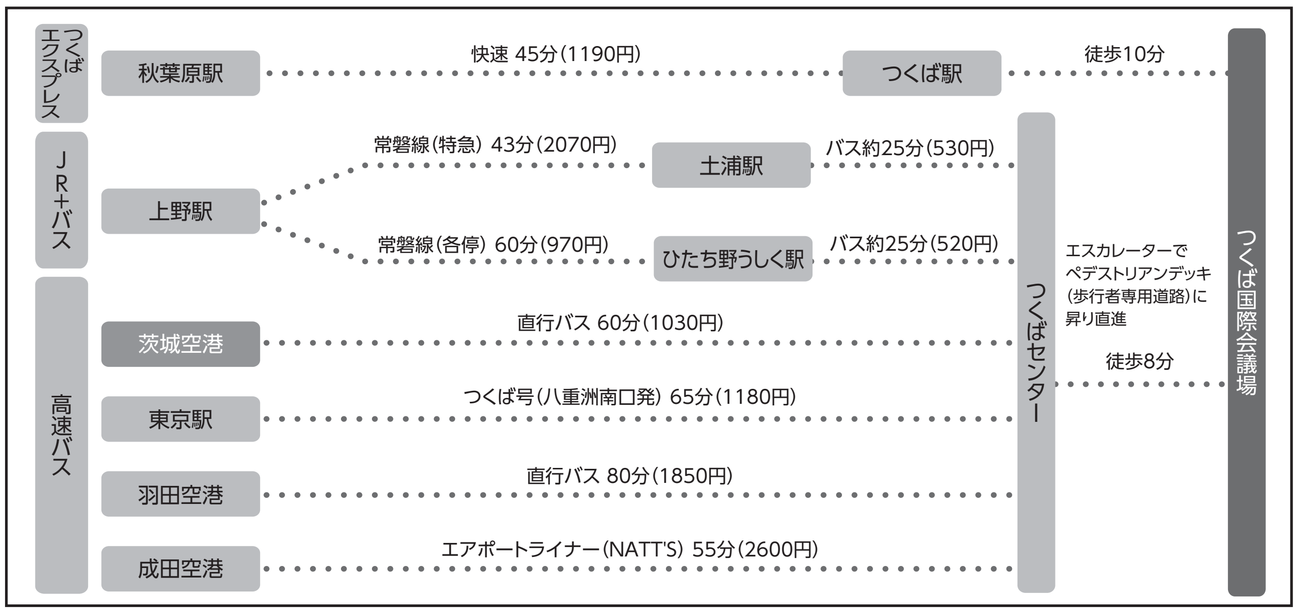
年会実行委員長	Chair
豊島 陽子（東京大学 大学院総合文化研究科）	Yoko Toyoshima（The University of Tokyo）

実行委員		Executive Committee Members	
新井 宗仁	（東京大学 大学院総合文化研究科）	Munehito Arai	（The University of Tokyo）
金子 邦彦	（東京大学 大学院総合文化研究科）	Kunihiko Kaneko	（The University of Tokyo）
加納 ふみ	（東京工業大学 科学技術創成研究院）	Fumi Kano	（Tokyo Institute of Technology）
上村 慎治	（中央大学 理工学部）	Shinji Kamimura	（Chuo University）
小林 琢也	（東京大学 大学院総合文化研究科）	Takuya Kobayashi	（The University of Tokyo）
佐藤 直樹	（東京大学 大学院総合文化研究科）	Naoki Sato	（The University of Tokyo）
澤井 哲	（東京大学 大学院総合文化研究科）	Satoshi Sawai	（The University of Tokyo）
庄田 耕一郎	（東京大学 大学院総合文化研究科）	Koh-ichiroh Shohda	（The University of Tokyo）
須河 光弘	（東京大学 大学院総合文化研究科）	Mitsuhiro Sugawa	（The University of Tokyo）
陶山 明	（東京大学 大学院総合文化研究科）	Akira Suyama	（The University of Tokyo）
千葉 かおり	（茨城工業高等専門学校 自然科学科）	Kaori Chiba	（National Institute of Technology, Ibaraki College）
豊田 太郎	（東京大学 大学院総合文化研究科）	Taro Toyota	（The University of Tokyo）
根岸 瑠美	（東京大学 分子細胞生物学研究所）	Lumi Negishi	（The University of Tokyo）
野口 誉之	（東京大学 大学院総合文化研究科）	Yoshiyuki Noguchi	（The University of Tokyo）
野地 博行	（東京大学 大学院工学系研究科）	Hiroyuki Noji	（The University of Tokyo）
畠山 哲央	（東京大学 大学院総合文化研究科）	Tetsuhiro Hatakeyama	（The University of Tokyo）
林 勇樹	（東京大学 大学院総合文化研究科）	Yuuki Hayashi	（The University of Tokyo）
樋口 秀男	（東京大学 大学院理学系研究科）	Hideo Higuchi	（The University of Tokyo）
村田 昌之	（東京大学 大学院総合文化研究科）	Masayuki Murata	（The University of Tokyo）
矢島 潤一郎	（東京大学 大学院総合文化研究科）	Junichiro Yajima	（The University of Tokyo）
若杉 桂輔	（東京大学 大学院総合文化研究科）	Keisuke Wakasugi	（The University of Tokyo）
若本 祐一	（東京大学 大学院総合文化研究科）	Yuichi Wakamoto	（The University of Tokyo）

※50音順。敬称略。

□ 交通のご案内

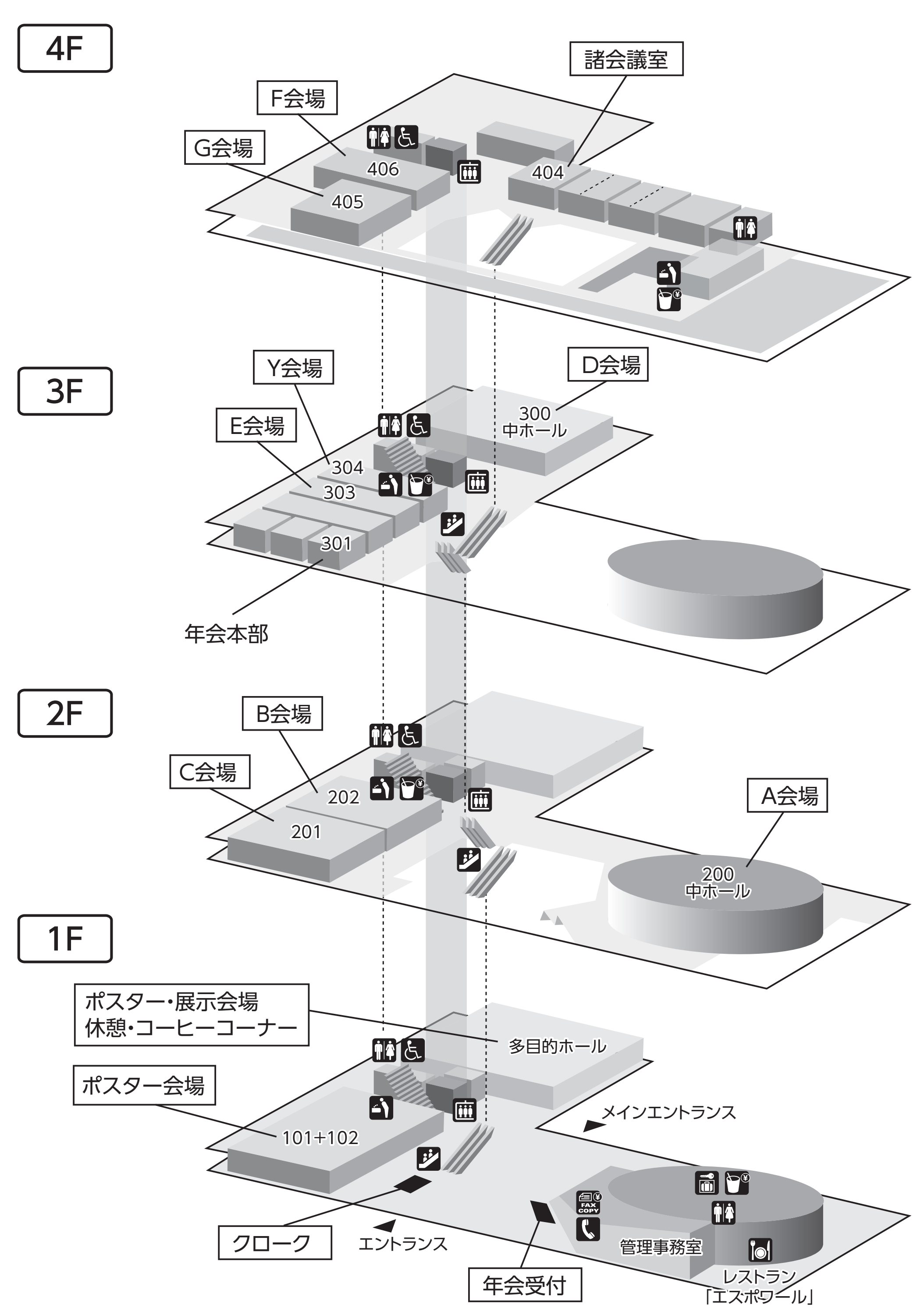
交通案内



周辺地図

至 下妻
至 筑波山
西大通り
東大通り
懇親会会場
ホテルグランド東雲
つくばセンター
中央公園
Bivi つくば
つくば駅
中央通り
つくばエクスプレス
Q't
オークラフロンティアホテルつくば
筑波学園郵便局
クレオ[イオン・西武]
ノバホール
土浦学園線
学園西
学園東
至 常総
つくば中央警察署
三井ビル
大清水公園
やまや
ヨークベニマル
カピオ
徒歩13分
デイズタウン
徒歩10分
オークラフロンティアホテル
つくば エポカル
竹園公園
つくば国際会議場
ミニストップ
研究交流センター
南大通東
南大通西
南大通り
物質・材料研究機構
至 牛久
至 桜・土浦インター
ペデストリアン(歩行者専用道路)

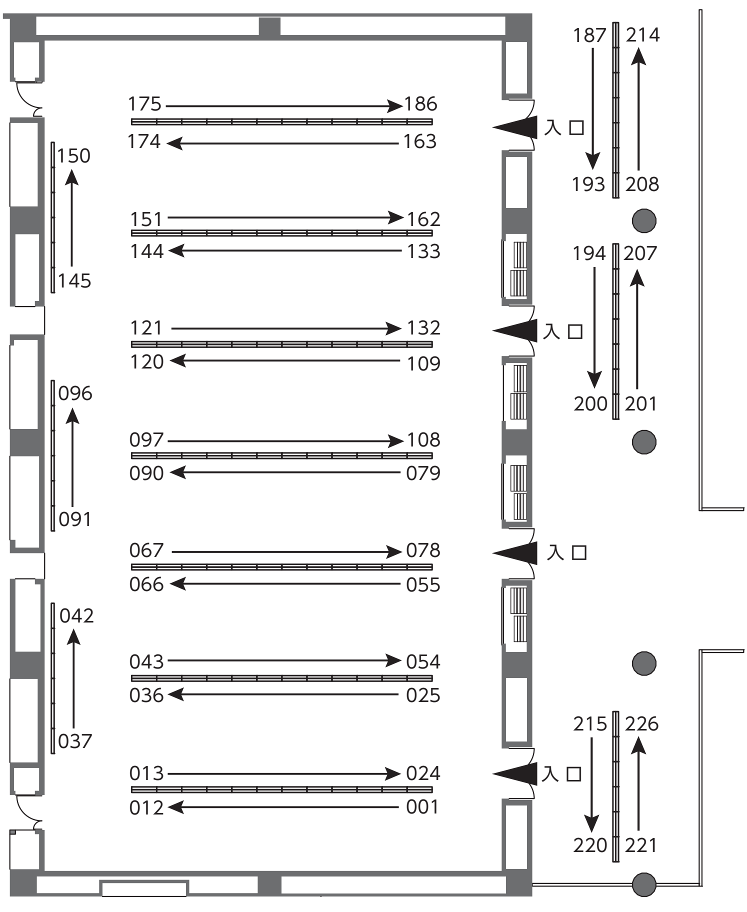
□ 会場のご案内



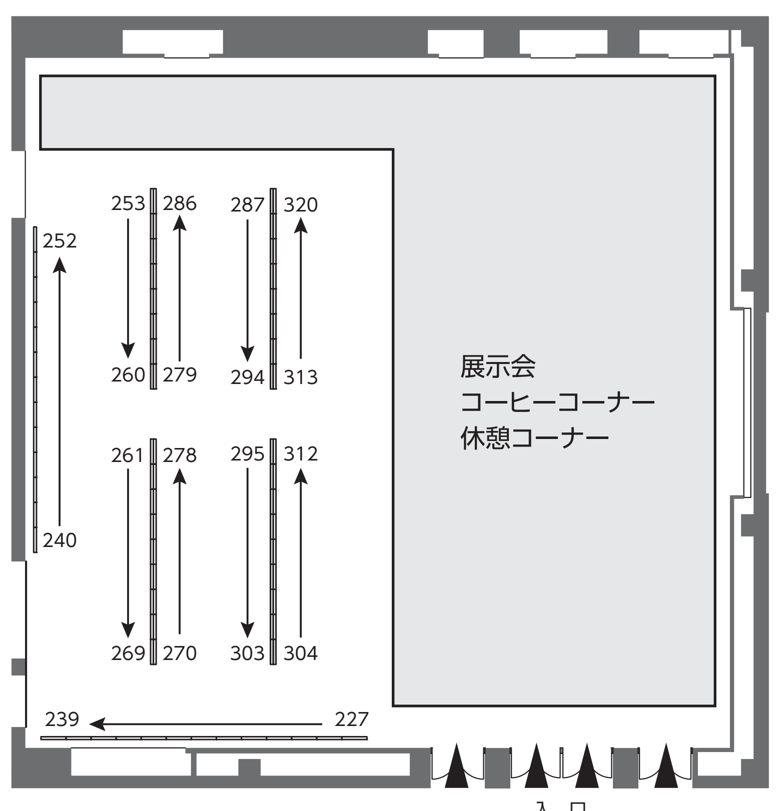
- 電話
- トイレ
- 車いす用トイレ
- エレベーター
- エスカレーター
- 給水所
- レストラン
- 飲料自動販売機
- コインロッカー
- ファックス・コピー(有料)

□ ポスター会場のご案内

○ 大会議室 101+102　ポスター番号 001～226



○ 多目的ホール　ポスター番号 227～320



2016年11月25日（金）：年会1日目

会場	階	部屋	会場名	9：00–11：30	11：45–12：35	12：45–13：45
つくば国際会議場	2F	中ホール200	A会場	1SAA 神経活動イメージングの最先端： 新規ツールとその活用 9：00－11：30	1LA 浜松ホトニクス 株式会社 11：45－12：35	学生発表賞フラッシュトーク1 12：45－13：45
	2F	中会議室202	B会場	1SBA 全細胞解析によるマイノリティ細胞の解明 9：00－11：30	1LB 株式会社菱化 システム 11：45－12：35	学生発表賞フラッシュトーク2 12：45－13：45
	2F	中会議室201	C会場	若手奨励賞招待講演 9：00－11：30	1LC 日本蛋白質構造 データバンク 11：45－12：35	学生発表賞フラッシュトーク3 12：45－13：45
	3F	中ホール300	D会場	1SDA 原子からいのちへ： 21世紀の新しい生命観を求めて 9：00－11：30	1LD オリンパス 株式会社 11：45－12：35	学生発表賞フラッシュトーク4 12：45－13：45
	3F	小会議室303	E会場	1SEA 運動性鞭毛・繊毛の最前線 ―生体ナノマシンの制御機構― 9：00－11：30		学生発表賞フラッシュトーク5 12：45－13：45
	3F	小会議室304	Y会場		男女共同参画・ 若手支援 シンポジウム 11：45　12：35	
	4F	中会議室406	F会場	1SFA 生物物理遺伝学： 生物物理学的ゲノム情報科学としての 9：00－11：30		学生発表賞フラッシュトーク6 12：45－13：45
	4F	小会議室405	G会場	1SGA 蛋白質工学を用いた会合と溶解性の最新の研究 9：00－11：30		学生発表賞フラッシュトーク7 12：45－13：45
	4F	小会議室404	諸会議室		出版委員会 11：45－12：35	第3回　理事会 12：45－13：45
	1F	大会議室 101+102	ポスター会場	ポスター貼付・掲示 8：30－13：55		
	1F	多目的ホール	ポスター会場	ポスター貼付・掲示 8：30－13：55		
	1F	多目的ホール	企業展示	機器・試薬・書籍展示 9：00－17：00		

14:00	15:00	16:00	17:00	18:00	19:00	20:00	21:00

16:00－16:30 (Biophysics and Physicobiology 論文賞受賞講演)

1SAP
細胞同士の絡み合いから理解する
集団運動の生物物理学
16:30－19:00

1SBP
モデル化と操作による高次生命現象の
解明への挑戦
16:30－19:00

1SCP
【学会本部企画Ⅰ】日本－中国 交流シンポジウム：
蛋白質設計とバイオテクノロジーへの応用
16:30－19:00

1SDP
モーターと細胞骨格の新展開
ステップから集団運動まで
16:30－19:10

1SEP
時空間精密構造解析による
生体分子活性サイトの機能解明
16:30－19:00

キャリア支援説明会
13:00－18:00

臨時社員総会
19:00－20:20

1SFP
細胞膜ナノ・メゾドメイン構造による
シグナル伝達の動的な制御機構
16:30－19:30

1SGP
可塑性とロバストネスの動的状態論
16:30－19:00

Biophysics and Physicobiology 編集委員会
13:50－14:50

若手奨励賞
選考委員会
15:00－16:00

ポスター討論
奇数
13:55－14:55

ポスター討論
偶数
14:55－15:55

ポスター掲示・撤去
15:55－17:00

ポスター討論
奇数
13:55－14:55

ポスター討論
偶数
14:55－15:55

ポスター掲示・撤去
15:55－17:00

2016年11月26日（土）：年会2日目

会場	階	部屋	会場名	9：00–11：30	11：45–12：35	12：35以降
つくば国際会議場	2F	中ホール200	A会場	2SAA 光遺伝学で活躍するタンパク質分子の生物物理学研究の展望 9：00−11：30	2LA キャノンマーケティングジャパン株式会社	
	2F	中会議室202	B会場	2SBA 構成的生物学の手法による生体分子，分子複合体，分子ネットワークの理解 9：00−11：30	2LB 日本エフイー・アイ株式会社	
	2F	中会議室201	C会場	2SCA 【学会本部企画Ⅱ】日本−韓国 交流シンポジウム：1分子生物物理学の最前線 9：00−11：30	2LC 国立研究開発法人日本医療研究開発機構（AMED）Human Frontier Science Program（HFSP）	
	3F	中ホール300	D会場	2SDA 温度生物学の挑戦 9：00−11：30	分野別専門委員会	会員総会・総会シンポジウム 12：35−13：55
	3F	小会議室303	E会場	2SEA 生命現象の理解を目指した立体構造インフォマティクスデータの活用 9：00−11：30		
	3F	小会議室304	Y会場	キャリア支援説明会 9：30−18：15		
	4F	中会議室406	F会場	2SFA 免疫学と生物物理の接点 9：00−11：30	2LF ライカマイクロシステムズ株式会社 11：45−12：35	
	4F	小会議室405	G会場	2SGA 電子顕微鏡が捉える生物アーキテクチャの解明 —高分解能化と多様な情報の融合— 9：00−11：30		
	4F	小会議室404	諸会議室			
	1F	大会議室101+102	ポスター会場	ポスター貼付・掲示 8：45−14：05		
	1F	多目的ホール	ポスター会場	ポスター貼付・掲示 8：45−14：05		
	1F	多目的ホール	企業展示	機器・試薬・書籍展示 9：00−17：00		

14：00	15：00	16：00	17：00	18：00	19：00	20：00	21：00

2SAP
生体分子ー電磁波間の共鳴を活用する
最先端バイオイメージング
16：15ー18：45

2SBP
ラマン散乱で探る bio. phys. chem. 三重点
16：15ー19：05

2SCP
【学会本部企画Ⅲ】日本ーオーストラリア
交流シンポジウム：
ライブセルイメージング
16：15ー18：45

2SDP
蛋白質の秩序化ー脱秩序化研究の最前線
16：15ー19：05

2SEP
新しい視点を創る光学顕微鏡技術
16：15ー18：45

2SFP
データ駆動科学（スパースモデリング）
による計測の進展
16：15ー18：45

2SGP
リン酸化ダイナミクスが支える
生命情報処理機構
16：15ー18：45

若手の会会議
17：45ー18：45

ポスター討論
奇数
14：05ー15：05

ポスター討論
偶数
15：05ー16：05

ポスター掲示・撤去
16：05ー17：30

ポスター討論
奇数
14：05ー15：05

ポスター討論
偶数
15：05ー16：05

ポスター掲示・撤去
16：05ー17：30

懇親会
ホテルグランド東雲
19：30ー21：30

2016年11月27日（日）：年会3日目

会場	階	部屋	会場名	プログラム
つくば国際会議場	2F	中ホール200	A会場	3SAA 蛍光・発光計測技術が拓く細胞生物学の新地平 9：45－12：15 / 3LA カールツァイスマイクロスコピー株式会社 12：30－13：20
	2F	中会議室202	B会場	3SBA 生体分子の柔らかさと機能をつなぐもの 9：45－12：15 / 3LB 株式会社オプトライン 12：30－13：20
	2F	中会議室201	C会場	3SCA 次世代研究者による動的構造生命 9：15－12：15 / 科研費説明会 12：30－13：20
	3F	中ホール300	D会場	3SDA 運動超分子マシナリーが織りなす調和と多様性 9：15－12：15
	3F	小会議室303	E会場	3SEA 多細胞合成生物学 9：15－12：15
	3F	小会議室304	Y会場	男女共同参画・若手支援委員会 12：30－13：20
	4F	中会議室406	F会場	3SFA ミトコンドリアの分子マシナリーと機能管理：合成、構造、機能、適応、そして淘汰 9：45－12：15 / 3LF 株式会社ニコンインステック 12：30－13：20
	4F	小会議室405	G会場	3SGA 人工生体プログラマブルシステム～精密構造設計から分子ロボティクスへ～ 9：45－12：15
	4F	小会議室404	諸会議室	企業との意見交流会 9：30－11：00
	1F	大会議室101+102	ポスター会場	ポスター貼付・掲示 9：00－13：35
	1F	多目的ホール	ポスター会場	ポスター貼付・掲示 9：00－13：35
	1F	多目的ホール	企業展示	機器・試薬・書籍展示 9：15－16：15

Time axis: 9：00 / 10：00 / 11：00 / 12：00 / 13：00

14：00	15：00	16：00	17：00	18：00	19：00	20：00	21：00

ポスター 討論 奇数 13：35 | 14：35	ポスター討論 偶数 14：35―15：35	ポスター 掲示・撤去 15：35 | 16：15
ポスター 討論 奇数 13：35 | 14：35	ポスター討論 偶数 14：35―15：35	ポスター 掲示・撤去 15：35 | 16：15

参加者へのご案内

1. 年会受付と参加登録

◇ 年会受付

場　　所: つくば国際会議場 エントランスホール(「会場のご案内」6 ページを参照)
受付時間: 11 月 25 日(金) 8:30 - 17:00
　　　　　26 日(土) 8:45 - 17:00
　　　　　27 日(日) 9:00 - 15:00

◆事前登録

事前登録が完了された方は、日本生物物理学会会員・非会員共に参加証および領収証、プログラム集冊子が事前送付されますので、会場での受付は不要です。当日は必ず参加証をお持ちください。
但し、海外からの参加者は、年会受付にて参加証とプログラム集冊子をお受け取りください。

※ネームタグホルダーを当日配布しますので、会場内では必ず参加証をご着用ください。
　配付場所: 年会受付付近

注意 1) 事前登録は 年会参加登録費(参加費)の振込後に完了します。振込がない場合、オンライン登録は無効となります。当日受付で当日参加費をお支払いください。
注意 2) 日本生物物理学会会員は年度会費を納めていない場合、参加証が送付されません。年度会費未納者・新規入会受付デスクにて年度会費をお支払いください。
注意 3) 参加費・年度会費ともに振込済みで、参加証が事前送付されていない場合は、総合受付デスクまでお越しください。
注意 4) 非会員のシンポジウム招待講演者については、会員である必要はなく、また、登録費は免除されます。懇親会に無料でご招待します。
注意 5) 海外の機関に所属する非会員については、ご入会いただかずに一般発表をしていただけます。

◆当日登録

事前登録が完了していない方は当日登録をしていただきます。
当日受付にお越しの上、参加費を現金でお支払いください。

◇ 当日年会諸費用 (一覧表)

当日参加	会員				非会員		
	正会員	シニア会員	大学院生	学部学生	一般	大学院生	学部学生
当日参加費	¥9,000	¥5,000	¥5,000	¥0	¥12,000	¥6,000	¥0
懇親会費	¥8,000	¥5,000	¥5,000	¥3,000	¥8,000	¥5,000	¥3,000

・参加のみの学部学生は参加費無料です。当日受付で学生証を提示してください。
　参加証とプログラム集冊子をお渡しします。ただし、懇親会は有料です。

・若手奨励賞招待講演者、Biophysics and Physicobiology 論文賞受賞講演者、Biophysics and Physicobiology Editors' Choice Award 受賞代表者は、懇親会に無料でご招待します。
　既に懇親会参加費を振り込まれている場合は、総合受付デスクで返金します。

◇ 参加証（名札）

参加証は会場内では必ずご着用ください。参加証のない方のご入場は固くお断りいたします。
事前送付された参加証は必ず会場にお持ちください（ネームタグホルダーは年会受付付近で配布いたします）。

◇ 領収書の発行

参加証とともに領収書をお渡しいたしますが、別の形式の領収書が必要な場合、お渡しした領収書と引き換えに総合受付デスクにて発行いたします。

◇ プログラム集冊子/オンライン予稿集【10 月 25 日（火）公開予定】

プログラム集冊子（前付・プログラム）は日本生物物理学会会員・事前登録が完了された非会員に事前に送付いたします（プログラム集冊子は総合受付デスクでも当日販売（3,500 円/税込）を行います）。なお予稿本文はプログラム集冊子には掲載されません。予稿本文は、オンライン予稿集をダウンロードして閲覧いただくことになります。

オンライン予稿集:
http://www.biophys.jp/dl/pro/54th_proceedings.pdf
ダウンロード ID: ambsj54
パスワード：tsukuba2016

プログラム（タイトル、発表者、所属）と予稿集は、年会ホームページにて公開します。
年会終了後は、半年ほど経て日本生物物理学会ホームページの年会の記録および J-Stage にて予稿集の pdf ファイルが公開されます。
日本生物物理学会ホームページの年会の記録 (http://www.biophys.jp/ann/ann02.html)
J-Stage の生物物理のページ (http://www.jstage.jst.go.jp/browse/biophys/-char/ja)

◇ プログラム検索（ウェブ版）【10 月 25 日（火）公開予定】

年会ホームページより「プログラム検索」を公開します。項目［演題タイトル（和文・英文）、発表者名（共著者含む）（漢字、カナ、ローマ字）、発表形式］から、演題番号、発表日、会場を検索・表示します。

◇ プログラム検索・予稿閲覧アプリ（無料）【11 月 21 日（月）公開予定】

スマートフォン(iPhone/Android)やタブレット(iPad/iPod Touch/Android)端末に対応した予稿閲覧アプリをご利用いただけます（演題検索、タイムテーブル一覧表示、ブックマーク登録等）。
App Store、Google Play よりダウンロードしてください（無料）。年会ホームページにもアプリ提供サイト(App Store, Google Play)を掲載しております。

アプリケーション名：第 54 回日本生物物理学会年会
検索ワード：bsj2016、生物物理、日本生物物理学会
アプリケーションの予稿閲覧パスワード：tsukuba2016

◇ 年度会費の支払いと入会の手続き

日本生物物理学会の年度会費が未納の場合は、年会受付の年度会費未納者・新規入会受付デスクでお支払いください。また、日本生物物理学会への新規入会も受け付けます。

2. 会場内のサービス・施設

◇ クローク

場　　所: つくば国際会議場エントランスホール（「会場のご案内」6 ページを参照）

利用時間: 11 月 25 日（金） 8:30 - 19:45
26 日（土） 8:45 - 19:15
27 日（日） 9:00 - 16:00

※貴重品や傘、またコンピュータなどについては、破損、紛失などの責任は負いかねますので、各自でお持ちください。

※懇親会への移動など会場を去られる際は荷物をお引き取りください。

◇ 昼食

ランチョンセミナー（1～3 日目）、男女共同参画・若手支援シンポジウム（1 日目）、科研費説明会（3 日目）でお弁当とお茶が無料で提供されます。当日の午前中に整理券を配布いたします。整理券のご利用方法は 18 ページ「ランチョンセミナー」をご参照ください。
この他、分野別専門委員会（次ページ参照）を開催し、お弁当とお茶が無料で提供されます（整理券なし・数量に限りがあります）。積極的にご参加ください。
また会期中、以下のレストランをご利用いただけます。
尚、つくば国際会議場周辺グルメマップを年会受付付近で配布しておりますのでご自由にお取りください。

■エスポワール （Tel: 029-850-3266）
場所: つくば国際会議場 1 階 （「会場のご案内」6 ページを参照）
9:00 - 17:00 （17 時以降のディナータイムは予約制となっております）
※但し 27 日は 14 時までの営業となっております。

◇ 呼び出し

会場内での呼び出しは、緊急の場合を除いて一切行いません。参加者間の連絡用として、年会受付付近に伝言板を設置しますので、ご利用ください。

◇ 駐車場

会場には 参加者用駐車場はございません 。会場へは公共交通機関をご利用ください。

◇ 宿泊

宿泊に関しては年会ホームページ「宿泊案内」をご参照ください。

◇ インターネット

講演会場外およびポスター会場外では無線 LAN をご利用いただけます。
※講演会場内およびポスター会場内ではご利用いただけません。
ネットワーク名: publics を選択してください。
パスワード等については、会場にてお知らせいたします。

◇ コーヒーコーナー

ポスター・展示会場休憩スペース（1 階、多目的ホール）をご利用ください。
ポスター発表が行われている時間帯はコーヒーを無料提供（杯数限定）しております。
その他の時間帯では有料で販売（100 円）となります。

◇ 託児所

年会期間中は、託児所を設置いたします。詳しくは年会ホームページをご覧ください。

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3. 年会行事・プログラム

◇ 会員総会・総会シンポジウム

一般社団法人日本生物物理学会第 3 回会員総会を年会 2 日目、11 月 26 日(土)12:35－13:55 に D 会場(中ホール 300)で開催しますのでご出席ください。また、総会シンポジウムを開催します。詳しくは7. 開催通知(22 ページ)をご覧ください。

◇ 若手奨励賞招待講演

日本生物物理学会若手奨励賞及び若手招待講演賞の選考会である講演会(若手奨励賞招待講演)を、年会 1 日目 11 月 25 日(金)9:00 - 11:30 に C 会場(中会議室 201)で開催します。

◇ 学生発表賞

日本生物物理学会学生発表賞の選考会であるフラッシュトーク(11 月 25 日(金) 12:45 - 13:45)とポスター発表(11 月 25 日(金)13:55 - 15:55、11 月 26 日(土)14:05 - 16:05)を開催します。

◇ Biophysics and Physicobiology 論文賞受賞講演

Biophysics and Physicobiology 論文賞受賞の講演会を、年会 1 日目 11 月 25 日(金)16:00 - 16:30 に A 会場(中ホール 200)で開催します。

◇ 分野別専門委員会

日時: 11 月 26 日(土)11:45 - 12:35
会場: D 会場(中ホール 300)
対象: 分野別専門委員
昼食: 委員の皆様にはお弁当とお茶を提供いたします(整理券なし)。

◇ 懇親会

日時: 11 月 26 日(土)19:30 - 21:30
会場: ホテルグランド東雲 2 階 東雲の間 (つくば国際会議場より 徒歩 13 分)
(茨城県つくば市小野崎 488ー1) Tel: 029-856-2211
※懇親会の当日参加も受け付けいたします(受付場所: 総合受付デスク、または懇親会会場前)。

◇ 男女共同参画・若手支援シンポジウム

日時: 11 月 25 日(金)11:45 - 12:35
会場: Y 会場(小会議室 304)
昼食: お弁当とお茶が無料で提供されます(整理券を配布いたします。次ページ「ランチョンセミナー」の項を参照)。

◇ キャリア支援説明会

日時: 11 月 25 日(金)13:00 – 18:00
　　　11 月 26 日(土) 9:30 – 18:15
　※詳細は 48 ページをご参照下さい。
会場: Y 会場(小会議室 304)
対象: 就職を考えておられる学生や研究者など

◇ 科研費説明会

日時: 11 月 27 日(日)12:30 – 13:20
会場: C 会場(中会議室 201)
昼食: お弁当とお茶が無料で提供されます(整理券を配布いたします。下記「ランチョンセミナー」参照)。

◇ ランチョンセミナー

昼食(お弁当とお茶、無料)をとりながらの協力企業によるセミナーにご参加ください。なお、お弁当の数に限りがあるため 当日の以下の時間帯に整理券を配布いたします。セミナー開始前に、会場入り口で整理券と引き換えにお弁当を受け取り、ご入場ください(整理券の発券方法は下記参照)。

◆整理券の発券について

ランチョンセミナー整理券を下記のように配布いたします。
時間: 11月25日(金)8:30 – 10:45、26日(土)8:45 – 10:45、27日(日)9:00 – 11:15
場所: つくば国際会議場エントランスホール 年会受付付近
※整理券はランチョンセミナー共催の企業、団体よりご提供いただく昼食の引換券になります。
当日開催されるセミナー分のみ発券いたします。券は枚数が無くなり次第終了となります。

◆整理券の注意事項

整理券は各日、セミナー開始後、無効となります。
午前のプログラム終了後、ランチョンセミナー開始時間までにご来場ください。
セミナー開始までにご来場されない場合、整理券は無効となり、お弁当は整理券をお持ちでない参加者に提供されますことをご了承ください。

◆ランチョンセミナー受講時のお願い

ランチョンセミナーは企業、団体等の共催によるセミナーです。参加される場合は最後までご聴講ください。また、共催者のアンケートには、できるだけ所属・氏名を記載して回答くださるよう、ご協力をお願いします。

◇ 機器・試薬・書籍等附設展示会

機器、試薬、ソフトウエア、書籍などの附設展示会をポスター・展示会場(1 階、多目的ホール)で行います。

◇ 市民講演会

テーマ: 「生命とは何かの普遍性に挑む〜理論物理と構成的実験からのアプローチ〜」
日　時: 11 月 5 日(土) 開場 13:30、 開演 14:00、 終演 16:00
会　場: 東京大学駒場 I キャンパス　21 KOMCEE East 地下 1 階ホール
　　　(東京都目黒区駒場 3-8-1、京王井の頭線「駒場東大前」駅)
参加費: 無料 (どなたでも自由に参加できます)

4. 禁止事項

◇ 撮影・録音

会場内でのカメラ、ビデオ、携帯電話などによる撮影や講演音声の録音などを禁止します。
一部、理事会の承認を得て、録画を行う場合があります。

◇ 喫煙・飲食

会場内は終日禁煙です。講演会場内での飲食はランチョンセミナー、男女共同参画・若手支援シンポジウム、科研費説明会、各種委員会など食事が提供される場合を除いて禁止します。

◇ 携帯電話

シンポジウム、ポスター発表等の会場内での携帯電話による通話を禁止します。講演会場内では電源をオフにするかマナーモードに設定し、呼び出し音が鳴らないようご注意ください。

5. 年会についての問い合わせ

◇ **会期中**　年会本部（会期中のみ通じます）Tel：050-3482-9415

◇ **会期外**　年会実行委員会　E-mail:bp_nenkai54@bio.c.u-tokyo.ac.jp

参加登録・演題登録　システムサポートデスク

〒602-8048　京都市上京区下立売通小川東入る
中西印刷株式会社内
E-mail：bsj2016sys-sprt@e-naf.jp

年会実行委員会サポート・展示・広告

〒101-0003　東京都千代田区一ツ橋 2-4-4 岩波書店一ツ橋別館 4F
株式会社エー・イー企画　Tel：03-3230-2744(代表)　Fax：03-3230-2479
実行委員会サポート　E-mail：jbp2016@aeplan.co.jp
広告・展示関連　E-mail：e_jbp54@aeplan.co.jp

6. 発表者へのご案内

◇ 使用言語

すべての発表は原則として英語をお使いください。

◇ 映写機器

会場にはパソコンを用意いたしません。ご自身のノートパソコンを必ずお持ちください。
発表に使用できる映写機器は、液晶プロジェクターのみです。音声出力には対応しません。
会場に備え付けの液晶プロジェクターにより、図等をスクリーンに映写して発表します。
使用ソフトはパワーポイント(米国マイクロソフト社)を標準とします。画像解像度は 1024×768 ピクセル(XGA)です。この環境下で発表データを作成ください。これより大きい画面サイズでデータを作成すると、スクリーン映写時に画面をはみ出す等の不具合が起こる可能性がある旨ご理解ください。

注意 1）会場スタッフがパソコンを会場に備え付けられた切り替え装置（セレクター）に接続いたします。
注意 2）切り替え装置に繋がるパソコンの映像出力端子は、「ミニ D-sub15 ピン端子（メス）」のみです。
端子の形状が異なる場合（Macintosh 等）、変換アダプターをお持ちください。
注意 3）発表に使用するパワーポイントファイルが入った USB メモリーを念のためにお持ちください。
注意 4）バッテリー切れに備え、必ず電源アダプターをお持ちください。
注意 5）発表中にスクリーンセーバーや省電力モードにならないよう、設定してください。

◇ シンポジウム、若手奨励賞招待講演のオーガナイザーの方へ

受付：　セッション開始の 15 分前までに各会場の「進行席」までお越しの上、係りの者に来場された旨をお伝えください。

進行：　一任いたしますので、講演者の講演時間を厳守し、円滑な運営にご協力ください。プログラムに記載されている各講演者の講演時間等に変更が生じた場合は、会場内の係りの者にご指示ください。
会場には時間を計測するスタッフを置いています。

◇ シンポジウム、若手奨励賞招待講演の講演者の方へ

受付：　セッション開始の 15 分前までに各会場の「PC 受付」にお越しください。
発表スライドをご確認いただいた後、会場スタッフがパソコンを切り替え装置（セレクター）に接続いたします。※スライドチェック用の試写室は設けておりません。

講演時間：シンポジウムの時間配分はオーガナイザーに一任しております。
若手奨励賞招待講演の講演時間は、発表 10 分、討論 3 分、パソコンの交換に 30 秒です。

◇ 学生発表賞フラッシュトークの講演者の方へ

集合：　年会初日 12:35 までにご自身の発表がある講演会場にお越しください（44～45 ページ目を参照）。

発表時間：発表の持ち時間は 2 分 30 秒です。2 分 30 秒経過時に進行係がベルを 2 回鳴らしますので、直ちに発表を終えてください。

使用言語：発表の使用言語は英語です。

発表スライド：　講演用のスライドは、事前に年会が用意する PC にインストールしております。スライドは、講演者にお送りした注意事項を守って pdf 形式で作製し、指定された期限までに学会事務局（bsj@nacos.com）にお送りください。スライドは所定の様式による表紙1枚を含めて、最大4枚まで使用できます。ただし、アニメーション効果を狙ってスライド数が多少増えることは認めます。ファイル容量は 10MB 以下にしてください。

◇ ポスター発表の方へ

ポスターの貼付・展示、説明・討論、撤去：

		11 月 25 日（金）	11 月 26 日（土）	11 月 27 日（日）
貼付・展示		8:30 – 13:55	8:45 – 14:05	9:00 – 13:35
説明・討論	奇数番号	13:55 – 14:55	14:05 – 15:05	13:35 – 14:35
	偶数番号	14:55 – 15:55	15:05 – 16:05	14:35 – 15:35
撤去		17:00 までに撤去	17:30 までに撤去	16:15 までに撤去

1. ポスターは日替わりで貼り替えてください。
2. ポスターボードの大きさは、幅 90 cm、高さ 210 cm。貼付に必要な押しピンは会場に用意します。
3. 撤去時間を過ぎて残ったポスターは年会事務局にて破棄しますので、ご了承ください。

◇ ポスター発表要項

ポスターは英語で作成してください。
ただし、タイトル、所属、著者名は、可能であれば日本語の併記もお願いいたします。
発表代表者の氏名には左肩に小さな○印を付けてください。

◇ 発表形式と演題番号（各予稿左上の番号）の見方

発表形式は、シンポジウム発表（Symposium Speech）、若手奨励賞招待講演（"Early Career Award in Biophysics" Candidate Presentations）、ポスター発表（Poster Presentation）、学生発表賞フラッシュトーク（Flash Talks for "Student Presentation Award"）があります。

シンポジウム発表：（例）1SAA-03
1 文字目は発表日（1:11 月 25 日(金)、2:11 月 26 日（土）、3:11 月 27 日（日））、2 文字目は Symposium (S)、3 文字目は会場名（A 会場）、4 文字目は午前・午後（AM,PM）、最後の 2 桁の数字は発表順です。

若手奨励賞招待講演：（例）1YC1045
1 文字目は発表日（1:11 月 25 日(金)）、2 文字目は Young（Y）、3 文字目は会場名（C 会場）、最後の 4 桁の数字は講演開始時刻です。

ポスター発表：（例）1Pos001
1 文字目は発表日（1:11 月 25 日(金)、2:11 月 26 日（土）、3:11 月 27 日（日））、2 文字目は Poster (Pos)、最後の 3 桁の数字はパネル番号を示します。

学生発表賞フラッシュトーク：（例）1Pos001*
ポスター発表の演題番号の右側に「*」がついています。

7. 一般社団法人日本生物物理学会第3回会員総会開催通知

日時: 11月26日(土)12:35 - 13:55

会場: D会場(中ホール300)

一般社団法人日本生物物理学会第3回会員総会を開催いたします。主な議題は下記の通りです。
是非ご出席ください。

議長: 会長 中村 春木

総会議題

(1)報告事項

平成27年度決算報告ならびに監査結果報告

平成28年度会計ならびに事業の中間報告と今後の計画

次期年会について

(2)第3回会員総会シンポジウム:1分子生物物理の勃興期から未来 木下一彦氏を偲んで

8. 理事会、会員総会、各種委員会の案内

委員会等	開催日程		会場
生物物理編集委員会	11月24日（木）	14:00 – 16:00	小会議室 404（諸会議室）
ホームページ編集委員会		16:00 – 18:00	小会議室 403（諸会議室）
Biophysics and Physicobiology 編集委員会		17:00 – 19:00	小会議室 404（諸会議室）
出版委員会	11月25日（金）	11:45 – 12:35	小会議室 404（諸会議室）
平成 28 年度第 3 回理事会（旧運営委員会）		12:45 – 13:45	小会議室 404（諸会議室）
Biophysics and Physicobiology 編集委員会		13:50 – 14:50	小会議室 404（諸会議室）
若手奨励賞選考委員会		15:00 – 16:00	小会議室 404（諸会議室）
臨時社員総会		19:00 – 20:20	小会議室 304（Y 会場）
分野別専門委員会	11月26日（土）	11:45 – 12:35	中ホール 300（D 会場）
会員総会・総会シンポジウム（総会）		12:35 – 13:55	中ホール 300（D 会場）
若手の会会議		17:45 – 18:45	小会議室 404（諸会議室）
企業との意見交換会	11月27日（日）	9:30 – 11:00	小会議室 404（諸会議室）
男女共同参画・若手支援委員会		12:30 – 13:20	小会議室 304（Y 会場）

（ ）は法人化前の名称

謝　辞

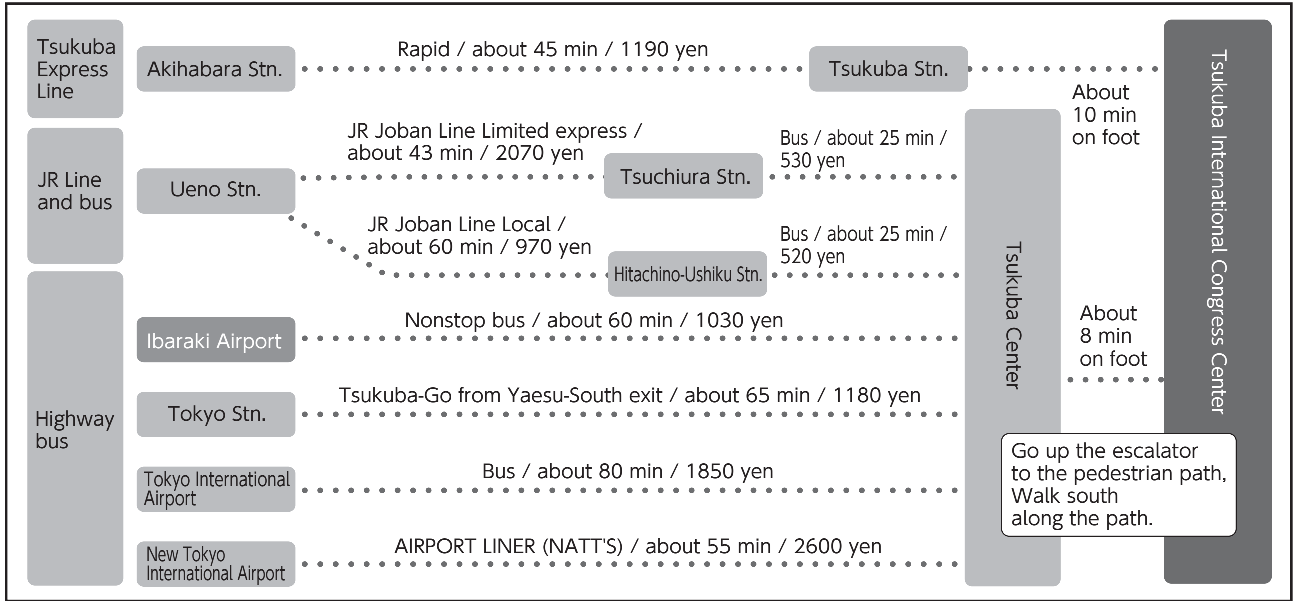
本年会の開催・運営に当たり、以下の団体よりご協力・ご援助いただきました。
関係者一同より御礼申し上げます。

新学術領域研究「シリア・中心体系による生体情報フローの制御」
新学術領域研究「ゆらぎと構造の協奏：非平衡系における普遍法則の確立」
新学術領域研究「3D 活性サイト科学」
新学術領域研究「温度を基軸とした生命現象の統合的理解」
新学術領域研究「共鳴誘導で革新するバイオイメージング」
新学術領域研究「スパースモデリングの深化と高次元データ駆動科学の創成」
新学術領域研究「理論と実験の協奏による柔らかな分子系の機能の科学」
新学術領域研究「動的構造生命科学を拓く新発想測定技術－タンパク質が動作する姿を活写する－」
新学術領域研究「運動超分子マシナリーが織りなす調和と多様性」
「複雑生命システム動態研究教育拠点」

第 54 回日本生物物理学会年会
実行委員長　豊島　陽子

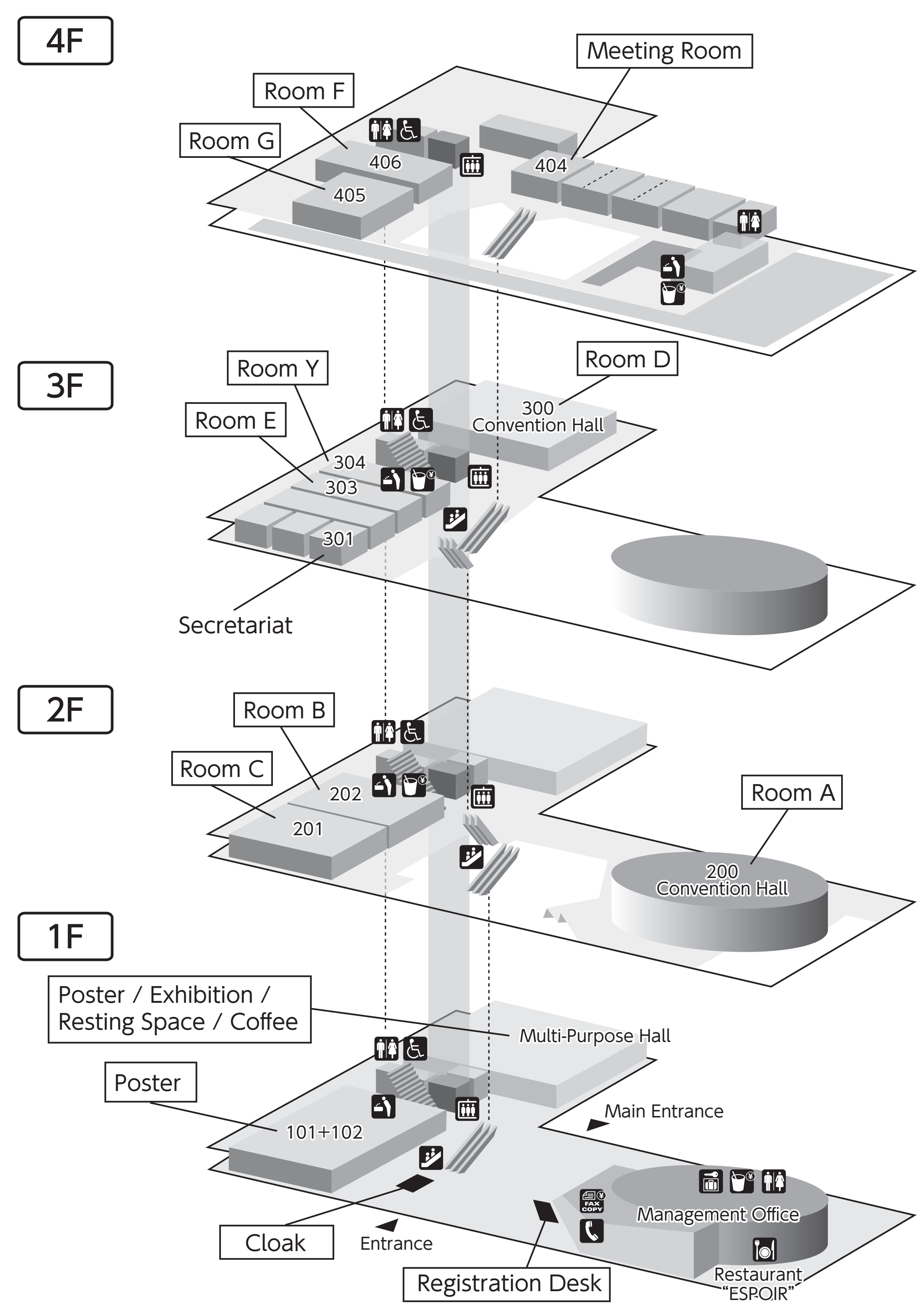
Access

Access Information



Map

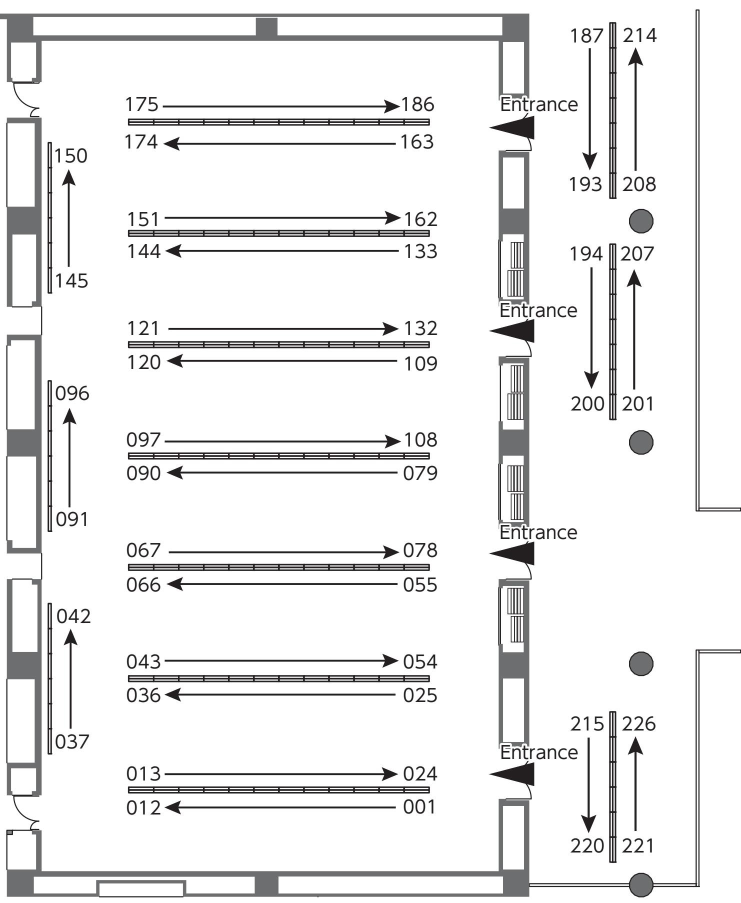
□ Floor Map



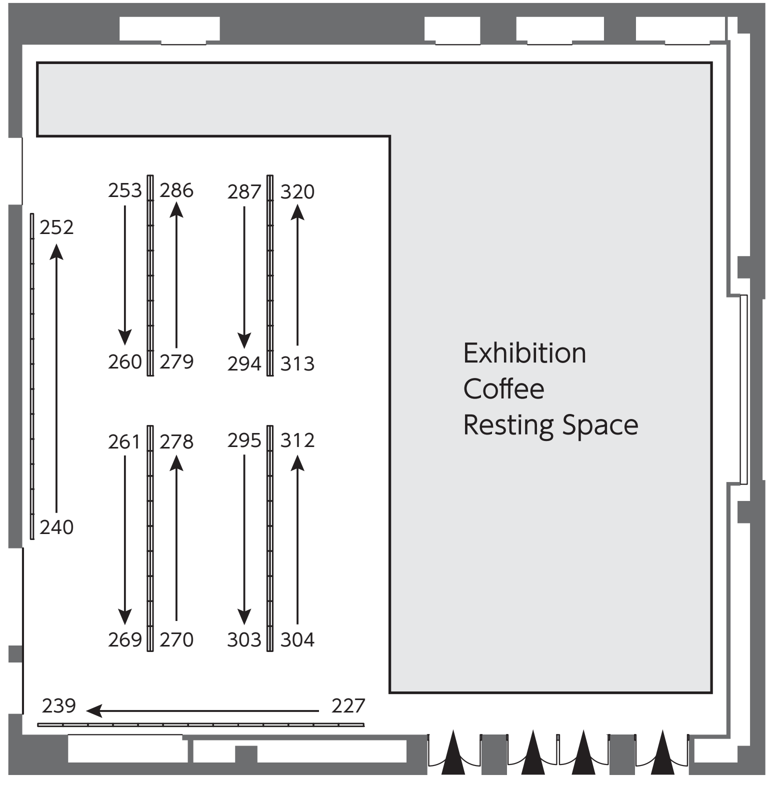
- Telephone
- Lavatory
- Lavatory for handicapped person
- Elevator
- Escalator
- Water supply place
- Restaurant
- Automatic vending machine (soft drinks)
- Coin-operated locker
- Facsimile & Copy (pay)

□ Poster Place

○ Conference Room 101+102 Poster No. 001～226



○ Multi-Purpose Hall Poster No. 227～320



November 25 (Fri.) Day 1

Tsukuba International Congress Center

Floor	Hall	Room	9：00–11：30	11：45–12：35	12：45–13：45
2F	Convention Hall 200	Room A	1SAA Advances in imaging neuronal activity: New tools and applications 9：00—11：30	1LA HAMAMATSU PHOTONICS K.K. 11：45—12：35	Flash Talks for Student Presentation Award 1 12：45—13：45
2F	Conference Room 202	Room B	1SBA Minority cell research enabled by exhaustive analyses of all cells 9：00—11：30	1LB Ryoka Systems Inc. 11：45—12：35	Flash Talks for Student Presentation Award 2 12：45—13：45
2F	Conference Room 201	Room C	"Early Career Award in Biophysics" Candidate Presentations 9：00—11：30	1LC Protein Data Bank Japan (PDBj) 11：45—12：35	Flash Talks for Student Presentation Award 3 12：45—13：45
3F	Convention Hall 300	Room D	1SDA From atoms to life: Exploring a new view of life in the 21st century 9：00—11：30	1LD Olympus Corporation 11：45—12：35	Flash Talks for Student Presentation Award 4 12：45—13：45
3F	Conference Room 303	Room E	1SEA Frontiers in motile cilia – regulatory mechanisms of bio-nanomachines – 9：00—11：30		Flash Talks for Student Presentation Award 5 12：45—13：45
3F	Conference Room 304	Room Y		11：45—12：35 (Gender Equality Promotion & Young Researchers Support Symposium)	
4F	Conference Room 406	Room F	1SFA Biophysical Genetics as a Genome Informatics Supported by Biophysics 9：00—11：30		Flash Talks for Student Presentation Award 6 12：45—13：45
4F	Conference Room 405	Room G	1SGA Advances in the engineering of protein oligomerization and solubility 9：00—11：30		Flash Talks for Student Presentation Award 7 12：45—13：45
4F	Conference Room 404	Meeting Room		Publications Meeting 11：45—12：35	3rd Board Meeting 12：45—13：45
1F	Conference Room 101+102	Poster Session	Poster Setup, Display 8：30—13：55		
1F	Multi-Purpose Hall	Poster Session	Poster Setup, Display 8：30—13：55		
1F	Multi-Purpose Hall	Exhibition	Exhibition 9：00—17：00		

14：00 | 15：00 | 16：00 | 17：00 | 18：00 | 19：00 | 20：00 | 21：00

16：00—16：30
Lecture by Biophysics and Physicobiology Outstanding Paper Awardees

1SAP
Biophysics of collective cell movement - From single-cell to multi-cell dynamics
16：30—19：00

1SBP
Modeling and Manupilation of Life: a Challenge to Unveil Its Complex Mechanism
16：30—19：00

1SCP
【BSJ Special Event Ⅰ】BSJ-BSC Joint Symposium: Protein Design and its Applications to Biotechnology
16：30—19：00

1SDP
New extremes of motor proteins and cytoskeleton: step into a new realm with steps and collective motions
16：30—19：10

1SEP
Understanding biochemical functions of the active sites in biomolecular systems by spatial–temporal analysis
16：30—19：00

Career Support Session
13：00—18：00

Extraordinary Member's General Meeting
19：00—20：20

1SFP
Unraveling the regulation mechanisms of signal transduction in nano- and meso-scale domains in cell membranes
16：30—19：30

1SGP
Dynamic state theory for plasticity and robustness of biological systems
16：30—19：00

Biophysics and Physicobiology Editorial Board Meeting
13：50—14：50

*ECAB Selection Meeting
15：00—16：00

Poster Presentation Odd numbers 13：55—14：55
Poster Presentation Even numbers 14：55—15：55
Poster Display, Removal 15：55—17：00

Poster Presentation Odd numbers 13：55—14：55
Poster Presentation Even numbers 14：55—15：55
Poster Display, Removal 15：55—17：00

＊ECAB：Early Career Award in Biophysics

November 26 (Sat.) Day 2

Tsukuba International Congress Center

Floor	Hall	Room	9:00 – 11:30	12:00 (11:45 – 12:35)	13:00
2F	Convention Hall 200	Room A	2SAA Perspective in biophysical studies on protein molecules applicable for optogenetics 9:00—11:30	2LA Canon Marketing Japan Inc.	
2F	Conference Room 202	Room B	2SBA Synthetic biology approaches to understand biological molecules, complexes, and networks 9:00—11:30	2LB FEI Company Japan Ltd.	
2F	Conference Room 201	Room C	2SCA 【BSJ Special Event Ⅱ】Korea-Japan Joint Symposium: Frontiers of Single Molecule Biophysics 9:00—11:30	2LC AMED・HFSP	
3F	Convention Hall 300	Room D	2SDA The Developing Field of Thermal Biology 9:00—11:30	Experts Committee	General Assembly / Assembly Symposium 12:35—13:55
3F	Conference Room 303	Room E	2SEA Applications of protein structure data for understanding biological phenomenon 9:00—11:30	11:45 – 12:35	
3F	Conference Room 304	Room Y	Career Support Session 9:30—18:15		
4F	Conference Room 406	Room F	2SFA Physical Immunology 9:00—11:30	2LF Leica Microsystems K.K. 11:45—12:35	
4F	Conference Room 405	Room G	2SGA Biological architecture elucidated by electron micrroscopy - Integration of highly-resolved strucutre and other various information - 9:00—11:30		
4F	Conference Room 404	Meeting Room			
1F	Conference Room 101+102	Poster Session	Poster Setup, Display 8:45—14:05		
1F	Multi-Purpose Hall	Poster Session	Poster Setup, Display 8:45—14:05		
1F	Multi-Purpose Hall	Exhibition	Exhibition 9:00—17:00		

14：00	15：00	16：00	17：00	18：00	19：00	20：00	21：00

2SAP
Advanced bioimaging utilizing resonance between electromagnetic waves and molecules for life
16：15－18：45

2SBP
Bio-Raman research seeking bio. phys. chem. about the triple point
16：15－19：05

2SCP
【BSJ Special Event Ⅲ】BSJ-ASB Joint Symposium: Live Cell Imaging
16：15－18：45

2SDP
Frontiers in protein organization and disorganization
16：15－19：05

2SEP
Taking a new look through the optical microscopy
16：15－18：45

2SFP
Advances in experimental measurements by data-driven science based on sparse modeling
16：15－18：45

2SGP
Information processing governed by dynamic protein phosphorylation
16：15－18：45

Young Researchers Society Meeting
17：45－18：45

Poster Presentation Odd numbers 14：05－15：05	Poster Presentation Even numbers 15：05－16：05	Poster Display, Removal 16：05－17：30
Poster Presentation Odd numbers 14：05－15：05	Poster Presentation Even numbers 15：05－16：05	Poster Display, Removal 16：05－17：30

Banquet
Hotel Grand Shinonome
19：30－21：30

November 27 (Sun.) Day 3

Tsukuba International Congress Center

Floor	Venue	Room	Session	Time
2F	Convention Hall 200	Room A	3SAA New fields of cell biology explored with fluorescence and bioluminescence techniques	9：45—12：15
2F	Convention Hall 200	Room A	3LA Carl Zeiss Microscopy Co., Ltd.	12：30—13：20
2F	Conference Room 202	Room B	3SBA What connects the softness of biomolecules to their functions?	9：45—12：15
2F	Conference Room 202	Room B	3LB OPTO-LINE, Inc.	12：30—13：20
2F	Conference Room 201	Room C	3SCA Dynamic structural biology by next-generation researchers	9：15—12：15
2F	Conference Room 201	Room C	KAKENHI Guide Meeting	12：30—13：20
3F	Convention Hall 300	Room D	3SDA Harmonized supramolecular motility machinery and its diversity	9：15—12：15
3F	Conference Room 303	Room E	3SEA Synthetic biology for multicellular system	9：15—12：15
3F	Conference Room 304	Room Y	Gender Equality Promotion & Young Researchers Committee Meeting	12：30—13：20
4F	Conference Room 406	Room F	3SFA Management of mitochondrial functions by molecular machineries: biogenesis, structure, function, adaptation, and elimination	9：45—12：15
4F	Conference Room 406	Room F	3LF NIKON INSTECH CO., LTD.	12：30—13：20
4F	Conference Room 405	Room G	3SGA Programmable bioinspired systems: Integration of precisely designed architectures towards molecular robots	9：45—12：15
4F	Conference Room 404	Meeting Room	Industry - Academia Discussion Meeting	9：30—11：00
1F	Conference Room 101+102	Poster Session	Poster Setup, Display	9：00—13：35
1F	Multi-Purpose Hall	Poster Session	Poster Setup, Display	9：00—13：35
1F	Multi-Purpose Hall	Exhibition	Exhibition	9：15—16：15

14：00		15：00		16：00		17：00		18：00		19：00		20：00		21：00
Poster Presentation Odd numbers 13：35 – 14：35	Poster Presentation Even numbers 14：35—15：35		Poster Display, Removal 15：35 – 16：15											
Poster Presentation Odd numbers 13：35 – 14：35	Poster Presentation Even numbers 14：35—15：35		Poster Display, Removal 15：35 – 16：15											

Information for Participants and Presenters

1. Registration

◇ Registration desk

Location: The Entrance Hall in the Tsukuba International Congress Center (Refer to the floor map page 26.)

Open Hours: Nov. 25(Fri) 8:30 - 17:00
Nov. 26(Sat) 8:45 - 17:00
Nov. 27(Sun) 9:00 - 15:00

◆Advance registration

For participants who have completed advance registration with full payment of the registration fee by the deadline, there is no need to stop by the registration desk. A name badge, a receipt and a program booklet have already been sent to these participants.

*Participants from overseas: Receive a name badge and a program booklet at the registration desk.
*Please wear your name badge throughout the meeting. Without it, you cannot enter the meeting site. (Name badge holders will be provided at a place near the registration desk.)

Note 1) Advance registration is completed only after the payment is done. In case your payment cannot be confirmed by the deadline, your registration is automatically cancelled. In this case, please register on-site at the registration desk.
Note 2) Name badges have not been sent to those who have not paid the BSJ annual membership fee. Please complete the payment at the BSJ desk of the meeting site.
Note 3) If you have already paid both registration and BSJ annual membership fees but not received a name badge, please visit the registration desk.
Note 4) For non-members who are invited to talk at a symposium, the membership is not required and your registration fee is waived. You are invited for free to the banquet.
Note 5) Attendees who belong to institutions outside of Japan can make a presentation without a membership.

◆On-site registration

Those who have not completed advance registration are required to register on-site at the registration desk. Only cash payment is acceptable.

◇ On-site registration fees

	BSJ Member				Non-Member		
	Regular	Senior	Student	Undergraduate student	Regular	Student	Undergraduate student
Registration fee	¥9,000	¥5,000	¥5,000	¥0	¥12,000	¥6,000	¥0
Banquet fee	¥8,000	¥5,000	¥5,000	¥3,000	¥8,000	¥5,000	¥3,000

・For undergraduate students, the registration fee is waived. You are required to present your student ID at the registration desk to receive a name badge and a program booklet. But the banquet fee is charged if you attend the banquet.

・The "Early Career Award in Biophysics" Candidate presenters, the Biophysics and Physicobiology Outstanding Paper Award presenter and the representative of Biophysics and Physicobiology Editors' Choice awardee are invited for free to the banquet. If you have already paid the banquet fee, you can get a full refund at the registration desk.

◇ Name badge

Please be sure to wear your name badge throughout the meeting. Entry without the badge is NOT acceptable. Remember to bring your name badge that was sent in advance. (Name badge holders will be provided at a place near the registration desk.)

◇ Receipt

A receipt is attached to the name badge. If you need another receipt form, it will be issued in exchange for the one attached to your name badge.

◇ Program booklet / Abstract online system 【Release date: Oct. 25 (Tue)】

A program booklet (a part of front matters, and program) will be sent in advance to BSJ members and non-members with advance registration. The abstracts will be released only on the online system. No printed abstract booklet will be issued. On the online system, you can browse, search and download abstracts.
Program booklets can be purchased at the meeting site: JPY3,500/booklet.

Abstract online system: http://www.biophys.jp/dl/pro/54th_proceedings.pdf Download ID: ambsj54 PW: tsukuba2016

The program (presentation title, presenter's name and affiliation) and the online abstracts will be released on the BSJ54 web site. A half year later after the meeting, the abstracts will be posted on the J-Stage web site which is linked from the BSJ web site.
BSJ web site: http://www.biophys.jp/ann/ann02.html
J-Stage web site: http://www.jstage.jst.go.jp/browse/biophys/-char/ja

◇ Program search system (Web version) 【Release date: Oct. 25 (Tue)】

Program search system will be released on the BSJ54 web site.

◇ Free app to search and browse Program & Abstracts 【Release date: Nov. 21 (Mon)】

A free applet for smart phones (iPhone /Android) and tablet computers (iPad /iPod Touch /Android) can be downloaded from App Store or Google Play.

App name: The 54th Annual Meeting of the Biophysical Society of Japan Search word: bsj2016 PW of the abstracts browsing system: tsukuba2016

◇ BSJ membership (payment of the annual membership fee, and admission procedures)

For those who have not yet paid their annual membership fee, you can pay at the BSJ desk. For non-members, you are welcome to sign up at the BSJ desk to become a new member. For non-members who are invited to talk at a symposium or belong to institutions outside of Japan, the BSJ membership is not required.

2. Services & Facilities

◇ Cloakroom

Location: The Entrance Hall in the Tsukuba International Congress Center (Refer to the floor map page 26.)

Open Hours: Nov. 25(Fri) 8:30 - 19:45
Nov. 26(Sat) 8:45 - 19:15
Nov. 27(Sun) 9:00 - 16:00

*Valuables or computers cannot be checked into the cloak since the society/meeting does not hold any responsibility for loss or damage of your items.
*Please pick your items up when you leave the meeting venue.

◇ Lunch

Free lunch:
Free lunch will be provided at luncheon seminars (day 1-3), Gender Equality & Young Researchers Support Symposium (day 1), and KAKENHI Guide (day 3). Lunch tickets will be distributed in the morning of day 1-3.
Please refer to luncheon seminar page.
Also a limited number of free lunch without lunch tickets will be provided at Experts Committee (day 2).

◆Restaurant

A Restaurant is available as follows:

■ Restaurant "ESPOIR"(Tsukuba International Congress Center, 1F)
9:00 - 17:00 (until 14:00 on Nov. 27)
*Dinner time, after 17:00, is reservation-only.

Please feel free to take a gourmet map near the registration desk.

◇ Paging service・bulletin board

No paging service is available to call an individual except for an emergency. Please use a bulletin board near the registration desk in order to contact with other participants.

◇ Parking

Parking lot is not available for participants.

◇ Accommodation

Please refer to "Accommodation" page of the BSJ2016 web site.

◇ Internet

A Wireless LAN service is available except in the Conference Rooms and Convention Halls.
An ID and password will be informed at the meeting venue.
Network name: publics

◇ Coffee

Coffee is available for free (limited quantities) during Poster Presentation at the poster/exhibition place (Multi-Purpose Hall, 1F). Please note that coffee is charged (JPY100) except during Poster Presentation.

3. Programs & Events

*Several programs and events (committee meetings, general assembly meeting and its associated workshop, and lecture for public) are omitted here.

◇ "Early Career Award in Biophysics" Candidate Presentations

Date & Time: Nov.25 (Fri) 9:00 – 11:30
Place: Room C (Conference Room 201)

◇ Student Presentation Award

・Flash Talk

Date & Time: Nov.25 (Fri) 12:45 – 13:45

・Poster Presentation

Date & Time: Nov.25 (Fri) 13:55 – 15:55 / Nov.26 (Sat) 14:05 – 16:05

◇ Lecture by Biophysics and Physicobiology Outstanding Paper Awardees

Date & Time: Nov. 25 (Fri) 16:00 – 16:30
Place: Room A (Convention Hall 200)

◇ Banquet

Date & Time: Nov. 26 (Sat) 19:30 – 21:30
Place: Hotel Grand Shinonome, Banquet hall "SHINONOME", 2F
(About 13-minute walk from Tsukuba International Congress Center)
Address: 488-1 Onozaki, Tsukuba, Ibaraki Tel: 029-856-2211
*On-site registration is available at the registration desk or the banquet reception desk.

◇ Luncheon seminar

Lunch tickets will be distributed at the luncheon seminar desk, as shown below.

◆Distribution of lunch tickets

Luncheon seminar desk
Hours: Nov. 25 (Fri) 8:30 – 10:45 / 26 (Sat) 8:45 – 10:45 / 27 (Sun) 9:00 – 11:15
*The desk will be closed when all the tickets are distributed.
Location: The Entrance Hall in the Tsukuba International Congress Center, near the registration desk

*Only tickets for the seminars on the day are provided on a first-come-first-served basis.
*Lunches are provided by courtesy of companies and groups co-sponsoring luncheon seminars.

◆Attention

Please note that the lunch tickets will become invalid when you do not come before the starting time of the seminars and that the resulting remaining lunches will be provided to those who are attending the seminars without lunch tickets.

◆Request

We kindly ask you to attend till the last.
Also, please enter your affiliation and name in the questionnaire provided at the luncheon seminar as possible as you can.

◇ Exhibition

Instruments, reagents, software, books, etc. are displayed at the exhibition hall (Multi-Purpose Hall, 1F).

4. Prohibited Items

◇ Photography & recording

Photography and recording with camera, video, mobile phone and any device is NOT allowed at the meeting site. Please note that some recording may be performed after obtaining the board meeting's approval.

◇ Smoking, drinking & eating

Smoking is NOT allowed at the meeting site. Drinking and eating is NOT allowed inside lecture rooms except for luncheon seminars and other seminars/meetings in which meals are served.

◇ Cell-phone use

Talking on a mobile phone in the lecture/presentation rooms is NOT allowed. Please set your mobile phone on the silent mode or off, and make sure it will not make noises during lectures/presentations.

5. Contact

◇ During the meeting

Secretariat (Tel: 050-3482-9415 *phone number reachable during the meeting)

◇ Before or after the meeting

The organizing committee of the BSJ54
bp_nenkai54@bio.c.u-tokyo.ac.jp
Registration and abstract submission support desk
Nakanishi Printing Company
Ogawa-higashiiru, Shimodachiuri-dori, Kamigyo-ku, Kyoto 602-8048
bsj2016sys-sprt@e-naf.jp
Support team, exhibition and advertisement secretariat
A & E planning Co., Ltd.
Iwanami Shoten Hitotsubashi Bekkan 4F, 2-4-4 Hitotsubashi, Chiyoda-ku, Tokyo 101-0003
Tel: 03-3230-2744 / Fax: 03-3230-2479
Support team: jbp2016@aeplan.co.jp
Exhibition and Advertisement secretariat: e_jbp2016@aeplan.co.jp

6. Information for Presenters

◇ Language

Prepare your slides in English and give your presentation in English.

◇ Projector

Please bring a laptop with you for your presentation. A projector is equipped in each lecture room.
A sound output is not accepted.

1) Please prepare your presentation file in Microsoft PowerPoint.
2) The output resolution should be XGA (1024 x 768). The higher resolutions would possibly lose some information.
3) Our staff will connect your laptop to a video switcher.
4) The video output connector of your laptop should be "miniD-sub15pin (female)". If your connector is a different type (for example, that of Macintosh computer), please bring a conversion adaptor.

5) Bring your PowerPoint file in a USB memory.
6) Bring your AC adaptor in case that your battery would die.
7) Deactivate the screen-saver and power saving mode of your laptop.

◇ For organizers of symposia & "Early Career Award in Biophysics" (ECAB) Candidate presentations

Please come to the assigned room by 15 minutes before the start of the session, and then tell our staff of your arrival. Keep the time schedule and make smooth progress in the program. As a time keeper, our staff will help you.

Time allocation:
Symposium: Time allocation will be controlled by chairpersons.

◇ For speakers of symposia & ECAB Candidate presentations

Please come to the "PC Reception Desk" in the assigned room by 15 minutes before the start of the session. Our staff will connect your computer to a video switcher.
*Please note that there is no preview room.

ECAB Candidate Presentation: Presentation 10min. + Discussion 3min. + Laptop change 30seconds

◇ For Speakers of Flash Talks for "Student Presentation Award"

Meeting place and time: Please come to your presentation room by 12:35 on the first day of the annual meeting (Refer to page 44 – 45).

Presentation time: The presentation time allotted to each presenter is 2 minutes and 30 seconds. Stop your presentation immediately when the time keeper rings the bell twice.

Presentation language: Prepare your slides in English and give your presentation in English.

Presentation slides: Your presentation slides will be installed in the PC prepared by the organizers. Please prepare the slides in the pdf format following the instructions sent to you and submit the file to the society office (bsj@nacos.com) by the deadline. You can use up to four pages including the title page prepared based on the template. If necessary, you can add a few pages to have an animation effect. The file size should be smaller than 10 MB.

◇ For poster presenters

		Day 1, Nov. 25	Day 2, Nov. 26	Day 3, Nov. 27
Setup, Display		8:30 – 13:55	8:45 – 14:05	9:00 – 13:35
Presentation Discussion	Odd Numbers	13:55 – 14:55	14:05 – 15:05	13:35 – 14:35
	Even Numbers	14:55 – 15:55	15:05 – 16:05	14:35 – 15:35
Removal		until 17:00	until 17:30	until 16:15

*Periods of poster display: Posters will be replaced every day for the next day's poster presentations.
*Panel size: 90cm wide x 210cm high. Push pins are available at the site.
*Removal: Any posters remaining on panels after the removal time will be discarded by the secretariat.

◇ Instructions for poster presentation

A poster must be written in English.
Put a small circle on the upper left of the presenter's name.

◇ Presentation types and how to read the presentation numbers

Presentation types are Symposium Speech, “Early Career Award in Biophysics” Candidate Presentations, Poster Presentations and Flash Talks for “Student Presentation Award”.

Speech at symposium: (Ex.) 1SAA-03

Presentation day (1, Nov. 25; 2, Nov. 26; 3, Nov. 27) + Symposium (S) + Session room (Room A) + AM (A) / PM (P) + Order of the talk

“Early Career Award in Biophysics” Candidate Presentations: (Ex.) 1YC1045

Presentation day (1, Nov. 25) + Young Scientists (Y) + Session room (Room C)
+ Starting time of the talk

Poster presentations: (Ex.) 1Pos001

Presentation day (1, Nov. 25; 2, Nov. 26; 3, Nov. 27) + Poster (Pos) + Panel number

Flash Talks for “Student Presentation Award”: (Ex.) 1Pos001*

“*” is attached on the right side of a poster number.

第 54 回日本生物物理学会年会　市民講演会
「生命とは何かの普遍性に挑む
～理論物理と構成的実験からのアプローチ～」

日　時：11 月 5 日（土）　開場 13 時 30 分、開演 14 時、終演 16 時

会　場：東京大学駒場 I キャンパス　21 KOMCEE East 地下 1 階ホール
（東京都目黒区駒場 3-8-1、京王井の頭線「駒場東大前」）

参加費：無料（どなたでも自由に参加できます）

主　催：第 54 回日本生物物理学会年会実行委員会

後　援：複雑生命システム動態研究教育拠点

世話人：澤井 哲（東京大学大学院総合文化研究科）
豊島 陽子（東京大学大学院総合文化研究科）

講演プログラム

「普遍生物学：変化しやすさと安定性の状態論」

金子 邦彦 教授（東京大学大学院総合文化研究科）

「生命とは何か」—量子力学の祖の一人、シュレーディンガーは、その著書で、情報を担う分子、DNA の性質を予言しました。以降、生物内の個々の分子の性質は調べ挙げられてきました。しかし、それら分子の集まった「生きている状態とは？」の答えには至っていません。要素（分子や細胞）の間の関係に着目して、「多様な成分を維持し成長し、適応して進化する」生物の普遍法則を構成的実験と理論物理で解き明かそうとしています。

「人工細胞デバイスはイノベーションをもたらすか？」

野地 博行 教授（東京大学大学院工学系研究科、JST）

人工細胞関係の研究者の好奇心に基づく成果を伸ばしつつ、その技術を社会実装するための取り組みとしてのプログラムを行っている。まだ始まったばかりだけど、そのきっかけになったエピソードなどをかいつまんで紹介しながら基礎と実用は意外と近いことを話したい。でも一番大事なことは好奇心に基づく自由な研究。これが無いとユニークなイノベーションを生まないし、そもそも面白くないってことも伝えたい。

講演は日本語で行われます。

第 3 回会員総会シンポジウム
「1 分子生物物理の勃興期から未来　木下一彦氏を偲んで」

オーガナイザー：日本生物物理学会　理事会

日　時：11 月 26 日（土）13:00 ～ 13:55（会員総会中）

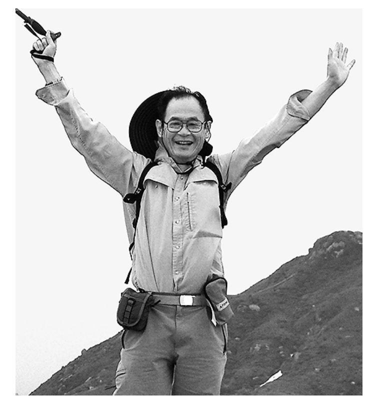
会　場：D 会場（中ホール 300）

司　会：野地博行

シンポジウム趣旨

1 分子生物物理学は、機能している生体分子を 1 分子単位で「見て」「操作する」ことでその機能を解明する学問領域として大きく発展しました。日本生物物理学会は、この 1 分子生物物理学の勃興期から極めて大きな貢献をしています。昨年 11 月初旬、この 1 分子生物物理学を牽引してきた木下一彦氏（当時早稲田大学教授）が南アルプスで亡くなられました（享年 69 歳）。木下氏は、リポソームの電気穿孔の高速イメージングから始まり、アクトミオシンや ATP 合成酵素の 1 分子計測で世界的成果を達成してきた 1 分子生物物理学の世界的パイオニアの 1 人です。木下氏逝去のニュースは、国内にとどまらず国外においても大きな衝撃を与えました。本シンポジウムは、本学会にも大きな影響を与えてきた木下氏の人柄と研究を振り返りながら、1 分子生物物理のこれからを議論することを目的とします。そこで、木下氏とともに 1 分子生物物理学を牽引してきた石渡信一氏と、木下研出身者で新しい研究を展開されている城口克之氏を演者としてお招きします。

木下一彦氏略歴



1946 年 10 月 22 日　愛知県豊川市　生まれ
1976 年 2 月　東京大学理学博士
1989 年 4 月　慶應義塾大学理工学部教授
2001 年 4 月　岡崎国立共同研究機構統合バイオサイエンスセンター教授
2005 年 4 月　早稲田大学理工学部物理学科教授
2015 年 11 月 逝去

プログラム

13:00–13:05　はじめに
13:05–13:30　石渡信一「木下一彦と 1 分子生理学」
13:30–13:45　城口克之「「1 個」を見たあと」
13:45–13:55　質疑応答

石渡信一　「木下一彦と 1 分子生理学」

木下一彦は昨年 2015 年 11 月 3 日、南アルプスの仙丈ケ岳（3032 m）の頂上付近で滑落し、突然我々の前からいなくなりました。すでにわが国の三百名山を踏破し、山で亡くなれば本望だと、数日前にも言っていた言葉が現実のものになるとは、思いもよらないことでした。学部時代から 50 年近く公私共に身近な友人であった木下（学生時代以来これ以外の呼び方をしたことがないので、呼び捨てにさせて下さい）の“1 分子生理学”について語ります。木下は自らの研究を好んで 1 分子生理学と呼びました。自らの研究をそう呼んだのは、細胞機能を担うタンパク質が 1 分子レベルでどのように働くか、動くか、1 分子機能の仕組みを

明らかにすること、そのために力によって機能を操作できるかを問い、あくまでも機能の仕組みの解明に力点を置いたということです。木下の業績の中では F_1-ATPase の回転運動の直視と、回転メカニズムの詳細な研究が有名ですが、そこに至る研究の道筋にも興味がもたれます。木下一彦という稀有な才能をもった研究者が辿った道筋を追います。

城口克之 「「1 個」を見たあと」

学部 3 年時の生物物理の講義の初日に、「生物の定義はなんですか？」と、木下先生に質問しました。率直に答えてくださったお人柄に無意識（？）に惹かれ、私は木下研を志望しました。ちょうど F_1-ATPase の回転運動が光学顕微鏡で観察されたすぐ後でした。他の研究室で学位を取得したのち、ポスドクとして再び木下研にお世話になりました。ミオシン V の“歩く”仕組みを解明しようと 1 分子観察に取り組み、木下さんから独特な励ましのお言葉を頂きながら、なぜ 1 個を見たいのか、見えたら何が分かりそうなのか、それっぽく見える動きは本当にミオシンの動きなのか、など、1 分子研究についてたくさんのことを考えさせられました。再び木下研を離れる時に、「新しい分野にチャレンジしたほうがいいよ」とアドバイスを頂きました。この宿題に四苦八苦している私ではありますが、なんとかせねばという思いを、本シンポジウムで皆様と共有させていただけましたら幸いです。

学生発表賞フラッシュトークの講演会場
Rooms for Flash Talks for "Student Presentation Award"

日　時：11 月 25 日（金）12:45 ～ 13:45

Date & Time: Day 1, Nov. 25 (Fri) 12:45 ～ 13:45

フラッシュトークは次の会場で行います。講演者は 12:35 までにご自身のポスター番号がある講演会場にお越しください。

Flash Talks will be given in the following rooms. Please come to your presentation room by 12:35.

A 会場（中ホール 200）

分野区分 1：蛋白質 : 構造 ／ 蛋白質 : 構造機能相関 ／ ヘム蛋白質

司会：木川隆則（理研）、昆　隆英（阪大）

Room A (Convention Hall 200)

Research Area 1: Protein: Structure, Protein: Structure & Function, Heme proteins

Chairpersons: Takanori Kigawa (RIKEN), Takahide Kon (Osaka Univ.)

ポスター番号（Poster Numbers）：

1Pos003*, 1Pos008*, 1Pos013*, 1Pos020*, 1Pos032*, 1Pos033*, 1Pos036*, 1Pos038*, 1Pos039*, 1Pos089*, 1Pos094*, 2Pos001*, 2Pos007*, 2Pos013*, 2Pos022*, 2Pos028*, 2Pos034*, 2Pos039*, 2Pos046*, 2Pos050*

B 会場（中会議室 202）

分野区分 2：蛋白質 : 物性 ／ 蛋白質 : 機能 ／ 蛋白質 : 計測・解析 ／ 蛋白質工学

司会：玉田太郎（QST）、石川春人（阪大）

Room B (Conference Room 202)

Research Area 2: Protein: Property, Protein: Function, Protein: Measurement & Analysis, Protein: Engineering

Chairpersons: Taro Tamada (QST), Haruto Ishikawa (Osaka Univ.)

ポスター番号（Poster Numbers）：

1Pos048*, 1Pos051*, 1Pos054*, 1Pos057*, 1Pos060*, 1Pos063*, 1Pos078*, 1Pos083*, 2Pos052*, 2Pos055*, 2Pos058*, 2Pos061*, 2Pos064*, 2Pos067*, 2Pos070*, 2Pos075*, 2Pos076*, 2Pos085*, 2Pos092*, 2Pos095*

C 会場（中会議室 201）

分野区分 3：核酸結合蛋白質 ／ 核酸 ／ 光生物

司会：神山　勉（名大）、河野秀俊（QST）

Room C (Conference Room 201)

Research Area 3: Nucleic acid binding protein, Nucleic acid, Photobiology

Chairpersons: Tsutomu Kouyama (Nagoya Univ.), Toshihide Kono (QST)

ポスター番号（Poster Numbers）：

1Pos110*, 1Pos113*, 1Pos117*, 1Pos238*, 1Pos240*, 1Pos243*, 1Pos245*, 1Pos248*, 1Pos252*, 1Pos255*, 1Pos263*, 2Pos098*, 2Pos103*, 2Pos114*, 2Pos229*, 2Pos232*, 2Pos240*, 2Pos245*, 2Pos250*, 2Pos251*

D 会場（中ホール 300）

分野区分 4：細胞生物学 ／ バイオイメージング

司会：竹居孝二（岡山大）、松崎勝巳（京大）

Room D (Convention Hall 300)

Research Area 4: Cell biology, Bioimaging

Chairpersons: Kohji Takei (Okayama Univ.), Katsumi Matsuzaki (Kyoto Univ.)

ポスター番号（Poster Numbers）：

1Pos161*, 1Pos166*, 1Pos172*, 1Pos175*, 1Pos179*, 1Pos188*, 1Pos306*, 1Pos311*, 1Pos315*, 1Pos318*, 2Pos174*, 2Pos189*, 2Pos192*, 2Pos193*, 2Pos194*, 2Pos195*, 2Pos297*, 2Pos298*, 2Pos301*

E 会場（小会議室 303）

分野区分 5：筋肉 ／ 分子モーター ／ 計測

司会：大岩和弘（NICT）、西山雅祥（京大）

Room E (Conference Room 303)

Research Area 5: Muscle, Molecular motor, Measurements

Chairpersons: Kazuhiro Oiwa (NICT), Masayoshi Nishiyama (Kyoto Univ.)

ポスター番号（Poster Numbers）：

1Pos130*, 1Pos132*, 1Pos137*, 1Pos142*, 1Pos145*, 1Pos152*, 1Pos297*, 2Pos142*, 2Pos143*, 2Pos145*, 2Pos146*, 2Pos153*, 2Pos162*, 2Pos166*, 2Pos167*, 2Pos277*, 2Pos282*

F 会場（中会議室 406）

分野区分 6：膜蛋白質 ／ 生体膜・人工膜 ／ 神経科学・感覚

司会：須藤雄気（岡山大）、湊元幹太（三重大）

Room F (Conference Room 406)

Research Area 6: Membrane proteins, Biological & Artificial membrane, Neuroscience & Sensory systems

Chairpersons: Yuki Sudo (Okayama Univ.), Kanta Tsumoto (Mie Univ.)

ポスター番号（Poster Numbers）：

1Pos095*, 1Pos101*, 1Pos102*, 1Pos190*, 1Pos227*, 1Pos228*, 1Pos230*, 1Pos231*, 2Pos201*, 2Pos204*, 2Pos213*, 2Pos214*, 2Pos215*, 2Pos225*, 2Pos226*

G 会場（小会議室 405）

分野区分 7: 水・水和・電解質 ／ 発生・分化 ／ 生命の起源・進化 ／ ゲノム生物学 ／ バイオインフォマティクス ／ 数理生物学 ／ 非平衡・生体リズム ／ バイオエンジニアリング

司会：飯野亮太（岡崎統合バイオ）、金城玲（阪大）

Room G (Conference Room 405)

Research Area 7: Water & Hydration & Electrolyte, Development & Differentiation, Origin of life & Evolution, Genome biology, Bioinformatics, Mathematical biology, Nonequilibrium state & Biological rhythm, Bioengineering

Chairpersons: Ryota Iino (OIIB, IMS), Akira Kinjo (Osaka Univ.)

ポスター番号（Poster Numbers）：

1Pos271*, 1Pos276*, 1Pos287*, 1Pos289*, 1Pos290*, 1Pos291*, 1Pos292*, 2Pos123*, 2Pos126*, 2Pos129*, 2Pos132*, 2Pos257*, 2Pos258*, 2Pos261*, 2Pos310*, 2Pos311*, 2Pos314*, 2Pos315*, 2Pos316*

一般社団法人日本生物物理学会　第 5 回 Biophysics and Physicobiology 論文賞受賞講演会
The 5th Award Seminar for outstanding Biophysics and Physicobiology paper

オーガナイザー：日本生物物理学会 Biophysics and Physicobiology 論文賞選考委員会

Organizers: Award committee for outstanding Biophysics and Physicobiology paper

日　時：11 月 25 日（金）16：00 〜 16：30 ／ Nov. 25 Fri.

場　所：A 会場（中ホール 200）／ Room A (Convention Hall 200)

形　式：講演会 ／ Lecture

第 5 回 Biophysics and Physicobiology 論文賞受賞者

BPPB Outstanding Paper Awardee

須藤雄気

Yuki Sudo

岡山大学医歯薬学総合研究科（薬学系）

Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama University

H^+ および Na^+ 透過型べん毛モーター固定子ユニット内のイオン透過経路の比較解析

Comparative study of the ion flux pathway in stator units of proton- and sodium-driven flagellar motors

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Organisms sense and respond to external stimuli to survive in the environments by changing their motile mode. Flagellar motors are essential for the motile microorganisms, and they are powered by the electrochemical potential gradient of specific ions across the membrane. Transmembrane proteins, MotA and MotB from *Escherichia coli*, work as a H^+ channel, while their homologs of *Vibrio alginolyticus*, PomA and PomB, work as a Na^+ channel. These MotA/B and PomA/B complexes play essential roles in torque generation as the stators. It is known that a conserved residue, Asp32 for MotB and Asp24 for PomB, forms one of the ion binding sites. Ala39 of MotB and Cys31 of PomB are located on the same sides as Asp32 of MotB and Asp24 of PomB, respectively, in a helical wheel diagram. In this study, a series of mutations were introduced into Ala39 of MotB and Cys31 of PomB. As a result, the motility of mutant cells was markedly decreased as the volume of the side chain increased. The loss of function was suppressed by mutations of M206S for MotA and L183F for PomA, respectively, and the increases in the volume caused by mutations of A39V for MotB and C31A for PomB were close to the decreases in the volume caused by mutations of M206S for MotA and L183F for PomA. From these results, we concluded that Ala39 of MotB and Cys31 of PomB form part of the ion flux pathway with Met206 of MotA and Leu183 of PomA in the MotA/B and PomA/B stator units, respectively.

男女共同参画・若手支援委員会企画シンポジウム
キャリアデザインの第一歩
―大学院生・研究者のための自己分析ワーク―
Academic Self-Analysis Workshop on Career Development

オーガナイザー：日本生物物理学会　男女共同参画・若手支援委員会

Organizers : Promotion of Gender Equality and Young Researchers Committee

日　時：11 月 25 日（金）11:45 ～ 12:35（ランチョンセミナーの時間帯）

会　場：Y 会場（小会議室 304）

昼　食：お弁当とお茶を無料で提供いたします。ただし、数に限りがあります。

形　式：ワークショップ（各自、筆記用具をご準備ください。）

概要：自身のキャリアを想像しキャリアアップの準備を進めること、すなわち「キャリアデザイン」を早期に行うことは、後悔のない人生設計のために非常に重要です。そこで今回は、キャリアデザインの最初のステップである「自己理解」をみなさんに実践していただくために、「大学院生･研究者のための自己分析ワーク」を体験できる企画をご用意しました。このワークは大学院生や研究者のためのキャリアアップ支援団体、Tokyo Ph.D. Lounge（TPL）が【研究者専用】に独自に考案したものです。大学院生活や研究生活を分析の主な軸として、キャリアを考える上で重要となる自分の強みや価値観を探ります。本企画を監修する TPL 代表の筒井香織さんは、生物物理学会出身の元研究者ですが、産業カウンセラーの資格を取得し、現在では大学でのキャリア教育や就職支援に携わっています。TPL では、先輩博士によるセミナーとグループワークで構成される「博士キャリア座談会」を定期的に開催し、アカデミック人材の自主的なキャリア構築をサポートしています。

本シンポジウムでは、参加者のみなさんに実際に「大学院生・研究者のための自己分析ワーク」に挑戦していただきます。当日会場で配布するワークシートの簡単な設問に回答することで、自身の強みや重視したいこと、将来の目標などを分析します。参加者のみなさんには二人ずつでペアを組み、ワークシートの内容をお互いに発表していただきます。自身の強み・価値観を整理して相手に伝え、相手からフィードバックを受けることで、これまで気づかなかった自身の内面や意外な一面の発見につなげることが狙いです。

今回の生物物理学会年会では、この男女共同参画若手支援委員会シンポジウムで「自己理解」を、年会期間中に開催されるキャリア支援説明会で「職業理解」「会社理解」を行います。本シンポジウムとキャリア支援説明会の両方を通じて、みなさんのキャリアデザインをサポートする内容となっていますので、興味をお持ちの方は奮ってご参加ください。

キャリア支援説明会

オーガナイザー：第 54 回年会実行委員会、日本生物物理学会 男女共同参画・若手支援委員会
日　時：11 月 25 日（金）13:00 ～ 18:00、11 月 26 日（土）9:30 ～ 18:15
会　場：Y 会場（小会議室 304）
形　式：セミナーおよび交流会

概要：若手研究者や学生の今後のキャリア構築の一助となるように、昨年の年会から「キャリア支援説明会」を開催しています。本年会では、学会本部の男女共同参画・若手支援委員会と年会実行委員会が協力して内容を刷新し、キャリアエージェントとして活躍されている（株）レゾナンスの前田恵一氏をお迎えして、以下のようにセミナー、交流会等を実施します。

プログラム：
11 月 25 日（金）
第 1 部　13:00–13:30　キャリアデザインセミナー「研究者に求められるビジネス指向性」
　経済産業省が発表している「社会人基礎力」をもとに、企業が求めている人材像を例示します。その上で研究者としての経験を持つ強みや特性を活かしながら、キャリアアップにどうつなげていくかを考えます。
第 2 部　13:40–15:40　業界説明、協賛企業の紹介および交流会
　10 社程度の企業を招待し交流会を実施します。協賛企業の人事採用担当者との情報交換を通して、キャリア意識の醸成を狙いつつ、業務イメージを持つことでキャリア形成の準備を行います。
第 3 部　16:00–18:00　企業分析術の基礎　「データが示す企業の特性（ファイナンス・会計）」
「ファイナンス」や「会計」は研究者が普段接することの少ない言葉ですが、企業の姿を自分の目で確認するために欠かせない基礎素養であり、幅広いキャリア形成に役立ちます。今回は「企業分析術」の基礎として、「ファイナンス」や「会計」各 1 時間ずつのレクチャーを行います。一般には見えにくい各企業の特徴をデータから解析するための基礎知識の習得を目指します。

11 月 26 日（土）
第 4 部　9:30–11:30　企業分析術の基礎　「データが示す企業の特性（ファイナンス・会計）」
（第 3 部と同じ内容）
第 5 部　14:05–16:05　業界説明、協賛企業の紹介および交流会
（第 2 部と同じ内容）
第 6 部　16:15–18:15　協賛企業によるブース形式の説明会
協賛企業が実際にブースを構え、企業紹介や業務説明を中心に個別説明をする場を設ける予定です。

＊説明はすべて日本語で行われます。

「科研費改革の動向」
Reorganization of KAKENHI : Current Activities of JSPS

世話人：豊島陽子（東京大学大学院総合文化研究科、日本学術振興会学術システム研究センター専門研究員）
Organizer： Yoko Toyoshima (Graduate School of Arts and Sciences, the University of Tokyo; Program Officer, Research Center for Science Systems, JSPS)

日　時：11 月 27 日（日）12:30 ～ 13:20（ランチョンセミナーの時間帯）
会　場：C 会場（中会議室 201）
昼　食：お弁当とお茶が無料で提供されます。ただし、数に限りがあります。
形　式：日本語による講演会

講師：日本学術振興会　研究助成企画課長　大鷲 正和

日本が将来にわたって卓越した研究成果を生み出し続けるために、科学技術・学術審議会において「科研費制度の抜本的改革」が提言され、科研費の研究種目・枠組みの見直しや審査システムの見直しが行われています。平成 30 年度科研費（平成 29 年 9 月公募）には「審査システム改革 2018」として、審査システムの完全移行が行われる予定であり、科研費業務の大部分を担当している日本学術振興会の学術システム研究センターが中心となり、2 年余りにわたり議論を進め、審査区分と審査方式とを一体的に見直す改革案をとりまとめているところです。今回は、このシステム改革の概要を中心に、ご説明をいただきます。

若手奨励賞招待講演　Early Career Award in Biophysics Candidate Presentations

第 1 日目（11 月 25 日（金））／Day 1（Nov. 25 Fri.）

9:00～11:30　C 会場／Room C：中会議室 201／Conference Room 201
1YC　日本生物物理学会若手奨励賞選考会
Early Career Award in Biophysics Candidate Presentations

オーガナイザー：男女共同参画・若手支援委員会
Organizer: Promotion of Gender Equality and Young Researchers Committee

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Since 2005, Biophysical Society of Japan (BSJ) has granted "Early Career Award in Biophysics" to young BSJ members for their excellent presentations that show great potential to contribute to the progress of biophysics. In this 12th year, we received 42 highly qualified applications. After the first round of competitive screening based on submitted documents, the following eleven applicants, including one student member, were selected as the young invited speakers. In this symposium, each speaker will make 10-minute presentation followed by 3-minute discussion as the second round of screening. Up to five winners will be selected and announced at the banquet held in the evening of the second day. We welcome all the BSJ members to attend this symposium to foresee the future of biophysics in Japan through the speakers and their researches.

09:00:00　阿部 淳　3Pos013
1YC0900　シアノバクテリア時計タンパク質 KaiC の AFM 観察
AFM observation of a ring-shaped structure of KaiC
○阿部 淳, 向山 厚, 古池 美彦, 秋山 修志（自然科学研究機構 分子科学研究所 協奏分子システム研究センター 階層分子システム解析研究部門）
Jun Abe, Atsushi Mukaiyama, Yoshihiko Furuike, Shuji Akiyama (*Division of Trans-Hierarchical Molecular Systems, Research Center of Integrative Molecular Systems (CIMoS), Institute for Molecular Science (IMS)*)

09:13:30　太田 禎生　2Pos293
1YC0913　人知を超える超高速・高精度蛍光形態サイトメトリー
Ghost Cytometry: fluorescence "imaging" cytometry beyond human's limit
○太田 禎生[1,2], 野地 博行[1,3]（[1]東大・工, [2]科学技術振興機構さきがけ, [3]科学技術振興機構ImPACT）
Sadao Ota[1,2], Hiroyuki Noji[1,3] ([1]*Sch. Eng., Univ. Tokyo*, [2]*JST, PRESTO*, [3]*JST, ImPACT*)

09:27:00　木下 佳昭　2Pos154
1YC0927　高度高塩菌ハロバクテリウムサリナラムのべん毛の回転とステップ運動の直接観察
Direct observation of rotation and steps of the archaellum in the swimming halophilic archaeon Halobacterium salinarum
○木下 佳昭[1], 内田 就也[2], 中根 大介[1], 西坂 崇之[1]（[1]学習院大学 理・物理, [2]東北大学 理・物理）
Yoshiaki Kinosita[1], Nariya Uchida[2], Daisuke Nakane[1], Takayuki Nishizaka[1] ([1]*Department of Physics, Gakushuin University*, [2]*Department of Physics, Tohoku University*)

09:40:30　寺島 浩行　3Pos174
1YC0940　細菌べん毛 III 型タンパク質輸送の *in vitro* 再構築
***In vitro* reconstitution of the bacterial flagellar type III protein export**
○寺島 浩行[1], 川本 晃大[2], 巽 千夏[1], 難波 啓一[2,3], 南野 徹[2], 今田 勝巳[1]（[1]阪大・院理, [2]阪大・院生命機能, [3]理研 生命システム）
Hiroyuki Terashima[1], Akihiro Kawamoto[2], Chinatsu Tatsumi[1], Keiichi Namba[2,3], Tohru Minamino[2], Katsumi Imada[1] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*, [3]*Quant. Bio. Cent., Riken.*)

09:54:00　永井 健　1Pos285
1YC0954　様々な回転する自走粒子の集団運動
Collective motion of various kinds of rotating self-propelled particle
○永井 健[1], 住野 豊[2], Hugues Chaté[3,4], 大岩 和弘[5,6], 杉 拓磨[7], 岩崎 秀雄[8]（[1]北陸先端大・先端理工, [2]東京理大・応物, [3]CEA-Saclay, [4]北京計算科学研, [5]NICT・未来ICT, [6]兵県大・院生命理工, [7]滋賀医大・分子神経科学研, [8]早稲田大・先端理工）
Ken Nagai[1], Yutaka Sumino[2], Chate Hugues[3,4], Kazuhiro Oiwa[5,6], Takuma Sugi[7], Hideo Iwasaki[8] ([1]*Sch. Mater. Sci., JAIST*, [2]*Dep. Appl. Phys., Tokyo Univ. Sci.*, [3]*CEA-Saclay*, [4]*Beijing Comp. Sci. Res. Ctr.*, [5]*Adv. ICT Res. Inst., NICT*, [6]*Grad. Sch. Sci., Univ. Hyogo*, [7]*Mol. Neurosci. Res. Ctr., Shiga Univ. of Med. Sci.*, [8]*Sch. Adv. Sci. Eng., Waseda Univ.*)

10:07:30 中島 昭彦 2Pos183

1YC1007 動的な場における時間空間知覚メカニズムの解析：走化性パラドクスの克服と細胞の整流作用

Delineating temporal and spatial sensing in migrating cells: chemotactic wave paradox and rectification of the leading edge response

○中島 昭彦[1], 石原 秀至[2], 石田 元彦[3], 井元 大輔[3], 澤井 哲[1,3]（[1]東大・院総文・複雑生命, [2]明治大・理工, [3]東大・院総文・広域科学）

Akihiko Nakajima[1], Shuji Ishihara[2], Motohiko Ishida[3], Daisuke Imoto[3], Satoshi Sawai[1,3] ([1]*Res. Cent. Comp. Sys. Biol., Grad. Sch. Arts Sci., Univ. Tokyo*, [2]*Sch. Sci. Tech., Meiji Univ.*, [3]*Dept. Basic Sci., Grad. Sch. Arts Sci., Univ. Tokyo*)

10:21:00 中山 義敬 1Pos215

1YC1021 コリネ細菌の機械受容チャネルによる細胞力覚とグルタミン酸放出機構

Bacterial mechanosensation and glutamate export by mechanosensitive channels in Corynebacterium glutamicum

○中山 義敬[1], 駒澤 光佑[2], Navid Bavi[1,3], 橋本 賢一[2], 川崎 寿[2], Boris Martinac[1,3]（[1]ビクターチャン心臓病研究所, [2]東京電機大学, [3]ニューサウスウェールズ大学）

Yoshitaka Nakayama[1], Kosuke Komazawa[2], Navid Bavi[1,3], Ken-ichi Hashimoto[2], Hisashi Kawasaki[2], Boris Martinac[1,3] ([1]*Victor Chang Cardiac Research Institute*, [2]*Tokyo Denki University*, [3]*University of New South Wales*)

10:34:30 畠山 哲央 1Pos288

1YC1034 生命システムの振動現象における頑健性と可塑性の互恵的関係

Reciprocity between robustness and plasticity in biological oscillators

○畠山 哲央, 金子 邦彦（東京大学総合文化研究科）

Tetsuhiro S. Hatakeyama, Kunihiko Kaneko (*Department of Basic Science, The University of Tokyo*)

10:48:00 原田 隆平 2Pos036

1YC1048 生物学的レアイベントを再現する効率的構造サンプリング手法の開発

Developments of conformational sampling methods for reproducing biologically rare events of proteins

○原田 隆平, 重田 育照（筑波大学計算科学研究センター）

Ryuhei Harada, Yasuteru Shigeta (*Center for Computational Sciences, University of Tsukuba*)

11:01:30 Eiji Yamamoto 2Pos212

1YC1101 **Investigating interactions and dynamics of pleckstrin homology domains on a lipid membrane surface**

Eiji Yamamoto[1], Antreas C. Kalli[2], Takuma Akimoto[1], Mark S.P. Sansom[2], Kenji Yasuoka[3] ([1]*Grad. Sch. Sci. Technol., Keio Univ.*, [2]*Dept. Biochem., Univ. Oxford*, [3]*Dept. Mech. Eng., Keio Univ.*)

11:15:00 渡邉 宙志 2Pos042

1YC1115 タンパク質の構造と機能の相関を利用した Channelrhodopsin と MtrF の戦略的立体構造モデリング

Strategic modeling of channelrhodopsins and MtrF based on the correlation between protein structures and functions

○渡邉 宙志[1,2], 山下 雄己[2], Marcus Elstner[3], 石北 央[1,2]（[1]東京大学 先端科学技術研究センター, [2]東京大学 工学部, [3]カールスルーエ工科大学）

Hiroshi C. Watanabe[1,2], Yuki Yamashita[2], Marcus Elstner[3], Hiroshi Ishikita[1,2] ([1]*UTokyo, RCAST*, [2]*UTokyo, School of Engineering*, [3]*Karlsruhe Institute of Technology*)

シンポジウム　Symposium

第1日目（11月25日（金））／Day 1（Nov. 25 Fri.）

9:00～11:30　A会場（中ホール200）／Room A (Convention Hall 200)
1SAA　神経活動イメージングの最先端：新規ツールとその活用
Advances in imaging neuronal activity: New tools and applications

オーガナイザー：ベアン クン（沖縄科学技術大学院大学学園），冨永 貴志（徳島文理大学）
Organizers: Kuhn Bernd (OIST Graduate University), Takashi Tominaga (Tokushima Bunri University)

Functional optical imaging is revolutionizing neuroscience. Every year new molecular or optical tools are added or improved and allow to study the biophysics of biological processes which were not accessible before. This symposium gives a snapshot of some of these current developments. Experts from probe design and methods development as well as from the interface of methods development and neuroscience will report their latest results. The symposium focuses on voltage and calcium probe design and imaging, two-photon optogenetics and FRET/FLIM, molecular orientation imaging, and birefringence imaging.

1SAA-01　「使える」膜電位感受性色素による神経回路解析法
“Conventional” voltage sensitive dye imaging of neural circuit activity
○冨永 貴志, 冨永 洋子（徳島文理大・神経研）
Takashi Tominaga, Yoko Tominaga (*Inst. Neurotic., Tokushima Bunri Univ.*)

1SAA-02　Tuning Genetically-Encoded Voltage Indicators to Better Resolve Different Types of Neuronal Activity
Bradley Baker (*KIST*)

1SAA-03　新規偏光顕微鏡を用いたマウス海馬スライスにおけるシナプス活動の非侵襲的計測
Imaging of neuronal activity in mice hippocampal slices by instantaneous polarized light microscopy
○小池（谷） 真紀[1], Mehta Shalin[1], Oldenburg Rudolf[1], 冨永 貴志[2], 谷 知己[1]（[1]ウッズホール海洋生物学研究所, [2]徳島文理大学）
Maki Koike-Tani[1], Shalin Mehta[1], Rudolf Oldenburg[1], Takashi Tominaga[2], Tomomi Tani[1] ([1]*Marine Biological Laboratory*, [2]*Tokushima Bunri University*)

1SAA-04　光活性化酵素制御とイメージング技術による cAMP/cGMP の時空間的機能探索
Two-photon optogenetic control and live imaging of postsynaptic cAMP/cGMP intracellular messengers
○岡本 賢一（LTRI, MSH）
Kenichi Okamoto (*LTRI, MSH*)

1SAA-05　in vivo calcium imaging with genetically encoded calcium indicators
Junichi Nakai[1,2], Keiko Gengyo-Ando[1,2], Masaaki Sato[1,2], Masamichi Ohkura[1,2] ([1]*Grad. Sch. Sci. Eng. Saitama Univ.*, [2]*BBSSI, Saitama Univ.*)

1SAA-06　Exploring input-output relations of neurons in awake mice
Christopher J. Roome, **Bernd Kuhn** (*Kuhn Unit, OIST*)

9:00～11:30　B会場（中会議室202）／Room B (Conference Room 202)
1SBA　全細胞解析によるマイノリティ細胞の解明
Minority cell research enabled by exhaustive analyses of all cells

オーガナイザー：永井 健治（大阪大学），上田 泰己（東京大学）
Organizers: Takeharu Nagai (Osaka University), Hiroki Ueda (The University of Tokyo)

If we carefully observe the cell population that at first glance looks uniform and homogeneous, we may find small number of heterogeneous cells with a different nature. Moreover, this minority cells would sometimes significantly alter the behavior of the whole cell population. In this symposium, we would like to discuss not only analytical methods for sensitive detection or visualization of such minority cells, but also the theories regarding principle or mechanism how the minority cells are generated and exert biological roles.

1SBA-01 全身・全脳透明化の先に見えてくるもの〜生命の『時間』の謎の解明に向けて〜
Toward Organism-level Systems Biology in Mammals〜Whole-body and whole-organ clearing and imaging with single-cell resolution〜
○上田 泰己[1,2]（[1]東京大学, [2]理化学研究所）
Hiroki R. Ueda[1,2] ([1]*The University of Tokyo*, [2]*RIKEN (QBiC)*)

1SBA-02 4K/8K CMOS イメージングによるマルチスケール生体全細胞解析
Multi-scale in vivo 4K/8K imaging analysis
○西村 智[1,2]（[1]自治医科大学, [2]東大）
Satoshi Nishimura[1,2] ([1]*Jichi Med. Univ.*, [2]*The Univ. of Tokyo*)

1SBA-03 マイノリティ細胞の同定と解析による自己免疫疾患発症制御機構の解明
Elucidation of pathomechanisms of autoimmunity by minority cell research
○岡崎 拓（徳島大学先端酵素学研究所免疫制御学分野）
Taku Okazaki (*Division of Immune Regulation, Institute for Genome Research, Tokushima University*)

1SBA-04 Raman spectroscopic approaches to label-free cell characterization and finding functional minorities
Katsumasa Fujita (*Osaka University*)

1SBA-05 マイノリティ細胞研究にむけた神経細胞および脳組織内在性グルタミン酸受容体の蛍光可視化
Visualization of native glutamate receptors in live neurons or neuronal tissues for minority cell study
○清中 茂樹（京大・院工）
Shigeki Kiyonaka (*Grad. Sch. Eng., Kyoto Univ.*)

1SBA-06 超解像生理機能イメージング法の開発とマイノリティ細胞の可視化の試み
Development of superresolution techniques for imaging physiological functions toward visualization of minority cells
○永井 健治（大阪大学産業科学研究所）
Takeharu Nagai (*ISIR, Osaka Univ.*)

1SBA-07 Finding genomic minority cells by sequencing
Katsuyuki Shiroguchi[1,2,3] ([1]*RIKEN Quantitative Biology Center*, [2]*RIKEN Center for Integrative Medical Sciences*, [3]*JST PRESTO*)

1SBA-08 1 細胞ラマン分光イメージングから如何にして細胞の個性を定量化するか？
How can one quantify cell individuality from Single Cell Raman Imaging?
○小松崎 民樹[1,2]（[1]北大 電子研 社会創造数学センター, [2]北大 生命）
Tamiki Komatsuzaki[1,2] ([1]*Hokkaido Univ., RIES, MSC*, [2]*Hokkaido Univ., Grad. Sch. Life Sci.*)

9:00〜11:30　D 会場（中ホール 300）／Room D (Convention Hall 300)
1SDA　原子からいのちへ：21 世紀の新しい生命観を求めて
From atoms to life: Exploring a new view of life in the 21st century

オーガナイザー：赤坂 一之（京都府立大学），伏見 譲（総合研究大学院大学）
Organizers: Kazuyuki Akasaka (Kyoto Prefectural University), Yuzuru Husimi (SOKENDAI)

Biomolecular science in the last century has revolutionized our approach to life: Today in biochemical and medical societies, crucial life phenomena are being discussed in terms of changes in macromolecular structures and interactions, and even of motions of individual atoms. How can the basically random thermal motions of atoms derive the macromolecular machinery into the dynamism of life? How in nature is the connection between atoms and life made generally possible? In this symposium, we intend to share our thoughts with the audience of all ages.

はじめに
赤坂 一之
Kazuyuki Akasaka

1SDA-01 蛋白質—無秩序な原子の動きを“命の動き”に変えるデバイス
Proteins-converting random motions of atoms into the dynamism of life
○赤坂 一之（京都府立大・院生命環境科学）
Kazuyuki Akasaka (*Kyoto Prefectural University*)

1SDA-02 分子と細胞、そして細胞と組織をつなぐメカニカルシグナル
Mechanical signals interface molecules with cells, and cells with tissues
○曽我部 正博（名大院・医）
Masahiro Sokabe (*Nagoya Univ. Grad. Sch. Med.*)

1SDA-03 分子情報システムとしての生命
Which parameters characterize“life”?
○美宅 成樹（サイエンスライター）
Shigeki Mitaku (*Science writer*)

1SDA-04 情報進化—原子といのちを結ぶ進化能的生命観—
Informational Evolution: An evolvability view point of life composed of atoms
○伏見 譲（総研大）
Yuzuru Husimi (*SOKENDAI*)

おわりに
伏見 譲
Yuzuru Husimi

9:00～11:30　E 会場（小会議室 303）／Room E (Conference Room 303)
1SEA　新学術領域研究「シリア・中心体系による生体情報フローの制御」共催
運動性鞭毛・繊毛の最前線—生体ナノマシンの制御機構—
Frontiers in motile cilia – regulatory mechanisms of bio-nanomachines –

オーガナイザー：小田 賢幸（山梨大学），若林 憲一（東京工業大学）
Organizers: Toshiyuki Oda (University of Yamanashi), Ken-ichi Wakabayashi (Tokyo Institute of Technology)

Cilia and flagella are conserved motile organelles that play essential roles in cellular motility of eukaryotes and development of higher organisms by generating fluid flow. The beating motion of cilia/flagella is driven by dyneins, whose activities are tightly regulated by complex molecular mechanisms. In this symposium, leading young scientists will present their recent findings regarding the ciliary/flagellar motility and its regulatory mechanisms in various model organisms.

opening remarks
若林 憲一
Ken-ichi Wakabayashi

1SEA-01 多細胞性緑藻ボルボックスの走光性：5000 の細胞が協調して泳ぐには？
Phototaxis in the multicellular green alga *Volvox*: How 5000 independent cells coordinate their motion?
○植木 紀子（東工大・化生研）
Noriko Ueki (*CLS, Tokyo Tech.*)

1SEA-02 繊毛の運動を支える細胞内構造の理解へ向けて
Towards understanding of cell structure that governs motion pattern of motile cilia
○篠原 恭介（東京農工大学）
Kyosuke Shinohara (*Tokyo University of Agriculture and Technology*)

1SEA-03 Roles of calcium in the regulation of sperm flagellar movement
Kogiku Shiba (*SMRC, Tsukuba Univ.*)

1SEA-04 IFT81 および IFT74 の N 末端領域によるチューブリンの鞭毛内輸送

The IFT81 and IFT74 N-termini together form the main module for intraflagellar transport (IFT) of tubulin

○久保 智広[1,4], Brown Jason[1,2], Bellve Karl[1], Craige Branch[1], Craft Julie[3], Forgarty Kevin[1], Lechtreck Karl[3], Witman George[1]（[1]マサチューセッツ大学・医, [2]セイラム州立大, [3]ジョージア大, [4]山梨大・医）

Tomohiro Kubo[1,4], Jason Brown[1,2], Karl Bellve[1], Branch Craige[1], Julie Craft[3], Kevin Forgarty[1], Karl Lechtreck[3], George Witman[1] ([1]*UMASS Med.*, [2]*Salem State Univ.*, [3]*Univ. of Georgia*, [4]*Univ. of Yamanashi Faculty of Medicine*)

1SEA-05 繊毛・鞭毛の中の動きを見る

Dynamics of molecules inside cilia and flagella

○高尾 大輔（遺伝研）

Daisuke Takao (*NIG*)

1SEA-06 脊椎動物運動性繊毛における PIH タンパク質の機能解析

The function of PIH proteins in the vertebrate motile cilium

○山口 博史[1,2], 山崎 陽祐[1], 小田 賢幸[1,3], 吉川 雅英[1], 武田 洋幸[2]（[1]東大・院医, [2]東大・院理, [3]山梨大・院医）

Hiroshi Yamaguchi[1,2], Yousuke Yamazaki[1], Toshiyuki Oda[1,3], Masahide Kikkawa[1], Hiroyuki Takeda[2] ([1]*Grad. Sch. Med., Univ. Tokyo*, [2]*Grad. Sch. Sci., Univ. Tokyo*, [3]*Grad. Sch. Med., Univ. Yamanashi*)

closing remarks

小田 賢幸

Toshiyuki Oda

9:00～11:30　F 会場（中会議室 406）／Room F (Conference Room 406)

1SFA　生物物理遺伝学：生物物理学的ゲノム情報科学としての

Biophysical Genetics as a Genome Informatics Supported by Biophysics

オーガナイザー：中井 謙太（東京大学），白井 剛（長浜バイオ大学）

Organizers: Kenta Nakai (The University of Tokyo), Tsuyoshi Shirai (Nagahama Institute of Bio-Science and Technology)

Since genetic information can be treated independently of the physical nature of its carrier DNA, the progress of genome information science has deviated to a certain extent from that of biophysics. The situation, however, has been changed recently because the importance of epigenome information, which is tightly linked with the 3D conformation of carrier DNA, i.e., the chromatin structure, has been recognized increasingly. Thus, in this symposium, we will celebrate the birth of a new field, biophysical genetics, inviting active researchers in it.

はじめに

中井 謙太

Kenta Nakai

1SFA-01 クロマチンの三次元構造と動的構造

Three dimensional structures and dynamics of chromatin

○胡桃坂 仁志（早稲田大学理工学術院 先進理工学部）

Hitoshi Kurumizaka (*Waseda University, Faculty of Science and Engineering*)

1SFA-02 Hi-C データを用いた遺伝子発現制御の理解

Using Hi-C data to understand gene regulation

○須山 幹太（九州大学 生体防御医学研究所）

Mikita Suyama (*Medical Institute of Bioregulation*)

1SFA-03 Waves of chromatin remodeling in mouse dendritic cells in response to LPS stimulation

Alexis Vandenbon (*IFReC, Osaka University*)

1SFA-04 出芽酵母 *yku70 esc1* 変異型における遺伝子発現の変化を引き起こすメカニズム

Mechanisms for the misregulated gene expression in the *yku70 esc1* mutant of budding yeast

○徳田 直子, 笹井 理生（名古屋大学）

Naoko Tokuda, Masaki Sasai (*Nagoya University*)

1SFA-05 刺激された血管内皮細胞における核内構造のダイナミクス
Dynamics of chromatin structure in stimulated vascular endothelial cells
○和田 洋一郎[1,2], 中田 庸一[3], 大田 佳宏[3], 井原 茂男[2,3]（[1]東京大学アイソトープ総合センター, [2]先端科学技術研究センター, [3]東京大学大学院数理科学研究科）
Youichiro Wada[1,2], Youichi Nakata[3], Yoshihiro Ohta[3], Sigeo Ihara[2,3] ([1]*Isotope Science Center, The University of Tokyo*, [2]*Research Center for Advanced Science and Technology, The University of Tokyo*, [3]*Graduate School of Mathematical Sciences, The University of Tokyo*)

総合討論, おわりに
白井 剛
Tsuyoshi Shirai

9:00〜11:30　G 会場（小会議室 405）／Room G (Conference Room 405)
1SGA　蛋白質工学を用いた会合と溶解性の最新の研究
Advances in the engineering of protein oligomerization and solubility

オーガナイザー：黒田 裕（東京農工大学），赤沼 哲史（早稲田大学）
Organizers: Yutaka Kuroda (Tokyo University of Agriculture and Technology), Satoshi Akanuma (Waseda University)

Aggregation, oligmorization, and solubility are important issues in protein research. However, much of the present research on these phenomena focuses on amyloidogenic or crystalline aggregation. This workshop will introduce recent studies on amorphous protein aggregation, protein solubility, as well as the control and design of protein oligomers. We hope that it will provide an opportunity to decipher biophysical mechanisms governing these phenomena, and that it will shed insight into mechanisms that are common to amyloidogenic/crystalline aggregation and those that are not.

1SGA-01 序論
Introduction
○赤沼 哲史（早大 人間）
Satoshi Akanuma (*Faculty of Hum. Sci., Waseda Univ.*)

1SGA-02 脂質膜のアミロイド線維形成への影響
The effects of lipid membranes on the fibrillation of amyloidogenic proteins
○寺川（鈴木） まゆ（ウィールコーネルメディスン）
Mayu S. Terakawa (*Weill Cornell Medicine, Biochemistry*)

1SGA-03 新規タンパク質分子間結合面の創成と人工タンパク質繊維の作成
De-novo design of a protein-protein interface and creation of protein fibrils
○八木 創太[1], 赤沼 哲史[2], 内田 達也[3], 山岸 明彦[1]（[1]東薬大 応用生命, [2]早大 人間, [3]東薬大 分子生命）
Sota Yagi[1], Satoshi Akanuma[2], Tatsuya Uchida[3], Akihiko Yamagishi[1] ([1]*Tokyo Univ. Pharm. Life Sci., Dep. Appl. Life Sci.*, [2]*Waseda Univ., Facul. Hum. Sci.*, [3]*Tokyo Univ. Pharm. Life Sci., Dep. Mol. Life Sci.*)

1SGA-04 人工タンパク質をブロックに見立てた超分子ナノ構造複合体の設計構築
Design and construction of supramolecular nanostructures by using *de novo* protein nanobuilding blocks
小林 直也, 木村 尚弥, ○新井 亮一（信州大・繊維・応用生物）
Naoya Kobayashi, Naoya Kimura, **Ryoichi Arai** (*Appl. Biol., Tex. Sci. & Tech., Shinshu Univ.*)

1SGA-05 時間分解小角 X 線小角散乱法を用いたフェリチンの会合機構の解析
Ferritin assembly mechanism studied by time-resolved small-angle X-ray scattering
○佐藤 大輔, 池口 雅道（創価大・理工）
Daisuke Sato, Masamichi Ikeguchi (*Fac. of Sci. and Eng., Soka Univ.*)

1SGA-06 ペプチド溶解性の全原子分子動力学シミュレーション及びその実験的検証
Large scale molecular dynamics of peptide solubility and its experimental assessment
○黒田 裕（東京農工大学工学部生命工学科）
Yutaka Kuroda (*Dept. Biotech. Life Sci., TUAT*)

1SGA-07 熱測定による高温で可逆的に形成される蛋白質の会合体の検出
High-temperature reversible oligomerization of proteins detected by calorimetry
○城所 俊一[1], 中村 成芳[1,2]（[1]長岡技科大・生物, [2]北九州高専・生産デザイン）
Shun-ichi Kidokoro[1], Shigeyoshi Nakamura[1,2] ([1]*Dept. Bioeng., Nagaoka Univ. Tech.*, [2]*Dept. Creat. Eeng., Natl. Inst. Tech. Kitakyushu College*)

1SGA-08 終わりに
Concluding Remarks
○有坂 文雄（日大生物資源科学）
Fumio Arisaka (*Nihon U. Biores. Sci.*)

16:30～19:00　A 会場（中ホール 200）／Room A (Convention Hall 200)
1SAP　細胞同士の絡み合いから理解する集団運動の生物物理学
Biophysics of collective cell movement - From single-cell to multi-cell dynamics

オーガナイザー：澤井 哲（東京大学），青木 一洋（自然科学研究機構 基礎生物学研究所）
Organizers: Satoshi Sawai (The University of Tokyo), Kazuhiro Aoki (National Institute for Basic Biology, National Institutes of Natural Sciences)

Collective cell movement forms the basis of morphogenesis, wound healing as well as cancer invasion. From what appears as random and variable traits that are specific to certain cell types and species, recent studies have uncovered some of the common elements that underlies the dynamics of cell shape, migration, cell-cell interactions and stemness. The symposium will focus on the dynamics that are highly coordinated between the cells and highlight the most recent and exciting progress by some of the younger scientists in this emerging field.

1SAP-01 Intercellular propagation of ERK activity orients collective cell migration
Kazuhiro Aoki (*OIIB, NIBB, Div. of Quantitative Biology*)

1SAP-02 外力が駆動する細胞集団運動を支えるアクチン細胞骨格制御の解明
Actin interacting protein 1 and cofilin sense the extrinsic stretching force and orient cell rearrangement in Drosophila wing
○杉村 薫[1,2], 井川 敬介[1]（[1]京都大学物質―細胞統合システム拠点, [2]JST・さきがけ）
Kaoru Sugimura[1,2], Keisuke Ikawa[1] ([1]*WPI-iCeMS, Kyoto Univ.*, [2]*JST PRESTO*)

1SAP-03 細胞外基質の粘弾性に応答する上皮細胞の集団運動と 3 次元形態形成
Collective Movement and 3D Morphogensis of Epithelial Cells Responding to Viscoelasticity of the Extracellular Matrix
○芳賀 永（北大・院・先端生命）
Hisashi Haga (*Faculty of Advanced Life Sci., Hokkaido Univ.*)

1SAP-04 マイクロ流体デバイスを用いた細胞性粘菌の集団的細胞運動の解析
Microfluidic analysis of group cell migration in *Dictyostelium*
藤森 大平[1], 中島 昭彦[2], 井元 大輔[1], 石原 秀至[4], ○澤井 哲[1,2,3]（[1]東京大学大学院総合文化研究科 広域科学専攻 相関基礎科学系, [2]東京大学大学院総合文化研究科 複雑系生命システム研究センター, [3]JST さきがけ, [4]明治大学 理工学部）
Taihei Fujimori[1], Akihiko Nakajima[2], Daisuke Imoto[1], Shuji Ishihara[4], **Satoshi Sawai**[1,2,3] ([1]*Dept. Basic Sci., Grad. School of Arts and Sci., Univ. of Tokyo*, [2]*Research Ctr. for Complex Systems Biology, Univ. of Tokyo*, [3]*JST PRESTO*, [4]*School of Sci. Eng., Meiji Univ.*)

1SAP-05 単一ヒト表皮幹細胞からの多層上皮構造の形成原理
A mechanistic principle of multilayered epithelial formation from single human epidermal stem cells
○難波 大輔（東京医科歯科大・難研・幹細胞医学）
Daisuke Nanba (*Tokyo Medical & Dental Univ., Med. Res. Inst., Dept. Stem Cell Biol.*)

1SAP-06 がん細胞の集団的浸潤：病理からの視点
Collective invasion of cancer cells: perspectives from pathology
○榎本 篤（名古屋大・院・医・腫瘍病理）
Atsushi Enomoto (*Dept. Pathol., Nagoya Univ. Grad. Sch. Med.*)

16:30～19:00　B 会場（中会議室 202）／Room B (Conference Room 202)
1SBP　モデル化と操作による高次生命現象の解明への挑戦
Modeling and Manupilation of Life: a Challenge to Unveil Its Complex Mechanism

オーガナイザー：茅 元司（東京大学），井上 尊生（ジョンズ・ホプキンス大学）
Organizers: Motoshi Kaya (The University of Tokyo), Takanari Inoue (Johns Hopkins University)

Measurements of molecular dynamics and decoding of genetic information have been progressively advanced and thus, provided a substantial amount of information in life science field. However, our biological system cannot be interpreted simply by superimposing individual functions revealed by these technologies. Rather, it is a complex system by cooperative interactions among cellular and molecular components. In this symposium, we focus on the constructive modeling approaches and experimental manipulations designed to unveil complex mechanisms of the biological events, such as cell division, blood vessel formation, cellular temperature control, immune reaction, and muscle contraction.

1SBP-01 骨格筋ミオシン間における力発生の同調現象を明らかにする
Molecular mechanism of synchronous force generations among skeletal myosins
○茅 元司（東京大学 大学院理学系研究科）
Motoshi Kaya (*University of Tokyo, Graduate School of Science*)

1SBP-02 体細胞分裂期における細胞質ダイニンの操作
Manipulation of cytoplasmic dynein during mitosis
○清光 智美（名古屋大学大学院理学研究科）
Tomomi Kiyomitsu (*Nagoya University*)

1SBP-03 Intracellular production of synthetic RNA granules by ligand-yielded multivalent enhancers
Takanari Inoue (*Johns Hopkins University*)

1SBP-04 単一細胞内局所加熱による細胞熱応答の原理の解明
The mechanisms of cellular response to temperature changes as revealed by local heating in single cells
○岡部 弘基[1,2], 時 ベイニ[1], 船津 高志[1]（[1]東京大学大学院薬学系研究科, [2]JST さきがけ）
Kohki Okabe[1,2], Beini Shi[1], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*PRESTO, JST*)

1SBP-05 血管のメカニカルストレスによるフィブロネクチンピラー形成
Vascular mechanical stress organizes Fibronectin into pillars bridging tissue gap
○佐藤 有紀[1,2]（[1]九州大学・医学研究院, [2]JST・さきがけ）
Yuki Sato[1,2] ([1]*Grad. Sch. Med. Sci., Kyushu Univ.*, [2]*JST, PRESTO*)

1SBP-06 チューブリンアイソタイプと微小管動態の多様性
Distinct contribution of different tubulin isotypes to microtubule dynamics
○杉本 亜砂子（東北大学・生命科学）
Asako Sugimoto (*Life Sciences, Tohoku Univ.*)

16:30～19:00　C 会場（中会議室 201）／Room C (Conference Room 201)
1SCP　［学会本部企画 I］日本－中国交流シンポジウム：蛋白質設計とバイオテクノロジーへの応用
[BSJ Special Event I] BSJ – BSC Joint Symposium: Protein Design and its Applications to Biotechnology

オーガナイザー：中村 春木（大阪大学），Xiyun Yan（Institute of Biophysics）
Organizers: Haruki Nakamura (Osaka University), Yan Xiyun (Institute of Biophysics)

In order to make much deeper collaborations between BSJ (Biophysical Society of Japan) and BSC (Biophysical society of China) for promotion of biophysics in a global manner, we start a Joint Bilateral Symposium inviting active researchers from both BSJ and BSC side. This year we focus on the theme “Protein Design and its Applications to Biotechnology”, and both societies invite three speakers, respectively. It is expected to provide a scope on the future biophysics studies in Japan and China.

1SCP-01 Computational design of catalytic triad based organophosphate capture proteins
Chu Wang (*Dept. Chem. Biol., CCME, Peking Univ.*)

1SCP-02 Chemical Probes with Fuorogenic Switches for Visualizing Modified Protein and DNA
Yuichiro Hori[1,2] ([1]*Grad. Sch. Eng., Osaka Univ.*, [2]*IFReC, Osaka Univ.*)

1SCP-03 Self-assembly of protein nanofibrils that display active enzymes
Sarah Perrett (*Inst. Biophys., CAS*)

1SCP-04 アミロイド線維形成初期過程のタンパク質構造化メカニズムの解析
Investigating early steps in amyloid fibril formation
○茶谷 絵理（神戸大院理）
Eri Chatani (*Grad. Sch. Sci., Kobe Univ.*)

1SCP-05 Nanozyme: discovery and its application in tumor diagnosis
Xiyun Yan (*Inst. Biophys., CAS*)

1SCP-06 蛋白質相互作用の熱力学：分子設計と創薬
Thermodynamics of protein interaction for molecular design and therapeutics
○津本 浩平（東京大学）
Kouhei Tsumoto (*The University of Tokyo*)

16:30～19:10　D 会場（中ホール 300）／Room D (Convention Hall 300)
1SDP　新学術領域研究「ゆらぎと構造の協奏：非平衡系における普遍法則の確立」共催
モーターと細胞骨格の新展開　ステップから集団運動まで
New extremes of motor proteins and cytoskeleton: step into a new realm with steps and collective motions

オーガナイザー：西坂 崇之（学習院大学），永井 健（北陸先端科学技術大学院大学）
Organizers: Takayuki Nishizaka (Gakushuin University), Ken H. Nagai (JAIST)

Novel two directions of motor proteins and cytoskeletons will be presented in this session. One is the collective motions of condensed or regulated cytoskeletons in vivo or in vitro, both of which are characterized by biophysics, non-equilibrium physics and developmental biology. The other extreme is the exploration of the molecular mechanism of new machineries including rotary motors. Also, this symposium briefly represents Dr. Kazuhiko Kinosita Jr's fruitful contribution in this field as its introduction, who passed away last November.

オープニング

1SDP-01 Single molecule analysis of F_oF_1-ATP synthase
Rikiya Watanabe[1,2] ([1]*Department of Applied Chemistry, The University of Tokyo*, [2]*PRESTO, JST*)

1SDP-02 *De novo* 設計軸の回転から明らかになったトルク発生機構
Rotation of *de novo* designed axis and the torque generation mechanism
○岸川 淳一, 馬場 みほ里, 中西 温子, 横山 謙（京産大・総合生命・生命シス）
Jun-ichi Kishikawa, Mihori Baba, Atsuko Nakanishi, Ken Yokoyama (*Dept. LifeSci, Kyoto Sangyo Univ.*)

1SDP-03 滑走バクテリアと遊泳アーキアの運動超分子マシナリーの単位ステップ観察
Unitary steps of supramolecular-motility machineries in gliding bacteria and swimming archaea
○木下 佳昭[1], 中根 大介[1], 内田 就也[2], 宮田 真人[3], 西坂 崇之[1]（[1]学習院大学 理・物理, [2]東北大学 理・物理, [3]大阪市立大学 院理・細胞機能）
Yoshiaki Kinosita[1], Daisuke Nakane[1], Nariya Uchida[2], Makoto Miyata[3], Takayuki Nishizaka[1] ([1]*Dept. Phys., Gakushuin University*, [2]*Dept. Phys., Tohoku University*, [3]*Dept. Biol., Graduate School of Science, Osaka City University*)

1SDP-04 A small stroke for an individual, but giant motion for a population: negative gravitaxis and bioconvection of Chlamydomonas reinhardtii
Azusa Kage (*Dept. Finemechanics, Tohoku Univ.*)

1SDP-05 インビトロ運動アッセイ中の自走する微小管の集団運動
Collective motion of running microtubules in in vitro motility assay
○永井 健（北陸先端大・先端理工）
Ken Nagai (*Sch. Mater. Sci., JAIST*)

1SDP-06 胚発生過程における細胞集団運動を担うアクトミオシンの制御機構
Local regulation of actomyosin for the globally orchestrated collective cell movement during tissue morphogenesis
○進藤 麻子[1], Wallingford John[2], 木下 専[1]（[1]名大・院・ 生命理学, [2]テキサス大）
Asako Shindo[1], John Wallingford[2], Makoto Kinoshita[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*UT Austin*)

1SDP-07 Shape Remodeling of Active Cytoskeletal Vesicles
Andreas Bausch (*Lehrstuhl für Biophysik, TU München*)

16:30～19:00　E 会場（小会議室 303）／Room E (Conference Room 303)
1SEP　新学術領域研究「3D 活性サイト科学」共催
時空間精密構造解析による生体分子活性サイトの機能解明
Understanding biochemical functions of the active sites in biomolecular systems by spatial–temporal analysis

オーガナイザー：鷹野 優（広島市立大学），久保 稔（理化学研究所）
Organizers: Yu Takano (Hiroshima City University), Minoru Kubo (RIKEN)

Biomolecules have a rich diversity of functional dynamics, from a large domain movement to a small local structural change. The latter dynamics includes a sub-angstrom change in the active site, which is crucial to control its electronic state and reactivity. Recent advances in crystallography, single-molecule imaging, spectroscopy, and computer simulation allow us to analyze the high-resolution structures, chemical properties, and complex dynamics of biomolecules, and to better understand the coupling between macroscopic and microscopic events. We discuss how these methods can describe the biochemical functions of the active sites.

1SEP-01 Elucidation of structure-function relationship of biological active sites by molecular simulation
Yu Takano[1,2], Yusuke Kanematsu[1], Yasuhiro Imada[2] ([1]*Grad. Sch. Info. Sci., Hiroshima City Univ.*, [2]*IPR, Osaka Univ.*)

1SEP-02 Structural analysis of photosystem II to reveal the mechanism of light-induced water-splitting
Fusamichi Akita[1], Michihiro Suga[1], Keitaro Yamashita[2], Go Ueno[2], Hironori Murakami[2], Yoshiki Nakajima[1], Yasufumi Umena[1], Kunio Hirata[2], Minoru Kubo[2], Kazuya Hasegawa[2], Masaki Yamamoto[2], Hideo Ago[2], Jian-Ren Shen[1] ([1]*RIIS, Okayama Univ.*, [2]*Riken Harima*)

1SEP-03 X 線 1 分子追跡法によるマルチマータンパク質・機能的運動の可視化
Active 3D Motion Visualization of Multimeric Proteins by X-ray Single Molecule Tracking
○関口 博史（高輝度光科学研究センター）
Hiroshi Sekiguchi (*JASRI/SPring-8*)

1SEP-04 蛍光 X 線ホログラフィーによるヘモグロビンの金属周辺構造の可視化
Visualization by X-ray fluorescence holography of metal environments in hemoglobin
○佐藤 文菜[1], 柴山 修哉[1], 八方 直久[2], 林 好一[3], 佐々木 裕次[4]（[1]自治医大, [2]広島市大, [3]名工大, [4]東大）
Ayana Sato-Tomita[1], Naoya Shibayama[1], Naohisa Happo[2], Kouichi Hayashi[3], Yuji C. Sasaki[4] ([1]*Jichi Med. Univ.*, [2]*Hiroshima City Univ.*, [3]*Nagoya Inst. Tech.*, [4]*Tokyo Univ.*)

1SEP-05 マイクロ流路デバイスを用いた時間分解分光法による膜タンパク質の活性サイトの中間体構造解析
Intermediate structures of the active site in membrane proteins revealed by time-resolved spectroscopy with micro-channel devices
○木村 哲就（神戸大・院理）
Tetsunari Kimura (*Grad. Sch. Sci., Kobe Univ.*)

1SEP-06 チトクロム *c* 酸化酵素の時間分解 XFEL 結晶構造解析：機能部位間の相互作用ダイナミクスの観測
Time-resolved XFEL crystallography of cytochrome *c* oxidase: Probing the interaction dynamics between two functional sites
○久保 稔（理研・播磨）
Minoru Kubo (*RIKEN SPring-8 Center*)

16:30～19:30　F 会場（中会議室 406）／Room F (Conference Room 406)

1SFP　細胞膜ナノ・メゾドメイン構造によるシグナル伝達の動的な制御機構

Unraveling the regulation mechanisms of signal transduction in nano- and meso-scale domains in cell membranes

オーガナイザー：森垣 憲一（神戸大学），鈴木 健一（京都大学）
Organizers: Kenichi Morigaki (Kobe University), Kenichi Suzuki (Kyoto University)

Membrane domains play critical roles in the cellular signal transduction. Recent studies on receptor oligomerization and lipid rafts have suggested that dynamic aggregation of molecules in nano- and mesoscopic domains are regulating the signal transduction cascade. However, the regulation mechanisms remain elusive. The present symposium intends to give an overview of the current understanding by providing the most up-to-date views from recent studies using cellular membranes and model systems to gain insight for the future directions.

オープニング
鈴木 健一
Kenichi Suzuki

1SFP-01　生細胞膜上で形成される G タンパク質共役型受容体の動的ダイマー：１分子観察法を用いたアプローチ
Dynamic dimer formation of G-protein coupled receptor in the live plasma membrane: An approach by using single molecule observation
○笠井 倫志[1], 楠見 明弘[1,2]（[1]京大再生研, [2]沖縄科技大院）
Rinshi Kasai[1], Akihiro Kusumi[1,2] ([1]*Inst. Front. Med. Sci., Kyoto Univ.*, [2]*Membrane Cooperativity Unit, OIST*)

1SFP-02　細胞膜の分子組織構造・反応カップリング
Coupling of reactions and molecular organizations in plasma membranes
○貝塚 芳久（物質・材料研究機構）
Yoshihisa Kaizuka (*NIMS*)

1SFP-03　Cytokine receptor dimerization: molecular determinants and cellular regulation
Jacob Piehler (*University of Osnabrueck*)

1SFP-04　マイクロクラスターは T 細胞受容体のエンドサイトーシスのシグナルユニットとして機能する
Microclusters as a signaling unit for T cell receptor endocytosis
○横須賀 忠（東京医大・免疫学）
Tadashi Yokosuka (*Dept. Immunol., Tokyo Medical Univ.*)

1SFP-05　BAR タンパク質による細胞膜の形態形成とファゴサイトーシスの関連
Plasma membrane morphogenesis by the BAR domain superfamily proteins for phagocytic cup formation
○末次 志郎（奈良先端科学技術大学院大学）
Shiro Suetsugu (*NAIST*)

1SFP-06　視細胞円板膜上のロドプシン多量体クラスターがつくる一過的メゾ領域
Transient meso-domains formed by oligomeric clusters of rhodopsin in retinal disk membrane
○林 文夫[1], 齋藤 夏美[1], 谷本 泰士[2], 森垣 憲一[2,3], 妹尾 圭司[4]（[1]神戸大·院理, [2]神戸大・院農, [3]神戸大・バイオ, [4]浜松医大）
Fumio Hayashi[1], Natsumi Saito[1], Yasushi Tanimoto[2], Kenich Morigaki[2,3], Keiji Seno[4] ([1]*Grad. Sch. Sci. Kobe Univ.*, [2]*Grad. Sch. Agri. Kobe Univ.*, [3]*Biosig. Res. Cent. Kobe Univ.*, [4]*Hamamatsu Univ. Med.*)

クロージング
森垣 憲一
Kenichi Morigaki

16:30～19:00　G 会場（小会議室 405）／Room G (Conference Room 405)
1SGP　「複雑生命システム動態研究教育拠点」共催
可塑性とロバストネスの動的状態論
Dynamic state theory for plasticity and robustness of biological systems

オーガナイザー：金子 邦彦（東京大学），古澤 力（理化学研究所）
Organizers: Kunihiko Kaneko (The University of Tokyo), Chikara Furusawa (RIKEN)

Biological systems exhibit robustness to various perturbations, including expression noise and environmental/genetic changes, while they are plastic to the surrounding environment, changing their state through processes like adaptation, evolution, and cell differentiation. Although the coexistence of robustness and plasticity can be understood as a dynamic property of biological systems, the mechanisms responsible for it are largely unknown. In this symposium, we will discuss how we can understand robustness and plasticity of biological systems based on both experimental and theoretical analysis.

1SGP-01　マイクロチャンバーと融合した大腸菌の生存
E. coli survival in inorganic chamber
○田端 和仁[1,2,3], 森泉 芳樹[1], 渡邊 力也[1], 芦川 裕樹[1], 野地 博行[1,3]（[1]東大院・工, [2]さきがけ・JST, [3]ImPACT・内閣府）
Kazuhito Tabata[1,2,3], Yoshiki Moriizumi[1], Rikiya Watanabe[1], Hiroki Ashikawa[1], Hiroyuki Noji[1,3] ([1]*Grad. sch. eng., Univ. of Tokyo*, [2]*PREST JST*, [3]*ImPACT Cabinet Office*)

1SGP-02　1 細胞レベルでの薬剤耐性獲得プロセス
Acquisition of drug resistance at the single-cell level
○若本 祐一（東大 院総合文化）
Yuichi Wakamoto (*Univ. of Tokyo*)

1SGP-03　Plasticity of developmental process that determines floral organ number
Miho Kitazawa[1,2], Koichi Fujimoto[2] ([1]*CELAS, Osaka Univ.*, [2]*Dept. Biol. Sci, Osaka Univ.*)

1SGP-04　生物システムの可塑性の理解に向けて：理論解析と実験進化
Toward Understanding of Biological Plasticity: Computational and Experimental analysis
○古澤 力[1,2]（[1]理研・生命システム, [2]東大・院理学）
Chikara Furusawa[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sci., Univ. Tokyo*)

1SGP-05　表現型適応と進化のマクロ現象論：揺動応答関係、遺伝的同化、スローマニフォルド仮説
Macroscopic Theory of Phenotypic Adaptation and Evolution: Fluctuation-response, Genetic Assimilation, and Slow-Manifold Hypothesis
○金子 邦彦（東京大学）
Kunihiko Kaneko (*University of Tokyo*)

第 2 日目（11 月 26 日（土））／Day 2（Nov. 26 Sat.）

9:00～11:30　A 会場（中ホール 200）／Room A (Convention Hall 200)
2SAA　光遺伝学で活躍するタンパク質分子の生物物理学研究の展望
Perspective in biophysical studies on protein molecules applicable for optogenetics

オーガナイザー：古谷 祐詞（自然科学研究機構 分子科学研究所），須藤 雄気（岡山大学）
Organizers: Yuji Furutani (Institute for Molecular Science, National Institutes of Natural Sciences), Yuki Sudo (Okayama University)

Optogenetics, a technology for controlling cellular activity by light, has rapidly expanded over the past decade, paving the way for experiments that would have once seemed impossible. Prior to this new trend, light-receptive proteins utilized for optogenetics have been extensively investigated in a variety of research fields, leading to the elucidation of the molecular mechanisms of them, which enabled us rational designs of optogenetics tools. This symposium focuses on recent advances of light-receptive proteins and their applications for optogenetics. New directions of the optogenetics in biophysics will be discussed.

2SAA-01 光生物分野における新区分の立ち上げ
Launching a new category in photobiology
○古谷 祐詞[1], 須藤 雄気[2]（[1]自然科学研究機構・分子研, [2]岡山大・院医歯薬）
Yuji Furutani[1], Yuki Sudo[2] ([1]*Inst. Mol. Sci, Nat. Inst. Nat. Sci.*, [2]*Grad. Sch. Med. Dent. Pharm. Sci., Okayama Univ.*)

2SAA-02 オプトジェネティクス革命
Optogenetic revolution
○八尾 寛[1,2]（[1]東北大学大学院生命科学研究科, [2]東北大学医学系研究科脳コアセンター）
Hiromu Yawo[1,2] ([1]*Tohoku University Graduate School of Life Sciences*, [2]*Center for Neuroscience, Tohoku University Graduate School of Medicine*)

2SAA-03 新規オプトジェネティクスツール探索：天然および人工の微生物型ロドプシン
Exploration of new optogenetic tools: natural and artificial microbial rhodopsins
○井上 圭一[1,2]（[1]名工大・院工, [2]JST・さきがけ）
Keiichi Inoue[1,2] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*JST PRESTO*)

2SAA-04 レチナールタンパク質を基盤とした光遺伝学ツールの開発に向けて
Towards production of retinal protein-based optogenetic tools
○須藤 雄気（岡山大学大学院医歯薬学総合研究科（薬学系））
Yuki Sudo (*Div. Pharm. Sci., Okayama Univ.*)

2SAA-05 Genetic, biochemical and biophysical studies on flavoprotein photoreceptors applicable for optogenetics
Shinji Masuda (*Center for Biological Resources & Informatics, Tokyo Institute of Technology*)

2SAA-06 Optogenetic potentials of bistable animal opsin-based pigments for regulating GPCR signalings
Mitsumasa Koyanagi[1,2,3] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*OCARINA, Osaka City Univ.*, [3]*JST PRESTO*)

2SAA-07 生体光操作技術の進展
Technological advances for optical control of living organisms
○七田 芳則（京大・院理・生物物理）
Yoshinori Shichida (*Dept. of Biophys., Grad. School of Sci., Kyoto Univ.*)

9:00～11:30　B 会場（中会議室 202）／Room B (Conference Room 202)
2SBA　構成的生物学の手法による生体分子，分子複合体，分子ネットワークの理解
Synthetic biology approaches to understand biological molecules, complexes, and networks

オーガナイザー：古田 健也（情報通信研究機構 未来 ICT 研究所），多田隈 尚史（京都大学）
Organizers: Ken’ya Furuta (Advanced ICT Research Institute, NICT), Hisashi Tadakuma (Kyoto University)

Synthetic biology approach has opened the new era of biology and biophysics. In this symposium, to unveil the secret of life phenomena, we focus on the de novo design of artificial molecules, complexes and networks: from redesign of enzymes to reconstitution of intracellular transport systems.

Introduction
古田 健也
Ken’ya Furuta

2SBA-01 Design of Nucleotide Binding Site Toward Controlling and Understanding Molecular Motor
Takahiro Kosugi (*Institute for Molecular Science*)

2SBA-02 タンパク質分子ブロックを用いた分子モーターのエンジニアリング
Engineering approaches to molecular motors based on protein building blocks
○古田 健也（(国研）NICT）
Ken’ya Furuta (*NICT*)

2SBA-03 Beyond DNA and RNA: synthetic genetic polymers
Alexander I. Taylor, Philipp Holliger (*MRC Laboratory of Molecular Biology*)

2SBA-04 Construction of DNA origami base gene transcription nano chip
Hisashi Tadakuma (*Kyoto Univ, iCeMS*)

2SBA-05 アクトミオシン細胞骨格の in vitro 再構成
In vitro reconstitution of contractile actomyosin cytoskeleton
○宮崎 牧人[1,2], 石渡 信一[1]（[1]早大・物理, [2]早大・WABIOS）
Makito Miyazaki[1,2], Shin'ichi Ishiwata[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*WABIOS, Waseda Univ.*)

2SBA-06 Biomolecular Motors: From Cellular Function to Nanotechnological Applications
Stefan Diez (*B CUBE, TU Dresden, Germany*)

9:00～11:30 C 会場（中会議室 201）／Room C (Conference Room 201)
2SCA ［学会本部企画 II］日本ー韓国交流シンポジウム：1 分子生物物理学の最前線
[BSJ Special Event II] Korea-Japan Joint Symposium: Frontiers of Single Molecule Biophysics

オーガナイザー：尹 兌榮（延世大學），榎 佐和子（東京大学）
Organizers: Tae-Young Yoon (Yonsei University), Sawako Enoki (The University of Tokyo)

Single molecule imaging and manipulation techniques are powerful tools to explore many biological phenomena. They are used to reveal the biological function, mechanics, intermolecular interactions, and dynamics of proteins and nucleic acids at single molecule level. Recently, the field of single molecule biophysics has heralded spectacular technical breakthroughs such as improvement of both spatial and temporal resolution, and development of optics for investigating complicated biological processes in living cells. This symposium provides a forum for world leading Korean and Japanese scientists to share recent advances in field of single molecule biophysics, and discuss future applications in both academic and medical settings.

opening remarks
尹 兌榮
Tae-Young Yoon

2SCA-01 ZMW 法による生命現象の可視化の展開
Expansion of biological applications using Zero-Mode Waveguides
○上村 想太郎（東京大学大学院理学系研究科生物科学専攻）
Sotaro Uemura (*Dept. of Biol. Sci., Grad. Sch. of Sci., The Univ. of Tokyo*)

2SCA-02 Observation of single membrane proteins under mechanical tension
Tae-Young Yoon (*Yonsei University*)

2SCA-03 High-speed angle-resolved imaging of catalytic subunit of F1-ATPase
Sawako Enoki[1], Ryota Iino[2], Yoshihiro Minagawa[1], Yamato Niitani[3], Michio Tomishige[3], Hiroyuki Noji[1] ([1]*Dept. Appl. Chem, Grad. Sch. Eng. Univ. of Tokyo*, [2]*Okazaki Inst. Integ. BioSci., NINS*, [3]*Dept. Appl. Phys, Grad. Sch. Eng. Univ. of Tokyo*)

2SCA-04 Stochastic Regulation of DNA Mismatch Repair
Jong-Bong Lee (*Dept. of Physics, POSTECH*)

2SCA-05 細胞内一分子計測で探るキネシンの制御機構
Dissecting kinesin regulation through single molecule in cellulo measurements
○岡田 康志[1,2]（[1]理研 生命システム研究センター, [2]東大・理・物理）
Yasushi Okada[1,2] ([1]*QBiC, RIKEN*, [2]*Dept. Phys., Grad. Sch. Sci., Univ. Tokyo*)

2SCA-06 Propagation of gene expression noise by RNA polymerase in living cells
Nam Ki Lee (*Dept. of Physics, POSTECH*)

closing remarks
榎 佐和子
Sawako Enoki

9:00～11:30　D 会場（中ホール 300）／Room D (Convention Hall 300)
2SDA　新学術領域研究「温度を基軸とした生命現象の統合的理解」共催
温度生物学の挑戦
The Developing Field of Thermal Biology

オーガナイザー：岡部 弘基（東京大学），原田 慶恵（大阪大学）
Organizers: Kohki Okabe (The University of Tokyo), Yoshie Harada (Osaka University)

Temperature, a key regulator of biochemical reactions, influences important physiological functions. Recently intracellular thermometry has revealed that there are significant temperature changes at the single cell level related directly to cellular events, which encouraged a novel field of biology focused solely on temperature, thermal biology, to emerge. This symposium will provide an overview of the latest developments in the field of thermal biology, revealing the relationship between temperature and life activities, and will explore how this fundamental physical parameter contributes to all molecular-based biologies.

はじめに
岡部 弘基
Kohki Okabe

2SDA-01　細胞生物学のためのオンチップ高感度熱量センサ
On-chip high sensitive thermal sensors for cell biology
○小野 崇人, 猪股 直生（東北大学）
Takahito Ono, Naoki Inomata (*Tohoku University*)

2SDA-02　蛍光センサーを利用した一細胞温度計測からわかること
What we see in single-cell thermometry by using fluorescent sensors
○鈴木 団[1,2]（[1]早大・WABIOS, [2]JSTさきがけ）
Madoka Suzuki[1,2] ([1]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*, [2]*JST, PRESTO*)

2SDA-03　様々な生物種の温度測定に利用でき且つ速い温度変化を測定可能な蛍光性温度プローブタンパク質
Genetically encoded ratiometric fluorescent thermometer with wide temperature range and rapid response
○中野 雅裕[1], 新井 由之[1], 小寺 一平[2], 岡部 弘基[3,4], 亀井 保博[5], 永井 健治[1]（[1]阪大・産研, [2]北大・電子研, [3]東大院・薬学系研究科, [4]JST, さきがけ, [5]基生研）
Masahiro Nakano[1], Yoshiyuki Arai[1], Ippei Kotera[2], Kohki Okabe[3,4], Yasuhiro Kamei[5], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ.*, [2]*RIES, Hokkaido Univ.*, [3]*Grad. Sch. Pharma., Univ. Tokyo*, [4]*JST, PRESTO*, [5]*NIBB*)

2SDA-04　機能性磁性ナノ粒子を用いたガン温熱療法
Hyperthermia using functional magnetite nanoparticles
○井藤 彰（九大・工・化工）
Akira Ito (*Dept. of Chem. Eng., Fac. of Eng., Kyushu Univ.*)

2SDA-05　人工再構成系を用いた温度感受性 TRP チャネルの機能解析
Single channel analysis of the thermosensitive TRP channels in bilayer lipid membrane
○内田 邦敏[1,2,3], Zakharian Eleonora[2], 富永 真琴[3], 山崎 純[1]（[1]福岡歯科大学, [2]イリノイ大学医学部, [3]生理学研究所）
Kunitoshi Uchida[1,2,3], Eleonora Zakharian[2], Makoto Tominaga[3], Jun Yamazaki[1] ([1]*Fukuoka Dent. Coll.*, [2]*Univ. of Illinois Coll. of Med.*, [3]*NIPS*)

2SDA-06　外温性および内温性動物の脳の発生と進化
Brain development and evolution of ectothermal and endothermal animals
○野村 真（京都府立医科大学生物学教室）
Tadashi Nomura (*Dept. Biol. Kyoto Pref. Univ. Med.*)

9:00～11:30　E 会場（小会議室 303）／Room E (Conference Room 303)
2SEA　生命現象の理解を目指した立体構造インフォマティクスデータの活用
Applications of protein structure data for understanding biological phenomenon

オーガナイザー：内古閑 伸之（中央大学），根本 航（東京電機大学）
Organizers: Nobuyuki Uchikoga (Chuo University), Wataru Nemoto (Tokyo Denki University)

Recently, huge amounts of various biological data are generated by various new technologies and available to biological researches for obtaining new biophysical views. For more understanding biology with increasing biological data, it is necessary to develop bioinformatic methods.
In this symposium, we introduce biological and bioinformatic studies mainly with protein structures, which can be some clue for deep understanding of biology.

2SEA-01　剛体ドッキングによるタンパク質間相互作用表面のプロファイル解析
Profile analysis of protein interaction surface with rigid-body docking decoys
○内古閑 伸之（中央大学 理工学部 物理学科）
Nobuyuki Uchikoga (*Dept. of Physics, Chuo Univ.*)

2SEA-02　マウスはやはりヒト炎症性疾患のモデルになる ーバイオインフォマティクス的手法によるマウスモデルの再評価ー
Genomic responses in mouse models greatly mimic human inflammatory diseases
○高雄 啓三[1,2,3]（[1]富山大・生命科学先端研究支援ユニット, [2]富山大院・医学薬学, [3]生理学研究所）
Keizo Takao[1,2,3] ([1]*Life Sci. Res. Ctr., Univ. Toyama*, [2]*Grad. Sch. Med. Pharm., Univ. Toyama*, [3]*NIPS*)

2SEA-03　Development of an Efficient Amino Acid Substitution Matrix: MIQS
Kentaro Tomii[1], Kazunori Yamada[1, 2] ([1]*AIST*, [2]*Tohoku University*)

2SEA-04　ドッキングモデル構造群を用いたタンパク質間相互作用予測
Rigid docking based protein-protein interaction prediction by using high scoring docking models
○松崎 由理（東工大・情生院）
Yuri Matsuzaki (*ACLS, Tokyo Tech.*)

2SEA-05　An index to collect homologous sequences with the same or similar biochemical functions
Wataru Nemoto[1], Shoichiro Kato[1], Hiroyuki Toh[2] ([1]*Div. of Life Sci. & Eng., Sch of Sci & Eng., Tokyo Denki Univ.*, [2]*Dep. of Biomed. Chem., Sch. of Sci. & Tec., Kwansei Gakuin Univ.*)

9:00～11:30　F 会場（中会議室 406）／Room F (Conference Room 406)
2SFA　免疫学と生物物理の接点
Physical Immunology

オーガナイザー：小林 徹也（東京大学），秋山 泰身（東京大学）
Organizers: Tetsuya J. Kobayashi (The University of Tokyo), Taishin Akiyama (The University of Tokyo)

Adaptive immunity is a highly evolved adaptive system in which fundamental biophysical processes such as molecular recognitions, chemotaxis, and collective responses play the crucial roles. Immunological system is, therefore, a good target to address the question how a complex adaptive system emerges out of the combinations of basic biophysical processes. In this symposium, we clarify the physical aspects of immunology, and discuss the potential contributions of biophysics and quantitative biology to the problems in immunology.

2SFA-01　Physical & quantitative aspects of immunology
Tetsuya J. Kobayashi[1,2], Taishin Akiyama[3] ([1]*IIS, Univ. Tokyo*, [2]*JST PRESTO*, [3]*Institute of Medical Science, The University of Tokyo*)

2SFA-02　Ｔ細胞活性化の一細胞分子イメージング
Single cell molecular imaging for T cell activation
○斉藤 隆[1,2]（[1]理研・IMS, [2]阪大 IFReC）
Takashi Saito[1,2] ([1]*RIKEN-IMS*, [2]*IFReC Osaka Univ.*)

2SFA-03 Application of stochastic models in quantitative immunology

Shunsuke Teraguchi, Yutaro Kumagai (*IFReC, Osaka Univ.*)

2SFA-04 動的な誘引場に対する免疫細胞の走化性に見られる共通性と特異性

Generality and specificity in chemotaxis response of immune cells in dynamic gradients of chemoattractant

○中島 昭彦[1], 石田 元彦[2], 澤井 哲[1,2]（[1]東大・院総文・複雑生命, [2]東大・院総文・広域科学）

Akihiko Nakajima[1], Motohiko Ishida[2], Satoshi Sawai[1,2] ([1]*Res. Cent. Comp. Sys. Biol., Grad. Sch. Arts Sci., Univ. Tokyo*, [2]*Dept. Basic Sci., Grad. Sch. Arts Sci., Univ. Tokyo*)

2SFA-05 適応免疫応答を調節するリンパ節内の細胞ダイナミクス

Cellular dynamics shaping adaptive immune responses in the lymph node

○岡田 峰陽[1,2,3]（[1]理化学研究所 統合生命医科学研究センター, [2]科学技術振興機構さきがけ, [3]横浜市立大学大学院生命医科学研究科）

Takaharu Okada[1,2,3] ([1]*RIKEN Center for Integrative Medical Sciences*, [2]*PRESTO, Japan Science and Technology Agency*, [3]*Graduate School of Medical Life Science, Yokohama City Univ.*)

2SFA-06 Quantitative analysis of T cell repertoire and homeostasis

Taishin Akiyama[1], Tetsuya J. Kobayashi[2] ([1]*Institute of Medical Science, The University of Tokyo*, [2]*Institute of Industrial Science, The University of Tokyo*)

9:00～11:30　G 会場（小会議室 405）／Room G (Conference Room 405)

2SGA　電子顕微鏡が捉える生物アーキテクチャの解明ー高分解能化と多様な情報の融合ー

Biological architecture elucidated by electron micrroscopy - Integration of highly-resolved strucutre and other various information -

オーガナイザー：安永 卓生（九州工業大学），岩崎 憲治（大阪大学）

Organizers: Takuo Yasunaga (Kyushu Institute of Technology), Kenji Iwasaki (Osaka University)

Recent progress of electron microscopy (EM) provides us a new era when we observe protein structure at a near atomic resolution and protein architecture in situ, such as in lipid bilayers, cellar organelles, cells, tissues and so on. Also, other imaging techniques as light microscopy and atomic force microscopy can be integrated with EM to elucidate organic architecture under physiological conditions. Here we introduce cutting-edge observations and discuss further potentials of EM.

2SGA-01 脂質二分子膜を隔てた情報変換をとらえるクライオ電子顕微鏡単粒子解析法

Single particle cryoEM to elucidate signal transduction through lipiid bilayer membrane

○重松 秀樹[1,2,3]（[1]理研CLST, [2]横浜市大院・生命医科学, [3]エール大・医）

Hideki Shigematsu[1,2,3] ([1]*RIKEN CLST*, [2]*Med. Life Sci., Yokohama City University*, [3]*Yale Univ. Sch. Med.*)

2SGA-02 極低温電子顕微鏡構造に基づいた胃プロトンポンプ―胃酸抑制剤結合モデル

Binding model of the acid suppressant to the gastric proton pump based on cryo-EM structure

○阿部 一啓[1,2]（[1]名大・細胞生理, [2]名大院・創薬）

Kazuhiro Abe[1,2] ([1]*Cellular and Structural Physiology Institute, Nagoya Univ.*, [2]*Grad. Sch. Pharm.*)

2SGA-03 電子顕微鏡を用いた繊毛の三次元構造解析

Three-dimensional electron microscopy of cilia

○小田 賢幸（山梨大・院医）

Toshiyuki Oda (*Grad. Sch. Med., Univ. Yamanashi*)

2SGA-04 CryoTEM のための CryoCLEM システムの最新アプリケーション

Latest application of CryoCLEM system for cryo-TEM

○石原 あゆみ[1], 荒牧 信二[2], 肥後 智也[2], 安永 卓生[2]（[1]ライカマイクロシステムズ株式会社, [2]九工大・院情報工・生命情報工）

Ayumi Ishihara[1], Shinji Aramaki[2], Tomoya Higo[2], Takuo Yasunaga[2] ([1]*Leica Microsystems K.K.*, [2]*Grad. Sch. Computer Sci. & Systems Eng., Kyushu Inst. of Tech.*)

2SGA-05 Correlative Atomic Force and Transmission Electron Microscopy

Katsuya Shimabukuro[1], Yutaro Yamada[1,2] ([1]*NIT, Ube College*, [2]*Dep. of Bio. Kanazawa Univ.*)

2SGA-06 Cryo-electron microscopy single particle analysis at near atomic resolution
Naoyuki Miyazaki, Kenji Iwasaki (*IPR, Osaka Univ.*)

16:15～18:45　A 会場（中ホール 200）／Room A (Convention Hall 200)
2SAP　新学術領域研究「共鳴誘導で革新するバイオイメージング」共催
生体分子ー電磁波間の共鳴を活用する最先端バイオイメージング
Advanced bioimaging utilizing resonance between electromagnetic waves and molecules for life

オーガナイザー：宮脇 敦史（理化学研究所），根本 知己（北海道大学）
Organizers: Atsushi Miyawaki (RIKEN), Tomomi Nemoto (Hokkaido University)

For the elucidation of biological emergent functions, multidimensional information is required to be investigated at each level of molecule, cell or organ by using optical imaging or optical manipulations. Recently, several epoch-making methodologies for such visualizations and manipulations have been proposed based on advanced light and laser technologies. Here, we serve an opportunity for "resonant" interactions among researchers controlling electromagnetic waves and ones controlling molecules, hoping that it will produce dramatic breakthroughs and broad-ranging discussions on their potentials for life sciences.

2SAP-01 Cruising inside cells
Atsushi Miyawaki[1,2] ([1]*RIKEN BSI*, [2]*RIKEN RAP*)

2SAP-02 NIR II/III (OTN-NIR)におけるバイオイメージング ー透明性を求めてー
Bioimaging in NIR II/III (OTN-NIR) seeking for transparency
○曽我 公平[1,2], 上村 真生[1,2]（[1]東理大・基礎工・材料工, [2]東理大・総研院・IFC）
Kohei Soga[1,2], Masao Kamimura[1,2] ([1]*Dept. Mater. Sci. & Tech., Tokyo Univ. of Sci.*, [2]*IFC, Tokyo Univ. of Sci.*)

2SAP-03 半導体レーザー高機能パルス光源による多光子イメージング
Advanced semiconductor-laser optical pulse sources for multiphoton microscopy
○横山 弘之（東北大学未来科学技術共同研究センター）
Hiroyuki Yokoyama (*New Industry Creation Hatchery Center (NICHe), Univ. Tohoku*)

2SAP-04 ベクトルビームを用いた共焦点顕微鏡法における分解能向上
Resolution enhancement in confocal microscopy with vector beams
○佐藤 俊一, 小澤 祐市（東北大・多元研）
Shunichi Sato, Yuichi Kozawa (*IMRAM, Tohoku Univ.*)

2SAP-05 白色レーザーによるコヒーレント非線形光学イメージング
Coherent nonlinear optical imaging using a white-ligtht laser source
○加納 英明（筑波大学・数理物質）
Hideaki Kano (*Inst. of Applied Physics, Univ. of Tsukuba*)

2SAP-06 光シート顕微鏡の改良と発生生物学への応用
Light-sheet microscopy: technical development and application for developmental biology
○野中 茂紀（基生研）
Shigenori Nonaka (*National Inst. for Basic Biol.*)

16:15～19:05　B 会場（中会議室 202）／Room B (Conference Room 202)
2SBP　ラマン散乱で探る bio. phys. chem. 三重点
Bio-Raman research seeking bio. phys. chem. about the triple point

オーガナイザー：盛田 伸一（東北大学），星野 由美（広島大学）
Organizers: Shin-ichi Morita (Tohoku University), Yumi Hoshino (Hiroshima University)

Raman microscope studies on live cells have attracted many researchers these past several years, providing cutting-edge applications, for instance, marking small molecules using alkyne based tags, estimating internal states of single cells, and observing tissues and small animals in a direct manner. Here, in this symposium, synthetic chemists and bio-physicists meet and discuss to find upcoming directions of bio-Raman research. The symposium therefore targets researchers who are interested in bio-Raman research not only the experts.

2SBP-01　ラマン分光等イメージング技術で紐解く生命現象と情報伝達過程
Bio-imaging without staining: Raman imaging and others
○岡 浩太郎（慶應大・理工・生命情報）
Kotaro Oka (*Dep. Biosci. & Infor., Keio Univ.*)

2SBP-02　細胞分化のバイオ・ラマン研究： 中間状態の検出
Bio-Raman Research on Cellular Differentiation to Detect the Reversible State
○盛田 伸一（東北大院理）
Shin-ichi Morita (*Tohoku Univ.*)

2SBP-03　蛍光プローブの精密設計による迅速癌検出
Rapid cancer imaging by rationally designed fluorescence probes
○神谷 真子[1,2], 浦野 泰照[1,3,4]（[1]東京大学大学院医学系研究科, [2]JST さきがけ, [3]東京大学大学院薬学系研究科, [4]AMED CREST）
Mako Kamiya[1,2], Yasuteru Urano[1,3,4] ([1]*Grad. Sch. of Med., Univ. of Tokyo*, [2]*JST PRESTO*, [3]*Grad. Sch. of Pharm. Sci, Univ. of Tokyo*, [4]*AMED CREST*)

2SBP-04　バイオラマン顕微鏡を用いた卵子のクオリティー評価
Oocyte evaluation using Bio-Raman microscope
○星野 由美（広島大学大学院生物圏科学研究科）
Yumi Hoshino (*Hiroshima University*)

2SBP-05　二本鎖 RNA オーバーハング構造結合選択性を有する合成蛍光プローブの開発と RNA 干渉研究への応用
Synthetic fluorescent probes capable of selective binding to 3'-overhanging structures in double-stranded RNAs for RNA interference study
○佐藤 雄介（東北大学大学院理学研究科化学専攻）
Yusuke Sato (*Department of Chemistry, Graduate School of Science, Tohoku University*)

2SBP-06　細胞膜分子動態が語る細胞の個性
What membrane molecule dynamics tell us about the cell
○坂内 博子[1,2], 丹羽 史尋[2,3], 有薗 美沙[2,4], 御子柴 克彦[2]（[1]JST・さきがけ・１細胞, [2]理研・脳センター, [3]パリ高等師範学校生物学研究所, INSERM, [4]ボルドー大学）
Hiroko Bannai[1,2], Fumihiro Niwa[2,3], Misa Arizono[2,4], Katsuhiko Mikoshiba[2] ([1]*JST PRESTO*, [2]*RIKEN BSI*, [3]*IBENS, INSERM*, [4]*Univ. of Bordeaux*)

16:15〜18:45　C 会場（中会議室 201）／Room C (Conference Room 201)
2SCP　［学会本部企画 III］日本ーオーストラリア交流シンポジウム：ライブセルイメージング
[BSJ Special Event III] BSJ-ASB Joint Symposium: Live Cell Imaging

オーガナイザー：林 久美子（東北大学），高橋 聡（東北大学）
Organizers: Kumiko Hayashi (Tohoku University), Satoshi Takahashi (Tohoku University)

We have this symposium on live cell imaging for the purpose of exchanges between Australian Society for Biophysics (ASB) and Biophysical Society of Japan (BSJ). Cutting-edge researches on fluorescence correlation spectroscopy and fluorescence probes to measure biochemical quantities in cells such as pH, ATP concentration and temperature are introduced. Structure analysis of cells using XFEL (X-ray Free Electron Laser) is also included as a new topic on live cell imaging.

2SCP-01　線虫胚における細胞質流動のイメージングとモデリング
Imaging and modeling of cytoplasmic streaming in the *C. elegans* embryo
○木村 暁[1,2]（[1]遺伝研・細胞建築, [2]総研大・遺伝学）
Akatsuki Kimura[1,2] ([1]*Cell Arch. Lab., Nat. Inst. Genet.*, [2]*Dept. Genet., SOKENDAI*)

2SCP-02　Profilin-1 membrane dynamics in live cells
Pierre Moens (*Univ. of New England*)

2SCP-03　細胞機能に関わる細胞内 pH の計測
Fluorescence imaging of cytoplasmic pH associated with cellular functions
○森本 雄祐[1], 上田 昌宏[1,2]（[1]理研・生命システム, [2]阪大・院生命機能）
Yusuke V. Morimoto[1], Masahiro Ueda[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*)

2SCP-04　Pair correlation microscopy reveals nanoparticle shape to control intracellular transport
Elizabeth Hinde (*Univ. of New South Wales*)

2SCP-05　Fluidic microenvironment in live cells revealed by standard molecules and nanoparticles
Chan-Gi Pack[1], Min-Kyo Jung[1], Sung-Sik Han[2] ([1]*University of Ulsan College of Medicine & AMC*, [2]*Korea University*)

2SCP-06　RGB カラーの蛍光タンパク質センサーによる細胞内 ATP の時空間イメージングと定量解析
Spatiotemporal imaging and quantitative analysis of subcellular ATP using RGB-colorful fluorescent protein based indicators
○新井 敏[1], 伊藤 秀城[2], Sudhaharan Thankiah[2], Lane E. Birgitte[2], 北口 哲也[1]（[1]早大・WABIOS, [2]IMB, A*STAR, Singapore）
Satoshi Arai[1], Hideki Ito[2], Thankiah Sudhaharan[2], E. Birgitte Lane[2], Tetsuya Kitaguchi[1] ([1]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*, [2]*Inst. of Med. Biol. (IMB), A*STAR, Singapore*)

2SCP-07　X 線レーザーによる生きた細胞のナノイメージング
Imaging live cell at the nanoscale by X-ray laser diffraction
○城地 保昌[1,2]（[1]JASRI, [2]理研RSC）
Yasumasa Joti[1,2] ([1]*JASRI*, [2]*RIKEN SPring-8 center*)

16:15〜19:05　D 会場（中ホール 300）／Room D (Convention Hall 300)
2SDP　蛋白質の秩序化−脱秩序化研究の最前線
Frontiers in protein organization and disorganization

オーガナイザー：伊野部 智由（富山大学），濵田 大三（神戸大学）
Organizers: Tomonao Inobe (University of Toyama), Daizo Hamada (Kobe University)

"Why and how the proteins can fold into well-ordered structures?" have been one of the most important questions in biology. Recent analysis has clarified that this complex process is also coupled with a variety of biological phenomena including protein translation, amyloid formation and degradation as well as protein-protein interactions. In this symposium, we recategorised these into "Protein Organization / Disorganization Problems" and will discuss future perspectives.

はじめに
濵田 大三
Daizo Hamada

2SDP-01 マイクロ秒分解一分子蛍光測定でみる変性タンパク質のダイナミクスとタンパク質折り畳み転移
Microsecond tracking of unfolded protein dynamics and protein folding transitions by single-molecule fluorescence spectroscopy
○小井川 浩之（東北大 多元研）
Hiroyuki Oikawa (*IMRAM, Tohoku Univ.*)

2SDP-02 タンパク質凝集体の表面から突出したポリペプチド鎖は分子シャペロンによる脱凝集効率に影響を与える
Polypeptides protruded from the surface of protein aggregation influence the efficiency of disaggregation by molecular chaperones
○渡辺 洋平[1,2], 山崎 孝史[1], 野島 達也[3], 小田 彰克[1]（[1]甲南大・理工・生物, [2]甲南大・統合ニューロ, [3]東工大・IIR）
Yo-hei Watanabe[1,2], Takashi Yamasaki[1], Tatsuya Nojima[3], Akiyoshi Oda[1] ([1]*Dept. Biol., Facult. Sci. Eng., Konan Univ.*, [2]*Inst. Integrated Neurobiol., Konan Univ.*, [3]*IIR, Tokyo Tech.*)

2SDP-03 Integrated in vivo and in vitro nascent chain profiling reveals widespread translational pausing
Yuhei Chadani[1,2], Tatsuya Niwa[1], Shinobu Chiba[2], Hideki Taguchi[1], Koreaki Ito[2] ([1]*Inst. of Innovative Research, Tokyo Inst. of Tech.*, [2]*Fac. of Life Sci., Kyoto Sangyo Univ.*)

2SDP-04 天然タンパク質の分子サイズに関する統計解析
Statistical analysis on the molecular size of native proteins
○河合 秀信, 高橋 大輔, 新井 宗仁（東大・総合文化・生命環境）
Hidenobu Kawai, Daisuke Takahashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

2SDP-05 ユビキチン化に伴う蛋白質の凝集体形成
Ubiquitylation-induced protein aggregation
○森本 大智[1], ヴァリンダ エリック[2], 深田 はるみ[3], 菅瀬 謙治[1], 星野 大[4], 藤井 高志[5], 難波 啓一[6], 小松 雅明[7], 田中 啓二[8], 白川 昌宏[1]（[1]京大・工, [2]京大・医, [3]大府大・生命環境, [4]京大・薬, [5]理研・QBiC, [6]阪大・生命機能, [7]新潟大・医, [8]東京都医学研・蛋白質代謝）
Daichi Morimoto[1], Erik Walinda[2], Harumi Fukada[3], Kenji Sugase[1], Masaru Hoshino[4], Takashi Fujii[5], Keiichi Namba[6], Masaaki Komatsu[7], Keiji Tanaka[8], Masahiro Shirakawa[1] ([1]*Eng., Kyoto Uni.*, [2]*Med., Kyoto Uni.*, [3]*Life Envi. Sci., Osaka Pref. Uni.*, [4]*Pharm., Kyoto Uni.*, [5]*Frontier Biosci., Osaka Uni.*, [6]*Frontier Biosci., Osaka Uni.*, [7]*Med., Niigata Uni.*, [8]*Lab. Protein Metabolism, Tokyo Metro. Ins. Med. Sci.*)

2SDP-06 分子シャペロンによるプロテアソームタンパク質分解の制御
Regulation of proteasomal degradation by molecular chaperone
○伊野部 智由（富山大･工･生命工）
Tomonao Inobe (*Grad. Sch. Sci. and Eng., Univ. Toyama*)

総合討論
伊野部 智由
Tomonao Inobe

16:15～18:45　E 会場（小会議室 303）／Room E (Conference Room 303)

2SEP　新しい視点を創る光学顕微鏡技術
Taking a new look through the optical microscopy

オーガナイザー：加藤 薫（産業技術総合研究所），西山 雅祥（京都大学）
Organizers: Kaoru Katoh (AIST), Masayoshi Nishiyama (Kyoto University)

Optical microscopy is an important tool for imaging and measurement in life sciences. This session focus on technical topics that can be seeds of future key technologies. Detail of each technology should be explained and biophysical application will be shown in the presentation. This session will show possibilities of future life sciences from a standing point of imaging technologies.

2SEP-01 高圧力顕微鏡法で生きた細胞内で働く分子機械を操作する
High-pressure microscopy for controlling molecular machines in living cells
○西山 雅祥（京大白眉セ）
Masayoshi Nishiyama (*The HAKUBI Center, Kyoto Univ.*)

2SEP-02 生細胞内における生体分子動態マッピングに向けて
Researchs towards bio-molecular dynamics mapping in cell
○山本 条太郎（北大院先端生命）
Johtaro Yamamoto (*Faculty of Adv. Life Sci., Hokkaido Univ.*)

2SEP-03 絶対零度で蛍光１分子を見る
Fluorescence microscopy of single molecules at a few K.
○藤芳 暁（東京工業大学 理学院）
Satoru Fujiyoshi (*Tokyo Tech*)

2SEP-04 位相差法による無染色での試料の同定法の開発
Apodized phase contrast imaging for identification of specimens without staining
○大瀧 達朗[1,2]（[1]ニコン・コアテク, [2]東北大・院医工学）
Tatsuro Otaki[1,2] ([1]*Core Technology, Nikon Corp.*, [2]*Grad. Sch. Biomed. Eng., Tohoku Univ.*)

2SEP-05 光の波面を制御して散乱体を透視する
Seeing through scattering media by controlling wavefront of light
○白井 智宏, 加藤 薫（産業技術総合研究所）
Tomohiro Shirai, Kaoru Katoh (*AIST*)

2SEP-06 構成分子の位置および向きの１分子観察から読み解く分子会合のダイナミクス
Dissection of molecular assembly dynamics by tracking orientation and position of single molecules in live cells
○谷 知己（ウッズホール海洋生物学研究所）
Tomomi Tani (*Marine Biological Laboratory*)

16:15～18:45　F 会場（中会議室 406）／Room F (Conference Room 406)
2SFP　新学術領域研究「スパースモデリングの深化と高次元データ駆動科学の創成」共催
データ駆動科学 （スパースモデリング）による計測の進展
Advances in experimental measurements by data-driven science based on sparse modeling

オーガナイザー：木川 隆則（理化学研究所），池谷 鉄兵（首都大学東京）
Organizers: Takanori Kigawa (RIKEN), Teppei Ikeya (Tokyo Metropolitan University)

Sparse modeling (SpM), which is a key technology in data-driven science, enables efficient extraction of the maximum amount of information from experimental measurements by exploiting the inherent sparseness that is common to all high-dimensional data. In this symposium, researchers who are achieving remarkable results by SpM will be presented in different research fields, information science, statistical mechanics, astronomy and structure biology. Clarifying the common principles that apply in the background of each case, the future perspectives of biomolecule measurements will be discussed.

Introduction
木川 隆則
Takanori Kigawa

2SFP-01 スパースモデリングとデータ駆動科学
Sparse modeling and date driven science
○岡田 真人（東大）
Masato Okada (*Univ. of Tokyo*)

2SFP-02 Recent development of Monte Carlo sampling techniques
Koji Hukushima (*Univ. of Tokyo*)

2SFP-03 Fourier imaging with sparse modeling: An application to black hole astronomy
Mareki Honma (*NAOJ Mizusawa*)

2SFP-04 ベイズ解析を用いるＸ線１分子観察
X-ray Single Molecule Observations using Bayesian Analysis
○佐々木 裕次（東京大学大学院 新領域創成科学研究科）
Yuji Sasaki (*Graduate School of Frontier Sciences, The University of Tokyo*)

2SFP-05 スパース NMR データを用いた細胞内蛋白質立体構造決定
Protein NMR structure determination for sparse data set dervied from living cells
○池谷 鉄兵[1,2], 池田 思朗[3], 木川 隆則[4], 伊藤 隆[1,2], Guentert Peter[5]（[1]首都大院 理工, [2]CREST, JST, [3]統計数理研, [4]理研 生命システム研究センター, [5]フランクフルトゲーテ大学）
Teppei Ikeya[1,2], Shiro Ikeda[3], Takanori Kigawa[4], Yutaka Ito[1,2], Peter Guentert[5] ([1]*Tokyo Metropolitan University, Graduate School of Science and Engineering*, [2]*CREST, JST*, [3]*The Institute of Statistical Mathematics*, [4]*RIKEN, QBiC*, [5]*Goethe University Frankfurt am Main*)

16:15～18:45　G 会場（小会議室 405）／Room G (Conference Room 405)

2SGP　リン酸化ダイナミクスが支える生命情報処理機構
Information processing governed by dynamic protein phosphorylation

オーガナイザー：大出 晃士（東京大学），小川 覚之（東京大学）
Organizers: Koji L. Ode (The University of Tokyo), Tadayuki Ogawa (The University of Tokyo)

This symposium aims to foster a deeper understanding of the significance of reversible protein phosphorylation driven by kinases and phosphatases in the regulation of dynamic information processing in cells, including frequency control in central nerve systems, spatiotemporal regulation of cell structure, rhythmic response driven by molecular oscillator, and signal processing through kinase cascade. From a cross-cutting perspective, a unified property of reversible phosphorylation that governs nonlinear and complex cellular dynamics will be discussed.

2SGP-01 クリプトクロム蛋白質の柔軟なループ構造への多重リン酸化は哺乳類概日時計の周期長を相加的に制御する
Multiple phosphorylation at flexible loops of cryptochrome additively modulates the period of mammalian circadian clock
○大出 晃士[1,2], 上田 泰己[1,2]（[1]東大・院医・システムズ薬理, [2]理研・生命システム研究センター）
Koji L. Ode[1,2], Hiroki R. Ueda[1,2] ([1]*Dept. of Sys. Pharm., Grad. Sch. of Med., the Univ. of Tokyo*, [2]*QBiC, RIKEN*)

2SGP-02 リン酸化で規定されるタンパク質コンフォメーションから細胞の応答性を予測する
Signaling protein conformation regulated by multiple phosphorylations points in the direction of cell fate
○日比野 佳代[1,2,3]（[1]遺伝研, [2]総研大, [3]理研）
Kayo Hibino[1,2,3] ([1]*NIG*, [2]*SOKENDAI*, [3]*RIKEN*)

2SGP-03 CaMKIIα とカルシニューリンによる神経入力情報のデコーディングと表現
Nonlinear Decoding and Asymmetric Representation of Neuronal Input Information by CaMKIIα and Calcineurin
○藤井 哉, 井上 昌俊, 尾藤 晴彦（東京大学大学院医学系研究科）
Hajime Fujii, Masatoshi Inoue, Haruhiko Bito (*Department of Neurochemistry, Grad. Sch. of Medicine, The Univ. of Tokyo*)

2SGP-04 リン酸化アイソタイプの定量解析によるシナプスリン酸化シグナル伝達の新知見
Novel insight of synaptic phosphorylation signal transduction by quantitative analysis of phosphoisotypes
○細川 智永（理研・脳科学）
Tomohisa Hosokawa (*RIKEN BSI*)

2SGP-05 動的蛋白質リン酸化による生命現象の時間スケール調節
Dynamic protein phosphorylation as a time “scale” machine
○畠山 哲央（東京大学総合文化研究科）
Tetsuhiro S. Hatakeyama (*Department of Basic Science, The University of Tokyo*)

2SGP-06 微小管ダイナミクスを制御する異なる特異的リン酸化カスケード
Site-specific Phosphorylation Cascades that Differentially Regulate Microtubule Dynamics in Neuron
○小川 覚之, 廣川 信隆（東大・院医）
Tadayuki Ogawa, Nobutaka Hirokawa (*Grad. Sch. Med., Univ. Tokyo*)

第 3 日目（11 月 27 日（日））／Day 3（Nov. 27 Sun.）

9:45～12:15　A 会場（中ホール 200）／Room A (Convention Hall 200)

3SAA　蛍光・発光計測技術が拓く細胞生物学の新地平

New fields of cell biology explored with fluorescence and bioluminescence techniques

オーガナイザー：今村 博臣（京都大学），小柴 琢己（九州大学）

Organizers: Hiromi Imamura (Kyoto University), Takumi Koshiba (Kyushu University)

Fluorescence techniques have played great roles in biological research. Especially, fluorescence imaging techniques have propelled cell biology research, and are still under rapid development. Recently, bioluminescence techniques also become important for cell biology. In this symposium, we will invite relatively young investigators who explore new fields of cell biology with fluorescence and bioluminescence techniques.

3SAA-01　Mitochondrial-mediated antiviral immunity and oxidative phosphorylation
Takumi Koshiba (*Dep of Biol., Fac. of Sci., Kyushu Univ.*)

3SAA-02　線虫の塩忌避学習による行動変化に関与する神経の同定及び神経回路の解析
Identification of neurons and analysis of the neuronal circuit involved in the learned salt-avoidance behavior in C. elegans
○張 文瑄[1,3], 豊島 有[1,3], 国友 博文[1,3], 金森 真奈美[1,3], 寺本 孝行[2,3], 石原 健[2,3], 飯野 雄一[1,3]（[1]東京大学大学院理学系研究科生物科学専攻, [2]九州大学大学院理研究院生物科学専攻, [3]CREST, JST）
MoonSun Jang[1,3], Yu Toyoshima[1,3], Hirofumi Kunitomo[1,3], Manami Kanamori[1,3], Takayuki Teramoto[2,3], Takeshi Ishihara[2,3], Yuichi Iino[1,3] ([1]*Department of Biological Sciences, Graduate School of Science, The University of Tokyo*, [2]*Department of Biology, Faculty of Science, Kyushu University*, [3]*CREST, Japan Science and Technology Agency*)

3SAA-03　蛍光イメージングで紐解くインフルエンザウイルス感染の分子基盤
The molecular basis of influenza virus infection unveiled by fluorescence imaging
○大場 雄介, 藤岡 容一朗, 西出 真也, 南保 明日香（北海道大学大学院医学研究科細胞生理学分野）
Yusuke Ohba, Yoichiro Fujioka, Shinya Nishide, Asuka Nanbo (*Department of Cell Physiology, Hokkaido University Graduated School of Medicine*)

3SAA-04　ATP イメージングにより明らかになったアポトーシス細胞における細胞内 ATP 濃度変化の仕組み
ATP imaging revealed a mechanism of intracellular ATP changes during apoptosis
○今村 博臣（京都大学 生命科学研究科）
Hiromi Imamura (*Graduate School of Biostudies, Kyoto University*)

3SAA-05　可溶性因子を介した免疫細胞相互作用の 1 細胞モニタリング
Monitoring immune-cell communication via soluble factors at single-cell resolution
○白崎 善隆[1,2]（[1]東大・院理, [2]理研・IMS）
Yoshitaka Shirasaki[1,2] ([1]*Grad. Sch. Sci., Tokyo Univ.*, [2]*IMS, RIKEN*)

3SAA-06　Imaging RNA in living neural circuits with hybridization-sensitive fluorescent probes
Dan Ohtan Wang[1,2] ([1]*Institute for Integrated Cell-Material Sciences, Kyoto University*, [2]*K-CONNEX*)

9:45～12:15　B 会場（中会議室 202）／Room B (Conference Room 202)
3SBA　新学術領域研究「理論と実験の協奏による柔らかな分子系の機能の科学」共催
生体分子の柔らかさと機能をつなぐもの
What connects the softness of biomolecules to their functions?

オーガナイザー：石井 邦彦（理化学研究所），井上 圭一（名古屋工業大学）
Organizers: Kunihiko Ishii (RIKEN), Keiichi Inoue (Nagoya Institute of Technology)

Many biomolecules achieve their functions through dynamically changing their conformations. Behind such dynamics-function couplings, there exist exquisite mechanisms which utilize the softness of the molecules, and each of them is accompanied with a characteristic controlling factor. In this symposium, presentations will be given by young researchers from theory and experiment focusing on various factors connecting molecular softness with biological functions. Through the discussion over a breadth of examples, we pursue a universal concept underlying the role of softness in biological systems.

3SBA-01　二次元蛍光寿命相関分光法で観るマイクロ秒領域の生体分子の熱ゆらぎ
Thermal fluctuation of biomolecular conformation on microsecond timescale detected by 2D fluorescence lifetime correlation spectroscopy
○石井 邦彦[1,2], 田原 太平[1,2]（[1]理研・田原分子分光, [2]理研・光量子工学領域）
Kunihiko Ishii[1,2], Tahei Tahara[1,2] ([1]*Molecular Spectroscopy Lab., RIKEN*, [2]*RIKEN Center for Advanced Photonics*)

3SBA-02　QM/MM RWFE-SCF 法とマイクロ秒 MD 計算によるタンパク質荷電性残基の pKa 予測
pKa prediction of ionizable residues in proteins by QM/MM RWFE-SCF method combined with microsecond-long MD simulations
○長谷川 太祐[1], 林 重彦[1]（[1]京大院理, [2]京大院理）
Taisuke Hasegawa[1], Shigehiko Hayashi[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Sci., Kyoto Univ.*)

3SBA-03　アンキリンリピートドメインと脂質の相互作用による TRPV1 チャネル活性の制御
Regulatory mechanism of TRPV1 channel activity by the interaction of ankyrin repeat domain with phospholipids
○竹村 和浩[1], 末次 志郎[2], 北尾 彰朗[1]（[1]東大分生研, [2]NAIST バイオサイエンス）
Kazuhiro Takemura[1], Shiro Suetsugu[2], Akio Kitao[1] ([1]*IMCB, Univ. of Tokyo*, [2]*Grad. Sch. Biol. Sci., NAIST*)

3SBA-04　タンパク質を基盤とした酸素およびヘム濃度プローブ分子の開発
Protein-based molecular probes for the local concentrations of oxygen and heme
○石川 春人（阪大院理）
Haruto Ishikawa (*Grad. Scl. Sci., Osaka Univ.*)

3SBA-05　フラビン結合タンパク質は目的の機能を示すことに対してどの程度「柔らかい」か？
How are flavoproteins“soft”for exhibiting intended functions?
○岩田 達也[1,2]（[1]名古屋工業大学大学院工学研究科生命・応用化学専攻, [2]名古屋工業大学オプトバイオテクノロジー研究センター）
Tatsuya Iwata[1,2] ([1]*Life Sci. Appl. Chem., Grad. Sch. Eng. NITech*, [2]*OptBioTech. Res. Ctr., NITech*)

3SBA-06　DNA 整列固定技術を用いた DNA 結合蛋白質の単分子機能解析
Single-molecule characterization of DNA-binding proteins with stretchable DNA array
○鎌形 清人（東北大多元研）
Kiyoto Kamagata (*IMRAM, Tohoku Univ.*)

9:15～12:15　C 会場（中会議室 201）／Room C (Conference Room 201)

3SCA　新学術領域研究「動的構造生命科学を拓く新発想測定技術―タンパク質が動作する姿を活写する―」共催 次世代研究者による動的構造生命

Dynamic structural biology by next-generation researchers

オーガナイザー：塚崎 智也（奈良先端科学技術大学院大学），西田 紀貴（東京大学）
Organizers: Tomoya Tsukazaki (Nara Institute of Science and Technology), Noritaka Nishida (The University of Tokyo)

The recent developments of innovative technologies in the fields of X-ray crystallography, NMR, cryo-EM, high-speed AFM, and MD simulations, provided the dynamic structural information that greatly contributed to the elucidation of protein functions. In this symposium, 8 prominent young investigators, who are expected to lead the next generation of structural life sciences, will present the latest achievements in their research.

opening remarks

3SCA-01　ナノディスクに再構成した AglB タンパク質の単粒子解析
Single particle analysis of the AglB protein embedded in nanodiscs
○川崎 由貴[1], 眞柳 浩太[1], Srivastava Ashutosh[2], Tama Florence[2,3], 神田 大輔[1]（[1]九大・生医研・構造生物, [2]名大・理学・物理, [3]理研・計算科学）
Yuki Kawasaki[1], Kouta Mayanagi[1], Ashutosh Srivastava[2], Florence Tama[2,3], Daisuke Kohda[1] ([1]*Div. Struct. Biol. of Med. Inst. Bioreg., Kyushu Univ*, [2]*Dept. of Phys., Grad sch. of Sci., Nagoya Univ*, [3]*AICS., RIKEN*)

3SCA-02　染色体分配を支える CENP-A licensing 複合体の構造基盤
Structural basis of the CENP-A licensing protein complex
○有吉 眞理子, 松田 麻理子, 白川 昌宏（京大・院工）
Mariko Ariyoshi, Mariko Matsuda, Masahiro Shirakwa (*Grad. Sch. Eng., Kyoto Univ.*)

3SCA-03　フェムト秒 X 線自由電子レーザーによって明らかにされた光化学系 II 複合体の中間体構造
Crystal structure of the oxygen evolving photosystem II in the intermediate state revealed by femtosecond X-ray free electron lasers
○菅 倫寛（岡山大・異分野基礎研）
Michi Suga (*RIIS, Okayama Univ.*)

3SCA-04　高分子量タンパク質の機能的運動性を解明するための多量子 NMR 解析法の開発と応用
Developments and applications of multiple quantum NMR methods to characterize functional dynamics of high molecular weight proteins
○外山 侑樹[1,2], 加納 花穂[1], 間瀬 瑤子[1], 横川 真梨子[1], 大澤 匡範[1], 嶋田 一夫[1]（[1]東大・院薬, [2]バイオ産業情報化コンソーシアム）
Yuki Toyama[1,2], Hanaho Kano[1], Yoko Mase[1], Mariko Yokogawa[1], Masanori Osawa[1], Ichio Shimada[1] ([1]*Grad. Sch. Pharm. Sci., the Univ. of Tokyo*, [2]*JBIC*)

3SCA-05　高速 AFM を用いてタンパク質が動作する姿を活写する
Visualization of protein molecules in action by high-speed atomic force microscopy
○柴田 幹大[1,2], 古寺 哲幸[2], 内橋 貴之[1,2], 安藤 敏夫[2]（[1]金沢大・理工, [2]バイオAFM FRC）
Mikihiro Shibata[1,2], Noriyuki Kodera[2], Takayuki Uchihashi[1,2], Toshio Ando[2] ([1]*Dept. Phys., Kanazawa Univ.*, [2]*Bio-AFM FRC*)

3SCA-06　タンパク質膜透過を駆動するモータータンパク質のスナップショット
Snapshots of a protein translocation motor
○古川 新[1], 吉海江 国仁[1], 森 貴治[2], 森 博幸[3], 森本 雄祐[2], 菅野 泰功[1], 岩木 薫大[1], 南野 徹[4], 杉田 有治[2], 田中 良樹[1], 塚崎 智也[1]（[1]奈良先端大・バイオ, [2]理研, [3]京大・ウイルス研, [4]阪大・院生命機能）
Arata Furukawa[1], Kunihito Yoshikaie[1], Takaharu Mori[2], Hiroyuki Mori[3], Yusuke Morimoto[2], Yasunori Sugano[1], Shigehiro Iwaki[1], Tooru Minamino[4], Yuji Sugita[2], Yoshiki Tanaka[1], Tomoya Tsukazaki[1] ([1]*NAIST*, [2]*RIKEN*, [3]*Kyoto Univ.*, [4]*Osaka Univ.*)

3SCA-07 分子シミュレーションによる SecDF プロトン透過機構の解明
Molecular mechanisms underlying proton transport in SecDF
○森 貴治[1,2], 田中 良樹[3], 吉海江 国仁[3], 塚崎 智也[3], 杉田 有治[1,2,4,5]（[1]理研 杉田理論分子科学, [2]理研 iTHES, [3]奈良先端大, [4]理研 AICS, [5]理研 QBiC）
Takaharu Mori[1,2], Yoshiki Tanaka[3], Kunihito Yoshikaie[3], Tomoya Tsukazaki[3], Yuji Sugita[1,2,4,5] ([1]*RIKEN Theor. Mol. Sci. Lab.*, [2]*RIKEN iTHES*, [3]*NAIST*, [4]*RIKEN AICS*, [5]*RIKEN QBiC*)

3SCA-08 フレキシブルフィッティングによる電子顕微鏡データからの構造モデリング
Structure Modeling from Cryo-EM Data using Flexible Fitting Approach
○宮下 治（理化学研究所計算科学研究機構）
Osamu Miyashita (*RIKEN AICS*)

closing remarks

9:15～12:15　D 会場（中ホール 300）／Room D (Convention Hall 300)
3SDA　新学術領域研究「運動超分子マシナリーが織りなす調和と多様性」共催
運動超分子マシナリーが織りなす調和と多様性
Harmonized supramolecular motility machinery and its diversity

オーガナイザー：宮田 真人（大阪市立大学），上田 太郎（早稲田大学）
Organizers: Makoto Miyata (Osaka City University), Taro QP Uyeda (Waseda University)

The molecular mechanism of force generation by "conventional" motor proteins, e.g. myosin, kinesin, and dynein, is now fairly well understood after decades of research. However, many mechanisms of motility cannot be explained using only conventional motor proteins. Such motilities are driven by highly organized structures, which we call "supramolecular motility machinery", and their diversity records the evolutional history of life on earth. In this symposium, we will discuss about the principle and the origin of motility, based on new knowledge about poorly characterized motility mechanisms.

3SDA-01 イオン駆動型回転モーターにおけるエネルギー変換マシナリーの分子解剖：細菌べん毛モーター固定子の機能と構造
Dissection of the energy-conversion machinery in the ion-driven rotary motor: structural and functional studies of the flagellar stator
○小嶋 誠司（名大・院理・生命理学）
Seiji Kojima (*Div. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

3SDA-02 好アルカリ性 *Bacillus* 属細菌と枯草菌がもつ Na^+駆動型べん毛モーターの中性環境での Na^+透過性の違いの解明
The elucidation of the Na^+-requirement mechanism for flagellar rotation between alkaliphilic and neutrophilic *Bacillus* at neutral pH
○高橋 優嘉[1], 伊藤 政博[1,2]（[1]東洋大学 バイオ・ナノ, [2]東洋大 生命科）
Yuka Takahashi[1], Masahiro Ito[1,2] ([1]*Bio-Nano., Toyo Univ.*, [2]*Faculty of Life Sciences, Toyo Univ.*)

3SDA-03 バクテリアべん毛モーターの回転方向切り替えメカニズム
Switching mechanism of the bacterial flagellar motor
○南野 徹（大阪大学大学院生命機能研究科）
Tohru Minamino (*Grad. Sch. Frontier Biosci, Osaka Univ.*)

3SDA-04 The actin-like cytoskeletal protein MamK plays a role in positioning of magnetic organelles for bacterial magnetotactic motility
Azuma Taoka, Yoshihiro Fukumori (*Col. Sci. and Eng., Kanazawa Univ.*)

3SDA-05 F-ATPase から進化したマイコプラズマ滑走運動
Mycoplasma gliding developed from F-type ATPase
○宮田 真人（大阪市立大学大学院理学研究科）
Makoto Miyata (*Osaka City University*)

3SDA-06 バクテロイデーテス細菌がスムーズに滑走する仕組み
Structure and mechanism of gliding motility of *Bacteroidetes*
○柴田 敏史（長崎大学 医歯薬学総合研究科 口腔病原微生物学）
Satoshi Shibata (*Graduate Sch. of Biomedical Science, Nagasaki Univ.*)

3SDA-07 アクチンフィラメントの構造多型性：アクチン結合タンパク質の制御および細胞運動への寄与
Structural polymorphism of actin filaments: its implication in regulation of actin binding proteins and cell motility
○上田 太郎[1,2], ンゴー キエン[1], 野口 太郎[3], 長崎 晃[2], 古寺 哲幸[4], 徳楽 清孝[5]（[1]早稲田大・物理, [2]産総研・バイオメディカル, [3]都城高専・物質工学, [4]金沢大・バイオAFM, [5]室蘭工大・工）
Taro Uyeda[1,2], Kien Ngo[1], Taro Noguchi[3], Akira Nagasaki[2], Noriyuki Kodera[4], Kiyotaka Tokuraku[5] ([1]*Dept. of Physics, Waseda Univ.*, [2]*Boomed. Res. Inst., AIST*, [3]*Dept. Chem. Sci. Eng., Natl. Inst. Tech., Miyakonojo Coll.*, [4]*Bio AFM Res. Ctr., Kanazawa Univ.*, [5]*Muroran Inst. Tech.*)

9:15～12:15　E 会場（小会議室 303）／Room E (Conference Room 303)
3SEA　多細胞合成生物学
Synthetic biology for multicellular system

オーガナイザー：木賀 大介（早稲田大学），戎家 美紀（理化学研究所）
Organizers: Daisuke Kiga (Waseda University), Miki Ebisuya (RIKEN)

Synthetic/reconstruction approach for reaction network in cells enables us to understand the network from systems science points of view. On top of construction of such network in a cell, a system with multi cell species each of which accommodates synthetic network has also been constructed in this manner. Developments in cell manipulation techniques in microfluidics or artificial organ with ES/iPS cells can be combined with the synthetic approach. In this symposium, we will introduce frontline of the approach and discuss future innovation from this field.

はじめに
木賀 大介
Daisuke Kiga

3SEA-01 Design and construction of synthetic microbial communities by combining synthetic biological subsystems
Shotaro Ayukawa (*ACLS, Tokyo Tech*)

3SEA-02 人工細胞パターン形成
Synthetic cell pattern formation
○戎家 美紀（理研QBiC）
Miki Ebisuya (*RIKEN QBiC*)

3SEA-03 Microfluidic droplet reactor for artificial/living cellular systems
Masahiro Takinoue[1,2] ([1]*Dept. Comput. Sci., Tokyo Tech*, [2]*PRESTO, JST*)

3SEA-04 Generation of a self-organizing kidney comprising multiple renal cell types
Minoru Takasato (*RIKEN CDB*)

3SEA-05 合成生物学研究のための哺乳類の in vitro 生命システム
An in vitro Living System in Mammals for Synthetic Biology Research
○田川 陽一（東京工業大学 生命理工学院）
Yoh-ichi Tagawa (*Tokyo Institute of Technology School of Life Science and Technology*)

9:45～12:15　F 会場（中会議室 406）／Room F (Conference Room 406)
3SFA　ミトコンドリアの分子マシナリーと機能管理: 合成、構造、機能、適応、そして淘汰
Management of mitochondrial functions by molecular machineries: biogenesis, structure, function, adaptation, and elimination

オーガナイザー：遠藤 斗志也（京都産業大学），鈴木 俊治（東京大学）
Organizers: Toshiya Endo (Kyoto-Sangyo University), Toshiharu Suzuki (The University of Tokyo)

In this symposium, we will discuss recent progress in the studies on mitochondrial protein machineries. Machineries for the transport of proteins and lipid (Endo) and for the cristae-formation (Oka) will be introduced, showing how the complicated mitochondrial architecture is generated. Structures and functions of the respiratory chain (Tsukihara and Kita) and FoF1-ATP synthase (Suzuki) will be discussed, emphasizing the power of X-ray analyses. The novel quality control mechanism for eliminating dysfunctional mitochondria will be shown, referring to the Parkinson-disease (Matsuda).

はじめに
鈴木 俊治
Toshiharu Suzuki

3SFA-01　タンパク質と脂質を運んでミトコンドリアをつくる仕組み
Mechanisms of mitochondrial biogenesis by protein and lipid transport
○遠藤 斗志也（京産大・総合生命）
Toshiya Endo (*Kyoto Sangyo Univ., Fac. Life Sci.*)

3SFA-02　精密 X 線結晶構造解析によるチトクロム酸化酵素の酸素還元・プロトンポンプ機構
Detailed crystal structural studies of bovine cytochrome oxidase to elucidate the coupling mechanism of dioxygen reduction and proton pump
○月原 冨武[1,2], 島田 敦広[1], 矢野 直峰[3], 村本 和優[1], 新澤- 伊藤 恭子[1], 山下 栄樹[2], 吉川 信也[1]（[1]兵県大・院生命理学, [2]阪大・蛋白研, [3]茨城大・フロンティアセンター）
Tomitake Tsukihara[1,2], Atsuhiro Shimada[1], Naomine Yano[3], Kazumasa Muramoto[1], Kyoko Shinzawa-Itoh[1], Eiki Yamashita[2], Shinya Yoshikawa[1] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Institute for Protein Research, Osaka Univ.*, [3]*Front. Res. Cen. Appli. Atom. Sci., Ibaraki Univ.*)

3SFA-03　環境適応における寄生虫ミトコンドリア呼吸鎖のリモデリング
Re-modeling of respiratory chain in the parasite mitochondria during their adaptation
北 潔（長崎大学）
Kiyoshi Kita (*Nagasaki University*)

3SFA-04　どのようにして哺乳類 F_1-ATPase は回転し、そして阻害されるのか? 顕微鏡一分子観察と X 線結晶構造解析による哺乳類 F_1 の角度分割解析
How does F_1-ATPase drive rotation? Angle-divided analysis of mammalian F_1-ATPases by single-molecule and X-ray crystallographic studies
○鈴木 俊治（東大・院・工・応化）
Toshiharu Suzuki (*School of Engineering, The University of Tokyo*)

3SFA-05　PINK1 と Parkin によるミトコンドリア品質管理機構は PKA を介した MIC60 のリン酸化により制御されている
PKA-dependent phosphorylation of MIC60 controls mitochondrial clearance regulated by PINK1 and Parkin
赤羽 しおり, 宇野 碧, 島崎 俊太, ○岡 敏彦（立教大学 理学部 生命理学科）
Shiori Akabane, Midori Uno, Shunta Shimazaki, **Toshihiko Oka** (*Department of Life Sicence, Rikkyo University*)

3SFA-06　ミトコンドリア品質管理マシナリーからパーキンソン病の発症機構を明らかにする
How mitochondrial quality control machinery resists a predisposition to Parkinson's disease
○松田 憲之（都医学総合研究所, ユビキチンプロジェクト）
Noriyuki Matsuda (*Ubiquitin Project, TMIMS*)

総括
遠藤 斗志也
Toshiya Endo

9:45～12:15　G 会場（小会議室 405）／Room G (Conference Room 405)

3SGA　人工生体プログラマブルシステム ～精密構造設計から分子ロボティクスへ～
Programmable bioinspired systems: Integration of precisely designed architectures towards molecular robots

オーガナイザー：石川 大輔（東京工業大学），鈴木 勇輝（東北大学）
Organizers: Daisuke Ishikawa (Tokyo Institute of Technology), Yuki Suzuki (Tohoku University)

It is one of the goal in biophysics to create artificially the dynamic structure or systems which lead the most suitable solution depending its environment and diverse functions like a cell. In recent years, there has been much efforts to construct molecular robots with sensing, computation and actuation by hybrid systems built on mechanical engineering and biology. In this symposium, we will discuss the approaches microscopically and macroscopically to integrate the dynamic systems based on a cell into a consistent system.

3SGA-01　DNA ナノ構造上に構築した化学的に制御可能なナノシステム
Chemically controllable nanosystems constructed in the DNA nanostructures
○遠藤 政幸（京都大学 物質－細胞統合システム拠点）
Masayuki Endo (*WPI-iCeMS, Kyoto University*)

3SGA-02　Organizing DNA origami components into crystalline structures at the lipid/aqueous solution interface
Yuki Suzuki (*FRIS, Tohoku Univ.*)

3SGA-03　生態模倣アクチュエータ作製に向けた試み：液晶中での微粒子運動
Bottom-up technologies for biomimetic actuators: motion of microbeads in liquid crystals
○武仲 能子[1,2]（[1]産総研機能化学, [2]JSTさきがけ）
Yoshiko Takenaka[1,2] ([1]*RI for Sustainable Chemistry, AIST*, [2]*JST PRESTO*)

3SGA-04　脂質修飾 DNA ナノ構造体の動的な集合体制御
Dynamic assembly control of lipid-modified DNA nanostructures
○与那嶺 雄介[1,2], セルバンテスーサルゲロ ケイテル[3], 中西 和嘉[2], 南 皓輔[2], 川又 生吹[3], 村田 智[3], 有賀 克彦[2]（[1]九大院工, [2]物材機構, [3]東北大院工）
Yusuke Yonamine[1,2], Keitel Cervantes-Salguero[3], Waka Nakanishi[2], Kosuke Minami[2], Ibuki Kawamata[3], Satoshi Murata[3], Katsuhiko Ariga[2] ([1]*Grad. Sch. of Eng., Kyushu Univ.*, [2]*NIMS*, [3]*Grad. Sch. of Eng., Tohoku Univ.*)

3SGA-05　DNA のプログラマビリティを利用したカプセル型分子ロボットの創製
Microcapsular robot based on programmability of DNA
○石川 大輔（東工大・情報理工）
Daisuke Ishikawa (*Sch. Comput., Tokyo Tech.*)

3SGA-06　自然知能システム：粘菌の計算パワーを活用する
Natural Intelligence System: Exploiting Computational Power of Amoeboid Organism
○青野 真士[1,2]（[1]東工大・地球生命研, [2]JSTさきがけ）
Masashi Aono[1,2] ([1]*Earth-Life Sci. Inst., Tokyo Tech*, [2]*PRESTO, JST*)

ポスター　Poster

第1日目（11月25日（金））／Day 1（Nov. 25 Fri.）大会議室 101 ＋102、多目的ホール／Conference Room 101+102, Multi-Purpose Hall

蛋白質：構造 / Protein: Structure

1Pos001 **Crystal structure of the 11-cis isomer of Pharaonis Halorhodopsin**
Siu Kit Chan[1], Haruki Kawaguchi[1], Hiroki Kubo[1], Kunio Ihara[3], Kosuke Maki[1], Tsutomu Kouyama[1,2] ([1]*Grad. Sch. of Sci., Nagoya Univ.*, [2]*RIKEN Harima Branch*, [3]*Center for Gene Research, Nagoya Univ.*)

1Pos002 **結晶構造から明らかになった、エンドセリン-1 によるエンドセリン受容体 B 型の活性化機構**
Crystal structures of the endothelin receptor type B reveal activation mechanism by endothelin-1
Wataru Shihoya[1,2], Tomohiro Nishizawa[3,4], Akiko Okuta[2], Kazutoshi Tani[2], Yoshinori Fujiyoshi[1,2], Osamu Nureki[3], Tomoko Doi[5] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Cellular and Structural Physiology Institute., Nagoya Univ.*, [3]*Grad. Sch. Sci., Univ. Tokyo*, [4]*JST PRESTO*, [5]*Grad. Sch. Sci., Kyoto Univ.*)

1Pos003* **X 線結晶構造解析による軸糸ダイニン軽鎖 1 の構造評価**
X-ray crystallographic characterization of the axonemal dynein light chain-1
Akiyuki Toda[1], Hideaki Tanaka[2], Yosuke Nishikawa[2], Toshiki Yagi[3], Genji Kurisu[2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Institute for Protein Research*, [3]*Facult. Life Environ., Pref. Univ. Hiroshsima*)

1Pos004 **固定子に作用するべん毛内膜蛋白質 FliL の構造解析**
Structural analysis of the Stator Associated Inner Membrane Protein FliL from Vibrio alginolyticus
Miyu Isumi[1], Yuuki Nishino[2], Mayuko Sakuma[2,3], Seiji Kojima[2], Michio Homma[2], Katsumi Imada[1] ([1]*Grad. Sch. of Sci., Osaka Univ.*, [2]*Grad. Sch. of Sci., Nagoya Univ.*, [3]*Radioisotpe Res. Cent.*)

1Pos005 **Oligomeric structure of the ExbB-ExbD complex revealed by X-ray crystallography and cryo-EM**
Saori Maki-Yonekura, Yoshiki Yamashita, Rei Matsuoka, Maiko Tanaka, Fumie Iwabuki, Koji Yonekura (*RIKEN SPring-8 center*)

1Pos006 **赤痢菌ニードル複合体の極低温電子顕微鏡による構造解析**
Structural analysis of needle complex from *shigella flexneri* by cryo electron microscopy
Naoko Kajimura[1,2], Takayuki Kato[1], Ariel J Blocker[3], Kei-ichi Namba[1,4] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*Res., Center for UHVEM, Osaka Univ.*, [3]*Sch. of Cell. & Mol. Med., Univ. of Bristol*, [4]*RIKEN, QBiC*)

1Pos007 **単一ミオシン結合状態のアクトミオシンの高分解能化**
F-actin structural changes induced by a single myosin head
Takahiro Namise, Kazuaki Yoshida, Takuo Yasunaga (*Kyushu Institue of Technology*)

1Pos008* **NMR analysis of C-terminal periplasmic domain of flagellar motor protein MotB and its active mutant L119P**
Gaby Almira[1], Ikumi Kawahara[1], Seiji Kojima[2], Katsumi Imada[3], Toshimichi Fujiwara[1], Michio Homma[2], Chojiro Kojima[1,4] ([1]*Inst. for Prot. Res., Osaka Univ.*, [2]*Div. of Biol. Sci., Grad. Sch. of Sci., Nagoya Univ.*, [3]*Dept. of Macromol. Sci., Grad. Sch. of Sci., Osaka Univ.*, [4]*Grad. Sch. of Eng., Yokohama National Univ.*)

1Pos009 **CS-Rosetta 法によるヒストン H2A-H2B ヘテロ二量体の溶液構造解析**
Determination of the solution structure of isolated histone H2A-H2B heterodimer by using CS-Rosetta
Tsutomu Yamane, Yoshihito Moriwaki, Hideaki Ohtomo, Mitsunori Ikeguchi, Jun-ichi Kurita, Masahiko Sato, Aritaka Nagadoi, Hideaki Shimojo, Yoshifumi Nishimura (*Graduate School of Medical Life Science, Yokohama City University*)

1Pos010 **時間分解 EPR 法によるヒトインスリンのアミロイド線維化による構造変化の観測**
Effects of amyloid fibrillations on geometries of human insulin as studied by time-resolved EPR spectroscopy
Tomoka Abe[1], Takashi Tachikawa[1], Eri Chatani[1], Paul Zierep[2], Stefan Weber[2], Toshifumi Mori[3], Shinji Saito[3], Yasuhiro Kobori[1] ([1]*Grad. Sch. Sci., Kobe Univ.*, [2]*Freiburg Univ.*, [3]*IMS*)

1Pos011 **溶液 NMR 法を用いた長距離情報の取得による Nrd1 のドメイン間配向の決定**
Structural analysis of a multi-domain protein using long-range distance information derived by solution NMR
Kan Nagai, Ayaho Kobayashi, Yutaka Ito, Masaki Mishima (*Graduate School of Science and Engineering, Tokyo Metropolitan University*)

1Pos012 **サイズ排除クロマトグラフィー／X 線小角散乱法に基づいたニトリラーゼ会合体プロトマーの構造特性**
Structural characterization on nitrilase protomers analyzed by size-exclusion chromatography/small-angle X-ray scattering (SEC-SAXS)
Masatoshi Usui[1,2], Ryo Ishiguro[1,2], Homare Yokota[1,2], Yusuke Takeda[1,2], Takaaki Hikima[2], Tetsuro Fujisawa[1,2] ([1]*Grad. Sch. Eng., Gifu Univ.*, [2]*SPring-8 Center, Harima Inst., RIKEN*)

1Pos013* **様々な炎症物質を認識する NLRP3-LRR ドメインの構造基盤の解明**
Investigation of molecular basis underlying the recognition of various inflammatory substances by NLRP3-LRR domain
Ryota Yamamoto[1], Kazuto Yamashita[1], Hiroshi Imamura[2], Motonari Tsubaki[1], Eri Chatani[1] ([1]*Grad. Sch. Sci., Univ. Kobe.*, [2]*AIST*)

1Pos014 **PRP の匂い分子結合における構造変化**
The structural changes of the peri-receptor protein (PRP) on the odorant-binding process
Xing Li[1], Durige Wen[1], Mitsuhiro Hirai[2], Noboru Ohta[3], Masaru Hojo[4], Mamiko Ozaki[4], Tatsuo Iwasa[1,5] ([1]*Div. Eng., Muroran Ins. of Tech.*, [2]*Dept. Phys., Gunma Univ.*, [3]*JASRI*, [4]*Dept. Biol., Grad. School Sci., Kobe Univ.*, [5]*Cen. Env. Sci. Dis. Mit. Adv. Res., Muroran Ins. of Tech.*)

1Pos015 **放射光小角散乱データに基づく対称性を考慮したニトリラーゼオリゴマーのモデリング**
Modeling of Nitrilase oligomer with flexible symmetry based on synchrotron small-angle scattering data
Tetsuro Fujisawa[1,2,3], Keiichi Kameyama[1], Ryo Ishiguro[1,2] ([1]*Dep. Chem. & Biomol. Sci., Fac. Eng., Gifu Univ.*, [2]*Spring-8 Center, RIKEN Hrima Inst.*, [3]*Synchro. Center, Nagoya Univ.*)

1Pos016 **β-シート中におけるアミノ酸トリプレットパターンの解析**
Analysis of amino acid triplet patterns in β-sheets
Hiromi Suzuki (*School of Agri., Meiji Univ.*)

1Pos017 **タンパク質の構造コンプライアンス特性とドメイン間運動の関係性解析**
Analysis of the Relationship Between Structural Compliance Properties and Inter-domain Motion of Proteins
Keisuke Arikawa (*Fcl. Eng., Kanagawa Inst. of Tech.*)

1Pos018 **自由エネルギー変分原理に基づく Pim-1 キナーゼ阻害剤系の相対的結合自由エネルギーの予測**
Prediction of the relative binding free energies for Pim-1 kinase - inhibitor systems based on the free energy variational principle
Anna Hirai (*Dept. of Bioinfo., Col. Life., Ritsumeikan Univ.*)

1Pos019 **Flexible docking between cyclin-dependent kinase 2 and its inhibitor using multicanonical MD**
Gert-Jan Bekker[1], Narutoshi Kamiya[2], Mitsugu Araki[3], Yasushi Okuno[4], Haruki Nakamura[1] ([1]*IPR, Osaka Univ.*, [2]*Grd. Sch. SS, Univ. Hyogo*, [3]*AICS, RIKEN*, [4]*Grd. Sch. Med., Kyoto Univ.*)

1Pos020* **高濃度リガンド条件による蛋白質-リガンド結合部位および経路の効率的探索**
Accurate and efficient protein-ligand docking method using all-atom molecular dynamics at high concentration of ligands
Chika Sato[1], Akio Kitao[1,2] ([1]*Grad. Sch. Front. Sci., Univ. Tokyo*, [2]*IMCB, Univ. Tokyo*)

1Pos021 **自由エネルギー変分原理を用いたタンパク - リガンド間相対的結合自由エネルギー計算の DHFR-TMP 系への応用**
Calculation of relative binding free energy between DHFR-TMP system on the basis of free energy variational principle
Naoto Nishimura (*Grad. Sch. bioinfo., Univ. Ritsumeikan*)

蛋白質：構造機能相関 / Protein: Structure & Function

1Pos022 **巨大ヘモグロビン酸素解離中間体の X 線結晶構造と分光学的解析**
Crystallographic and spectroscopic analysis of the oxygen-dissociation intermediate of the giant hemoglobin
Nobutaka Numoto[1], Taro Nakagawa[2], Nobutoshi Ito[1], Yoshihiro Fukumori[3], Kunio Miki[4] ([1]*Med. Res. Inst., Tokyo Med. & Dent. Univ.*, [2]*Nagahama Inst. of Bio-Sci. & Tech.*, [3]*Coll. of Sci. & Eng., Kanazawa Univ.*, [4]*Grad. Sch. of Sci., Kyoto Univ.*)

1Pos023 **Structural analysis of Calredoxin from *Chlamydomonas reinhardtii***
Ratana Charoenwattanasatien[1,2], Risa Mutoh[2], Hideaki Tanaka[2], Takashi Matsumoto[3], Takashi Oda[4], Mamoru Sato[4], Michael Hippler[5], Genji Kurisu[1,2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Inst. Protein Res, Osaka Univ.*, [3]*Rigaku*, [4]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*, [5]*Inst. Plant Biol. and Biotech., Univ. of Munster*)

1Pos024 **分子動力学による溶液中 *Agaricus brasiliensis* 由来 β-グルカンの構造解析**
Conformational analysis of β-glucans from *Agaricus brasiliensis* revealed by molecular dynamics in solution
Yoshitaka Matsumura[1], Kodai Inoue[1], Makoto Suminokura[1], Mikako Kubo[1], Mariko Demura[1], Takayuki Ichioka[1], Yasumasa Morimoto[1], Mitsuru Tashiro[2], Ken-ichi Ishibashi[3], Naohito Ohno[3], Masaki Kojima[1] ([1]*Sch. of Life Sci., Tokyo Univ. of Pharm. and Life Sci.*, [2]*Dept. of Chem., Coll. of Sci. and Tech., Meisei Univ.*, [3]*Sch. of Pharm., Tokyo Univ. of Pharm. and Life Sci.*)

1Pos025 **Aβ conformation on a hydrophilic/hydrophobic interface by molecular dynamics simulations**
Satoru Itoh[1,2], Hisashi Okumura[1,2] ([1]*IMS*, [2]*Sokendai*)

1Pos026 **分子動力学シミュレーションを用いた四量体型サルコシン酸化酵素における生成物の選択的移動の解明**
Selective transport of product in heterotetrameric sarcosine oxidase by molecular dynamics simulation
Go Watanabe[1], Takami Saito[2], Daisuke Nakajima[1], Akinori Hiroshima[1], Haruo Suzuki[1], Shigetaka Yoneda[1] ([1]*Sch. Sci., Kitasato Univ.*, [2]*Grad. Sch. Sci., Kitasato Univ.*)

1Pos027 **レオニン合成酵素における生成物支援機構の理論的解明**
Theoretical elucidation on the product assisted catalysis of threonine synthase
Yuzuru Ujiie[1], **Mitsuo Shoji**[1], Ryuhei Harada[1], Takeshi Murakawa[2], Yasuteru Shigeta[1], Hideyuki Hayashi[2] ([1]*Univ. of Tsukuba*, [2]*Osaka Medical College*)

1Pos028 **Hybrid ab initio molecular dynamical simulation of cytochrome c oxidase: Mechanisms of structural changes by dynamical ligand recognition**
Ryuichiro Terada, Kang Jiyoung, Masaru Tateno (*Grad. Sch. Sci., Univ. Hyogo*)

1Pos029 **スレオニル tRNA 合成酵素におけるアミノ酸選択機構の理論的研究**
Theoretical study on the molecular mechanism of amino-acid selection in threonyl-tRNA synthetase
Yoshiharu Mori[1], Hisashi Okumura[1,2] ([1]*IMS*, [2]*SOKENDAI*)

1Pos030 **量子化学計算ソフトウェアへのレプリカ交換法の導入**
The implementation the Replica-Exchange Umbrella Sampling in the quantum mechanical simulation packages
Shingo Ito[1], Yuko Okamoto[3], Stephan Irle[1,2] ([1]*Department of Chemistry, Graduate School of Science, Nagoya University*, [2]*Institute of Transformative Bio-Molecules (WPI-ITbM), Nagoya University*, [3]*Department of Physics, Graduate School of Science, Nagoya University*)

1Pos031 **Hydrogenase : ab initio quantum mechanics study of oxygen-tolerance mechanism**
Jae Hyun Kim, Jiyoung Kang, Masaru Tateno (*Graduate school of Life Science,University of Hyogo*)

1Pos032* アルカン合成酵素ADのNMRと分子動力学シミュレーションによるダイナミクス解析
Structural dynamics of an alkae synthase, AD, studied by NMR and molecular dynamics simulations
Yuma Suematsu[1], Yuji O. Kamatari[2], Yuuki Hayashi[1], Munehito Arai[1] ([1]*Dept. of Life Sci., Univ. of Tokyo*, [2]*Life Sci. Res. Center, Gifu Univ.*)

1Pos033* エンド-1,3-β-グルカナーゼ触媒ドメインの構造ダイナミクス解析
Structural dynamics analysis of catalytic domain of endo-1,3-β-glucanase
Ayako Miki[1], Satomi Inaba[1], Kazumasa Sakurai[2], Masayuki Oda[1] ([1]*Grad. Sch. of Life and Environ. Sci., Kyoto Pref. Univ.*, [2]*High Pressure Protein Res. Center, Kindai Univ.*)

1Pos034 カルモジュリン結合ペプチドはミッドカインに親和性を示し、その一アミノ酸変異による立体構造変化が結合親和性の向上に寄与する
Secondary structure change by single alanine substitution in Calmodulin-binding peptide improved the binding affinity with Midkine
Hidenao Arai, Koji Matsuoka, Naoto Nemoto (*Grad. Sch. Sci. Eng., Saitama Univ.*)

1Pos035 ラン藻由来アルカン合成酵素のアラニンスキャン変異解析
Alanine scanning mutagenesis of a cyanobacterial alkane synthase
Keigo Shimba, Fumitaka Yasugi, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

1Pos036* 複数のエピトープを認識するG2の1本鎖抗体の構造解析
Structural analysis of a single-chain Fv antibody of G2 that recognizes multiple epitopes
Daiki Usui[1], Yuji O. Kamatari[2], Satomi Inaba[1], Masayuki Oda[1] ([1]*Grad. Sch. of Life and Environ. Sci., Kyoto Pref. Univ.*, [2]*Life Sci. Res. Ctr., Gifu Univ.*)

1Pos037 複数の抗原を特異的に認識する抗体G2の3つめのエピトープの同定
Identification of the third epitope recognized by multispecific antibody G2
Md. Nuruddin Mahmud[1], Yasuo Inoshima[1], Naotaka Ishiguro[1], Yuji O. Kamatari[2] ([1]*United Grad. Sch. Veterinary Sci., Gifu Univ.*, [2]*Life Sci. Res. Ctr, Gifu Univ.*)

1Pos038* ラン藻でのアルカン合成に必要な2つの酵素間の結合部位の探索
Search for the binding sites between two enzymes essential for cyanobacterial alkane biosynthesis
Mari Chang[1], Keigo Shimba[2], Yuuki Hayashi[2], Munehito Arai[1,2] ([1]*Department of Physics, University of Tokyo*, [2]*Department of Life Sciences, University of Tokyo*)

1Pos039* ケモカイン受容体制御因子FROUNTー制御化合物間の立体構造情報に基づく相互作用解析
Structure-based analyses of the interaction between the chemokine receptor-regulator FROUNT and anti-inflammatory compounds
Soichiro Ezaki[1], Sosuke Yoshinaga[1], Norihito Ishida[1], Mitsuhiro Takeda[1], Kaori Yunoki[1], Yuya Terashima[2], Etsuko Toda[2], Kouji Matsushima[2], Hiroaki Terasawa[1] ([1]*Faculty of Life Sciences, Kumamoto University*, [2]*Graduate School of Medicine, The University of Tokyo*)

1Pos040 CD28とSH2ドメインとの相互作用における構造熱力学的解析
Structural and thermodynamic analysis of interactions between CD28 and SH2 domains
Satomi Inaba[1], Nobutaka Numoto[2], Shuhei Ogawa[3], Hisayuki Morii[4], Teikichi Ikura[2], Ryo Abe[3], Nobutoshi Ito[2], Masayuki Oda[1] ([1]*Grad. Sch. Life Environ. Sci., Kyoto Pref. Univ.*, [2]*Med. Res. Inst., Tokyo Med. Dent. Univ. (TMDU)*, [3]*Res. Inst. Biomed. Sci., Tokyo Univ. Sci.*, [4]*College Liberal Arts Sci., Tokyo Med. Dent. Univ.*)

1Pos041 T細胞受容体による特異的および交差反応的な抗原認識機構の解明
Analyses of the structural mechanisms of specific and crossreactive recognitions of peptide-MHC by TCRs
Yuko Tsuchiya[1], Yoshiki Namiuchi[2], Hiroshi Wako[3], Hiromichi Tsurui[4] ([1]*IPR, Osaka Univ.*, [2]*QBiC, RIKEN*, [3]*Sch. of Social Sci., Waseda Univ.*, [4]*Sch. of Med., Juntendo Univ.*)

1Pos042 レプリカ交換分子動力学シミュレーションによって明らかになったHP1αCD/histone H3 tail複合体形成の仕組み
Mechanism of the complex formation of HP1αCD/histone H3 tail revealed by the replica-exchange molecular dynamics simulations
Satoshi Omori[1], Nobuto Hashiguchi[2], Kei Moritsugu[2], Yoshifumi Nishimura[2], Akinori Kidera[2] ([1]*GSIS, Tohoku Univ.*, [2]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*)

1Pos043 Go-likeモデルを用いたプラストシアニンとシトクロムf複合体の構造安定性に関する理論的研究
Theoretical study on the structural stability of plastocyanin and cytochrome f complex by using Go-like model
Satoshi Nakagawa, Shogo Kinoshita, Makoto Wada, Kazutomo Kawaguchi, Hidemi Nagao (*Grad. Sch. Nat. Sci. Tech., Kanazawa Univ.*)

1Pos044 粗視化分子動力学シミュレーションを用いたMVMの生物物理学的特性の解明
Elucidating biophysical properties of the Minute Virus of Mice capsid: Coarse-Grained Molecular simulation
Koji Ono, Shoji Takada (*Dept. Biophys., Grad. Sch. Sci., Kyoto Univ.*)

1Pos045 粗視化力場を用いたタンパク質-リガンド結合シミュレーション：結合経路上の変異がリガンド結合に及ぼす影響の解析
Coarse-grained simulations of protein-ligand binding: effect of mutations near the ligand-binding pathways
Tatsuki Negami, Tohru Terada, Kentaro Shimizu (*Grad. Sch. of Agri. and Life. Sci., The Univ. of Tokyo*)

1Pos046 タンパク質-リガンドドッキング計算における最適パラメータの同定による分子設計の拡張
Identification of optimal parameter values in ligand-receptor docking calculation to extend applicability
Takuya Sumi[1], Hiroshi Yamaguchi[2], Ryuichiro Terada[1], Jiyoung Kang[1], Masaru Tateno[1] ([1]*Grad. Sch. Sci., Univ. of Hyogo*, [2]*Grad. Sch. Med., Nagoya Univ.*)

1Pos047 **Analysis of protein complexes structures towards rational design of inhibitors of Protein-protein interactions (PPIs)**
Daisuke Kobayashi, George Chikenji (*Nagoya Univ.*)

蛋白質：物性 / Protein: Property

1Pos048* **天然タンパク質の立体構造物性に関する統計解析**
Statistical analysis on the structural properties of native proteins
Hidenobu Kawai, Daisuke Takahashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

1Pos049 **膜貫通βバレルにおけるβストランドのねじれと曲りに関する解析**
Twisting and bending of β-strand in the transmembrane β-barrel
Nobuaki Kikuchi, Shinichi Ebisawa, Yuka Watanabe, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. Bioinfo., Grad. Sch. Eng., Soka Univ.*)

1Pos050 **球状蛋白質の構造的性質とフォールディング速度との相関**
Relationship between the Folding Rate and Structure-based Properties of Globular Proteins
Balachandran Manavalan[2,3], **Kunihiro Kuwajima**[1,2], Jooyoung Lee[2,3] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Comput. Sci., KIAS*, [3]*Center In-Silico Protein Sci., KIAS*)

1Pos051* **金属イオンがα-ラクトアルブミンのフォールディング中間体の熱力学的安定性に与える影響**
Effects of metal ions on thermodynamic stability of folding intermediates of α-lactalbumin
Reina Shinozaki, Michio Iwaoka (*Dep. Chem., Sch. Sci., Tokai Univ.*)

1Pos052 **ウマアポミオグロビンのpHによるフォールディング機構**
Mechanism of pH-induced folding of horse apomyoglobin studied by a statistical mechanical model
Takuya Mizukami, Yosuke Sakuma, **Kosuke Maki** (*Grad. Sch. Sci., Nagoya Univ.*)

1Pos053 **SWAXS解析によるトレハロースがミオグロビン構造へ与える効果の解明**
SWAXS analysis on effect of trehalose on myoglobin structure
Satoshi Ajito, Mitsuhiro Hirai (*Grad. Sch. Sci., Univ. Gunma*)

1Pos054* **Conformational Diversity in the Intrinsically Disordered HIV-1 Tat Protein induced by Zinc and pH**
Tomoko Kunihara, Yuuki Hayashi, Hisashi Kudo, Hidenobu Kawai, Yoshiki Oka, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

1Pos055 **ＮＭＲ測定と同期したアゾベンゼン架橋剤の光異性化反応によるＧＢ１タンパク質のフォールディング操作**
Manipulating Protein GB1 Folding Using Photoisomerization of an Azobenzene Cross-Linker Synchronously with NMR Observation
Toshio Nagashima, Keisuke Ueda, Toshio Yamazaki (*RIKEN CLST*)

1Pos056 **レプリカ交換分子動力学シミュレーションによるpHに依存したポリグルタミン酸の構造変化の研究**
Replica-exchange molecular dynamics study of pH dependent structural changes of polyglutamic acids
Ryosuke Iwai[1], Tetsuro Nagai[2], Kota Kasahara[3], Takuya Takahashi[3] ([1]*Grad. Sci. Life Sci., Ritsumeikan Univ.*, [2]*Dept. of Phys., Nagoya Univ.*, [3]*Coll. Life. Sci., Ritsumeikan Univ.*)

1Pos057* **Secondary structural change of glucagon during fibril formation process with DMPC lipid bilayers as revealed by ^{13}C solid-state NMR**
Kazumi Haya, Izuru Kawamura, Akira Naito (*Grad. Sch. Eng., Yokohama Natl. Univ.*)

1Pos058 **天然変性タンパク質c-Junと転写コアクチベータCBPのKIXドメインの相互作用**
Interaction of the intrinsically disordered c-Jun with the KIX domain of the transcriptional coactivator CBP
Satoru Yoshizaki[1], Tomoko Kunihara[2], Yuuki Hayashi[1,2], Munehito Arai[1,2] ([1]*Dept. Integrated Sci., Univ. Tokyo*, [2]*Dept. Life Sci., Univ. Tokyo*)

1Pos059 **ヒトα-シヌクレインのダイナミクスとアミロイド線維形成のしやすさの関係**
Relationship between the dynamics of human α-synuclein and its propensity to form amyloid fibrils
Fumiaki Kono[1], Tatsuhito Matsuo[1], Taiki Tominaga[2], Kaoru Shibata[3], Katsuya Araki[4], Hideki Mochizuki[4], Satoru Fujiwara[1] ([1]*QuBS, QST*, [2]*CROSS-Tokai*, [3]*J-PARC Center*, [4]*Osaka Univ. Grad. Sch. Med.*)

1Pos060* **中間状態で阻害するフィブリノーゲンのアミロイド線維化抑制効果**
Fibrinogen inhibits amyloid fibrillation by stopping at the stage of intermediates
Taiki Akai (*Grad. Sch. of Sci., Kobe Univ.*)

1Pos061 **ポリグルタミン酸の構造特性に関する陽溶媒における効率的な分子動力学による研究**
Structural feature of polyglutamic acids studied by enhanced molecular dynamics with explicit solvent
Tetsuro Nagai[1], Ryosuke Iwai[2] ([1]*Dept. of Phys., Nagoya Univ.*, [2]*Grad. Sci. Life Sci., Ritsumeikan Univ.*)

1Pos062 **Control and biophysical characterization of soluble protein oligomers using short peptide tags**
Md. Golam Kabir[1], Mohammad Monirul Islam[2], Tomonori Saotome[1], Yutaka Kuroda[1] ([1]*Tokyo Univ. Agri. Eng. Kuroda lab Biotechnology and Life science*, [2]*University of Chittagong, BANGLADESH*)

1Pos063* **複数のアミロイド性ペプチドを含む複雑な系におけるアミロイド線維形成**
Amyloid Fibrillation in Promiscuous Systems Containing Various Amyloidogenic Peptides
Hiroya Muta[1], Masatomo So[1], Kazumasa Sakurai[2], Yuji Goto[1] ([1]*IPR, Osaka Univ.*, [2]*High Pressure Protein Res. Cent., Inst. for Advanced Tech., Kinki Univ.*)

蛋白質：機能 / Protein: Function

1Pos064 **光合成生物及び非光合成生物由来ferredoxin-NADPH酸化還元酵素触媒反応の可逆性**
Reversibility of the redox reactions catalyzed by ferredoxin-NADPH oxidoreductases from phototroph and heterotroph
Daisuke Seo (*Nat. Sci. Tec., Kanazawa Univ.*)

1Pos065 **高活性型 CaMKIδ(1-299)のキナーゼ研究への活用**
Application of high active form CaMKIδ(1-299) for the study of protein kinase
Yukako Senga[1], Kazutoshi Akizuki[2], Syouichi Katayama[3], Yasushi Shigeri[4], Isamu Kameshita[2], Atsuhiko Ishida[5], Noriyuki Sueyoshi[2] ([1]*BMRI, AIST*, [2]*Dept. Appl. Biol. Sci., Fac. Agr., Kagawa Univ.*, [3]*Dept. of Pharm., Coll. of Pharm., Ritsumeikan Univ.*, [4]*HRI, AIST*, [5]*Grad. Sch. Integr. Arts Sci., Hiroshima Univ.*)

1Pos066 **システイン残基修飾によるピルビン酸デヒドロゲナーゼキナーゼ 2 の動的構造変化**
Dynamical structural changes of pyruvate dehydrogenase kinase 2 by modification of cysteine
Kyoka Kaiya[1], Yasuhiro Fuzino[2], Katumi Doi[3], Etuko Nishimoto[3], Yasuaki Hiromasa[3] ([1]*Grad. Sch. Bioresour. Bioenviron. Sci., Kyushu Univ.*, [2]*Div. Arts and Science, Kyushu Univ.*, [3]*Fac. Agr., Kyushu Univ.*)

1Pos067 **光照射を利用した硫酸還元菌由来[NiFe]ヒドロゲナーゼの活性化機構の FT-IR 研究**
FT-IR studies on the activation mechanism of [NiFe] hydrogenase from *Desulfovibrio vulgaris* Miyazaki F using light irradiation
Hulin Tai[1,2], Liyang Xu[1], Seiya Inoue[3], Koji Nishikawa[3], Yoshiki Higuchi[2,3], Shun Hirota[1,2] ([1]*Grad. Sch. Mat. Sci., NAIST*, [2]*CREST, JST*, [3]*Grad. Sch. Life Sci., Univ. Hyogo*)

1Pos068 **アフリカツメガエル由来(6−4)光回復酵素の 4 番目の電子移動トリプトファンの解析**
Analysis of the fourth electron-transferring tryptophan in Xenopus laevis (6-4) photolyase
Takahiro Kanda, Junpei Yamamoto, Shigenori Iwai (*Grad. Sch. Eng of Sci., Univ., Osaka*)

1Pos069 **分子シミュレーションによるエピジェネティックな酵素に対する基質の結合選択性の研究**
Study for the Ligand Binding Selectivity of Epigenetic Enzymes by using Molecular Simulations
Shuichiro Tsukamoto[1,3], Yoshitake Sakae[1], Yukihiro Itoh[2,3], Takayoshi Suzuki[2,3], Yuko Okamoto[1,3,4,5,6] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Grad. Sch. Med. Sci., Kyoto Pref. Univ. Med.*, [3]*JST-CREST*, [4]*Struc. Bio. Res. Cen., Grad. Sch. Sci., Nagoya Univ.*, [5]*Cen. Comput. Sci., Grad. Sch. Eng., Nagoya Univ.*, [6]*Info. Tech. Cen., Nagoya Univ.*)

1Pos070 **分子動力学シミュレーションによる MutS の homoduplex DNA と mismatch DNA の認識メカニズム解析**
Analysis of recognition of homoduplex and mismatched DNA by MutS by MD simulations
Hisashi Ishida, Atsushi Matsumoto (*National Institutes for Quantum and Radiological Science and Technology, Molecular Modeling and Simulation Group*)

1Pos071 **ONIOM 法を用いたアデニル酸キナーゼ反応機構に関する計算化学的研究**
Computational Study on the Reaction Mechanism of Adenylate Kinase with ONIOM method
Kenshu Kamiya (*Dept. of Phys., Sch. of Sci., Kitasato Univ.*)

1Pos072 **Evolutionary optimisation of elastic network structures: Models of allosteric proteins**
Holger Flechsig (*Hiroshima University*)

蛋白質工学 / Protein: Engineering

1Pos073 **バクテリオロドプシンの構造・機能特性に対する物理架橋 PVA ハイドロゲル中への固定の影響**
Effects of Immobilization of Bacteriorhodopsin with Poly(Vinyl Alcohol) Hydrogels on Its Structural and Functional Properties
Hikaru Tanaka[1], Yasunori Yokoyama[1], Hiroshi Takahashi[2], Takashi Kikukawa[3], Masashi Sonoyama[2], Koshi Takenaka[1] ([1]*Grad. Sch. Eng., Nagoya Univ.*, [2]*Grad. Sch. Sci. & Tech., Gunma Univ.*, [3]*Grad. Sch. Life. Sci., Hokkaido Univ.*)

1Pos074 **Thermo-induced phase separation dynamics of a biopolymer model on water-in-oil droplets**
Keitaro Horii[1], Kazunari Yoshida[2], Azusa Saito[3], Akito Takashima[1], Izumi Nishio[1] ([1]*Grad. Sch. of Sci. & Eng., Univ. of Aoyama*, [2]*Grad. Sch. of Med., Univ. of Tokyo*, [3]*Grad. Sch. of Sci. & Eng., Univ. of Yamagata*)

1Pos075 **タンパク質中に生成した金ナノクラスターの発光特性**
Emission property of Au Nanoclusters Formed in Protein
Takuma Dezawa[1], Hamza Al-kindi[1], Izabela Rzeznicka[2], Hiroshi Fukumura[1], Yutaka Shibata[1] ([1]*Grad. Sch. Sci., Univ. Tohoku*, [2]*Grad. Sch. Eng. & Sci., ShibauraInst Tech.*)

1Pos076 **青色光によるバクテリオロドプシン色素再生に対する脂質膜相転移の影響**
Effects of Lipid Phase Transition on Chromophore Regeneration of Bleached Bacteriorhodopsin in Bilayer Vesicles by Blue Light Irradiation
Shunsuke Yano[1], Kentarou Motegi[1], Hikaru Tanaka[1], Yasunori Yokoyama[1], Masashi Sonoyama[2], Koshi Takenaka[1] ([1]*Grad. Sch. Eng., Nagoya Univ.*, [2]*Grad. Sch. Sci. & Tech., Gunma Univ.*)

1Pos077 **ATP 結合タンパクのゼロからのデザイン**
Design of ATP-binding protein from scratch
Kengo Nakamura[1,2], Takahiro Kosugi[1,2], Nobuyasu Koga[1,2,3] ([1]*IMS CIMoS*, [2]*SOKENDAI*, [3]*JST PRESTO*)

1Pos078* **合理的設計による抗体精製用リガンド FPA の開発**
Rational design of FPA, a ligand for antibody purification
Yoshiki Oka[1], Taihei Sawada[1], Takahiro Watanabe[1], Hisashi Kudo[1], Manami Wada[1], Hidenobu Kawai[1], Mari Chang[2], Yuuki Hayashi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Phys., Univ. Tokyo*)

1Pos079 **新規酸化還元応答蛍光タンパク質の作成**
New design of redox sensitive fluorescence proteins
Kazunori Sugiura[1,2], Akiyoshi Higo[1,2], Toru Hisabori[1,2] ([1]*CLS, Tokyo Tech.*, [2]*CREST, JST*)

1Pos080 自然界のタンパク質を大きく改造して創るヘム結合タンパク質
Computational design of heme-binding proteins by largely remodeling naturally occurring proteins
Yoshitaka Moriwaki[1], Nobuyasu Koga[1,2] ([1]*CiMoS, IMS*, [2]*JST, PRESTO*)

1Pos081 **Selection of Ru(bpy)32+ motifs from a randomized peptide library**
Marziyeh Karimiavargani[1], Seiichi Tada[2], Noriko Minagawa[2], Takuji Hirose[2], Yoshihiro Ito[2], Takanori Uzawa[2] ([1]*Graduate school of Science and Engineering, Saitama University*, [2]*Nano Medical Engineering Laboratory, RIKEN*)

1Pos082 複数の遺伝子群の共進化を可能とする完全試験管内選択系の開発
Development of a totally in vitro selection system for co-evolution of plural genes
Asuka Ueki, Kei Fujiwara, Nobuhide Doi (*Grad. Sch. Sci. Tech., Keio Univ.*)

1Pos083* ファージディスプレイ法を用いたタンパク質デザインへの応用を目指した蛍光一分子ソーターの開発
Development of a single-molecular sorting system based on fluorescence detection for protein design using phage display method
Yuki Shimizu[1,2], Naoki Mikoshiba[1,3], Seiji Sakamoto[1,2], Hiroyuki Oikawa[1,2,3], Kiyoto Kamagata[1,2,3], Takehiko Wada[1,2], Satoshi Takahashi[1,2,3] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Sci., Tohoku Univ.*, [3]*Grad. Sch. Life Sci., Tohoku Univ.*)

1Pos084 進化分子工学に向けたスクリーニングシステムの開発
Development of an integrated femtoliter chamber array system for directed evolution of protein molecules
Yi Zhang, Hiroto Kizoe, Yoshihiro Minagawa, Kazuhito Tabata, Hiroyuki Noji (*Grad. Sch. Eng., Univ. Tokyo*)

ヘム蛋白質 / Heme proteins

1Pos085 チトクロム *c* とチトクロム酸化酵素の複合体構造が示す新しいタンパク質間相互作用様式
Complex structure of cytochrome *c* and cytochrome *c* oxidase shows a novel inter-protein interaction mode
Satoru Shimada[1], Kyoko Shinzawa-Itoh[1], Junpei Baba[1], Shimpei Aoe[1], Atsuhiro Shimada[1], Eiki Yamashita[2], Jiyoung Kang[1], Masaru Tateno[1], Shinya Yoshikawa[1], Tomitake Tsukihara[1,2] ([1]*Picobiology Inst., Grad. Sch. Life Sci., Univ. Hyogo*, [2]*Inst. Protein Res., Osaka Univ.*)

1Pos086 蛍光偏光解消度を利用したシトクロム c-シトクロム c 酸化酵素間電子伝達複合体形成における相互作用解析
Interaction analysis of electron transfer complex formation between cytochrome c-cytochrome c oxidase using fluorescence anisotropy
Hiroshi Kagaya[1], Wataru Sato[1], Takeshi Uchida[1,2], Kyoko Itoh－Shinzawa[3], Shinya Yoshikawa[3], Koichiro Ishimori[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*, [3]*Grad. Sch. of Life Sci., Univ. of Hyogo*)

1Pos087 チトクロム c 酸化酵素の水素結合状態変化の酸素還元反応への影響
The effect of the hydrogen bond network on the oxygen reduction of cytochrome c oxidase
Yudai Aoyagi, Tatsuhito Nishiguchi, Kyoko Shinzawa-Itoh, Shinya Yoshikawa, Satoru Nakashima, Takashi Ogura (*Grad. Sch. Lif. Sci., Univ. Hyogo*)

1Pos088 時間分解共鳴ラマン分光法によるチトクローム酸化酵素の共役機構
Coupling mechanism of Cytochrome c oxidase studied by time-resolved resonance Raman spectroscopy
Satoru Nakashima, Yoshiyuki Nakagawa, Kyoko Itoh-Shinzawa, Shinya Yoshikawa, Takashi Ogura (*Grad. Sch. Sci., Univ. Hyogo*)

1Pos089* シトクロム c–シトクロム c 酸化酵素間の電子伝達複合体形成における脱水和の機能的意義
Functional significance of dehydration for formation of electron transfer complex between cytochrome c and cytochrome c oxidase
Wataru Sato[1], Kyoko Shinzawa-Itoh[3], Takeshi Uchida[2], Peter Brzezinski[4], Shinya Yoshikawa[3], Koichiro Ishimori[2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*, [3]*Grad. Sch. of Life Sci., Hyogo Univ.*, [4]*Dept. of Biochem. and Biophys., Stockholm Univ.*)

1Pos090 呼吸鎖ヘム・銅酸素還元酵素スーパーファミリーのプロトン輸送経路の構造解析
Structural analysis of the proton transfer pathway in respiratory heme-copper oxygen reductase superfamily
Kazumasa Muramoto (*Grad. Sch. of Life Sci., Univ. of Hyogo*)

1Pos091 ナノリットルフロー時間分解可視・赤外分光法を用いた一酸化窒素還元酵素の短寿命反応過渡種の計測
Detection of Short-Lived Reaction Species of Nitric Oxide Reductase Using Nanoliter-Flow Time-Resolved Visible/IR Spectroscopy
Hanae Takeda[1], Tetsunari Kimura[2], Shoko Ishii[1], Takehiko Tosha[3], Yoshitsugu Shiro[1,3], Minoru Kubo[3,4] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Grad. Sch. Sci., Kobe Univ.*, [3]*SPring-8 Center, RIKEN*, [4]*JST PRESTO*)

1Pos092 金電極上に固定化した一酸化窒素還元酵素の電気化学的還元活性
Electrochemical reduction activity of nitric oxide reductase immobilized on Au electrodes
Shogo Nakagawa[1], Masaru Kato[1,2], Takehiko Tosya[3], Ichizo Yagi[1,2] ([1]*Grad. Sch. Environ. Sci., Hokkaido Univ.*, [2]*Faculty of Environ. Earth Sci., Hokkaido Univ.*, [3]*RIKEN*)

1Pos093 ナノディスクに再構成した Vibrio cholerae 由来シトクロム cbb3 の構造，機能的評価
Structural and functional characterization of nanodisc-reconstituted cytochrome cbb3 oxidase from Vibrio cholerae
Masanao Inoue[1], Akihiro Shibata[1], Mizue Imai[1], Takeshi Uchida[2], Kazumasa Muramoto[3], Noritsugu Shiro[3], Shinya Furukawa[3], Koichiro Ishimori[2] ([1]*Graduate School of Science, Hokkaido University*, [2]*Graduate School of Science, Hokkaido University*, [3]*Graduate School of Life Science, Hyogo University*)

1Pos094* シトクロム *c* とカルジオリピン含有バイセルの相互作用の溶液 NMR 解析
Solution NMR characterization of the interaction between cyt *c* and cardiolipin-incorporated bicelles
Hisashi Kobayashi, Satoshi Nagao, Shun Hirota (*Grad. Sch. Mat. Sci., Nara Inst. Sci. Tech.*)

膜蛋白質 / Membrane proteins

1Pos095* **Molecular mechanism of the ATP-dependent modulation of the Mg^{2+} channel MgtE for Mg^{2+} homeositasis**
Atsuhiro Tomita[1], Mingfeng Zhang[2], Hironori Takeda[3], Fei Jin[2], Tatsuro Maruyama[4], Masanori Osawa[4], Ryuichiro Ishitani[1], Ichio Shimada[4], Zhiqiang Yan[2], Motoyuki Hattori[2], Osamu Nureki[1] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Sch. Life Sci., Fudan Univ.*, [3]*Fac. Life Sci., Kyoto Sangyo Univ.*, [4]*Grad. Sch. Pharm., Univ. Tokyo*)

1Pos096 **チトクロム酸化酵素の高分解能結晶構造から明らかとなった高効率プロトンポンプ機構**
High-resolution crystal structure of cytochrome c oxidase reveals the mechanism of highly efficient proton pumping
Atsuhiro Shimada[1], Naomine Yano[1], Kazumasa Muramoto[1], Eiki Yamashita[2,3], Kyoko Shinzawa-Itoh[1], Tomitake Tsukihara[1,2], Shinya Yoshikawa[1] ([1]*Picobiol. Inst., Univ. Hyogo*, [2]*Inst. Protein Res., Osaka Univ.*, [3]*SPring-8, RIKEN*)

1Pos097 **立体構造に基づいた原核生物由来ナトリウムチャネルにおける選択性フィルターの変異体解析**
Structural and mutational analysis of the selectivity filter of prokaryotic sodium channel
Katsumasa Irie[1,2], Yukari Haga[2], Shun Nakamura[2], Yoshinori Fujiyoshi[1,2] ([1]*CeSPI, Nagoya Univ.*, [2]*Grad. Sch. Pharm. Nagoya Univ.*)

1Pos098 **X 線 1 分子動態計測法への試料温度ジャンプシステムの導入**
The introduction of temperature-jump system to the Diffracted X-ray Tracking (DXT)
Hirofumi Shimizu, Masayuki Iwamoto (*Univ. Fukui. Fac. Med. Sci.*)

1Pos099 **原子分解能のシミュレーションによって明らかになった ADP/ATP 膜輸送体の交互アクセス機構**
Deciphering Alternating Access Mechanism of a Mitochondrial ADP/ATP Membrane Transporter with Atomistic Simulations
Koichi Tamura[1], Shigehiko Hayashi[2] ([1]*RIKEN AICS*, [2]*Grad. Sch. Sci., Kyoto Univ.*)

1Pos100 **サーモフィリックロドプシンの極めて高い熱安定性に対する統計熱力学**
Statistical Thermodynamics for Remarkably High Thermal Stability of Thermophilic Rhodopsin
Satoshi Yasuda[1,2,3], Yuta Kajiwara[4], Takeshi Murata[1,2,5], Masahiro Kinoshita[3] ([1]*Grad. Sch. Sci., Chiba Univ.*, [2]*MCRC, Chiba Univ.*, [3]*IAE, Kyoto Univ.*, [4]*Grad. Sch. Ener. Sci., Kyoto Univ.*, [5]*PRESTO*)

1Pos101* **脂質二分子膜中におけるポア形成ペプチドの分子メカニズム解明に向けたモデルペプチドのチャネル電流測定**
Systematically designed model pore-forming peptides study on molecular mechanism in lipid bilayers using channel current recording
Yusuke Sekiya[1], Hirokazu Watanabe[1], Kenji Usui[2], Ryuji Kawano[1] ([1]*Tokyo Univ. Agr. Tech.*, [2]*Konan Univ.*)

1Pos102* **補酵素フラビンの置換による微生物外膜シトクロムのプロトン移動の発見**
Proton transfer reaction in outer-membrane flavocytochromes revealed by replacement of flavin cofactor
Yoshihide Tokunou[1], Kazuhito Hashimoto[2], Akihiro Okamoto[2] ([1]*Dept. of Appl. Chem., Univ. Tokyo*, [2]*Natl. Inst. for Mater. Sci.*)

1Pos103 **Toward the elucidation of structure/function relationship of transport proteins**
Naoki Soga[1], Rikiya Watanabe[1,2], Hiroyuki Noji[1] ([1]*Dept. of App. Chem., The University of Tokyo*, [2]*PRESTO, JST*)

1Pos104 **生細胞における G タンパク質共役型受容体の拡散・機能連関の比較解析**
Comparative analysis of diffusion-function relationship of G protein-coupled receptors on the living cell surface
Masataka Yanagawa[1], Michio Hiroshima[1,2], Yuichi Togashi[3], Takahiro Yamashita[4], Yoshinori Shichida[4], Masayuki Murata[5], Masahiro Ueda[2,6], Yasushi Sako[1] ([1]*Cellular Informatics Lab., RIKEN*, [2]*QBiC, RIKEN*, [3]*RcMcD, Hiroshima Univ.*, [4]*Dept. Biophys., Grad. Sci., Kyoto Univ.*, [5]*Dept. Life Sci., Grad. Arts and Sci., Univ. Tokyo*, [6]*Grad. Frontier Biosci., Osaka Univ.*)

1Pos105 **Trafficking of endocytic PAR-1 carrier vesicles in cancer cell**
Seohyun Lee[1], Kohsuke Gonda[2], Hideo Higuchi[1] ([1]*Graduate school of science, University of Tokyo*, [2]*Graduate school of medicine, Tohoku university*)

1Pos106 **ミトコンドリア内膜タンパク質のマイクロ流路デバイスによる実時間解析**
The real-time analysis of respiratory chain complex I on mitochondrial inner membrane by using microfluidic device
Yuji Kimura, Sayaka Kazami, Yu Hashimoto, Hiroyasu Itoh (*Tsukuba Research Center, Hamamatsu Photonics KK*)

1Pos107 **細胞シグナリングに関与する上皮成長因子受容体クラスターのコレステロールを介した形成メカニズム**
Cholesterol Mediated Mechanism for Signaling Cluster Formation of Epidmal Growth Factor Receptorer
Michio Hiroshima[1,2], Masahiro Ueda[1], Yasushi Sako[2] ([1]*RIKEN QBiC*, [2]*RIKEN Cellular Informatics Laboratory*)

1Pos108 **ヨクトリットルスケール空間において粘性が DNA の運動に与える影響の評価**
Evaluation of viscosity effect on DNA movement in yocto (10^{-24}) liter space
Masaki Matsushita, Hirokazu Watanabe, Masayuki Ohara, Ryuji Kawano (*Life Sci. Biotech., Tokyo Univ. Agri. Tech.*)

核酸結合蛋白質 / Nucleic acid binding proteins

1Pos109 **ヌクレオソームスライディングの分子機構に関する分子シミュレーション研究**
Molecular Mechanisms of Nucleosome Sliding Revealed by Coarse-Grained Molecular Dynamics Simulation
Toru Niina, Shoji Takada (*Graduate School of Science, Kyoto Univ.*)

1Pos110* **多分子及び一分子測定により解明されたがん抑制タンパク質 p53 の超高速セグメント間移動**
Ultrafast intersegmental transfer of a tumor suppressor p53 investigated by ensemble and single-molecule measurements
Yuji Itoh[1,2], Agato Murata[1,2], Satoshi Takahashi[1,2], Kiyoto Kamagata[1,2] ([1]*IMRAM, Univ. Tohoku*, [2]*Grad. Sch. Sci., Univ. Tohoku*)

1Pos111 **大腸菌非六量体型 DNA ヘリカーゼ UvrD 多量体の 1 分子 FRET イメージング**
Single-molecule FRET imaging of the oligomeric form of the non-hexameric *Escherichia coli* helicase UvrD
Hiroaki Yokota (*Biophotonics lab, GPI*)

1Pos112 Identification of initial ES complex of topoisomerase IIβ and target DNA employing molecular dynamics docking simulation
Kakeru Sakabe, Hiroshi Nisihigami, Jiyoung Kan, Masaru Tateno (*Grad. Sch. Sci., Univ. Hyogo*)

1Pos113* Elongation of Intrinsically Disordered Linker in p53 and the Effects on DNA Binding and Sliding Ability
Dwiky Rendra Graha Subekti[1,2], Agato Murata[1,2], Yuji Ito[1,2], Satoshi Takahashi[1], Kiyoto Kamagata[1] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Sci., Tohoku Univ.*)

1Pos114 Nucleoprotein Filament Assembly Dynamics of Dmc1 and Rad51 Recombinases
Sheng-Yao Lin[1], Wen-Hsuan Chang[1], Chih-Yuan Kao[2], Hung-Yuan Chi[2], Hung-Wen Li[1] ([1]*Department of Chemistry, National Taiwan University, Taipei, Taiwan*, [2]*Institute of Biochemical Sciences, National Taiwan University, Taipei, Taiwan*)

1Pos115 リバースジャイレースによるバブル DNA 超らせん導入の物理機構
Physical mechanism of introducing positive supercoils into bubble DNA by reverse gyrase
Ryota Moritake, Takato Sato, Yuta Suzuki, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

1Pos116 Single molecule FRET measurements of Cas9 conformation change
Kazushi Isomura[1], Shohei Kajimoto[2], Saki Osuka[3], Hiroshi Nishimasu[2], Tomohiro Shima[1], Tomotaka Komori[1], Osamu Nureki[2], Sotaro Uemura[1] ([1]*Uemura lab., Grad. Sch. Sci., Univ. Tokyo*, [2]*Nureki lab., Grad. Sch. Sci., Univ. Tokyo*, [3]*Dep. Bio., Sci., Univ. Tokyo*)

1Pos117* Investigate how mSWI5-SFR1 complex facilitates mRAD51 presynaptic filament formation using single-molecule approaches
Chih-Hao Lu[1], Guan-Chin Su[2], Peter Chi[2], Hung-Wen Li[1] ([1]*Dept. of Chemistry, Nat'l Taiwan Univ.*, [2]*Institute of Biochemical Science, Nat'l Taiwan Univ.*)

1Pos118 粗視化シミュレーションによる障害物存在下での DNA 結合タンパク質挙動研究
The movement of DNA binding protein including obstacles along DNA
Mami Saito[1], Shoji Takada[1], Tsuyoshi Terakawa[2] ([1]*Grad. Sch. Sci., Uni. Kyoto*, [2]*Medical Center, Uni. Columbia*)

核酸：構造・物性 / Nucleic acid: Structure & Property

1Pos119 結晶構造中で観察される DNA 構造ゆらぎの網羅解析
DNA conformational transition inferred from re-evaluation of m|Fo|-D|Fc| electron density maps
Tomoko Sunami[1], Toshiyuki Chatake[2], Hidetoshi Kono[1] ([1]*National Institutes for Quantum and Radiological Science and Technology*, [2]*Kyoto University Research Reactor Institute*)

1Pos120 ＤＮＡ高次構造の捩じれ速度依存性
Dependence of twisting velocity on higher order structure of DNA
Kotaro Yoshida, Yoshihiro Murayama (*Tokyo Univ. of Agri. and Tech.*)

1Pos121 マイクロ液滴界面を利用した RNA 転写配列を有する DNA マイクロ構造体の構築
Construction of DNA micro-structures with RNA transcription sequences using the interface of microdroplets
Risa Watanabe[1], Masamune Morita[1], Miho Yanagisawa[2], Masahiro Takinoue[1] ([1]*Dept. Comput. Sci., Tokyo Tech.*, [2]*Dept. Appl. Phys., Tokyo Univ. Agri. Tech.*)

1Pos122 Phase transition of genomic DNA molecules in solutions with different concentration of propanol
Yue Ma[1], Yuko Yoshikawa[2], Koichiro Sadakane[1], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Doshisha University*, [2]*Ritsumeikan University*)

1Pos123 Nucleic Acid Folding Revealed From Replica Exchange Molecular Dynamics
Jacob Swadling (*University of Tokyo*)

1Pos124 荷電脂質膜表面上での自己組織化 DNA マイクロ構造の形成
Formation of self-assembled DNA microarchitectures on a cationic lipid membrane surface
Masamune Morita[1], M. Shin-ichiro Nomura[2], Satoshi Murata[2], Miho Yanagisawa[3], Masahiro Takinoue[1] ([1]*Dept. Comput. Sci., Tokyo Tech.*, [2]*Dept. Robotics, Tohoku University*, [3]*Dept. Appl. Phys., Tokyo Univ. Agri. Tech.*)

1Pos125 DNA 光修復活性を有する DNA 酵素の赤外分光解析
FTIR spectroscopic analysis of a DNAzyme possessing DNA photorepair activity
Yuhi Kurahashi, Wijaya I M. Mahaputra, Tatsuya Iwata, Hideki Kandori (*Nagoya Institute of Technology*)

1Pos126 Sub-millisecond folding dynamics of preQ$_1$ riboswitch studied by two-dimensional fluorescence lifetime correlation spectroscopy (2D FLCS)
Bidyut Sarkar[1], Kunihiko Ishii[1,2], Tahei Tahara[1,2] ([1]*Molecular Spectroscopy Laboratory, RIKEN*, [2]*RIKEN Center for Advanced Photonics*)

分子モーター / Molecular motor

1Pos127 F$_1$-ATPase の回転における加水分解待ち状態から ATP 結合待ち状態への構造遷移
Conformational transition from catalytic dwell to ATP-binding dwell in F$_1$-ATPase rotation
Kei-ichi Okazaki[1,2], Mitsuhiro Sugawa[3], Gerhard Hummer[2] ([1]*IMS*, [2]*MPI Biophysics*, [3]*Univ. of Tokyo*)

1Pos128 a-subunit ヘリックスが傾いた新構造における FO 回転分子モーターのイオン伝導経路解析
Analysis of the ion pathway of FO molecular motor using the revised structure with tilted a-subunit helices
Kota Tezuka, Ryoichi Kiyama, Daiki Yamakoshi, Dan Parkin, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

1Pos129 好熱菌由来の回転モーター F$_1$ の燐酸解離のタイミング
Timing of Pi release in the rotary motor thermophilic F$_1$ (TF$_1$)
Kengo Adachi[1], Kazuhiro Oiwa[2], Masasuke Yoshida[3], Taro Uyeda[1], Kazuhiko Kinosita, Jr.[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*Adv. ICT Res. Inst., NICT*, [3]*Dept. Mol. Biosci., Kyoto Sangyo Univ.*)

1Pos130* **F1-ATPase の制御因子としての ε サブユニットの ATP 解離反応における役割**
Role of ε subunit on ATP dissociation as a regulator for F1-ATPase
Makoto Genda[1], Rikiya Watanabe[1], Yasuyuki Yamada[2], Hiroyuki Noji[1] ([1]*Graduate School of Engineering, University of Tokyo*, [2]*Department of Life Science, Rikkyo University*)

1Pos131 **F1-ATPase の P-loop 変異体におけるリン酸解離の機構**
The kinetics of Pi release in F1-ATPase investigated with P-loop mutations
Hiroka Narita[1], Hitoshi Hoshina[1], Hikaru Yoshida[1], Yohei Nakayama[1], Shoichi Toyabe[2], Hiroshi Ueno[3], Eiro Muneyuki[1] ([1]*Dept. of Phys., Chuo Univ.*, [2]*Dept. Appl. Phys., Tohoku Univ.*, [3]*Dept. Appl. Chem., Tokyo Univ.*)

1Pos132* **腸球菌 V-ATPase の Na^+濃度依存 ATPase 活性を促進する化合物の同定**
Identification of accelerators on Na^+-depending ATPase activity of Enterococcus hirae V-ATPase
Senka Gi[1], Lica Fabiana Yakushiji[1], Hiroshi Ueno[2], Hiroyuki Noji[2], Takayoshi Arai[1], Katsuhiko Moriyama[1], Hideo Togo[1], Takeshi Murata[1,3] ([1]*Grad. Sch. Sci., Univ. Chiba*, [2]*Grad. Sch. Eng., Univ. Tokyo*, [3]*PRESTO, JST*)

1Pos133 **キネシンによる微小管の構造変化**
A novel function of kinesin-1: changing microtubule conformation that accelerates successive kinesin binding
Tomohiro Shima[1,2], Manatsu Morikawa[3], Junichi Kaneshiro[1], Taketoshi Kambara[1], Shinji Kamimura[4], Toshiki Yagi[5], Hiroyuki Iwamoto[6], Taro Ichimura[1], Tomonobu Watanabe[1], Sotaro Uemura[2], Ryo Nitta[7], Yasushi Okada[1,2], Nobutaka Hirokawa[3] ([1]*RIKEN QBiC*, [2]*Grad. Sch. Sci., Univ. Tokyo*, [3]*Grad. Sch. Med., Univ. Tokyo*, [4]*Dept. Biol. Sci., Chuo Univ.*, [5]*Dept. Life Sci., Pref. Univ. Hiroshima*, [6]*SPring-8, JASRI*, [7]*RIKEN CLST*)

1Pos134 **二量体分子モーターの歩行に関する統一モデル**
A unified walking model for dimeric motor proteins
Kazuo Sasaki[1], Motoshi Kaya[2], Hideo Higuchi[2] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Sci., Univ. Tokyo*)

1Pos135 **タンデムに２つの頭部をつないだキネシンを用いた選択的な前方へのステップの研究**
Preferential forward stepping mechanism of kinesin-1 studied using tandemly joined two-headed monomer
Kohei Matsuzaki[1], Hiroshi Isojima[1], Sawako Enoki[2], Hiroyuki Noji[2], Michio Tomishige[1] ([1]*Dept. Appl. Phys., Grad. Sch. Eng., Univ. Tokyo*, [2]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*)

1Pos136 **光応答性蛋白質 Dronpa を利用したキネシン運動活性の光可逆的阻害**
Photo-reversible inhibition of kinesin motor activity utilizing photochromic protein Dronpa
Kohei Uchida, Shinsaku Maruta, Kazunori Kondo (*Grad. Sch. Bioinfo., Univ. Soka*)

1Pos137* **ゆらぎの定理を用いた非侵襲な力測定によるメラニン色素顆粒輸送のメカニズムの解明**
Application of the fluctuation theorem for the non-invasive measurement of force to pigment transport in melanophores
Shin Hasegawa[1], Kazuho Ikeda[2], Takashi Sagawa[3], Yasushi Okada[2,4], Kumiko Hayashi[1,5] ([1]*Sch. Eng., Tohoku Univ.*, [2]*QBiC, RIKEN*, [3]*NICT*, [4]*Sch. Sci., Univ. Tokyo*, [5]*AMED, PRIME*)

1Pos138 **DNA オリガミを用いた速度の異なるキネシンによる協調運動の観察**
Cooperative transport by two kinesin motors with different velocities studied using programmable DNA origami
Ryosuke Masuda[1], Tsuyoshi Omi[1], Yamato Niitani[1], Mitsuhiro Iwaki[2], Michio Tomishige[1] ([1]*Department of Applied Physics, Univ. of Tokyo*, [2]*QBiC, RIKEN*)

1Pos139 **Inhibitory mechanism for photochromic kinesin Eg5 inhibitor composed of spiropyran derivative**
Ryoma Yamamoto, Kei Sadakane, Shinsaku Maruta (*Grad. Sch. Bioinfo., Univ. Soka*)

1Pos140 **1 分子 FRET 観察による細胞質ダイニンの構造変化の計測**
Single molecule FRET observation of cytoplasmic dynein's conformational change
Mikiya Sakata[1], Takuya Kobayashi[1], Mitsuhiro Sugawa[1], Tomohiro Shima[2], Junichiro Yajima[1], Yoko Y. Toyoshima[1] ([1]*Grad. Sch. Arts and Sci., Univ. of Tokyo*, [2]*Grad. Sch. Sci., Univ. of Tokyo*)

1Pos141 **微小管系モーターダイニンを基に新規アクチン系モーターをエンジニアリングする**
Engineering Novel Actin-Based Molecular Motors from the Microtubule-based Motor Dynein
Akane Furuta, Kazuhiro Oiwa, Hiroaki Kojima, Ken'ya Furuta (*Frontier Research Lab, NICT*)

1Pos142* **細胞質ダイニンのマルチスケールシミュレーション:全原子から連続体へ**
Multiscale Simulations of Cytoplasmic Dynein: From All-atom to Continuum Mechanics
Shinji Iida[1,3], Benjamin Hanson[4], Narutoshi Kamiya[2], Genji Kurisu[1], Takahide Kon[3], Haruki Nakamura[1], Sarah Harris[4] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. SS, Univ. Hyogo*, [3]*Grad. Sch. Sci., Osaka Univ.*, [4]*Sch. Phys. Astro., Univ. Leeds*)

1Pos143 **Diffusive Component in Directed Movements of Cytoplasmic Dynein**
Takayuki Torisawa, Ken'ya Furuta, Kazuhiro Oiwa, Hiroaki Kojima (*NICT, Advanced ICT Research Institute*)

1Pos144 **細胞質ダイニンの構造変化を伴う運動メカニズムに関する分子シミュレーション研究**
Molecular simulation study on the working mechanism with structural changes of cytoplasmic dynein
Shintaroh Kubo, Shoji Takada (*Grad. Sci., Univ. Kyoto*)

1Pos145* **ヒト細胞質ダイニン１分子のパワーストローク運動距離の測定**
Measurement of the power stroke distance of cytoplasmic dynein motor
Yoshimi Kinoshita[1], Taketoshi Kambara[1,2], Kaori Nishikawa[1], Motoshi Kaya[1], Hideo Higuchi[1] ([1]*The University of Tokyo*, [2]*QBiC, Riken*)

1Pos146 **ミオシン S1 によって誘起されるアクチンフィラメントの協同的構造変化の高速 AFM による観察と、その生理的意義**
High-speed AFM demonstration of cooperative structural changes in actin filaments induced by myosin S1 and physiological implication
Kien Xuan Ngo[1,2], Noriyuki Kodera[3], Toshio Ando[3], Taro Ueda[1,2] ([1]*Biomed. Res. Inst., AIST*, [2]*Dept. Phys., Waseda Univ.*, [3]*Dept. Phys., & Bio-AFM FRC, Kanazawa Univ.*)

1Pos147 **単一フィラメントにおけるアクチン重合・脱重合ダイナミクスの力学的制御**
Mechanical manipulation of polymerization dynamics of individual actin filaments
Hiroaki Kubota[1], Makito Miyazaki[1,2], Taisaku Ogawa[3], Togo Shimozawa[4], Kazuhiko Kinosita Jr.[1], Shin'ichi Ishiwata[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*Waseda Bioscience Research Institute in Singapore, Waseda Univ.*, [3]*QBiC, Riken*, [4]*Dept. Life Sci. Med. Biosci., Waseda Univ.*)

1Pos148 **Actomyosin contraction with a contractile ring related cross-linker in an *in vitro* active gel model system**
Kyohei Matsuda, Takuya Kobayashi, Mitsuhiro Sugawa, Yoko Y. Toyoshima, Junichiro Yajima (*Department of Life Sciences, Graduate School of Arts & Sciences, The University of Tokyo*)

1Pos149 **混み合い環境でのアクチン線維の集団運動による秩序構造の出現**
Appearance of ordered structure by collective motion of actin filaments in crowded environments
Takahiro Iwase, Yasuhiko Sasaki, Kuniyuki Hatori (*Dept. Bio-Systems Eng., Yamagata Univ.*)

1Pos150 **Self-organizations of actin filament networks in confined spaces: A simulation study**
Takahiro Nitta (*Gifu Univ.*)

1Pos151 **蛍光顕微鏡および高速 AFM によるミオシンと F-アクチン間の協同的結合の経時的観察**
Real-time observation of cooperative binding between myosin and F-actin by fluorescence microscopy and high-speed atomic force microscopy
Rika Hirakawa[1], Hiroaki Ueno[1], Noriyuki Kodera[2], Taro Q.P. Uyeda[3], Kiyotaka Tokuraku[1] ([1]*Muroran Inst. Tech.*, [2]*Bio-AFM FRC, Inst. Sci. &Eng., Kanazawa Univ.*, [3]*Waseda Univ.*)

1Pos152* **枯渇力が誘起する微小管集団運動に関する研究**
Study of the collective motion of microtubules induced by depletion force
Ai Saito[1], Ryuhei Suzuki[1], Tamanna Ishrat Fahana[1], Arif Md. Rashedul Kabir[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*)

細胞生物学 / Cell biology

1Pos153 **神経細胞における細胞骨格アクチンの修復の分子メカニズム**
Molecular mechanism of cytoskeletal Actin repairing in nerve cells
Tomboy Higo[1], Ayumi Ishihara[2], Shinji Aramaki[1], Yoshiko Itou[2], Takuo Yasunaga[1] ([1]*Kyushu Institute of Technology*, [2]*Leica Microsystems*)

1Pos154 **超解像光学顕微鏡で観察した収縮環の計測と解析**
Measurement and analysis of contractile ring observed with STED and Sim
Kaoru Katoh[1,2], Keijyu Kamijo[3], Minami Tanaka[2], Masayuki Takahashi[4], Issei Mabuchi[5], Hiroshi Hosoya[6] ([1]*BioMed. Res. Inst., AIST*, [2]*Gad. Sch. of Life & Environ. Sci., Univ. Tsukuba*, [3]*Sch. of Med., Tohoku Med. Pharm. Univ.*, [4]*Dept. of Chem, Fac. of Sci., Hokkaido Univ.*, [5]*Dept. of Life Sci. Gakushuin Univ.*, [6]*Dept. of Biol., Fac. of Sci., Kanagawa Univ.*)

1Pos155 **アクチンフィラメントの協同的構造変化を阻害するアクチン変異の遺伝子内サプレッサー解析**
Intragenic suppressor analysis of actin mutation that impairs cooperative conformational change of actin filament
Tenji Yumoto[1,4], Takehiko Yoko-o[2], Keiko Hirose[3], Taro Uyeda[4] ([1]*Grad. Sch. Life & Env. Sci., Univ. of Tsukuba*, [2]*Bioprod. Res. Inst., AIST*, [3]*Biomed. Res. Inst., AIST*, [4]*Dept. Phys., Waseda Univ.*)

1Pos156 **クライオ電子線トモグラフィ法で明らかにした，フィロポディア内におけるファシンによるアクチンフィラメント束化メカニズム**
F-actin bundling mechanisms by fascin in filopodia was revealed by cryo-ET
Shinji Aramaki[1], Kouta Mayanagi[2], Kazuhiro Aoyama[3,4], Takuo Yasunaga[1] ([1]*Department of Bioscience and Bioinformatics*, [2]*Medical Institute of Bioregulation, Kyushu University*, [3]*FEI Japan*, [4]*Research Centre for Ultra-High Voltage Electron Microscopy, Osaka University*)

1Pos157 **MEA システムを用いたニワトリ胚由来心臓組織片の薬剤応答**
Drug response of embryonic chick heart tissue pieces using multi electrode array system
Yosuke Kamei[1], Toshiyuki Mitsui[2], Tomoyuki Kaneko[1] ([1]*LaRC,Grad. Sci. Eng., Hosei Univ.*, [2]*Dept. Math. Phys., Col. Sci. Eng., Aoyama Univ.*)

1Pos158 **心筋細胞の集合体に対する機械的刺激の影響**
Influence of mechanical stimulus on embryonic chick heart cell aggregates
Shin Arai, Ayaha Tsuyuki, Takahiro Uehara, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

1Pos159 **フィードバックマイクロレオロジーによる細胞骨格の非線形力学挙動計測**
Nonlinear mechanical properties of Cytoskeletons measured with Dual-Feedback Microrheology
Natsuki Honda, Kenji Nishizawa, Takayuki Ariga, Daisuke Mizuno (*Kyushu University, Department of Physics*)

1Pos160 **ケラトサイトの運動方向を決定する２つのメカノセンシング機構**
Keratocytes have hybrid mechanosensing system to decide their migration direction
Chika Okimura, Yoshiaki Iwadate (*Fac. Sci., Yamaguchi Univ.*)

1Pos161* **非熱的な力に駆動された細胞内部の混み合い状態**
Intracellular crowding mechanics driven by athermal force
Kenji Nishizawa, Daisuke Mizuno (*Grad. Sch. Sci., Kyushu Univ.*)

1Pos162 **心筋細胞の集合体群に与えるマルチプローブ機械的刺激の影響**
Effect of Multi-probe stimuli on cardiac cell aggregates with spontaneous beat
Ayaha Tsuyuki, Shin Arai, Takahiro Uehara, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

1Pos163 **魚類表皮細胞ケラトサイトのかたち・サイズと牽引力**
Relationship between traction forces, and shape and size of keratocytes
Ayane Sonoda, Chika Okimura, **Yoshiaki Iwadate** (*Fac. Sci., Yamaguchi Univ.*)

1Pos164 **アメーバ運動する線虫精子の牽引力測定**
Measurement of Traction Force generated by Amoeboid Sperm of C. elegans
Midori Yoshimura[1], Hikaru Emoto[1], Chika Okimura[2], Yoshiaki Iwadate[2], Katsuya Shimabukuro[1] ([1]*Dep. of Chem. and Bio. Eng., NIT, Ube College*, [2]*Faculty of Sci., Yamaguchi University*)

1Pos165 **細胞周期進行に伴う細胞内部環境のダイナミクス**
The dynamics of Intracellular Environments during Cell-cycle progression
Katsuhiro Umeda, Kenji Nishizawa, Daisuke Mizuno (*Grad. Sci., Univ. Kyushu*)

1Pos166* **Actin Cytoskeleton Remodeling Dynamics of Adherent Cells Under Mechanical Strain of Gelatin Substrate**
Kwokhoi Ng[1], Kentaro Iketaki[1], Ryuzo Kawamura[1], Seiichiro Nakabayashi[1], Yosuke Yoneyama[3], Fumihiko Hakuno[3], Shin-Ichiro Takahashi[3], Fumiki Yanagawa[2], Toshiyuki Takagi[2], Shinji Sugiura[2], Toshiyuki Kanamori[2], Hiroshi Yoshikawa[1] ([1]*Dept. Chem.,Saitama Univ.*, [2]*BRD.,AIST.*, [3]*GASLS., The Univ. of Tokyo*)

1Pos167 **血管壁内力学環境を考慮したコラーゲン微細溝基質による血管平滑筋細胞の分化制御**
Control of vascular smooth muscle cell differentiation using a novel micro-grooved collagen substrate
Kazuaki Nagayama, Keiichi Uchida, Saki Takeuchi (*Micro-Nano Biomechanics Laboratory, Department of Intelligent Systems Engineering, Ibaraki University*)

1Pos168 **力学的強度の制御を可能とする光架橋性コラーゲンゲルの開発**
Development of photo-cross-linked collagen gels with tunable mechanical property
Takahiro Fujisawa[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*IMCE, Kyushu Univ.*)

1Pos169 **組織切片の伸展性応答: ひび割れパターンと病態**
Response of Tissue Slice to Mechanical Stretching: Characteristic Cracking Pattern Reflecting Disease State
Keisuke Danno[1], Takuto Nakamura[1], Naohiko Nakamura[2], Kota Iguchi[2], Masaya Ikegawa[3], Kenichi Yoshikawa[3] ([1]*Doshisha Univ.*, [2]*Kyoto Univ.*, [3]*Doshisha Univ.*)

1Pos170 **タリンとビンキュリンによる力と硬さの感知**
Force- and rigidity-sensing by talin and vinculin
Hiroaki Hirata[1,2,3], Keng-Hwee Chiam[3], Hitoshi Tatsumi[4], Chwee Teck Lim[3], Masahiro Sokabe[1,3] ([1]*Nagoya Univ. Grad. Sch. Med., Mechanobiology Lab*, [2]*R-Pharm Japan*, [3]*Mechanobiology Inst., Natl. Univ. Singapore*, [4]*Kanazawa Inst. Tech.*)

1Pos171 **マイクロメートルスケールの足場構造に依存した細胞性粘菌の細胞遊走**
Migration of Dictyostelium cells on micro-scale ridge structures
Gen Honda[1], Akihiko Nakajima[2], Satoshi Sawai[1,2,3] ([1]*Graduate School of Arts and Sciences, University of Tokyo*, [2]*Research Center for Complex Systems Biology*, [3]*PRESTO, JST*)

1Pos172* **膜タンパク質による細胞間相互作用の定量的解析**
The quantitative analysis of the intercellular interaction by membrane proteins
Takumi Miyatake[1,2], Yoshihisa Kaizuka[2] ([1]*Graduate School of Pure and Applied Sciences, University of Tsukuba*, [2]*National Institute for Materials Science*)

1Pos173 **重力下での形態形成・維持に対するアクトミオ シンネットワークの寄与**
Teoretical study of contribution of YAP-dependent actomysin network to morphogenesis under gravity
Kazunori Takamiya[1], Hiraku Nishimori[1,2], Akinori Awazu[1,2] ([1]*Grad. Sch. Sci., Univ. Hiroshima*, [2]*RcMcD*)

1Pos174 **Divergence of structural strategies for E-cadherin homophilic binding among bilaterians**
Shigetaka Nishiguchi[1,2,3], Akira Yagi[3], Nobuaki Sakai[3], Hiroki Oda[1,2] ([1]*JT BRH*, [2]*Osaka Univ.*, [3]*Olympus Co.*)

1Pos175* **DNA hybridization を介した細胞-細胞間接着ダイナミクスの解明**
Dynamics of cell-cell adhesion via DNA hybridization
Ken Sato[1], Yuji Teramura[2], Ryuzo Kawamura[1], Naritaka Kobayashi[1], Seiichiro Nakabayashi[1], Hiroshi Yoshikawa[1] ([1]*Dept. Chem., Saitama Univ.*, [2]*Dept. Bioeng., Tokyo Univ.*)

1Pos176 **膵島 α 細胞の分泌顆粒動態に及ぼす接着分子 CADM1 の影響**
Effect of cell adhesion molecule 1 expression on intracellular granule movement in pancreatic α cells
Tadahide Furuno[1], Satoru Yokawa[1,2], Takanari Ikeda[1], Yoshikazu Inoh[1], Ryo Suzuki[2], Takahiro Suzuki[3], Naohide Hirashima[2] ([1]*Sch. Pharm., Aichi Gakuin Univ.*, [2]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [3]*Sch. Dent., Aichi Gakuin Univ.*)

1Pos177 **分散培養 iPS 細胞の増殖応答性に対するハイドロゲル表面へのラミニン修飾状態の本質的効果**
Essential role of mannter of laminin-modification for hydrogel surface on the prolifelation activity of dissociated iPS cells
Kenta Mizumoto[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Univ. Kyushu*, [2]*IMCE., Univ. Kyushu*)

1Pos178 **細胞内ナノ粒子導入・細胞間相互作用制御のための材料工学**
Materials engineering approaches to modulate cell membrane structures for nanoparticles delivery and regulation of cell-cell interactions
Yoshihisa Kaizuka, Tomoto Ura, Hidenobu Nakao (*NIMS*)

1Pos179* **脂質膜に覆われた細胞サイズ生体高分子ゲルの弾性率**
Elasticity of biopolymer gel in cell-sized droplet covered with a lipid membrane
Atsushi Sakai, Yoshihiro Murayama, Miho Yanagisawa (*Tokyo university of Agriculture and Technology*)

1Pos180 **原始真核生物の細胞内小器官の 3D 構造モデルから得られる生物の新たな情報**
New obvious information obtained from cell organelle 3D-structural models of primitive eukaryote
Atsuko H. Iwane[1,2], Rina Nagai[1,2], Hikari Mori[1], Takako Ichinose[1,2] ([1]*Cell Field Struc., QBiC, Riken*, [2]*Grad. Sch. Fronti., Biosci., Osaka Univ.*)

1Pos181 **Accumulation of cargo proteins can physically trigger vesiculation in membrane trafficking system**
Masashi Tachikawa (*RIKEN*)

1Pos182 **多突起型チューブリン封入リポソームの形態形成メカニズム**
Morphogenetic mechanism of tubulin-encapsulating giant liposomes with a hundred of membrane spines
Masahito Hayashi, Kingo Takiguchi (*Grad. Sch. Sci., Nagoya Univ.*)

1Pos183 **アクトミオシンネットワークの収縮による細胞サイズ液滴の運動**
Directed motion of cell-sized droplets driven by actomyosin network contraction
Yuto Sano[1], Makito Miyazaki[1,2], Kozue Hamao[3], Shin'ichi Ishiwata[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*Waseda Bioscience Research Institute in Singapore, Waseda Univ.*, [3]*Dept. Bio. Sci., Hiroshima Univ.*)

1Pos184 **高濃度アクチン繊維が引き起こす立体的集団運動**
The collective motion and band pattern formations of sliding actin filaments driven by HMM
Yuuji Setoguchi, Hirotaka Taomori, Masayuki Hoshida, Yu Ichinose, Hajime Honda (*Dept. Bioeng., Nagaoka Univ. Tech.*)

1Pos185 **細胞に優しい三次元組織体の構築：レーザートラップと高分子の混雑効果の活用**
Constructing stable cellular assembly in the absence of artificial scaffold by use of laser tweezers
Shoto Tsuji, Aoi Yoshida, Taeko Ohta, Hiroaki Taniguchi, Kenichi Yoshikawa (*Doshisha University*)

1Pos186 **ヒト疾患診断マーカーとして有用なエキソソームの生物物理解析に適した新規分離調製法の開発**
A novel isolation and preparation method for the biophysical analyses of useful exosomes as diagnostic markers for human diseases
Noriyuki Ishii, Mitsushi J. Ikemoto, Takayuki Odahara (*Biomedical Research Institute, National Institute of Advanced Industrial Science and Technology (AIST)*)

1Pos188* **アクチンの示すミクロ顆粒系での局在性転移：細胞の混雑環境モデリング**
Selective Localization of Actin in Micro-Domains under Molecular Crowding: Difference among Monomeric, Linear-Polymeric and Bundling State
Naoki Nakatani[1], Chen-Yang Shew[2], Kanta Tsumoto[3], Kingo Takiguchi[4], Masahito Hayashi[4], Shunsuke Tanaka[4], Kenichi Yoshikawa[1] ([1]*Grad. Sch. Life and Medical Sciences, Doshisha Univ.*, [2]*Division of Science & Technology, College of Staten Island, New York City Univ.*, [3]*Grad. Sch. Engineering, Mie Univ.*, [4]*Grad. Sch. Science, Nagoya Univ.*)

生体膜・人工膜：ダイナミクス / Biological & Artificial membrane: Dynamics

1Pos189 **ジャイアントベシクルにおけるアミロイド繊維の形成**
Amyloid fibril formation in giant vesicle
Tong Zhu[1], Kensuke Kurihara[1,2,3] ([1]*Okazaki Institute for Integrative Bioscience*, [2]*Institute for Molecular Science*, [3]*Research Center for Complex Systems Biology, The Univ. of Tokyo*)

1Pos190* **人工細胞モデルを用いた生体膜融合機構の解明**
Biophysical principle of membrane fusion revealed by artificial lipid vesicles
Yui Suzuki[1], Ken Nagai[1], Anatoly Zinchenko[2], Tsutomu Hamada[1] ([1]*JAIST*, [2]*Grad. Sch. of Environmental Studies, Nagoya Univ.*)

1Pos191 ***in vitro* １分子イメージング解析により明らかになった PI(4,5)P2 依存的な PTEN の膜結合の促進・安定化**
Phosphatidylinositol lipid PI(4,5)P2 enhances membrane binding of PTEN revealed by *in vitro* single-molecule imaging analysis
Daisuke Yoshioka[1], Seiya Fukushima[1,3], Daichi Okuno[3], Satomi Matsuoka[3], Toru Ide[4], Masahiro Ueda[2,3] ([1]*Dep. Biol. Sci., Grad. Sch. of Sci., Osaka Univ.*, [2]*Grad. Sch. of Front. Biosci., Osaka Univ.*, [3]*RIKEN QBiC*, [4]*Grad. Sch. of Nat. Sci. and Tech., Okayama Univ.*)

1Pos192 **Measurements of mitochondrial motility in cell body**
Hyunjin Choi, Yuki Sugimoto, Yoshihiro Ohta (*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*)

1Pos193 **人工脂質膜小胞内における PIP3/PTEN traveling wave の再構成**
Reconstitution of traveling wave of PIP3/PTEN on membrane in GUVs
Hitomi Matsubara (*Lab. Single Molecule Biology, Grad. FBS., Osaka Univ.*)

1Pos194 **細胞透過ペプチド・オリゴアルギニンの単一ベシクルへの侵入に対する脂質組成の効果**
Effects of lipid compositions on the entry of cell-penetrating peptide oligoarginine into single vesicles
Sabrina Sharmin[1], Md Zahidul Islam[1], Mohammad Abu Sayem Karal[1], Sayed Ul Alam Shibly[1], Hideo Dohra[2], Masahito Yamazaki[1,3,4] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Green Sci. Tech., Shizuoka University*, [3]*Res. Inst. Ele., Shizuoka Univ.*, [4]*Grad. Sch. Sci., Shizuoka Univ.*)

1Pos195 **Functional significance of trimerization in Cl^- pumping properties of halorhodopsin examined by nanodisc reconstitution**
Eri Hashimoto[1], Kenshiro Suzuki[1], Ayumi Yamamoto[1], Takashi Tsukamoto[2], Takeshi Uchida[1,3], Takashi Kikukawa[2], Makoto Demura[2], Koichiro Ishimori[1,3] ([1]*Grad. Sch. of Chem. Sci. and Eng. Hokkaido Univ.*, [2]*Grad. Sch. of Life Sci. Hokkaido Univ.*, [3]*Fac. of Sci. Hokkaido Univ.*)

1Pos196 **脂質膜の伸展の分光学的研究**
Spectroscopic Investigation of Osmotic Pressure-Induced Membrane Stretching
Chiranjib Ghatak[1], Sayed Ul Alam Shibly[2], Masahito Yamazaki[1,2,3] ([1]*Res. Inst. Ele., Shizuoka Univ.*, [2]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*)

1Pos197 **細菌-膜小胞間相互作用の動態と情報伝達機構の解析**
Dynamics and signal transduction of the interaction between bacteria and membrane vesicles
Yosuke Tashiro, Yusuke Hasegawa, Kotaro Takaki, Hiroyuki Futamata (*Dept. of Eng., Shizuoka Univ.*)

1Pos198 **マガイニン２が誘起する脂質膜中のポア形成に対する抗菌ペプチド・PGLa の効果**
Effect of Antimicrobial Peptide PGLa on Magainin 2-Induced Pore Formation in Lipid Membranes
Farliza Parvez[1], Md Jahangir Alam[2], Hideo Dohra[3], Masahito Yamazaki[1,2,4] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Ele., Shizuoka Univ.*, [3]*Res. Inst. Green Sci. Tech., Shizuoka Univ.*, [4]*Grad. Sch. Sci., Shizuoka Univ.*)

生体膜·人工膜：輸送 / Biological & Artificial membrane: Transport

1Pos199 **大腸菌異物排出系トランスポーター MdtB, MdtC 会合の可視化**
Assembly of the xenobiotic efflux transporters MdtB and MdtC of *Escherichia coli*
Megumi Yamazaki[1], Kentaro Yamamoto[1], Masatoshi Nishikawa[1], Yoshiyuki Sowa[1,2], **Ikuro Kawagishi**[1,2] ([1]*Dept. Frontier Biosci., Hosei Univ.*, [2]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

1Pos200 **外部環境を感知する磁場駆動型リポソームの創製**
Magnetically-driven moving liposomes that sense environmental information
Mika Ebihara[1], Taro Toyota[2], Naoto Nemoto[1] ([1]*Grad. Sch. Sci. Eng., Saitama Univ.*, [2]*Grad. Sch. Arts Sci., Univ. of Tokyo*)

1Pos201 **ミトコンドリア密集による ATP 産生の効率化**
Effective ATP generation by closely located mitochondria
Yusho Kuraoka[1], Daiki Yoshimatu[1], Takuya Takahashi[2], Yoshihiro Ohta[1] ([1]*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*, [2]*Dept. of Biosci. and Bioinformatics, Ritsumeikan Univ.*)

生体膜・人工膜：構造・物性 / Biological & Artificial membrane: Structure & Property

1Pos202 **高いトポロジー種数を持つベシクルの形態：核膜形状の形成**
Morphology of high-genus vesicles under pore-size constraint: Construction of nuclear envelope shape
Hiroshi Noguchi (*ISSP, Univ. Tokyo*)

1Pos203 **両親媒性 DNA オリガミによる W/O マイクロエマルションの光応答性観察**
Photo-responsive water-in-oil microemulsion made of amphiphilic DNA origami
Misato Tsuchiya[1], Daisuke Ishikawa[1], Yuki Suzuki[2], Masayuki Endo[3], Masahiro Takinoue[1] ([1]*Dept. Comput. Sci., Tokyo Tech.*, [2]*Fronti. Res. Inst. Interdiscip. Sci., Tohoku Univ.*, [3]*WPI-iCeMS., Kyoto Univ.*)

1Pos204 **合成高分子による膜曲率の認識**
Recognition of membrane curvature by synthetic amphiphilic polymers
Naho Sunagawa[1], Manami Tsukamoto[1], Kenichi Kuroda[2], Jun-ichi Kikuchi[1], Kazuma Yasuhara[1] ([1]*Grad. Sch. Mat. Sci., Nara Inst. Sci. Tech.*, [2]*Sch. Dentistry Univ. Michigan*)

1Pos205 **膜貫通タンパク質の細胞膜上二次元拡散における二段階緩和の理論解析**
Theoretical analysis of a two-step relaxation on protein diffusion in the plasma membranes
Tomonari Sumi[1], Atsushi Okumoto[2], Hitoshi Goto[2], Hideo Sekino[2] ([1]*Res. Inst. Interdisciplinary Sci., Okayama Univ.*, [2]*Toyohashi Univ. Tech.*)

1Pos206 **肺サーファクタントタンパク質 B の N 末端による脂質単分子膜の構造変化**
Morphology changes in lipid monolayers induced by the N-terminal segments of surfactant protein B
Hideyuki Nagatsuka, Masahiro Hibino (*Div. Sust. Enviro. Eng., Muroran Inst. Tech.*)

1Pos207 **シトクロム P450 基質薬剤クロルゾキサゾンとホスファチジルエタノールアミン・モデル膜との相互作用**
Interaction between cytochrome P450 substrate drug chlorzoxazone and phosphatidylethanolamine model membranes
Hiroshi Takahashi (*Grad. Sch. Sci. Tech., Gunma Univ.*)

1Pos208 **脂質キュービック相間の相転移における方位関係**
Orientational Relationships In Transformations Between Three Inverse Bicontinuous Cubic Phases of a Lipid
Toshihiko Oka[1,2] ([1]*Faculty of Science, Shizuoka University*, [2]*Research Institute of Electronics, Shizuoka University*)

1Pos209 **ジミリストイルホスファチジルコリン–コレステロール二成分二分子膜中における構成脂質の部分モル体積**
Partial molar volumes of constituent lipids in the binary bilayer of dimyristoylphosphatidylcholine and cholesterol
Nobutake Tamai[1], Naohiro Takeshita[2], Masaki Goto[1], Hitoshi Matsuki[1] ([1]*Grad. Sch. Biosci. Bioind., Tokushima Univ.*, [2]*Grad. Sch. Adv. Tech. Sci., Tokushima Univ.*)

1Pos210 **部分フッ素化リン脂質と DMPC の二成分系混合膜物性に関する系統的研究**
Mixing behaviors in the binary membrane of DMPC and its partially fluorinated analogues with different perfluoroalkyl chain lengths
Miki Horikoshi[1], Kohei Morita[1], Toshinori Motegi[1], Hiroshi Takahashi[1], Hideki Amii[1], Toshiyuki Takagi[2], Toshiyuki Kanamori[2], Masashi Sonoyama[1] ([1]*Div. Mol. Sci., Gunma Univ.*, [2]*AIST*)

1Pos211 **多電極アレイ上でのエレクトロフォーメーション法によるリポソームの作製**
Preparation of liposomes by electro formation method on multi electrode array
Hayato Akizuki, Tomoyuki Kaneko (*LaRC, Grad. Sci. Eng., Hosei Univ.*)

1Pos212 **PEG 脂質を導入した支持脂質二重膜の拡散特性**
Effect of PEG-lipid on diffusion properties of supported lipid bilayer
Moeko Saruta, Takuhiro Otosu, Shoichi Yamaguchi (*Grad. Sch. Sci. Eng., Saitama Univ.*)

1Pos213 プログラマブルな性質を有する DNA ナノプレートからなるマイクロカプセルの形成

Microcapsular compartments composed of programmable DNA nanoplates

Daisuke Ishikawa[1], Yuki Suzuki[2], Chikako Kurokawa[3], Masayuki Ohara[4], Masamune Morita[1], Miho Yanagisawa[3], Ryuji Kawano[4], Masayuki Endo[5], Masahiro Takinoue[1] ([1]*Sch. Comput., Tokyo Tech.*, [2]*FRIS, Tohoku Univ.*, [3]*Dept. Appl. Phys., Tokyo Univ. of Agri. and Tech.*, [4]*Dept. Life Sci. and Biotech., Tokyo Univ. of Agri. and Tech.*, [5]*WPI-iCeMS, Kyoto Univ.*)

生体膜･人工膜：興奮・チャネル / Biological & Artificial membrane: Excitation & Channels

1Pos215 コリネ細菌の機械受容チャネルによる細胞力覚とグルタミン酸放出機構

Bacterial mechanosensation and glutamate export by mechanosensitive channels in Corynebacterium glutamicum

Yoshitaka Nakayama[1], Kosuke Komazawa[2], Navid Bavi[1,3], Ken-ichi Hashimoto[2], Hisashi Kawasaki[2], Boris Martinac[1,3] ([1]*Victor Chang Cardiac Research Institute*, [2]*Tokyo Denki University*, [3]*University of New South Wales*)

1Pos216 全反射赤外分光で見る電位依存性プロトンチャネル VSOP への金属結合

Metal binding to the voltage-gated proton channel VSOP studied by ATR-FTIR

Masayo Iwaki[1], Kohei Takeshita[2,3,4], Yasushi Okamura[5], Atushi Nakagawa[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*Inst. Protein Res., Osaka Univ.*, [3]*Inst. Acad. Initiat., Osaka Univ.*, [4]*JST-PRESTO*, [5]*Grad. Sch. Med., Osaka Univ.*)

1Pos217 ナノキャビティでの K^+の占有が Kv1.2 チャネルを通る K^+の滑走を引き起こす

Occupancy of a K^+ in the nanocavity induces K^+ ions' run through the Kv1.2 channel

Takashi Sumikama, Shigetoshi Oiki (*Univ. of Fukui*)

1Pos218 KcsA チャネルの細胞内領域の荷電状態がチャネル開閉に与える影響

Effects of the electrostatic state of the cytoplasmic domain in the KcsA channel on its gating

Minako Hirano[1], Toru Ide[2] ([1]*GPI*, [2]*Okayama Univ.*)

1Pos219 **Discrimination between mitochondrial rounding and permeability transition**

Takahiro Shibata, Yoshihiro Ohta (*Grad. Sch. Life Sci. & Bio Tech., TUAT*)

1Pos223 **Artificial bilayers formed on a solid substrate for ion-channel recordings**

Toru Ide[1,2], Saki Nomura[1], Minako Hirano[2], Junnya Ichinose[1], Hiroaki Yokota[2] ([1]*Grad. Schl Sci. Tech., Okayama Univ.*, [2]*GPI*)

1Pos224 電位依存性ホスファターゼ VSP の酵素ドメインにおける膜相互作用部位の役割

The role of membrane interacting region of phosphatase domain in voltage-sensing phosphatase (VSP)

Akira Kawanabe[1], Masaki Hashimoto[1], Tomoko Yonezawa[1], Yuka Jinno[2], Souhei Sakata[2], Yasushi Okamura[1] ([1]*Osaka Univ.*, [2]*Osaka Med. Col.*)

1Pos225 イオンチャネル機能に対する膜脂質効果の解析に向けた脂質二重膜組成の迅速変更法

Rapid replacement of the lipid bilayer composition for the analysis of the lipid-effect on the ion channel function

Masayuki Iwamoto, Shigetoshi Oiki (*Dept. Mol. Physiol. & Biophys., Univ. Fukui Facult. Med. Sci.*)

1Pos226 高速原子間力顕微鏡による K^+チャネル KcsA とポア結合性サソリ毒アジトキシン-2 の結合ダイナミクスの一分子解析

Single-molecule blocking dynamics of a scorpion toxin on the KcsA potassium channel revealed by HS-AFM

Ayumi Sumino[1,2], Takayuki Uchihashi[3], Takashi Sumikama[2], Shigetoshi Oiki[2] ([1]*JST/PRESTO*, [2]*Facult. Med. Sci., Univ. Fukui*, [3]*Depart. Phys., Kanazawa Univ.*)

神経科学･感覚 / Neuroscience & Sensory systems

1Pos227* 線虫（C. elegans）嗅覚感覚神経細胞内の領域特異的なにおいに対する cGMP 応答

Compartmentalized cGMP responses to odor in Caenorhabditis elegans' olfactory sensory neurons

Hisashi Shidara, Keita Ashida, Kohji Hotta, Kotaro Oka (*Grad. Sch. Sci. and Tech., Keio Univ.*)

1Pos228* *C. elegans* の低温適応における温度情報伝達の分子ロジック

Molecular logic for temperature signaling in cold tolerance of *C. elegans*

Tomoyo Ujisawa[1], Misato Uda[1], Akane Ohta[1], Katsushi Arisaka[2], Atsushi Kuhara[1] ([1]*Inst. for Integrative Neurobiology, Konan University.*, [2]*Dept. of Physics and Astronomy, UCLA, U.S.A.*)

1Pos229 蛍光温度計シートを用いた神経細胞の熱発生計測

Detection of Neural Thermogenesis with Fluorescent Thermometer Sheet

Mizuho Gotoh[1,2,3], Kotaro Oyama[1,4], Yuki Kawamura[1], Hideki Itoh[1,5], Shin'ichi Ishiwata[1,6] ([1]*Sch. Adv. Sci. Eng., Waseda Univ.*, [2]*Grad. Sch. Comp. Human Sci., Tsukuba Univ.*, [3]*HIRI, AIST*, [4]*Cell Physiol., Jikei Univ.*, [5]*Inst. Med. Biol., A*STAR, Singapore*, [6]*WABIOS, Waseda Univ., Singapore*)

1Pos230* 線虫の早期嗅覚順応における感覚・介在神経細胞の部分特異的可塑性

Compartmentalized modulations of sensory and interneurons for early adaptation in C. elegans

Keita Ashida, Hisashi Shidara, Kohji Hotta, Kotaro Oka (*Keio University*)

1Pos231* イベルメクチンによる α7 ニコチン性アセチルコリン受容体の分子内動態増大の発見

Discovery of the internal motion enhancement of α7 nAChR with Ivermectin

Tomoyuki Baba[1], Keigo Ikezaki[1], Hiroshi Sekiguchi[2], Tai Kubo[3], Yuji C. Sasaki[1,2] ([1]*Grad. Sch. Front. Sci., Univ. Tokyo*, [2]*JASRI/SPring-8*, [3]*MolprofRC/AIST*)

1Pos232 ミミズ体壁刺激に関する慣れとその回復

Establishment and recovery of habituation by repeated tactile stimulus in earthworm

Yoshiichiro Kitamura[1], Hitoshi Aonuma[2], Hiroto Ogawa[3], Kotaro Oka[4] ([1]*Dept. Math Sci. Phys, Kanto Gakuin Univ.*, [2]*Res Inst Elect Sci., Hokkaido Univ.*, [3]*Dept. Biol. Sci., Hokkaido Univ.*, [4]*Dept. Biosci Info, Keio Univ.*)

1Pos233 **シャルコマリートゥース病の原因遺伝子の一つであるダイナミン２の変異は細胞の異常なアクチン動態とラメリポディア形成の減少をもたらす**

Expression of a dynamin 2 mutant associated with Charcot-Marie-Tooth disease leads to aberrant actin dynamics and lamellipodia formation

Hiroshi Yamada, Kinue Kobayashi, Yubai Zhang, Tetsuya Takeda, Kohji Takei (*Dep. of Neurosci., Grad. Sch. of Med., Dent., and Pharm. Sci., Okayama Univ.*)

1Pos234 **ヒト苦味受容体の基質認識の分子機構研究**

Ligand-induced structural changes of human bitter taste receptor

Mayu Hioki[1], Masayo Iwaki[1], Rei Abe-Yoshizumi[1], Hiroo Imai[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*Primate Res. Inst.*)

1Pos235 **性ホルモンによる海馬神経シナプスの制御：オスとメスの性差**

Effect of estrogen and androgen on hippocampal synapses : gender difference of male and female

Asami Kato[1], Yasushi Hojo[2], Yoshitaka Hasegawa[1], Yusuke Hatanaka[1], Suguru Kawato[1,3,4] ([1]*Grad. Sch. Univ. of Tokyo.*, [2]*Dept. Biochem.,Saitama-Med. Univ.*, [3]*Dept. of Urology, Juntendo Univ.*, [4]*Dept. of Urology, Teikyo Univ.*)

光生物：視覚·光受容 / Photobiology: Vision & Photoreception

1Pos236 **ナトリウムポンプ型ロドプシンの光反応中間体の発色団構造**

Structure of retinal chromophore of the photointermediates in sodium ion pump rhodopsin

Nao Nishimura[1], Misao Mizuno[1], Hideki Kandori[2], Yasuhisa Mizutani[1] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Eng., Nagoya Inst. Tech.*)

1Pos237 **固体 NMR を用いたミドルロドプシンのレチナール結合ポケットの構造解析**

Solid-state NMR structural study of retinal-binding pocket in middle rhodopsin

Izuru Kawamura[1], Hayato Seki[1], Arisu Shigeta[1], Yoshiteru Makino[1], Takashi Okitsu[2], Akimori Wada[2], Yuki Sudo[3] ([1]*Grad. Sch. Eng., Yokohama Natl. Univ.*, [2]*Kobe Pharm. Univ.*, [3]*Okayama Univ.*)

1Pos238* **酸性及び中性におけるナトリウムイオンポンプ KR2 のレチナール結合ポケットの固体 NMR 構造解析**

Solid-state NMR analysis of retinal binding pocket structure of sodium ion pump, KR2, at acidic and neutral pH

Arisu Shigeta[1], Shota Ito[2], Takashi Okitsu[3], Akimori Wada[3], Keiichi Inoue[2,4], Hideki Kandori[2], Izuru Kawamura[1] ([1]*Graduate School of Engineering, Yokohama National University*, [2]*Nagoya Institute of Technology*, [3]*Kobe Pharmaceutical University*, [4]*JST PRESTO*)

1Pos239 **Na+/H+ハイブリッドポンプロドプシン KR2 における His30 の役割**

Role of His30 in Na+/H+ Hybrid Pumping Rhodopsin KR2

Sahoko Tomida[1], Shota Ito[1], Rei Abe-Yoshizumi[1], Keiichi Inoue[1,2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech. Kandori Laboratory*, [2]*PRESTO, JST*)

1Pos240* **FTIR 分光法によって明らかになった光駆動内向きプロトンポンプの輸送機構**

Transport mechanism of light-driven inward proton pump revealed by FTIR spectroscopy

Shota Ito[1], Sahoko Tomida[1], Yoshitaka Kato[1], Yurika Nomura[1], Satoshi Tsunoda[1], Keiichi Inoue[1,2], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*PRESTO, JST*)

1Pos241 **H^+ポンプ型ロドプシンの比較研究：H^+ donor 残基の相互置換による検討**

Replacements of “donor” residues in the light-driven H^+-pump rhodopsins

Koki Nishiya[1], Syogo Sasaki[2], Jun Tamogami[3], Takashi Kikukawa[2], Tomoyasu Aizawa[2], Naoki Kamo[2], Makoto Demura[2] ([1]*Facu. Sci., Univ. Hokkaido*, [2]*Grad. Sch. Life Sci., Univ. Hokkaido*, [3]*Facu. Phar., Univ. Matsuyama*)

1Pos242 **アセタブラリアロドプシン II のプロトン移動における D92 および C218 残基間の相互作用の役割**

Role of the interaction between D92 and C218 in the proton transfer reaction in *Acetabularia* rhodopsin II

Jun Tamogami[1], Takashi Kikukawa[2], Keisuke Okawa[1], Noboru Ohsawa[3,4], Kohei Date[1], Toshifumi Nara[1], Makoto Demura[2], Tomomi Kimura-Someya[3,4], Mikako Shirouzu[3,4], Shigeyuki Yokoyama[3,5], Seiji Miyauchi[6], Kazumi Shimono[6], Naoki Kamo[2] ([1]*College Pharm. Sci., Matsuyama Univ.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*, [3]*RIKEN SSBC*, [4]*RIKEN CLST*, [5]*RIKEN Structural Biology Laboratory*, [6]*Fac. Pharm. Sci., Toho Univ.*)

1Pos243* **固体 NMR によるバクテリオロドプシンの暗順応状態における Tyr185 の構造解析**

Structure of Tyr185 in dark-adapted bacteriorhodopsin as studied by solid-state NMR

Yuto Otani[1], Arisu Shigeta[1], Yoko Kebukawa[1], Kensei Kobayashi[1], Takashi Okitsu[2], Akimori Wada[2], Satoru Tuzi[3], Akira Naito[1,3], Izuru Kawamura[1] ([1]*Grad. Sch. Eng, Yokohama Natl Univ.*, [2]*Kobe Pharm. Univ.*, [3]*Univ. of Hyogo*)

1Pos244 **光駆動ナトリウムポンプ KR2 の多量体形成に重要なアミノ酸残基**

Oligomerization of light-driven sodium pump KR2 is important for ion transport activity

Rei Abe-Yoshizumi[1], Shota Ito[1], Mikihiro Shibata[3], Keiichi Inoue[1,2], Takayuki Uchihashi[3], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*JST PRESTO*, [3]*Dept. Physics, Kanazawa Univ.*)

1Pos245* **Theoretical study on molecular mechanism of a light-driven ion transport of Halorhodopsin**

Ryo Oyama, Shigehiko Hayashi (*Grad. Sch. Sci., Kyoto Univ.*)

1Pos246 **Rhodobacter capsulatus PYP の複合体形成に伴う構造変化**

Complex induced structural changes of Rhodobacter capsulatus Photoactive Yellow Protein

Yoichi Yamazaki, Yohei Shibata, Hironari Kamikubo (*Grad. Sch. Mat. Sci. NAIST*)

1Pos247 **Rc-PYP の光依存的に形成する複数種の複合体解析**

Analysis of light dependent multiple complex formation of Rc-PYP

Yohei Shibata, Yoichi Yamazaki, Keito Yoshida, Shoki Nakata, Hironari Kamikubo (*Analysis of light dependent multiple complex formation of Rc-PYP*)

1Pos248* **同位体標識試料を用いたBLUFドメインの水素結合環境の解明**
Analysis of a hydrogen bonding network of the BLUF domain using isotope-labeled samples
Takashi Nagai[1], Tatsuya Iwata[1], Shota Ito[1], Mineo Iseki[2], Masakatsu Watanabe[3], Masashi Unno[4], Shinya Kitagawa[1], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*Dept. Pharmacol., Toho Univ.*, [3]*Grad. Sch. for Creation of Photonics Indust.*, [4]*Dept. of Chem. and Applied Chem. Saga Univ.*)

1Pos249 **ホタルオキシルシフェリン吸収・蛍光スペクトルにおける水和効果**
Hydration effects on absorption and fluorescence spectra of firefly oxyluciferin
Miyabi Hiyama[1], Yoshifumi Noguchi[1], Hidefumi Akiyama[1], Kenta Yamada[2], Nobuaki Koga[2] ([1]*ISSP, Univ. Tokyo*, [2]*Grad. Sch. Info. Sci., Nagoya Univ.*)

光生物：光合成 / Photobiology: Photosynthesis

1Pos250 **光化学系IIにおけるクロロフィル励起三重項状態の赤外分光解析**
FTIR analysis on the localization of the excited triplet state of chlorophyll in photosystem II
Tatsuya Mitomi, Ryo Nagao, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

1Pos251 **光合成水分解反応におけるメタノール阻害機構の赤外分光解析**
FTIR study on the mechanism of methanol inhibition in the S-state cycle of photosynthetic water oxidation
Haruna Yata, Tatsuki Shimizu, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

1Pos252* **Structure and Function of Novel Carbonyl-Carotenoid bound to Light-Harvesting Complex II from Transplastomic Lettuce**
Nami Yamano[1], Kentaro Ifuku[2], Hideki Hashimoto[1,3,5], Norihiko Misawa[4], Ritsuko Fujii[1,5] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*Grad. Sch. Biostudy, Kyoto Univ.*, [3]*Sch. Sci. Tech., Kwansei Gakuin Univ.*, [4]*Res. Inst. Biores. Biotech., Ishikawa prefectural Univ.*, [5]*OCARINA, Osaka City Univ.*)

1Pos253 **偏光全反射赤外分光法による光合成水分解Mnクラスター周辺のプロトン化構造の解析**
Protonation structure around the water-oxidizing Mn cluster in photosystem II revealed by polarized ATR-FTIR spectroscopy
Shin Nakamura, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

1Pos254 **酸素発生系マンガンクラスターの配位水分子の化学的性質**
Chemical properties of terminal water ligands of the Mn cluster
Hiroki Nagashima, Hiroyuki Mino (*Grad. Sch. Sci. Nagoya Univ.*)

1Pos255* **光化学系IIと阻害剤アジ化物イオンとの共結晶化とX線結晶構造解析**
Co-crystallization of photosystem II with an inhibitor NaN_3, and its structural analysis
Shoya Tamaru[1], Yasufumi Umena[2], Jian-Ren Shen[2] ([1]*Graduate School of Natural Science and Technology, Okayama University.*, [2]*Reseach Institute for Interdisciplinary Science, Okayama University.*)

1Pos256 **時間分解EPR法を用いたPSII反応中心に生成する初期電荷分離構造・電子的相互作用の解析**
Time resolved EPR study on orientations and electronic couplings of the primary charge-separated state in the PSII reaction center
Reina Minobe[1], Masashi Hasegawa[1], Shusuke Katagiri[3], Takahiro Sakai[2], Hiroki Nagashima[2], Takashi Tachikawa[1], Hiroyuki Mino[2], Yasuhiro Kobori[1] ([1]*Graduate School of Science, Kobe Univ.*, [2]*Graduate School of Science, Nagoya Univ.*, [3]*Graduate School of Science, Shizuoka Univ.*)

1Pos257 **FTIR分光電気化学法を用いた光化学系IIにおける第一キノン電子受容体 Q_A の酸化還元電位計測**
Measurement of the redox potential of the primary quinone electron accepter Q_A in photosystem II by FTIR spectroelectrochemistry
Ayaka Ohira, Ryo Nagao, Takumi Noguchi, Yuki Kato (*Grad. Sch. Sci., Nagoya Univ.*)

1Pos258 **光化学系IIにおけるキノン電子受容体の電子移動制御機構**
Regulation mechanism of electron transfer between quinone electron acceptors in Photosystem II
Yosuke Nozawa, Takumi Noguchi (*Division of Material Science, Graduate School of Science, Nagoya University*)

1Pos259 **Energy gap dependence for the exciton relaxation rate using Time-dependent renormalized Redfield theory**
Akihiro Kimura (*Graduate School of Science, Nagoya Univeristy*)

光生物：光遺伝学･光制御 / Photobiology: Optogenetics & Optical Control

1Pos260 **Photozipper-DNA複合体平衡の定量的モデル**
Quantitative modeling of the equilibria among Photozipper-DNA complexes
Yoichi Nakatani, Osamu Hisatomi (*Grad. Sch. Sci., Osaka Univ.*)

1Pos261 **光制御型bZIPモジュールPhotozipperの構造変化の変異体解析**
Mutational analyses of the conformational switching of a light-regulated bZIP module, Photozipper
Osamu Hisatomi (*Graduate School of Science, Osaka University*)

1Pos262 **水晶微量天秤による光制御型bZipモジュールphotozipperのDNA結合の解析**
The DNA-binding of a light-regulated bZIP module, photozipper, analyzed by quartz crystal microbalance
Samu Tateyama, Osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

1Pos263* **真正細菌のポンプ型ロドプシンの機能転換およびその分子メカニズムについての研究**
Functional conversion of eubacterial pump rhodopsins and the investigation of the molecular mechanism
Yurika Nomura[1], Keiichi Inoue[1,2], Shota Ito[1], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*JST PRESTO*)

1Pos264 **光駆動内向きプロトンポンプの発見**
Natural light-driven inward proton pump
Keiichi Inoue[1,2], Shota Ito[1], Yoshitaka Kato[1], Yurika Nomura[1], Mikihiro Shibata[3,4], Takayuki Uchihashi[3,4], Satoshi Tsunoda[1], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*JST PRESTO*, [3]*Faculty Sci., Kanazawa Univ.*, [4]*Bio-AFM Frontier Research Center, Kanazawa Univ.*)

1Pos265 **電気生理実験により解析した光駆動型ナトリウムポンプの輸送機構**
Transport mechanism of NaRs studied by electrophysiology measurement
Yuko Kozaki[1], Satoshi Tsunoda[1], Keiichi Inoue[1,2], Rei Abe-Yoshizumi[1], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*JST PRESTO*)

バイオインフォマティクス：構造ゲノミクス / Bioinformatics: Structural genomics

1Pos266 **An index to select homologous sequences with the same functional region**
Shoichiro Kato[1], Hiroyuki Toh[2], Wataru Nemoto[1] ([1]*Life Sci. & Eng., Grad. Sch. of Sci. & Eng*, [2]*Dept. Biomed. Chem., Scl. of Sci. & Tech., Kwansei Gakuin Univ.*)

1Pos267 **GWAS データによる疾患関連複合体モデルの予測**
Prediction of disease related supramolecule models using GWAS data
Toshiyuki Tsuji[1,2], Atsushi Hijikata[1], Takao Yoda[1], Tsuyoshi Shirai[1] ([1]*Nagahama Institute of Bio-Science and Technology*, [2]*MITA International School*)

1Pos268 **Predictions of cancer causing mutations that potentially affect GPCR-GPCR interaction**
Shunsuke Fujishiro[1], Vachiranee Limviphuvadh[2], Sebastian Maurer-Stroh[2], Yoshihiro Yamanishi[3], Hiroyuki Toh[4], Wataru Nemoto[1] ([1]*Life Sci. & Eng., Grad. Sch. of Sci. & Eng., Tokyo Denki Univ.*, [2]*BII, A*STAR.*, [3]*Med. Ins. of Bioreg., Kyushu Univ.*, [4]*Dept. Biomed. Chem., Sci. & Tech., Kwansei Gakuin Univ.*)

1Pos269 **ベクトル表現化したアミノ酸残基のマッチングによるタンパク質ーリガンド結合予測**
A new approach for protein-ligand binding predictions based on matching of vector-represented amino acid residues
Atsushi Hijikata, Masafumi Shionyu, Tsuyoshi Shirai (*Nagahama Inst. Bio-Sci. Tech.*)

1Pos270 **β-Trefoil タンパクのフォールディングに重要な残基に関する残基間平均距離統計に基づく解析**
Analysis of residues significant for folding of beta-trefoil proteins based on the inter-residue average distance statistics
Takuya Kirioka, Takeshi Kikuchi (*Dept. of Bioinfo., Col. Life Sci., Ritsumeikan Univ.*)

1Pos271* **多剤認識転写因子 LmrR における薬剤分子認識機構の計算化学的解析**
Computational study on the mechanism of multidrug recognition by a transcriptional repressor LmrR
Kazuho Cryershinozuka, Tadaomi Furuta, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech.*)

1Pos272 **Lysozyme superfamily のアミノ酸配列解析によるフォールディング領域予測**
Folding region predictions by amino acid analysis of lysozyme superfamily proteins
Takuto Nakashima, Michiro Kabata, Takeshi Kikuchi (*Dept. of Bioinfo., Col. Life Sci., Ritsumeikan Univ.*)

1Pos273 **二次構造順序の変化によって起こる蛋白質フォールドの多様化**
Loop connectivity change drives protein fold divergence
Shintaro Minami[1], George Chikenji[2], Motonori Ota[1] ([1]*Grad. Sch. of Comput. Sci., Nagoya Univ.*, [2]*Grad. Sch. of Eng., Nagoya Univ.*)

1Pos274 **タンパク質の立体構造とアミノ酸配列間の疎水性の関係**
The relationship between hydrophobicity in amino acid sequence and three-dimensional structure of a protein
Kohei Ohnishi, Takeshi Kikuchi (*Dept. of Bioinf., Col. of Life Sci., Ritsumeikan Univ.*)

数理生物学 / Mathematical biology

1Pos275 **Role of interdomain communication in pacemaking circadian rhythm studied by a single molecule model of KaiC**
Shota Hashimoto, Sumita Das, Masaki Sasai, Tomoki P. Terada (*Dept. Comput. Sci. Eng., Grad. Sch. Eng., Nagoya Univ.*)

1Pos276* **A stochastic simulation study on the correlation between circadian oscillation and ATPase activity of KaiC hexamer**
Sumita Das, Shota Hashimoto, Tomoki P. Terada, Masaki Sasai (*Department of Computational Science and Engineering, Nagoya University,Nagoya*)

1Pos277 **数理モデルによる心筋細胞の集団効果の解析**
Community effect of cardiomyocytes in beating rhythms is ruled by stable cells
Tatsuya Hayashi[1], Tetsuji Tokihiro[1,2], Hiroki Kurihara[2,3], Fumimasa Nomura[4], Kenji Yasuda[2,5] ([1]*Grad. Sch. Math. Sci., The Univ. of Tokyo*, [2]*JST, CREST*, [3]*Grad. Sch. Med., The Univ. of Tokyo*, [4]*Inst. Biomat. Bioeng., Tokyo Medical and Dental Univ.*, [5]*Fac. Sci. Eng., Waseda Univ.*)

1Pos278 **Discreteness-induced transition in multi-body reaction systems**
Yohei Saito, Yuki Sughiyama, Tetsuya Kobayashi (*IIS, Univ. Tokyo*)

1Pos279 **Probability Eddy currents in stochastic gene expression dynamics in eukaryotes**
Bhaswati Bhattacharyya[1], Masaki Sasai[1,2] ([1]*Department of Computational science and engineering, Nagoya University*, [2]*Department of Applied Physics, Nagoya University*)

1Pos280 **集団増殖系における定常状態熱力学**
Steady State Thermodynamics in Population Dynamics
Yuki Sughiyama, Tetsuya J. Kobayashi (*IIS, Univ. Tokyo*)

1Pos281 **上皮陥入過程における三次元多細胞動態の力学制御機構**
Mechanical regulatory mechanism of 3D multicellular dynamics during epithelial invagination
Satoru Okuda, Mototsugu Eiraku (*RIKEN Center for Developmental Biology*)

1Pos282 **Modeling folding of epithelial cell sheets**
FuLai Wen[1], YuChiun Wang[2], Tatsuo Shibata[1] ([1]*RIKEN Quantitative Biology Center*, [2]*RIKEN Center for Developmental Biology*)

1Pos283 **等方的なアクチンミオシン細胞骨格におけるモーター誘起応力に関する理論**
Theory on motor-induced stress in an isotropic actomyosin cytoskeleton
Tetsuya Hiraiwa (*Dept. Sci., Univ. Tokyo*)

1Pos284 **Phase-field simulations of the basic cell-cell effects of adhesion and chemoattractant on multi-cellular interaction**
Daisuke Imoto[1,5], Satoshi Sawai[1,2,3], Shuji Ishihara[4] ([1]*Dept. Basic Sci., Grad. School of Arts and Sci., Univ. of Tokyo*, [2]*Research Ctr for Complex Systems Biology, Univ. of Tokyo*, [3]*JST PRESTO*, [4]*School of Sci. Eng., Meiji Univ.*, [5]*National Research Institute of Police Science*)

1Pos285 **様々な回転する自走粒子の集団運動**
Collective motion of various kinds of rotating self-propelled particle
Ken Nagai[1], Yutaka Sumino[2], Chate Hugues[3,4], Kazuhiro Oiwa[5,6], Takuma Sugi[7], Hideo Iwasaki[8] ([1]*Sch. Mater. Sci., JAIST*, [2]*Dep. Appl. Phys., Tokyo Univ. Sci.*, [3]*CEA-Saclay*, [4]*Beijing Comp. Sci. Res. Ctr.*, [5]*Adv. ICT Res. Inst., NICT*, [6]*Grad. Sch. Sci., Univ. Hyogo*, [7]*Mol. Neurosci. Res. Ctr., Shiga Univ. of Med. Sci.*, [8]*Sch. Adv. Sci. Eng., Waseda Univ.*)

非平衡･生体リズム / Nonequilibrium state & Biological rhythm

1Pos286 **蛍光分光法による時計タンパク質 KaiC の動的構造変化の解析**
Spectroscopic characterization of the conformational change of the cyanobacterial clock protein KaiC
Atsushi Mukaiyama[1,2], Jun Abe[1], Yoshihiko Furuike[1,2], Eiki Yamashita[3], Takao Kondo[4], Shuji Akiyama[1,2] ([1]*IMS, CIMoS*, [2]*SOKENDAI*, [3]*IPR*, [4]*Naogya Univ.*)

1Pos287* **高速原子間力顕微鏡によって明らかにする Kai タンパク質間の動的相互作用のリン酸化状態依存性**
HS-AFM images reveal dynamic interaction between Kai proteins dependent on phosphorylation states of KaiC
Shogo Sugiyama[1], Tetsuya Mori[2], Takayuki Uchihashi[1,3], Johnson Carl H.[2], Toshio Ando[1,3] ([1]*Dept. of phys., Kanazawa Univ.*, [2]*Dept. of Biol. Sci., Univ. Vanderbilt*, [3]*Bio-AFM FRC., Kanazawa Univ.*)

1Pos288 **生命システムの振動現象における頑健性と可塑性の互恵的関係**
Reciprocity between robustness and plasticity in biological oscillators
Tetsuhiro S. Hatakeyama, Kunihiko Kaneko (*Department of Basic Science, The University of Tokyo*)

1Pos289* **自律的な振動運動を示す微小管リング状集合体**
Mechanical Oscillation of Dynamic Microtubule Rings
Masaki Ito[1], Kabir Arif Md. Rashedul[2], Md. Sirajul Islam[1], Daisuke Inoue[2], Shoki Wada[1], Kazuki Sada[1,2], Akihiko Konagaya[3], Akira Kakugo[1,2] ([1]*Grad. of CSE, Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*, [3]*DIS, TITECH*)

1Pos290* **キネシン駆動微小管のパターン形成と局所相互作用**
Configuring Dynamic Patterns of Microtubules Driven by Kinesins
Sakurako Tanida[1], Ken'ya Furuta[2], Kaori Nishikawa[1], Hiroaki Kojima[2], Masaki Sano[1] ([1]*Graduate School of Science, The University of Tokyo*, [2]*National Institute of Information and Communications Technology*)

1Pos291* **離散的な相互作用を行う振動子ネットワークの解析**
Analysis of nonlinear oscillator network with discrete interactions
Manami Ito[1], Masahiro Takinoue[1,2] ([1]*Dept. Comput. Intell. Syst. Sci., Tokyo Tech.*, [2]*Dept. Comput. Sci., Tokyo Tech.*)

1Pos292* **混み合い状況下におけるマイクロ粒子のラチェット輸送**
Ratchet transport of microparticles in crowded conditions
Masayuki Hayakawa[1], Yusuke Kishino[2], Masahiro Takinoue[1,2,3] ([1]*Dept. of Comput. Intell. and Syst. Sci., Tokyo Tech.*, [2]*Dept. of Engineering, Tokyo Tech.*, [3]*Dept. of Computer Science, Tokyo Tech.*)

1Pos293 **「ゆらぎ」が創り出す「秩序構造」：細胞の混雑環境のモデリング**
Fluctuation Creates Exotic Spatial-Order: Verification with a Simple Crowding Cellular-Model
Soutaro Oda[1], Chwen-Yang Shew[2], Kenichi Yoshikawa[1] ([1]*Faculty of Life and Medical Sciences, Doshisha University*, [2]*Department of Chemistry, CSI, City University of New York*)

1Pos294 **置かれた環境を感応する自発運動系：化学的非平衡性により駆動する生物らしさを示す実空間モデル**
Smart response of chemically driven self-motile object: Real-world modeling
Shiho Sato[1], Hiroki Sakuta[2], Kenichi Yoshikawa[1,2] ([1]*Facul. Life Med. Sci., Univ. Doshisha*, [2]*Grad. Sch. Life Med. Sci., Univ. Doshisha*)

計測 / Measurements

1Pos295 **Shot noise free number and brightness 解析法による生細胞内グルココルチコイド受容体二量体化過程の時空間分布解析**
Spatio-temporal distribution analysis of glucocorticoid receptor dimerization in cells by shot noise free number and brightness analysis
Ryosuke Fukushima[1], Jotaro Yamamoto[2], Masataka Kinjo[2] ([1]*Grad. Sch. Life Sci., Hokkaido Univ.*, [2]*Faculty of Adv. Life Sci., Hokkaido Univ.*)

1Pos296 **蛍光異方性を用いた免疫センサの試作**
Development of Fluorescence Anisotropy Immunosensor
Seiichi Suzuki, Sena Hasegawa, Maki Takagi, Takuya Ito, Toshinori Kojima (*Faculty Sci. Tech., Seikei Univ.*)

1Pos297* 実時間選択的回収による免疫細胞の１細胞遺伝子発現解析
Single cell gene expression analysis of stimulated immune cells with real-time selection
Yumiko Tanaka[1], Yoshitaka Shirasaki[1,2], Mai Yamagishi[1,2], Kaede Miyata[1], Nobutake Suzuki[1], Osamu Ohara[2], Kazuyo Moro[2], Sotaro Uemura[1] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*IMS., Riken*)

1Pos298 カップ形状 AFM チップを用いた簡便な細胞間相互作用計測法の開発
Easy Measurement of Cell-Cell Interactions Using Cup-Shaped AFM Chip
Hyonchol Kim[1], Ayana Yamagishi[1], Miku Imaizumi[2], Chikashi Nakamura[1,2] ([1]*Biomed. Res. Inst., AIST*, [2]*Grad. Sch. Eng., Tokyo Univ. Agric. Technol.*)

1Pos299 アンルーフ法を用いた水溶液環境下における細胞内骨格のＡＦＭイメージング
An Unroofing Method to Observe the Cytoskeleton Directly at Molecular Resolution Using Atomic Force Microscopy
Eiji Usukura[1], Akihiro Narita[1], Akira Yagi[2], Shuichi Ito[2], Jiro Usukura[1] ([1]*Grad. Sch. Sci., Univ. Nagoya*, [2]*Olympus Co., Ltd.*)

1Pos300 原子間力顕微鏡による細胞機能と力学特性の単一細胞相関解析法
Atomic force microscopy for single-cell correlation analysis between cellular function and cell mechanical property
Ryosuke Tanaka[1], Yoshikatsu Akiyama[2], Jun Kobayashi[2], Masayuki Yamato[2], Okajima Takaharu[1] ([1]*Grad. Sch. Info. Tech. Univ. Hokkaido*, [2]*Inst. Adv. BioMed. Eng. Sci. Univ. Tokyo Women's Med.*)

1Pos301 デジタルマイクロ流体技術によるデジタルバイオアッセイ
Digital bioassay in digital microfluidic platform
Ryohei Kobayashi[1], Sadao Ota[1,2], Hiroyuki Noji[1,3] ([1]*Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*JST, PRESTO*, [3]*ImPACT, JST*)

1Pos302 左右両耳内部における脈波計測
Measurements of Pulse Waves in the Both Ears
Yoshitomi Morikawa (*AIST*)

1Pos303 ラボＸ線光源を用いたＸ線１分子動態観察
X-ray Single Molecule Observations using Laboratory X-ray Generator
Keigo Ikezaki[1], Ken Matsubara[1], Yuhuku Matsushita[1], Jae-won Chang[1], Hiroshi Sekiguchi[2], Yuji Sasaki[1] ([1]*University of Tokyo*, [2]*Spring-8/JASRI*)

1Pos304 MALDI 法におけるマトリクスの分光学的解析
Spectroscopic analysis of matrices in ionization process of matrix-assisted laser desorption/ionization
Noritaka Masaki, Shigetoshi Okazaki (*Dept. Med. Spec., Hamamatsu Univ. Sch. Med.*)

1Pos305 HbA1c の蛍光相関分析に及ぼすヘモグロビン光吸収の影響
Effects of hemoglobin absorption on fluorescence correlation analysis for HbA1c
Atsushi Matsuo, Yasutomo Nomura, Mayuka Chiba, Misaki Naraoka (*Maebashi Institute of technology*)

バイオイメージング / Bioimaging

1Pos306* 高速原子間力顕微鏡を用いた癌細胞の核膜孔動態の可視化
High-speed atomic force microscopy visualization of the nuclear pores dynamics in cancer cells
Mahmoud Shaaban Mohamed[1,2,3], Yosuke Kikuchi[4], Watanabe-Nakayama Takahiro[2], Azuma Taoka[4], Akiko Kobayashi[1,2,3], Masaharu Hazawa[1,2,3], Noriyuki Kodera[2], Takayuki Uchihashi[2], Yoshihiro Fukumori[4], Toshio Ando[2], Richard Wong[1,2,3] ([1]*Cell-Bionomics Research Unit, Kanazawa University*, [2]*Bio-AFM Frontier Research Center, Kanazawa University*, [3]*Lab of Mol. Cell Biol. Institute of Science and Engineering, Kanazawa University*, [4]*Institute of Science and Engineering, Kanazawa University*)

1Pos307 高速 AFM による抗体分子の動的観察
The dynamic behaviors of antibody molecules
Yoko Kawamoto-Ozaki, Norito Kotani, Kumaresan Ramanujam, Aya Murakami, Takashi Morii, Takao Okada (*Research Institute of Biomolecule Metrology Co.,Ltd.*)

1Pos308 高速 AFM によるタンパク質の動的観察に向けた立体パターン基板の作製
Fabrication of 3D-patterned Substrate as a Platform for HS-AFM Observation of Protein Dynamics
Akane Goto[1], Shin'nosuke Yamanaka[1], Mikihiro Shibata[1,2], Takayuki Uchihashi[1,2], Noriyuki Kodera[1,2], Toshio Ando[2] ([1]*Dept. of phys., Kanazawa Univ.*, [2]*Bio-AFM FRC*)

1Pos309 高速スイッチング蛍光タンパク質と改良された SPoD-ExPAN による超解像イメージング
Superresolution imaging of live cells by fast photoswitching fluorescent protein and improved SPoD-ExPAN microscopy
Tetsuichi Wazawa[1,2], Yoshiyuki Arai[1,2], Tomoki Matsuda[1,2], Hiroki Takauchi[1], Yoshinobu Kawahara[1,2], Takashi Washio[1,2], Takeharu Nagai[1,2] ([1]*ISIR, Osaka Univ.*, [2]*CREST, JST*)

1Pos310 生細胞核内における INO80 クロマチン再構成複合体の 1 分子イメージング
Single-molecule imaging of the INO80 chromatin remodeling complex in the living cell nucleus
Yuma Ito[1], Masahiko Harata[2], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*Grad. Sch. Agr. Sci., Tohoku Univ.*)

1Pos311* アロディニア特異的な痛みに対する鎮痛薬評価系の確立に関する fMRI 研究
An fMRI study to establish an evaluation system of analgesic agents on allodynia-specific pain
Naoya Yuzuriha[1], Sosuke Yoshinaga[1], Mitsuhiro Takeda[1], Hiroshi Sato[2], Hiroaki Terasawa[1] ([1]*Fac. Life Sci., Kumamoto Univ.*, [2]*Bruker Biospin K.K.*)

1Pos312 **CLIP-170 phosphorylation mediates repositioning of microtubule-organizing center during T cell activation**
Wei Ming Lim, Yuma Ito, Makio Tokunaga, Kumiko Sakata-Sogawa (*Sch. Life Sci. Tech., Tokyo Inst. Tech.*)

1Pos313 **核小体構成タンパク質動態の１分子イメージング定量解析**
Single molecule imaging and quantitative analysis of Nucleolar-localized protein dynamics
Daiki Matsumoto[1], Yuma Ito[1], Noriko Saitoh[2], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*IMEG, Kumamoto Univ.*)

1Pos314 **高速超解像光学顕微鏡を用いた出芽酵母の膜交通の観察**
Observations of the membrane traffic in living yeast cells via the high-speed super-resolution optical microscope
Daisuke Miyashiro[1], Kazuo Kurokawa[1], Akihiko Nakano[1,2] ([1]*Riken, RAP*, [2]*University of Tokyo, Dep. Bio. Sci.*)

1Pos315* **新規微分干渉顕微法を用いた生細胞ヘテロクロマチンにおける物質密度のイメージング**
“Density” imaging of heterochromatin in live mouse cells using OI-DIC microscopy
Ryosuke Imai[1,2], Tadasu Nozaki[1], Tomomi Tani[3], Kayo Hibino[1,2], Michael Shribak[3], Kazuhiro Maeshima[1,2] ([1]*Natl. Inst. of Genet.*, [2]*SOKENDAI*, [3]*MBL, Woods Hole, USA*)

1Pos316 **線虫 *C. elegans* 胚発生における細胞動態の個体差定量解析**
Quantitative analysis of variability of cellular dynamics in *C. elegans* embryogenesis
Yusuke Azuma, Shuichi Onami (*RIKEN QBiC*)

1Pos317 **神経分化時における神経細胞内温度イメージング**
Imaging of intracellular temperature in PC12 cell nerve differentiation
Masaki Kinoshita[1,2], Kohki Okabe[3,4], Hisashi Tadakuma[2], Yoshie Harada[1,2,5] ([1]*Grad. Sch. Bio., Kyoto Univ.*, [2]*iCeMS, Kyoto Univ.*, [3]*Grad. Sch. Pharm, Tokyo Univ.*, [4]*PRESTO, JST*, [5]*IPR, Osaka Univ.*)

1Pos318* **ヒト免疫応答の１細胞実時間イメージングによるアレルギー診断の可能性**
Potential allergy diagnosis by real-time single-cell secretion imaging of human immune response
Kaede Miyata[1], Yoshitaka Shirasaki[1,2], Nobutake Suzuki[1], Hiroki Kabata[3], Mai Yamagishi[1,2], Osamu Ohara[2], Koichi Fukunaga[3], Kazuyo Moro[2], Sotaro Uemura[1] ([1]*Department of Bological Sciences, Graduate school of Tokyo*, [2]*Institute of Physical and Chemical Research, IMS.*, [3]*Division of Pulmonary Medicine, Keio University*)

1Pos319 **ソフトウェア「閻魔」と EMC アルゴリズムを用いたタンパク質３次元電子密度分布の再構成：XFEL-CXDI 実験を想定したシミュレーション**
Reconstruction of three-dimensional structures of a protein with software ENMA and EMC algorithm: A simulation for XFEL-CXDI experiment
Takashi Yoshidome[1], Yuki Sekiguchi[2,3], Tomotaka Oroguchi[2,3], Masayoshi Nakasako[2,3], Mitsunori Ikeguchi[4] ([1]*Dep. of Appl. Phys., Tohoku Univ.*, [2]*Fac. of Sci. and Tech., Keio Univ.*, [3]*RIKEN SPring-8 Center*, [4]*Grad. Sch. of Med. Life Sci. Yokohama City Univ.*)

1Pos320 **X 線自由電子レーザーを用いた低温コヒーレントＸ線回折イメージングによるシアノバクテリアの三次元構造解析**
Three-dimensional structure of a cyanobacterium visualized by cryogenic coherent X-ray diffraction imaging using X-ray free-electron laser
Amane Kobayashi[1,2], Yuki Sekiguchi[1,2], Koji Okajima[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Yayoi Inui[3], Takeshi Hirakawa[3], Sachihiro Matsunaga[3], Masaki Yamamoto[2] ([1]*Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*, [3]*Sci. Tech., Tokyo Univ. Sci.*)

第2 日目（11 月26 日（土））／Day 2 (Nov. 26 Sat.) 大会議室 101 ＋102、多目的ホール／Conference Room 101+102, Multi-Purpose Hall

蛋白質：構造 / Protein: Structure

2Pos001* **カルシウム依存的な鞭毛運動の制御に関わるタンパク質カラクシンの構造解析**
Structural analysis of calaxin, calcium-dependent flagellar movement regulator
Tomoki Shojima[1], Feng Hou[1], Yusuke Takahashi[1], Masahiko Okai[1], Katsutoshi Mizuno[2], Kazuo Inaba[2], Takuya Miyakawa[1], Masaru Tanokura[1] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*Shimoda Marine Research Center, Univ. Tsukuba*)

2Pos002 **Porphyromonas gingivalis の T9SS によって分泌される PGN_0123 の構造**
Structure of PGN_0123, a Type IX secretion substrate of Porphyromonas gingivalis
Yusuke Handa[1], Keiko Sato[2], Koji Nakayama[2], Katsumi Imada[1] ([1]*Grad. Sch. Sci. Osaka Univ.*, [2]*Grad. Sch. Biomedical Sci., Univ. Nagasaki*)

2Pos003 **時計タンパク質 KaiC のリン酸化状態と脱リン酸化状態における構造上の差異**
Structural Differences between Phosphorylated and Dephosphorylated States of Clock Protein KaiC
Yoshihiko Furuike[1], Jun Abe[1], Eiki Yamashita[3], Takao Kondo[4], Shuji Akiyama[1,2] ([1]*Research Center of Integrative Molecular Systems (CIMoS), Institute for Molecular Science (IMS)*, [2]*Department of Functional Molecular Science, SOKENDAI (The Graduate University for Advanced Studies)*, [3]*Institute for Protein Research, Osaka University*, [4]*Graduate School of Science, Nagoya University*)

2Pos004 **Structural characterization of Hsp104 from a thermophilic fungus, Chaetomium thermophilum**
Yosuke Inoue (*Tokyo University of Agriculture and Technology*)

2Pos005 **圧力応答を示す YFP 挿入変異体の高圧下での結晶構造**
Crystal structure of a pressure sensitive YFP mutant under high pressure
Mika Tsujii[1], Takayuki Nagae[2], Keiko Yoshizawa[3], Tomonobu Watanabe[3], Masahiro Nishiyama[4], Nobuhisa Watanabe[2], Tatsuya Kawaguchi[1], Katsumi Imada[1] ([1]*Grad. Sch. Sci., Univ. Osaka*, [2]*SRRC. Nagoya Univ.*, [3]*QBiC, Riken.*, [4]*Grad. Sch. Sci., Univ. Kyoto*)

2Pos006 **Crystallization of Hepatitis B virus Core Protein in genotype C**
Katsumi Omagari (*Nagoya City University*)

2Pos007* **巨大タンパク質会合体ヘモシアニンの多孔質性結晶を用いた生体分子の包摂**
Encapsulation of biomacromolecules into porous crystal of a huge protein complex hemocyanin
Asuka Matsuno[1], Ye Yuxin[2], Yuki Ohnishi[2], Akira Kitamura[1,2], Masataka Kinjo[1,2], Satoshi Abe[4], Takafumi Ueno[4], Yoshikazu Tanaka[1,2,3], Min Yao[1,2] ([1]*Graduate School of Life Science, Hokkaido University*, [2]*Faculty of Advanced Life Science, Hokkaido University*, [3]*JST, PRESTO*, [4]*Department of Life Science and Technology, Tokyo Institute of Technology*)

2Pos008 **蛋白質結晶中の分子間静電相互作用計算**
Calculation of inter-molecular electrostatic interactions in protein crystals
Takuya Takahashi[1], Shigeru Endo[2], Masanori Ootaki[3], Yoko Sugawara[2] ([1]*Bioinfo., Coll. Life. Sci., Ritsumeikan Univ.*, [2]*Dept. Physics, Sch. Sci., Kitasato Univ.*, [3]*Dep. Pharmacology, St. Marianna Univ. School of Medicine*)

2Pos009 **X 線自由電子レーザー（XFEL）回折像からの生体分子三次元構造の復元プログラムの開発**
Development of 3D reconstruction program for coherent diffraction patterns obtained by XFEL
Miki Nakano[1], Osamu Miyashita[1], Slavica Jonic[2], Atsushi Tokuhisa[1], Daewoon Nam[3], Yasumasa Joti[4], Changyong Song[3], Florence Tama[1,5] ([1]*RIKEN AICS*, [2]*IMPMC, Sorbonne University - CNRS UMR 7590, UPMC Univ. Paris 6, MNHN, IRD UMR 206*, [3]*POSTECH, Korea*, [4]*JASRI XFEL*, [5]*Grad. Sch. Science, Nagoya Univ.*)

2Pos010 **Hsp90 の構造変化に関する理論的研究**
Theoretical study of a conformational change in Hsp90
Kazutomo Kawaguchi, Hidemi Nagao (*Inst. Sci. Eng., Kanazawa Univ.*)

2Pos011 **Building a database of 3D biological shapes for the interpretation of XFEL diffraction patterns**
Sandhya Tiwari[1], Osamu Miyashita[1], Florence Tama[1,2] ([1]*Riken Advanced Institute for Computational Science*, [2]*Nagoya University*)

2Pos012 **胆汁酸輸送体の分子動力学シミュレーション**
Molecular dynamics simulation of the bile acid transporter
Shin-ichiro Tasaki, Ryunosuke Yoshino, Yoshitaka Moriwaki, Kentaro Shimizu, Tohru Terada (*Grad. Sch. of Agri. Life Sci., Univ. of Tokyo*)

2Pos013* **フレキシィボタンパク質ータンパク質ドクイン： PaCS−MD の応用**
Flexible-Body Protein-Protein Docking: an Application of Parallel Cascade Selection Molecular Dynamics
Duy P. Tran[1], Akio Kitao[1,2] ([1]*UTokyo, GSFS*, [2]*UTokyo, IMCB*)

2Pos014 **分子動力学シミュレーションを用いた Hras-GTP/GDP 複合体と溶媒水との水素結合の動きの解析**
Analysis of dynamics of hydrogen bond between the solvent water and the Hras-GTP/GDP complexes by molecular dynamics simulations
Takeshi Miyakawa[1], Ryota Morikawa[1], Masako Takasu[1], Kimikazu Sugimori[2], Kazutomo Kawaguchi[2], Hidemi Nagao[2] ([1]*Tokyo Univ. of Pharm. & Life Sci.*, [2]*Kanazawa Univ.*)

2Pos015 **分子動力学シミュレーションによる抗 HIV 中和抗体 PG16 の CDR-H3 における構造剛性の解析**
Molecular dynamics study of the structural rigidity of CDR-H3 of anti-HIV neutralizing antibody PG16
Ryo Kiribayashi[1], Hiroko Kondo[1], Daisuke Kuroda[2], Toru Saito[1], Jiro Kohda[1], Akimitsu Kugimiya[1], Yasuhisa Nakano[1], Yu Takano[1] ([1]*Hiroshima City Univ.*, [2]*Showa Univ.*)

2Pos016 **Molecular dynamics simulations of the basic amyloidogenic unit of IAPP**
Richa Tambi[1], Satoshi Kosuda[1], Gentaro Morimoto[2], Makoto Taiji[2], Yutaka Kuroda[1] ([1]*Tokyo University of Agriculture and Technology*, [2]*Quantitative Biology Center, RIKEN*)

2Pos017 **レプリカ交換モンテカルロ SAAP3D 法による C-ペプチドと Trp ケージの分子シミュレーション**
Molecular simulation of C-peptide and Trp-cage by SAAP3D-REMC method
Michio Iwaoka, Natsuki Babe, Yuya Shoji (*Tokai University, Department of Chemistry*)

2Pos018 **REST 法による TRP-cage のフォールディングシミュレーション**
In silico folding simulation of Trp-cage using the REST method and its variants
Motoshi Kamiya[1], Yuji Sugita[1,2,3] ([1]*AICS, RIKEN*, [2]*RIKEN*, [3]*QBiC, RIKEN*)

2Pos019 **タンパク質のフォールディング過程における階層性と不均一性の分子論的起源**
Molecular origin of heterogeneity and hierarchy behind protein folding
Toshifumi Mori[1,2], Shinji Saito[1,2] ([1]*IMS*, [2]*SOKENDAI*)

2Pos020 **プロリン型人工アミノ酸を含むペプチドの分子動力学計算**
Molecular dynamics simulation of peptide oligomers bearing the proline-type artificial amino acid
Yuko Otani[1], Satoshi Watanabe[1], Akio Kitao[2], Tomohiko Ohwada[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*IMCB, Univ. Tokyo*)

2Pos021 **Theoretical study of diffusion of plastocyanin with Langevin equation**
Makoto Wada, Satoshi Nakagawa, Shogo Kinoshita, Kurniawan Isman, Kouichi Kodama, Kazutomo Kawaguchi, Hidemi Nagao (*Nat. Sci. Kanazawa Univ.*)

2Pos022* **HIV-1 protease の触媒的加水分解反応に関する理論的研究**
Theoretical study on catalytic hydrolysis of HIV-1 protease
Masahiro Kaneso, Shigehiko Hayashi (*Grad. Sch. Sci., Kyoto. Univ.*)

蛋白質：構造機能相関 / Protein: Structure & Function

2Pos023 **ホタルルシフェラーゼの全原子を考慮した発光基質オキシルシフェリンの光吸収のｐＨ依存性の定量解析**
Quantitative Analysis of pH Effect on Absorption Peaks of Oxyluciferin by Considering All Atoms of Firefly Luciferase
Hironori Sakai[1], Itsuki Kaji[2], **Naohisa Wada**[2] ([1]*Insti. of Fluid Science, Tohoku Univ.*, [2]*Food Life Sciences, Toyo Univ.*)

2Pos024 **糖転移酵素の糖選択性とタンパク質認識に関わるアミノ酸の解析**
Sequence and structure analysis of glycosyltransferases for understanding the sugar selectivity and target recognition mechanisms
Go Miyasaka[1], Kenji Etchuya[2], Yuri Mukai[1,2] ([1]*Sch. Sci. & Tech., Meiji Univ.*, [2]*Grad. Sch. Sci. & Tech., Meiji Univ.*)

2Pos025 **アルカン合成関連酵素の機能発現における保存部位の役割**
Alanine scanning mutagenesis reveals functional roles of conserved residues in an enzyme for alkane biosynthesis
Masashi Nomura, Hisashi Kudo, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

2Pos026 **示差走査型蛍光定量法を用いたアルドケト還元酵素の化合物選択性の評価**
Evaluation of compound selectivity of aldo-keto reductases using differential scanning fluorimetry
Kabir Aurangazeb[1], Satoshi Endo[2], Naoki Toyooka[3], Mayuko Fukuoka[1], Kazuo Kuwata[1,4], **Yuji Kamatari**[5] ([1]*United Grad. Sch. Drug Dis. Med. Inf. Sci., Gifu Univ.*, [2]*Lab. Biochem., Gifu Pharm. Univ.*, [3]*Grad. Sch. Sci. Tech. Res., Univ. Toyama*, [4]*Grad. Sch. Med., Gifu Univ., Gifu Univ.*, [5]*Life Sci. Res. Ctr.*)

2Pos027 **Unique mechanism for broad substrate specificity of human MTH1**
Shaimaa Ali, **Teruya Nakamura**, Keisuke Hirata, Mami Chirifu, Shinji Ikemizu, Yuriko Yamagata (*Grad. Sch. Pharmaceut. Sci., Kumamoto Univ.*)

2Pos028* **ラン藻由来アルカン合成関連酵素の構造機能解析**
Structural and functional analysis of a cyanobacterial enzyme for alkane biosynthesis
Hisashi Kudo[1], Ryota Nawa[2], Yuuki Hayashi[1,2], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo.*, [2]*Dept. Pure & Applied Sci., Univ. Tokyo.*)

2Pos029 **クジラミオグロビンの分子進化**
Tracing evolution of whale myoglobin by resurrecting ancient proteins
Yasuhiro Isogai[1], Hiroshi Imamura[2], Setsu Nakae[3], Tomonari Sumi[4], Ken-ichi Takahashi[3], Taro Nakagawa[3], Antonio Tsuneshige[5], Tsuyoshi Sirai[3] ([1]*Dept. Biotech., Toyama Pref. Univ.*, [2]*Biomedical Res. Inst., AIST*, [3]*Dept. Comp. Bio-Sci., Nagahama Inst. Bio-Sci. Tech.*, [4]*Dept. Chem., Okayama Univ.*, [5]*Nano-Tech. Center, Hosei Univ.*)

2Pos030 **MEK1 リン酸化に伴う構造変化**
Structual dynamics of MEK1 activation through phosphorylation
Minami Ando, Kei Moritsugu, Akinori Kidera (*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*)

2Pos031 **アロステリーの概念拡張に向けて：トロンビンのアロステリック制御・再訪**
Toward Expanding the Concept of Allostery: Thrombin Allosteric Regulation, Revisited
Ikuo Kurisaki[1,2], Masayoshi Takayanagi[1,2], Barberot Chantal[1,2], Masataka Nagaoka[1,2] ([1]*Grad. Sch. Info. Sci. Univ. Nagoya*, [2]*JST-CREST*)

2Pos032 **サルコシン酸化酵素の反応生成物は４つの水チャネルの１つを選択的に移動する：平均力ポテンシャルによる検証**
Potential of mean force shows that the reaction product of sarcosine oxidase selectively exits from one of four water channels
Takami Saito[1], Go Watanabe[2], Daisuke Nakajima[2], Haruo Suzuki[2], Shigetaka Yoneda[2] ([1]*Grad. Sch. Sci., Kitasato Univ.*, [2]*Sch. Sci., Kitasato Univ.*)

2Pos033 **Characterizing NO diffusion in nitrite reductase: nitric oxide reductase complex**
Po-hung Wang[1], Kenta Yamada[1], Takehiko Tosha[2], Yoshitsugu Shiro[2,3], Yuji Sugita[1,4,5,6] ([1]*RIKEN Theoretical Molecular Science Laboratory*, [2]*RIKEN SPring-8 Center*, [3]*Graduate School of Life Science, University of Hyogo*, [4]*RIKEN Advanced Institute for Computational Science*, [5]*RIKEN Quantitative Biology Center*, [6]*RIKEN iTHES*)

2Pos034* **二段階緩和モード解析による蛋白質シミュレーションの動的解析**
Dynamical analysis of protein simulations by using two-step relaxation mode analysis
Naoyuki Karasawa[1], Ayori Mitsutake[1,2], Hiroshi Takano[1] ([1]*Grad. Sch. Sci. Technol., Keio Univ.*, [2]*JST, PRESTO*)

2Pos035 **MSES 法による EGFR キナーゼドメイン活性化の全原子構造解析**
Structural basis for activation of EGFR kinase domain at atomistic resolution revealed by multiscale enhanced sampling
Kei Moritsugu[1], Tohru Terada[2], Akinori Kidera[1] ([1]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*, [2]*Grad. Sch. of Agri. and Life Sci., Univ. of Tokyo*)

2Pos036 **生物学的レアイベントを再現する効率的構造サンプリング手法の開発**
Developments of conformational sampling methods for reproducing biologically rare events of proteins
Ryuhei Harada, Yasuteru Shigeta (*Center for Computational Sciences, University of Tsukuba*)

2Pos037 **タンパク質の長時間シミュレーションに関する緩和モード解析**
Relaxation mode analysis for long time simulations of proteins
Ayori Mitsutake[1,2], Hiroshi Takano[1] ([1]*Dept. Physics, Keio Univ.*, [2]*JST, PRESTO*)

2Pos038 **Molecular basis for Hsp104-mediated prion propagation in yeast**
Yoshiko Nakagawa[1,2], Hideki Taguchi[3], Motomasa Tanaka[1] ([1]*RIKEN Brain Science Institute*, [2]*Tokyo Institute of Technology*, [3]*Institute of Innovative Research, Tokyo Institute of Technology*)

2Pos039* **HSP70 のフタの構造動態は基質結合にいかに関与するか？**
How the lid exploits its structure dynamics in grasping the substrate in HSP70
Kohei Umehara[1], Naoya Tochio[2], Miho Hoshikawa[1], Shoji Ueki[3], Shin-ichi Tate[1,2] ([1]*Dept. Math. and Life Sci., Hiroshima Univ.*, [2]*RcMcD, Hiroshima Univ.*, [3]*Kagawa Sch. Pharm. Sci., Tokushima bunri Univ.*)

2Pos040 **New insights into high molecular weight complex formation of 2-Cys peroxiredoxin and its chaperone function**
Takamitsu Haruyama[1], Takayuki Uchihashi[1,2], Noriyuki Kodera[1], Toshio Ando[1], Hiroki Konno[1] ([1]*Bio-AFM FRC, Coll. Sci. & Eng., Kanazawa Univ.*, [2]*Coll. Sci. & Eng., Kanazawa Univ.*)

2Pos041 **Spectral characteristics of chimeric channelrhodopsins implicate the molecular identity involved in desensitization**
Alemeh Zamani, Toru Ishizuka, Hiromu Yawo (*Tohoku University*)

2Pos042 **タンパク質の構造と機能の相関を利用した Channelrhodopsin と MtrF の戦略的立体構造モデリング**
Strategic modeling of channelrhodopsins and MtrF based on the correlation between protein structures and functions
Hiroshi C. Watanabe[1,2], Yuki Yamashita[2], Marcus Elstner[3], Hiroshi Ishikita[1,2] ([1]*UTokyo, RCAST*, [2]*UTokyo, School of Engineering*, [3]*Karlsruhe Institute of Technology*)

2Pos043 **機械受容チャネル MscL のゲーティングにおいてメカノセンサーとゲートは密接に連動する**
Mechanosensor and the gate are tightly coupled in the mechano-gating of the bacterial mechanosensitive channel MscL
Yasuyuki Sawada[1], Takeshi Nomura[2], Masahiro Sokabe[1] ([1]*Mechanobiology Lab Nagoya Univ. Grad. Sch. Med.*, [2]*Physical Therapy Grad. Sch. Health Sciences Kyushu Nutrition Welfare Univ.*)

2Pos044 **電位依存性プロトンチャネル VSOP/Hv1 における亜鉛イオンの結合様式の解析**
A detailed analysis of the binding mode of a zinc ion to the voltage-gated proton channel VSOP/Hv1
Hiroko X. Kondo[1,2], Masayo Iwaki[3], Yusuke Kanematsu[1], Matsuyuki Shirota[2,4,5], Yasushige Yonezawa[6], Kengo Kinoshita[2,5,7], Hideki Kandori[3], Yu Takano[1] ([1]*GSIS, Hiroshima City Univ.*, [2]*GSIS, Tohoku Univ.*, [3]*Grad. Sch. Eng, Nagoya Inst tech*, [4]*Grad. Sch. Med., Tohoku Univ.*, [5]*ToMMo, Tohoku Univ.*, [6]*IAT, Kinki Univ.*, [7]*IDAC, Tohoku Univ.*)

2Pos045 **Molecular dynamics investigation of the full maltose transporter with and without the maltose binding protein MalE**
WeiLin Hsu, Tadaomi Furuta, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech.*)

2Pos046* **Binding and conformational dynamics of TOM20 and mitochondrial targeting signals using computational methods**
Arpita Srivastava[1], Osamu Miyashita[2], Florence Tama[1,3] ([1]*Dept. Phys., Sch. Sci., Nagoya Univ.*, [2]*RIKEN Adv. Inst. Comp. Sci.*, [3]*ITbM, Nagoya Univ.*)

2Pos047 **細胞間接着結合における α カテニン分子の力-構造-機能ダイナミクス**
Mechanical, structural and functional dynamics of α-catenin molecule at intercellular adherens junctions
Koichiro Maki[1,2], Taiji Adachi[1,2] ([1]*Institute for Frontier Medical Sciences, Kyoto University*, [2]*Department of Micro Engineering, Graduate School of Engineering, Kyoto University*)

2Pos048 **高速 AFM によるバクテリアコンデンシン複合体 MukBEF の構造動態の研究**
Structural dynamics of bacterial condensin complex MukBEF studied by HS-AFM
Hironori Yoneda[1], Kouichi Yano[2], Noriyuki Kodera[3,4], Kenta Yagi[1], Hironori Niki[2], Toshio Ando[1,3,5] ([1]*Div. of Math & Phys. Sci., Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ.*, [2]*Natl. Inst. of Genet.*, [3]*Bio-AFM FRC, Inst. of Sci. & Eng., Kanazawa Univ.*, [4]*PRESTO, JST*, [5]*CREST, JST*)

2Pos049 **高速 AFM によるダイナミン 1−アンフィファイジン複合体の動態観察**
High-Speed AFM imaging of dynamics of Dynamin1-Amphiphysin1 complexes
Daiki Ishikuro[1], Tetsuya Takeda[3], Toshiya Kozai[1], Yusuke Kumagai[1], Kaho Seyama[3], Huiran Yang[3], Hiroshi Yamada[3], Takayuki Uchihashi[1,2], Toshio Ando[1,2], Kohji Takei[3] ([1]*College of Science and Engineering, Kanazawa Univ.*, [2]*Bio-AFM FRC, Kanazawa Univ.*, [3]*Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama Univ.*)

2Pos050* **過飽和条件下におけるタンパク質分子内二次構造変化と芳香族アミノ酸立体配置**
Internal Secondary Structural Changes and Aromatic Rings Conformation of Protein under Supersaturated Condition
Yufuku Matsushita[1], Hiroshi Sekiguchi[2], Jae-won Chang[1], Keigo Ikezaki[1], Masaki Nishijima[3], Daizo Hamada[4], Yuji Goto[5], Yuji Sasaki[1,2] ([1]*Grad. Sch. Front. Sci., Univ. Tokyo*, [2]*JASRI/SPring-8*, [3]*UIC, Osaka Univ.*, [4]*Int. Res. Cent., Kobe Univ.*, [5]*IPR, Osaka Univ.*)

2Pos051 **二次元蛍光寿命相関分光法による脂質膜上シトクロム *c* の環境に依存した構造ゆらぎの研究**
Study of environment dependent dynamics of cytochrome *c* on a lipid membrane by 2D fluorescence lifetime correlation spectroscopy
Miyuki Sakaguchi[1], Masaru Yamanaka[2], Shun Hirota[2], Kunihiko Ishii[1,3], Tahei Tahara[1,3] ([1]*Molecular Spectroscopy Lab. RIKEN*, [2]*NAIST Graduate School of Materials Science*, [3]*RIKEN Center for Advanced Photonics*)

蛋白質：物性 / Protein: Property

2Pos052* **一分子蛍光分光測定により明らかになったユビキチンの変性状態の不均一性とゆっくりとしたダイナミクス**
Significant Heterogeneity and Slow dynamics of the Unfolded Ubiquitin Detected by Single-Molecule Fluorescence Spectroscopy
Masataka Saito[1,2], Kamonprasertsuk Supawich[1], Keiichiro Kushiro[3], Madoka Takai[3], Eric H.-L. Chen[4], Po-Ting Chen[4], Rita P.-Y. Chen[4], Hiroyuki Oikawa[1,2], Satoshi Takahashi[1,2] ([1]*Grad. Sch. Sci., Tohoku Univ.*, [2]*IMRAM, Tohoku Univ.*, [3]*Sch. Eng., Univ. Tokyo.*, [4]*IBC, Academia Sinica*)

2Pos053 **高活性型 DHFR 変異体の構造揺らぎの NMR 解析**
NMR analysis of structural fluctuation of a highly active mutant of *Escherichia coli* dihydrofolate reductase
Takuro Nobe, Yuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

2Pos054 **C-ペプチド領域をもたないウシ膵臓インスリンの二本鎖酸化的フォールディング経路**
Double-chain oxidative folding pathways of bovine pancreatic insulin without C-peptide region
Kenta Arai[1], Toshiki Takei[1,2], Reina Shinozaki[1], Yuya Asahina[2], Hironobu Hojo[2], Michio Iwaoka[1] ([1]*Tokai Univ.*, [2]*Inst. for Prot. Res., Osaka Univ.*)

2Pos055* **圧力ジャンプ FTIR 法を用いた β ラクトグロブリンのフォールディング反応解析**
A pressure-jump FTIR study of the folding reaction of β-Lactoglobulin
Satoshi Hayakawa, Tsubasa Yamamoto, Minoru Kato (*Dept. Applied Chemistry, Ritsumeikan Univ.*)

2Pos056 **700 MPa 高圧下で観測された蛍光蛋白質 Akane families の特異な蛍光挙動**
Unique properties of a GFP-like protein Akane families, observed under 700 MPa high-pressure
Akihiro Maeno[1], Yuko Kato[2], Mitsuru Jimbo[3], Kei Amada[4], Kazuyuki Akasaka[5] ([1]*Chem., Kansai Med. Univ.*, [2]*Electro. Res. Lab., Fukuoka Inst. Tech.*, [3]*Marine Biosci., Kitasato Univ.*, [4]*Life, Environ. and Mat. Sci., Fukuoka Inst. Tech.*, [5]*Grad. Sch. Life and Environ. Sci., Kyoto pref. Univ.*)

2Pos057 **タンパク質ジスルフィドイソメラーゼ様触媒活性を有する新規低分子ジセレニド化合物を用いた酸化的フォールディングに関する研究**
Study on the oxidative folding by using a novel small-molecular diselenide compound having protein disulfide isomerase-like activity
Yuki Asano, Haruhito Ueno, Michio Iwaoka, Kenta Arai (*Tokai Univ.*)

2Pos058* **分子内ジスルフィド結合がリポカリン型プロスタグランジン D 合成酵素の熱安定性及び構造安定性に与える影響**
Effects of an intramolecular disulfide bond on the thermal and conformational stability of lipocalin-type prostaglandin D synthase
Shogo Atsuji[1], Yoshiaki Teraoka[1], Young-Ho Lee[2], Yuji Goto[2], Takashi Inui[1] ([1]*Grad. Sch. Life & Envi. Sci., Osaka Pref. Univ.*, [2]*Inst. Prot. Res., Osaka Univ.*)

2Pos059 **タンパク質の翻訳時フォールディングにおけるリボソーム効果の粗視化分子動力学シミュレーション研究**
Ribosomal effects on protein cotranslatoinal folding studied by coarse-grained molecular dynamics simulation
Kazushi Mochizuki, Shoji Takada (*Graduate School of Science, Kyoto Univ.*)

2Pos060 **蛋白質の相互作用とダイナミクスに及ぼす高分子混み合いの影響：全原子分子動力学法による研究**
Influence of Macromolecular Crowding on the Dynamics and Interactions of proteins: All-atom Molecular Dynamics Study
Isseki Yu[1], Tadashi Ando[2], Takaharu Mori[1], Jaewoon Jung[3], Ryuhei Harada[3], Yuji Sugita[1,2,3], Michael Feig[4] ([1]*RIKEN*, [2]*RIKEN QBIC*, [3]*RIKEN AICS*, [4]*Michigan State Univ.*)

2Pos061* **高温条件下で変性した DEN4 ED3 の可逆的なオリゴマー形成の一残基置換による阻害**
Unusual reversible oligomerization of unfolded Dengue envelope protein domain 3 at high temperature and its abolition by a point mutation
Tomonori Saotome[1], Shigeyoshi Nakamura[2], Mohammad M. Islam[1], Akiko Nakazawa[2], Mariano Dellarole[3], Fumio Arisaka[4], Shun-ichi Kidokoro[2], Yutaka Kuroda[1] ([1]*Dept. of Biotech. and Life Sci., Tokyo Univ. of Agric. and Tech.*, [2]*Dept. of Bioeng., Nagaoka Univ. of Tech.*, [3]*CBS, Univ. of Montpellier*, [4]*Coll. of Biores. Sci., Univ. of Nihon*)

2Pos062 **球殻状超分子集合における局所的相互作用の役割**
The role of local interactions on the spherical shell-shaped supermoleculer assembly
Daisuke Sato[1], Hideaki Ohtomo[2], Atsushi Kurobe[2], Kazuo Fujiwara[1], Masamichi Ikeguchi[1] ([1]*Fac. of Sci. and Eng., Soka Univ.*, [2]*Fac. of Eng., Soka Univ.*)

2Pos063 **赤外分光法によるカルシウム結合タンパク質の金属配位構造解析 - 合成ペプチドアナログの凝集による問題**
Infrared study of the Ca^{2+}-coordination structures of Ca^{2+}-binding proteins: the problem of aggregation of synthetic peptide analogues
Masayuki Nara[1], Hisayuki Morii[2], Takuya Miyakawa[3], Masaru Tanokura[3] ([1]*TMDU*, [2]*AIST*, [3]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*)

2Pos064* **競争的凝集形成機構に基づいた蛋白質異常凝集の理解**
Understanding of aberrant protein aggregation based on the competitive aggregation mechanism
Masayuki Adachi, Masatomo So, Yuji Goto (*Inst. Protein Res., Osaka Univ.*)

2Pos065 **ヘパリンによるアミロイド線維形成の促進と抑制の分子機構**
Molecular mechanism underlying the heparin-induced acceleration and inhibition of amyloid fibrillation
Ayame Nitani, Hiroya Muta, Masayuki Adachi, Masatomo So, Yuji Goto (*Inst. Protein res., osaka Univ.*)

2Pos066 **キメラカルシトニンによるヒトカルシトニンアミロイド凝集阻害機構の解明**
Analysis of amyloid formation and inhibition mechanisms of human calcitonin by chimera calcitonin
Chiaki Ota[1], Hiroko Tanaka[1], Tomoyasu Aizawa[2], Yoichi Yamazaki[1], Mikio Kataoka[1], Hironari Kamikubo[1] ([1]*Grad. Sch. Mat. Sci., NAIST*, [2]*Grad. Sch. Life Sci., Hokkaido Univ.*)

2Pos067* **プリオンタンパク質とプリオンタンパク質を標的とする RNA 分子の Aβ 線維化への影響**
The effects of prion protein and a RNA molecule that binds to prion protein on Aβ fibrillation
Mamiko Iida[1,2], Tsukasa Mashima[1,2], Yudai Yamaoki[2], Takashi Nagata[1,2], Masato Katahira[1,2] ([1]*Grad. Sch. of Energy Sci., Kyoto Univ.*, [2]*Inst. of Adv. Energy, Kyoto Univ.*)

蛋白質：機能 / Protein: Function

2Pos068 **High-level expression, purification and characterization of the plant antimicrobial peptide snakin-1 in *Pichia pastoris***
Md. Ruhul Kuddus[1,2], Farhana Rumi[1], Motosuke Tsutsumi[1], Megumi Yamano[1], Masakatsu Kamiya[1], Takashi Kikukawa[1], Makoto Demura[1], Tomoyasu Aizawa[1] ([1]*Grad. Sch. Life Sci., Hokkaido Univ.*, [2]*Dhaka Univ.*)

2Pos069 **A study on tryptophan-dependent translation termination arrest of TnaC**
Tomoki Shinozawa, Ryo Iizuka, Zhuohao Yang, Takashi Funatsu (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

2Pos070* **1 分子不凍タンパク質の温度依存ダイナミクスと AgI との吸着関係**
Temperature Dynamics of Single Molecular AntiFreeze Proteins and adsorption to AgI interaction
Rio Okada[1], Tatsuya Arai[2], Yuhuku Matushita[1], Jae-won Chang[1], Hiroshi Sekiguchi[3], Tadashi Mori[4], Masaki Nishijima[4], Keigo Ikezaki[1], Sakae Tsuda[2], Yuji Sasaki[1,3] ([1]*Frontier Science., Adv. Material Science., Tokyo Univ.*, [2]*AIST / Grad. Schl. of Life Sci., Hokkaido Univ.*, [3]*JASRI*, [4]*Grad. Schl. of Eng., Div. of Ap. Chem., Osaka Univ.*)

2Pos071 **青色光センサータンパク BlrP1 の反応の光強度依存性**
Light intensity determines photoreaction of blue-light sensor protein BlrP1
Kosei Shibata, Yusuke Nakasone, Masahide Terazima (*Grad. Sch. Sci. Unv. Kyoto*)

2Pos072 反転膜小胞を用いたべん毛軸構造蛋白質の輸送順序の解析

Secretion order of the Class-II flagellar axial proteins analyzed by inverted membrane vesicles (IMV)

Yudai Matsumoto[1], Chinatsu Tatsumi[1], Hiroyuki Terashima[1], Tohru Minamino[2], Katsumi Imada[1] ([1]*Grad. Sch. of Sci., Osaka Univ.*, [2]*Grad. Sch. of Front. BioSci., Osaka Univ.*)

2Pos073 Alp7/TACC-Alp14/TOG protein complex promotes assembly of *S. pombe* microtubules

Douglas Drummond[1], Frauke Hussmann[2], Daniel Peet[2], Douglas Martin[3], Robert Cross[2] ([1]*Faculty of Agriculture, Kyushu Univ.*, [2]*Warwick Medical Sch., UK*, [3]*Lawrence Univ., USA*)

2Pos074 タウタンパク質に対する Pin1 由来のプロテアーゼの活性

Activity of a protease derived from Pin1 for tau protein

Teikichi Ikura, Nobutoshi Ito (*Med. Res. Inst., Tokyo Med. Dent. Univ.*)

2Pos075* アミロイド線維形成の圧力依存性を解明するための計算研究

Computational Research to Reveal the Pressure Dependency of the Formation of Amyloid Fibrils

Naohiro Nishikawa[1,2], Yoshiharu Mori[2], Yuko Okamoto[1,3,4,5], Hisashi Okumura[2,6] ([1]*Grad. Sch. of Sci., Nagoya Univ.*, [2]*IMS*, [3]*Str. Biol. Res. Cent., Grad. Sch. of Sci., Nagoya Univ.*, [4]*Cent. for Comp. Sci., Grad. Sch. of Eng., Nagoya Univ.*, [5]*Info. Tech. Cent., Nagoya Univ.*, [6]*SOKENDAI*)

蛋白質：計測·解析 / Protein: Measurement & Analysis

2Pos076* ESI-QTOF MS 法を用いたアミロイドベータペプチドのオリゴマー形成機構の解析

ESI-QTOF MS analyses of the oligomerization mechanism of amyloid β peptides

Shintaro Yoshida[1], Sosuke Yoshinaga[1], Mitsuhiro Takeda[1], Ayumi Tanaka[1], Takashi Hamaguchi[1], Hitomi Yamaguchi[1], Shigeto Iwamoto[1], Takashi Saito[2], Yoshihiko Takinami[3], Toshiyuki Kohno[4], Takaomi C. Saido[2], Hiroaki Terasawa[1] ([1]*Fac. Life Sci., Kumamoto Univ.*, [2]*RIKEN BSI*, [3]*Bruker Daltonics*, [4]*Kitasato Univ. Sch. Med.*)

2Pos077 Processing of XFEL still images with a reference oscillation data set for crystal structural analyses of Cytochrome c Oxidase

Luo Fangjia[1], Atsuhiro Shimada[1], Keitaro Yamashita[2], Kunio Hirata[2], Masaki Yamamoto[2], Kyoko Itoh-Shinzawa[1], Shinya Yoshikawa[1], Tomitake Tsukihara[1] ([1]*Picobiology INST, Grad. Sch. of Life Sci., Univ. of Hyogo*, [2]*RIKEN SPring-8 Center*)

2Pos078 中性子タンパク質結晶構造解析での水素高感度検出のための動的核偏極法の予備的結果

The preliminary result of Dynamic Nuclear Polarization method for more sensitive detection of hydrogen in Neutron Protein Crystallography

Naoya Komatsuzaki[1], Ichiro Tanaka[1,2], Takahiro Iwata[3], Daisuke Miura[3], Yoshiyuki Miyachi[3], Genki Nukazuka[3], Hiroki Matsuda[3], Toshiyuki Chatake[4], Katsuhiro Kusaka[2], Nobuo Niimura[2] ([1]*Coll. of Eng., Ibaraki Univ.*, [2]*Frontier Res. Ctr., Ibaraki Univ.*, [3]*Faculty of Sci., Yamagata Univ.*, [4]*RRI, Kyoto Univ.*)

2Pos079 Elucidating the mechanisms of proton pumping in cytochrome c oxidase by time resolved IR spectroscopy

Chen Li, Tatsuhito Nishiguchi, Shun Yamauchi, Kyoko Shinzawa-Itoh, Shinya Yoshikawa, Satoru Nakashima, Takashi Ogura (*Grad. Sch. Sci., Univ. Hyogo*)

2Pos080 ストップトフロー装置と過渡回折格子法を組み合わせたタンパク質反応検出手法の開発

Time-resolved detection of the transient grating signal using stopped flow system

Shunki Takaramoto, Yusuke Nakasone, Masahide Terazima (*Dept. of chem, Univ. Kyoto*)

2Pos081 顕微ラマン分光法による微小液滴内での酵素活性検出系の開発

Detection of enzymatic activity in femtliter droplets using micro-Raman spectroscopy

Hironobu Yamashita[1], Kazuhito Tabata V.[1,2], Hiroshi Ueno[1], Hiroyuki Noji[1,3] ([1]*Department of Applied Chemistry, the University of Tokyo*, [2]*PRESTO, JST*, [3]*ImPACT, CAO, Govt*)

2Pos082 蛋白質周囲の水分子運動の測定を目的とした蛍光アップコンバージョン測定装置の開発

Development of fluorescence up-conversion apparatus to investigate hydration dynamics around the protein surface

Asahi Fukuda, Tomotaka Oroguchi, Masayoshi Nakasako (*Grad. Sci. Tech., Keio Univ.*)

2Pos083 Grid inhomogeneous solvation theory を用いた血液凝固因子 Xa の水和解析

Hydration analysis of Coagulation Factor Xa using grid inhomogeneous solvation theory

Hiroyuki Sato, Azuma Matsuura (*Fujitsu Laboratories Ltd.*)

2Pos084 キナーゼと ATP 競争阻害剤との結合自由エネルギー計算における、薬剤結合サイトの立体構造柔軟性がもたらす影響

The effect of conformational flexibility on the binding free energy calculation between kinases and their ATP-competitive inhibitors

Mitsugu Araki[1], Narutoshi Kamiya[1,2], Miwa Sato[3], Masahiko Nakatsui[1,5], Takatsugu Hirokawa[4], Yasushi Okuno[1,5] ([1]*RIKEN, AICS*, [2]*Grad. Sch. Sim. St., Univ. Hyogo*, [3]*Mitsui Knowledge Industry Co., Ltd.*, [4]*AIST, Molecular Profiling Research Center for Drug Discovery*, [5]*Grad. Sch. Med., Kyoto Univ.*)

2Pos085* 溶質構造エントロピー計算法の理論的研究

Theoretical study for solute configurational entropy calculation methods

Simon Hikiri[1], Takashi Yoshidome[1,2], Mitsunori Ikeguchi[1] ([1]*Grad. Sch. of Med. Life Sci. Yokohama city Univ.*, [2]*Dept. of Appl. Phys., Tohoku Univ.*)

2Pos086 分子シミュレーションにおける静電相互作用計算法：零多重極子法の理論と実際

Computational method for electrostatic interactions in molecular simulation: theory and practice in the zero-multipole summation

Ikuo Fukuda[1], Han Wang[2], Narutoshi Kamiya[3], Kota Kasahara[4], Tohru Terada[5], Shun Sakuraba[6], Haruki Nakamura[1] ([1]*IPR, Osaka Univ.*, [2]*Freie Universitaet Berlin*, [3]*Univ. of Hyogo*, [4]*Col. Life Sci., Ritsumeikan Univ.*, [5]*Grad. Sch. Agr. Life Sci. Univ. Tokyo*, [6]*Grad. Sch. Frontier Sci., Univ. Tokyo*)

蛋白質工学 / Protein: Engineering

2Pos087 **天然状態と似たそして異なるアポミオグロビン折り畳み中間体の構造**
Native-like and non-native structures in the folding intermediate of apomyoglobin
Chiaki Nishimura (*Fac. Pharm. Sci., Teikyo Heisei Univ.*)

2Pos088 **理想タンパク質の安定性のオリジンを探る**
Stability for de novo designed ideal proteins revisited
Mami Yamamoto[1,2], Rie Koga[1], Takahiro Kosugi[1,2], Nobuyasu Koga[1,2,3] ([1]*CIMoS, IMS.*, [2]*SOKENDAI*, [3]*JST, PRESTO*)

2Pos089 **c-Myb- KIX 間相互作用を阻害するペプチドの合理的設計**
Rational design of a peptide inhibitor of the c-Myb-KIX interaction
Shunji Suetaka[1], Yoshiki Oka[2], Yuuki Hayashi[1,2], Munehito Arai[1,2] ([1]*Dept. Integrated Sci., Univ. Tokyo*, [2]*Dept. Life Sci., Univ. Tokyo*)

2Pos090 **β シートモデルタンパク質 OspA への αB クリスタリンのアミロイド形成配列移植と評価**
Grafting of a short amyloid forming sequence from αB crystalline into β-rich model protein, OspA
Yuki Hori[1], Kenta Hongo[2], Norio Yoshida[3], **Koki Makabe**[1] ([1]*Graduate School of Science and Engineering, Yamagata University*, [2]*School of Information Science, JAIST*, [3]*Department of Chemistry, Graduate School of Sciences, Kyushu University*)

2Pos091 **レアメタルに特異的に結合するペプチドの設計**
Design of peptides that specifically bind to the rare metal
Yoshihiro Iida, Atsuo Tamura (*Grad. Sci., Univ. Kobe*)

2Pos092* **進化分子工学によるフィチン酸塩加水分解酵素の活性向上**
Improving activity of a phytate-hydrolyzing enzyme by directed evolution
Manami Wada, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

2Pos093 **Toward Directed Evolution of Bacterial Biosensor for Arsenite Detection by Compartmentalized Partnered Replication**
Seaim Lwin Aye, Asuka Ueki, Kei Fujiwara, Nobuhide Doi (*Grad. Sch. Sci. Tech., Keio Univ.*)

2Pos094 **フェムトリットルチャンバーアレイを用いたスクリーニングシステムによるアルカリフォスファターゼの進化分子工学**
Directed evolution of alkaline phosphatase by femtoliter chamber array screening system
Makoto Kato, Yi Zhang, Kazuhito Tabata, Hiroyuki Noji (*Grad. Sch. Eng., Univ. Tokyo*)

2Pos095* **ファージ選別実験への応用を目指したスプリット GFP による蛍光ファージの作製**
Construction of fluorescent phages based on split GFP for the phage sorting technique
Naoki Mikoshiba[1,2], Yuki Shimizu[1,3], Rie Kiriguchi[1,2], Seiji Sakamoto[1,3], Hiroyuki Oikawa[1,2,3], Kiyoto Kamagata[1,2,3], Takehiko Wada[1,3], Satoshi Takahashi[1,2,3] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Life Sci., Tohoku Univ.*, [3]*Grad. Sch. Sci., Tohoku Univ.*)

2Pos096 **Toward design of diverse all-α protein structures**
Kouya Sakuma[1,2], Rie Koga[1], Takahiro Kosugi[1,2], Nobuyasu Koga[1,2,3] ([1]*CIMoS, IMS.*, [2]*SOKENDAI*, [3]*JST, PRESTO*)

2Pos097 **エクソンペプチドの構造予測と解析**
Prediction of Exon-Peptide Structures and Analysis
Michiko Nosaka (*National Institute of Technology, Sasebo College*)

核酸結合蛋白質 / Nucleic acid binding proteins

2Pos098* **粗視化分子動力学計算によるサブヌクレオソームの構造解析**
Structural modeling of the subnucleosome using coarse-grained molecular dynamics simulations
Masahiro Shimizu, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

2Pos099 **Holliday junction DNA facilitates RuvA-RuvB complex formation**
Yong-Woon Han[1], Reiko Yamamoto[1], Kimiko Nakao[1], Hisashi Tadakuma[1], Yoshie Harada[1,2,3] ([1]*Kyoto Univ., iCeMS*, [2]*Grad. Sch. Biostudies, Kyoto Univ.*, [3]*Inst. for Protein Res., Osaka Univ.*)

2Pos100 **定量的イメージング法を用いた単一細胞由来のグルココルチコイド受容体のホモ二量体と転写活性の関連解析**
Quantification of homodimeric glucocorticoid receptor and transcriptional activity from single cell using quantitative imaging techniques
Sho Oasa[1], Akira Sasaki[2], Shintaro Mikuni[1], Johtaro Yamamoto[1], Masataka Kinjo[1] ([1]*Fac. Adv. Life Sci., Hokkaido Univ.*, [2]*AIST*)

2Pos101 **(6-4)光回復酵素の T(6-4)T と T(6-4)C の光活性及び光修復における 赤外分光解析の比較**
Comparative FTIR study of photoactivation and photorepair of T(6-4)T and T(6-4)C photoproducts by *Xenopus* (6-4) photolyase
Mai Kumagai[1], Daichi Yamada[1,2], Tatsuya Iwata[1], Elizabeth D Getzoff[4], Junpei Yamamoto[3], Shigenori Iwai[3], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*Ochanomizu University*, [3]*Osaka University*, [4]*The Scripps Research Institute*)

2Pos102 **3 本鎖 DNA 結合蛋白質が 3 本鎖 DNA を認識する分子機構**
Molecular mechanism of the triplex DNA-binding protein to recognize triplex DNA
Kazuki Kiuchi[1], Kohta Sugiyama[1], Ryotaro Kishi[1], Satoru Unzai[2,3], **Hidetaka Torigoe**[1] ([1]*Dept. Applied Chem., Fac. Sci., Tokyo Univ. Sci.*, [2]*Grad. Sch. Medical Life Sci., Yokohama City Univ.*, [3]*Fac. Biosci. Applied Chem., Hosei Univ.*)

2Pos103* **Characterization of the deamination activity of APOBEC3B by real-time NMR, which is distinct from that of APOBEC3G**
Li Wan[1], Takashi Nagata[1,2], Ryo Morishita[3], Akifumi Takaori[4], Masato Katahira[1,2] ([1]*Grad. Sch. of Energy Sci., Kyoto Univ.*, [2]*Inst. of Adv. Energy, Kyoto Univ.*, [3]*CellFree Sciences Co., Ltd.*, [4]*Dept. of Hematology and Oncology, Grad. Sch. of Med., Kyoto Uni.*)

2Pos104 **Sequence-Specific Protein-DNA Interactions for Molecular Simulations Modeled by Position Weight Matrix**
Cheng Tan, Shoji Takada (*Dept. Biophysics, Graduate School of Science, Kyoto University*)

2Pos105 **Mechanistic studies of transcriptional regulation by non-CpG methylated DNA**
Jianshi Jin[1,2,4], Tengfei Lian[1,2], Chan Gu[1,3], Kai Yu[1,2], Yi Qin Gao[1,3], Xiao-Dong Su[1,2] ([1]*Biodynamic Optical Imaging Center(BIOPIC), Peking University*, [2]*School of Life Sciences, Peking University*, [3]*College of Chemistry and Molecular Engineering, Peking University*, [4]*RIKEN Quantitative Biology Center (QBiC), Laboratory for Integrative Omics*)

2Pos106 **転写調節機構の統計的解釈〜コンセンサス配列でわかること、わからないこと〜**
Global Statistical Control of Transcriptional Pausing by Repetitive Genomic Sequences
Masahiko Imashimizu[1,2,3], Ariel Afek[4], Hiroki Takahashi[2], David Lukatsky[4] ([1]*IMSUT*, [2]*MMRC, Chiba Univ.*, [3]*NIH/NCI*, [4]*Ben-Gurion Univ. of the Negev*)

2Pos107 **Molecular dynamics of transcription factor-nucleosome interactions**
Giovanni Brandani, Shoji Takada (*Kyoto University*)

核酸：構造・物性 / Nucleic acid: Structure & Property

2Pos108 **Structured RNAs Induce Intersubunit Rolling and Codon-Anticodon Weakening During Ribosomal Frameshifting**
Kaichun Chang[1], Emmanuel Salawu[2,3], Jin-Der Wen[1], **Lee-Wei Yang**[2,3,4] ([1]*National Taiwan University*, [2]*National Tsing Hua University*, [3]*Bioinformatics Program, Academia Sinica*, [4]*National Center of Theoretical Science*)

2Pos109 **リンカー DNA により繋がっているダイヌクレオソーム構造のサンプリング**
Sampling di-nucleosome structures connected by linker DNA of various lengths
Hiroo Kenzaki[1], Shoji Takada[2] ([1]*ACCC, RIKEN*, [2]*Grad. Sch. Sci., Univ. Kyoto*)

2Pos110 **酵母間期染色体の力学モデルと核内構造・動態の解析**
Analysis of interface chromosome dynamics of yeast by course-grained models
Takamasa Yamamoto[1], Hiraku Nishimori[1,2], Akinori Awazu[1,2] ([1]*Dept. Math and Life Sci. Hiroshima Univ.*, [2]*RcMcD*)

2Pos111 **細胞分裂およびアポトーシス過程におけるクロマチンダイナミクスの 1 分子解析**
Chromatin dynamics in mitosis and apoptosis
Kayo Hibino[1,2], Kazuhiro Maeshima[1,2] ([1]*NIG*, [2]*SOKENDAI*)

2Pos112 **Grab & Watch: Correlative optical Tweezers-Fluorescence Microscopy (CTFM) as a versatile tool for chromatin studies**
Andrea Candelli[1,2], Gerrit Sitters[1,2], **Rosalie Driessen**[1,2], Willem Peutz[1,2], Olivier Heyning[1,2], Gijs Wuite[1,2], Erwin Peterman[1,2] ([1]*LUMICKS*, [2]*VU University, Amsterdam*)

2Pos113 **クロマチンのエピジェネティック状態を用いたヒト間期核内における染色体三次元構造のシミュレーション**
Simulating three-dimensional organization of chromosomes in human interphase nucleus using epigenetic state of chromatin
Shin Fujishiro, Masaki Sasai (*Nagoya Univ.*)

核酸：相互作用·複合体形成 / Nucleic acid: Interaction & Complex formation

2Pos114* **蛍光相互相関分光法を用いたグルココルチコイド受容体-DNA 間相互作用の定量化**
The different interaction affinity of monomeric or dimeric GR on DNA determined by fluorescence cross-correlation spectroscopy
Daisuke Yamashita, Mari Saito, Sho Oasa, Shintaro Mikuni, Masataka Kinjo (*Grad. Sch. Life Sci., Univ. Hokkaido*)

2Pos115 **Effects of cumulative acetylation in histone H3 tail studied by an enhanced conformational sampling MD simulation**
Jinzen Ikebe[1], Shun Sakuraba[2], Hidetoshi Kono[1] ([1]*QST, MMS*, [2]*Grad. School of Frontier Sci., Univ. Tokyo*)

2Pos116 **Photo-control of the ribosome movement along mRNA using a reversible photo-cross-linking probe**
Shunsuke Yamashiro[1], Ryo Iizuka[2], Takashi Funatsu[2] ([1]*Fac. of Pharm. Sci., The Univ. of Tokyo*, [2]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

2Pos117 **転写によって誘起されるクロマチンブラシの相分離**
Transcription driven phase separation in chromatin brush
Tetsuya Yamamoto[1], Helmut Schiessel[2] ([1]*National Composite Center, Nagoya University*, [2]*Instituut-Lorentz for Theoretical Physics, Leiden University*)

2Pos118 **蛍光消光現象を利用した単層カーボンナノチューブ表面上での一本鎖 DNA 吸着過程の観察**
Monitoring adsorption process of single stranded DNA to single-walled carbon nanotubes surfaces by fluorescence quenching
Shizuma Sato[1], Gilbert Bustamante[2], Jing Yong Ye[2], Kazuo Umemura[1] ([1]*Tokyo Univ. of Sci.*, [2]*UTSA*)

2Pos119 **DNA の網目構造が微粒子の拡散に及ぼす影響**
Effect of a mesh structure of DNA on diffusion of a small particle
Masaya Tanoguchi, Yoshihiro Murayama (*Tokyo Univ. of Agri. and Tech.*)

水·水和·電解質 / Water & Hydration & Electrolyte

2Pos120 **イオン液体ー水混合溶液中の β-ラクトグロブリンの会合構造**
Aggregation of β-lactoglobulin in alkylammonium-based nitrate ionic liquid-water mixture
Koji Yoshida, Ayako Fujiyoshi, Toshio Yamaguchi (*Fukuoka Univ.*)

2Pos121 **OH 伸縮振動のラマン分光によるアミノ酸水和層の解析**
Hydration analysis of amino acids by Raman OH-stretching spectroscopy
Yasutaka Naito[1], Yuki Ochiai[2], George Mogami[2], Makoto Suzuki[2] ([1]*Sch. Eng., Univ. Tohoku*, [2]*Grad. Sch. Eng., Univ. Tohoku*)

2Pos122 **広帯域誘電緩和分光を用いた球状および膜タンパク質における水和と熱活性効果の研究**

Effects of hydration and thermal excitation of globular and membrane proteins studied by broadband dielectric spectroscopy

Naoki Yamamoto[1], Shota Ito[2], Eri Chatani[1], Hideki Kandori[2], Keisuke Tominaga[1,3] ([1]*Graduate School of Science, Kobe University*, [2]*Graduate School of Engineering, Nagoya Institute of Technology*, [3]*Molecular Photoscience Research Center, Kobe University*)

2Pos123* **分子動力学法を用いた蛋白質周囲の水和ダイナミクスの検討：溶媒条件と蛋白質構造の影響について**

Effects of solvent pH and protein conformations on water dynamics around a denatured protein with molecular dynamics simulation

Takafumi Fujiyoshi[1], Naoki Ogasawara[1], Yuji Ezaki[2], Yuta Nonaka[2], Kota Kasahara[2], Takuya Takahashi[2] ([1]*Graduate School of Life Sciences, Ritsumeikan University*, [2]*College of Life Sciences, Ritsumeikan University*)

2Pos124 **ミオシン周囲の局所誘電環境**

Local dielectric environment around myosin

Takato Sato, Tohru Sasaki, Jun Ohnuki, Mitsunori Takano (*Dept. of Pure. & Appl. Phys., Waseda Univ.*)

2Pos125 **水溶液中における芳香環間相互作用の統計熱力学**

Statistical thermodynamics of aromatic-aromatic interactions in aqueous solution

Tomohiko Hayashi, Masahiro Kinoshita (*Inst. Adv. Energy, Kyoto Univ.*)

2Pos126* **Lennard-Jones ポテンシャルのパラメータの変更による溶質周囲の水分子のダイナミクスの探求**

Effects of Lennard-Jones potentials on the dynamics of water molecules around a solute

Yuki Takimoto[1], Kou Sakuma[2], Nana Okita[2], Kota Kasahara[2], Takuya Takahashi[2] ([1]*Grad. Sch. Life. Sci., Ritsumeikan Univ.*, [2]*Col. Life. Sci., Ritsumeikan Univ.*)

2Pos127 **親水性アミノ酸残基周辺の水和構造における酸素原子ローンペアの影響**

Effects of lone pairs of oxygen atoms on hydration structures around polar amino-acid residues: MD simulation study

Tomotaka Oroguchi, Masayoshi Nakasako (*Sci. Tech., Keio Univ.*)

2Pos128 **機械学習によるタンパク質親水／疎水表面における水分子の動的振る舞いの解析**

Analysis of water behavior near the protein hydrophobic/hydrophilic surface by machine learning techniques

Taku Mizukami[1], Nguyen Viet Cuong[3], Ho Tu Bao[2], Dam Hieu Chi[2] ([1]*JAIST, Materials Science*, [2]*JAIST, Knowledge Science*, [3]*HPC SYSTEMS Inc.*)

2Pos129* **MM/3D-RISM 法を用いた Pim1-リガンド系における結合自由エネルギーの予測**

Estimation of binding free energy based on the MM/3D-RISM method for the Pim1-ligand system

Takeshi Hasegawa[1], Masatake Sugita[1], Takeshi Kikuchi[1], Fumio Hiata[2] ([1]*Dept. of Bioinfo., Col. Life Sci., Ritsumeikan Univ.*, [2]*Toyota Physical and Chemical Research Institute*)

発生･分化 / Development & Differentiation

2Pos130 **ROXS によって mCherry の安定性と明るさが向上する**

in-vivo ROXS improve the brightness and photostability of a red fluorescent protein, mCherry in *C. elegans* embryos

Yukinobu Arata, Yasushi Sako (*Cell. Info. Lab. RIKEN*)

2Pos131 **Axon bundle regulates cortical tissue stiffness in the developing brain**

Misato Iwashita, Yoichi Kosodo (*Korea Brain Research Institute*)

2Pos132* **Mouse-ferret differences in the mechanical property of the developing cerebral cortex: tissue-level and single cell-level assessments**

Arata Nagasaka, Tomoyasu Shinoda, Takaki Miyata (*Grad. Sch. Med., Univ. Nagoya*)

2Pos133 **Macroscopic dynamics of vascular endothelial cells in angiogenesis**

Naoko Takubo[1], Kazuaki Naekura[1], Ryo Yoshida[2], Terumasa Tokunaga[3], Osamu Hirose[4], Yasunobu Uchijima[1], Yukiko Kurihara[1], Hiroki Kurihara[1] ([1]*Graduate School of Medicine, The University of Tokyo*, [2]*The Institute of Statistical Mathematics, Research Organization of Information and Systems*, [3]*Faculty of Computer Science and Systems Engineering, Kyushu Institute of Technology*, [4]*College of Science and Engineering, Kanazawa University*)

2Pos134 **ヒトの原腸形成時の細胞運動を in vitro で一細胞解析**

Single cell tracking of migration during human gastrulation in vitro

Shota Miyazaki[1], Yuta Yamamoto[1], Kohei Nakazono[1], Minh N. T. Le[1], Shuji Fuji[1,2], Kiyoshi Ohnuma[1,3] ([1]*Department of Bioengineering, Nagaoka University of Technology*, [2]*Department of Materials Science and Technology, Nagaoka University of Technology*, [3]*Department of Science of Technology Innovation, Nagaoka University of Technology*)

2Pos135 **多細胞運動におけるソリトン様運動関連遺伝子の探索**

Searching the responsible genes for biological soliton in multicellular movement

Kentaro Yoshida[1], Hiroki Takahashi[2], Yoshitoshi Ogura[3], Yutaka Suzuki[4], Tetsuya Hayashi[3], Hidekazu Kuwayama[1] ([1]*Fac. Life and Env. Sci., Univ. Tsukuba*, [2]*Medi. Myco. Res. Center, Chiba Univ.*, [3]*Dep. Bact., Fac. Med. Sci., Kyushu Univ.*, [4]*Dep. Med. Genome Sci., Grad. School Front. Sci., Univ. Tokyo*)

2Pos136 **A model for analyzing phenomena in multicellular organisms with multivariable polynomials**

Hiroshi Yoshida (*Dep. Math., Kyushu Univ.*)

2Pos137 ***C. elegans* の受精における二相性カルシウム波の分子基盤**

The molecular underpinnings of the biphasic calcium wave during fertilization in *C. elegans*

Jun Takayama, Shuichi Onami (*RIKEN QBiC*)

筋肉 / Muscle

2Pos138 **機能する昆虫飛翔筋-ウサギ骨格筋ハイブリッド筋線維の再構成**
Reconstitution of functional insect flight- and rabbit skeletal hybrid muscle fibers as monitored by X-ray diffraction
Hiroyuki Iwamoto (*SPring-8, JASRI*)

2Pos139 **アクチン骨格阻害剤 Latrunculin A はアクチンを脱重合させ重合を阻害する**
Latrunculin-A; a drug to inhibit actin cytoskeleton, depolymerizes and inhibits actin polymerization under TIRF microscopy
Ikuko Fujiwara[1], Thomas Pollard[2] ([1]*URA, NITech.*, [2]*MCDB, Yale Univ.*)

2Pos140 **バキュロウイルス-昆虫細胞を用いた、組換え β-アクチンの発現精製系の構築**
Expression and Purification of Recombinant Human β-Actin in Insect Cells
Mizuki Matsuzaki[1], Sae Kashima[1], Kayo Maeda[1], Mahito Kikumoto[1], Tomoharu Matsumoto[1], Kotaro Tanaka[1], Motonori Ota[2], Akihiro Narita[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Grad. Sch. Info. Sci., Nagoya Univ.*)

2Pos141 **結合した HMM 濃度の違いによる F-アクチンの状態変化**
HMM induced structural changes of actin monitored by in vitro fluorescence along single filaments
Marie Kobayashi, Satoshi Nakamura, Takahiro Hayashizaki, Hajime Honda (*Dept. Bioeng., Nagaoka Univ. Tech.*)

2Pos142* **超解像イメージング法を用いた骨格筋ミオシン分子動態の直接計測**
Direct measurement of dynamics of individual skeletal myosin by using a super-resolution localization method
ZhiYuan Zhang, Yuto Ashida, Masahito Ueda, Hideo Higuchi, Motoshi Kaya (*Department of Physics, Graduate School of Science, The University of Tokyo*)

分子モーター / Molecular motor

2Pos143* **HS-AFM で一方向的な運動が観測されたローターレス Enterococus hirae V_1-ATPase の結晶構造解析**
Crystal structures of rotorless Enterococus hirae V_1-ATPase which shows unidirectional dynamics by HS-AFM
Shintaro Maruyama[1], Kazuya Nakamoto[1], Kano Suzuki[1], Fabiana. Lica Yakushiji[1], Kenji Mizutani[1], Motonori Imamura[2], Takayuki Uchihashi[2,3,4], Takeshi Murata[1,5] ([1]*Grad. Sch. Sci., Univ. Chiba*, [2]*Bio-AFM FRC, Kanazawa Univ.*, [3]*Dept. Phys., Kanazawa Univ.*, [4]*CREST, JST*, [5]*PRESTO, JST*)

2Pos144 **F_1-ATPase の中心軸回転を駆動する触媒サブユニットの１分子構造変化観察**
Single-molecule detection of conformational changes in the catalytic subunit of F_1-ATPase that correlate with rotation of the shaft
Ryuichi Yokota[1], Ryota Yanagida[1], Yuta Nomura[1], Nagisa Mikami[2], Rinako Nakayama[2,3], Takayuki Nishizaka[2], Tomoko Masaike[1,3,4] ([1]*Dept. Appl. Biol. Sci., Tokyo Univ. of Sci.*, [2]*Dept. Phys., Gakushuin Univ.*, [3]*Res. Inst. for Sci. and Tech., Tokyo Univ. of Sci.*, [4]*PRESTO, JST*)

2Pos145* **V_1 回転分子モーターでの外来タンパク質の回転**
Rotation of endogenous proteins in V_1 rotary motor
Mihori Baba, Atsuko Nakanishi, Jun-ichi Kishikawa, Ken Yokoyama (*Kyoto Sangyo Univ. LifeSci.*)

2Pos146* **１分子観察とドッキングシミュレーションで蛍光基質を通じて明らかになった化学反応に伴う酵素の構造変化**
Conformational change of the rotary motor F_1-ATPase revealed by single-molecule imaging and docking simulation
Nagisa Mikami[1], Yuko Ito[2], Mitsuhiro Sugawa[1], Mitsunori Ikeguchi[2], Takayuki Nishizaka[1] ([1]*Dept. phys., Gakushuin Univ.*, [2]*Medical Life Sci., Yokohama City Univ.*)

2Pos147 **光渦トラップを使った力学測定システムの開発：DNA オリガミを使った F_1-ATPase の回転可視化**
Development of optical vortex trapping system with DNA origami for the precise measurements of torque generated by F_1-ATPase
Yu Hashimoto[1], Sayaka Kazami[1], Yuji Kimura[1], Tomoko Hyodo-Otsu[2], Taro Ando[2], Hiroyasu Itoh[1,2] ([1]*Tsukuba Research Lab., Hamamatsu Photonics K.K.*, [2]*Central Research Lab., Hamamatsu Photonics K.K.*)

2Pos148 ***Enterococcus hirae* 由来 V_1-ATPase の回転とヌクレオチド結合解離の１分子同時観察**
Single-molecule simultaneous observation of rotation and nucleotide binding/release of *Enterococcus hirae* V_1-ATPase
Yoshihiro Minagawa[1], Hiroshi Ueno[1], Hiroyuki Noji[1], Takeshi Murata[2], **Ryota Iino**[3,4] ([1]*Univ. Tokyo*, [2]*Chiba Univ.*, [3]*OIIB and IMS, NINS*, [4]*SOKENDAI*)

2Pos149 **高熱菌 *Bacillus* PS3 由来 F_oF_1-ATP 合成酵素のプロトン輸送活性および H^+/ATP の決定**
Determination of the proton pump activity and the H^+/ATP ratio of thermophilic *Bacillus* PS3 F_oF_1-ATP synthase
Naoya Iida[1], Yuzo Kasuya[1], Naoki Soga[2], Toshiharu Suzuki[2], Taro Uyeda[1], Masasuke Yoshida[3], Kazuhiko Kinosita[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*Dept. Eng, Univ. of Tokyo*, [3]*Dept. Mol Biochem, Kyoto Sangyo Univ.*)

2Pos150 **細菌べん毛モーターの回転方向変換制御機構の解明**
Elucidation of the directional switching mechanism of the bacterial flagellar motor by electron cryomicroscopy
Tomoko Miyata[1], Takayuki Kato[1], Akihiro Kawamoto[1], Keiichi Namba[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*QBiC, RIKEN*)

2Pos151 **Molecular dynamics study of pressure effects on unbinding of the CheY-FliM complex**
Hiroaki Hata[1], Yasutaka Nishihara[1], Masayoshi Nishiyama[2], Ikuro Kawagishi[3], Akio Kitao[1] ([1]*IMCB, UTokyo*, [2]*The HAKUBI Center, Kyoto Univ.*, [3]*Dept. of Frontier Biosci., Hosei Univ.*)

2Pos152 **海洋性ビブリオ菌 FliG の EHPQR-motif 周辺構造によるべん毛の回転方向決定**
Direction of flagellar motor rotation determined by the structure around EHPQR-motif of FliG in marine *Vibrio*
Tatsuro Nishikino[1], Atsushi Hijikata[2], Yasuhiro Onoue[1], Tsuyoshi Shirai[2], Michio Homma[1] ([1]*Div. Biol. Sci. Grad. Sch. Sci., Nagoya Univ.*, [2]*Dep. Biosci., Nagahama Inst. of Bio-Sci. Tec.*)

2Pos153* 2種イオン駆動型べん毛モーターの入力と出力の関係
Input-output relationship of dual ion driven flagellar motor
Kenta Arai[1], Taishi Kasai[2], Yuka Takahashi[3], Masahiro Ito[3], Yoshiyuki Sowa[1,2] ([1]*Hosei Univ.*, [2]*Research Center for Micro-Nano Tech. Hosei Univ.*, [3]*Toyo Univ.*)

2Pos154 高度高塩菌ハロバクテリウムサリナラムのべん毛の回転とステップ運動の直接観察
Direct observation of rotation and steps of the archaellum in the swimming halophilic archaeon Halobacterium salinarum
Yoshiaki Kinosita[1], Nariya Uchida[2], Daisuke Nakane[1], Takayuki Nishizaka[1] ([1]*Department of Physics, Gakushuin University*, [2]*Department of Physics, Tohoku University*)

2Pos155 キネシンネックリンカーの構造変化に伴う自由エネルギー変化
Measuring of energy at neck linker docking of single kinesin molecule
Yuichi Kondo, Hideo Higuchi (*Grad. Sch. Sci., Univ. of Tokyo*)

2Pos156 LZMWを利用した高濃度蛍光ATP存在下でのキネシン運動とATP結合の同時蛍光1分子計測
Simultaneous fluorescent observation of kinesin motility and ATP occupancy with high concentration of fluorescent labeled ATP using LZMW
Kazuya Fujimoto[1], Yuki Morita[1], Hirofumi Shintaku[1], Michio Tomishige[2], Ryota Iino[3], Hidetoshi Kotera[1], Ryuji Yokokawa[1] ([1]*Kyoto University Department of Micro Engineering*, [2]*The University of Tokyo Department of Applied Physics*, [3]*National Institute of Natural Science Institute for Molecular Science*)

2Pos157 CYK-4による kinesin-6の回転運動揺らぎ
CYK-4 induces the large fluctuations of the left-handed rotational movement of dimeric kinesin-6
Yohei Maruyama[1], Akihiko Sato[1], Tim Davis[2], Toshihisa Osaki[3], Shin Yamaguchi[1], Shoji Takeuchi[3], Masanori Mishima[2], Junichiro Yajima[1] ([1]*Dept. Life Sci., Grad. Sch. of Arts and Sci., Univ. of Tokyo*, [2]*CMCB at Warwick Med. Sch., Univ. of Warwick*, [3]*Inst. of Ind. Sci., Univ. of Tokyo*)

2Pos158 微小管と光応答性DNAによる物質輸送システムの構築
Construction of a nano-transportation system by using microtubules and photoresponsive DNA
Kentaro Kayano[1], Ryuuhei Suzuki[2], A.M.R. Kabir[2], Kazuki Sada[2,3], Akinori Kuzuya[4], Hiroyuki Asanuma[5], Akira Kakugo[2,3] ([1]*Dept. Chem. Fac. Sci., Hokkaido Univ.*, [2]*Grad. Sch. Chem. Sci. Eng., Hokkaido Univ.*, [3]*Fac. Sci., Hokkaido Univ.*, [4]*Fac. Chem. Mater. Bioeng., Kansai Univ.*, [5]*Grad. Sch. Eng., Nagoya Univ.*)

2Pos159 キネシンの非平衡熱散逸
Non-equilibrium dissipation of kinesin
Takayuki Ariga[1], Michio Tomishige[2], Daisuke Mizuno[1] ([1]*Dept. of Phys., Kyushu Univ.*, [2]*Dept. of Appl. phys., Univ. Tokyo*)

2Pos160 生細胞内における微小管へのキネシン結合速度定数の直接計測
Direct measurement of the binding rate constant of kinesin to microtubules in living cells
Taketoshi Kambara, Yasushi Okada (*RIKEN, QBiC*)

2Pos161 *Flavobacterium johnsoniae* の滑走に関与するマルチレール構造
The multi-rail structure contributes to gliding motility of *Flavobacterium johnsoniae*
Satoshi Shibata, Koji Nakayama (*Graduate Sch. of Biomedical Sciencces, Nagasaki Univ.*)

2Pos162* マイコプラズマ・モービレ滑走運動におけるシングルユニットが発生する力
Force generated by single unit in *Mycoplasma mobile* gliding
Masaki Mizutani[1], Isil Tulum[1], Yoshiaki Kinosita[2], Takayuki Nishizaka[2], Makoto Miyata[1] ([1]*Osaka City Univ., Grad. sch. Sci.*, [2]*Gakushuin Univ., Fac. Sci.*)

2Pos163 **Dynamics of Type IV pili controlled by light direction in unicellular cyanobacteria**
Daisuke Nakane, Takayuki Nishizaka (*Dept. of Phys., Gakushuin Univ.*)

2Pos164 マイコプラズマモービレ由来滑走タンパク質Gli349の構造ドメインの探索とその構造解析
Determination of domain boundaries and analysis of domain structures of the gliding protein Gli349 from Mycoplasma mobile
Yuuki Hayashi[1,2], Yoshihiro Nomura[2], Manami Wada[1], Tasuku Hamaguchi[3], Aya Takamori[3], Masato Miyata[3], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Integrated Sci., Univ. Tokyo*, [3]*Dept. Biol., Osaka City Univ.*)

2Pos165 高圧力顕微鏡法による深海微生物の遊泳運動観察
Swimming motility of deep-sea bacteria measured by high-pressure microscopy
Masayoshi Nishiyama[1], Chiaki Kato[2], Hiroshi Imai[3], Shinji Kamimura[3], Yoshie Harada[4] ([1]*The HAKUBI Center, Kyoto Univ.*, [2]*JAMSTEC*, [3]*Chuo Univ.*, [4]*Osaka Univ.*)

2Pos166* *Mycoplasma mobile* の滑走装置に局在するペアになったF型ATPアーゼのパラログ
Paired F-type ATPase paralog in gliding machinery of *Mycoplasma mobile*
Takuma Toyonaga[1], Yuhei Tahara[1], Noriyuki Kodera[2], Tasuku Hamaguchi[1], Toshio Ando[2], Makoto Miyata[1] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*Bio AFM-FRC., Kanazawa Univ.*)

2Pos167* **Dynamics and heterogeneity of ATP production and consumption in single C2C12 myotubes**
Naoki Matsuda[1], Katsuyuki Kunida[1], Takumi Wada[1], Haruki Inoue[2], Daisuke Hoshino[1], Shinya Kuroda[1,2] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Grad. Sch. Frontier Sci., Univ. Tokyo*)

2Pos168 **C 型インフルエンザウイルスの運動機構**
Motile mechanism of influenza C virus
Tatsuya Sakai[1], Yasushi Muraki[2], Mineki Saito[1] ([1]*Department of Microbiology, Kawasaki Medical School*, [2]*Division of Infectious Diseases and Immunology, Department of Microbiology, School of Medicine, Iwate Medical University*)

2Pos169 **鞭毛・繊毛の表面運動：現象の普遍性と膜タンパク質のダイナミクス**
Surface motility in eukaryote cilia/flagella: Generality and membrane protein dynamics
Ritsu Kamiya[1,3], Kogiku Shiba[2], Kazuo Inaba[2], Takako Kato-Minoura[3] ([1]*Gakushuin Univ., Fac. Sci.*, [2]*Tsukuba Univ., Shimoda Marine Res. Ctr.*, [3]*Chuo Univ., Fac. Sci. Eng.*)

2Pos170 **微小管ネットワークによって引き起こされる細胞質回転流動**
Spatial confinement of active microtubule networks induces large-scale rotational cytoplasmic flow
Kazuya Suzuki[1,2], Makito Miyazaki[1,2], Jun Takagi[3], Takeshi Itabashi[1,2], Shin'ichi Ishiwata[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*Waseda Bioscience Research Institute in Singapor, Waseda Univ.*, [3]*Quantitative Mechanobiology Lab., NIG*)

2Pos171 **軸糸直径サイズ変化による鞭毛繊毛の屈曲運動の制御**
Regulation of cilia and flagella bending movements through the change of axoneme diameter
Toshiki Yagi[1], Shinji Kamimura[2], Hiroyuki Iwamoto[3] ([1]*Dept. Life Sci., Prefectural Univ. of Hiroshima*, [2]*Dept. Biol. Sci., Chuo Univ.*, [3]*SPring-8 JASRI*)

2Pos172 **中心子の普遍的 9 回対称性構造の構築機構**
Assembly mechanisms of the nine-fold symmetry of the centriole structure
Masafumi Hirono (*Frontier Biosci. Hosei Univ.*)

2Pos173 **X 線繊維回折を用いた真核生物鞭毛軸糸の構造ダイナミクス解析**
X-ray fiber diffraction study on structural dynamics of flagellar axonemes of Chlamydomonas
Kazuhiro Oiwa[1,2], Hiroyuki Iwamoto[3], Junya Kirima[2], Yu Yamano[2] ([1]*Adv. ICT Res. Inst. NICT*, [2]*Univ. Hyogo*, [3]*SPring8, JASRI*)

2Pos174* **3-D measurement of bending dependency of the maximum force of the single tracheal cilium**
Takanobu A Katoh[1], Koji Ikegami[2], Toshihito Iwase[3], Tomoko Masaike[3,4], Mitsutoshi Setou[2], Takayuki Nishizaka[1] ([1]*Dept. Phys., Gakushuin Univ.*, [2]*Dept. Cell Biol. and Anat., Hamamatsu Univ. Sch. Med.*, [3]*Dept. Appl. Biol. Sci., Tokyo Univ. of Sci.*, [4]*PRESTO, JST*)

2Pos175 **アルファシヌクレインタンパク質による輸送性微小管の制御機構**
Alpha-synuclein binds unconventional microtubules that have a unique function
Shiori Toba[1], Mingyue Jin[1], Masami Yamada[1], Takuo Yasunaga[2], Yuko Fukunaga[3,4], Atsuo Miyazawa[3,4], Kyoko Itoh[5], Shinji Fushiki[5], Hiroaki Kojima[6], Hideki Wanibuchi[7], Yoshiyuki Arai[8], Takeharu Nagai[8], Shinji Hirotsune[1] ([1]*Dept. of Genetic Disease Research, Osaka City Univ. Graduate School of Medicine*, [2]*Faculty of Computer Science and Systems Engineering, Kyushu Institute of Technology*, [3]*Graduate School of Life Science, Univ. of Hyogo*, [4]*RSC-University of Hyogo Leading Program Center, RIKEN SPring-8 Center*, [5]*Kyoto Prefectural Univ. of Medicine Graduate School of Medical Sciences*, [6]*Advanced ICT Research Institute, National Institute of Information and Communications Technology*, [7]*Dept. of Pathology, Osaka City Univ. Graduate School of Medicine*, [8]*Institute of Scientific and Industrial Research, Osaka Univ.*)

2Pos176 **マウス Tppp (Tubulin Polymerization Promoting Protein)の機能解析**
Analysis of Tppp (Tubulin Polymerization Promoting Protein)
Masahiro Kawakita[1], Arashi Seki[1], Katsuyoshi Takaoka[2], Hiroshi Hamada[3], Kyosuke Shinohara[1] ([1]*Tokyo University of Agriculture & Technology*, [2]*EMBL Heidelberg*, [3]*RIKEN Center for Developmental Biology*)

2Pos177 **マウス織毛細胞における Dpcd の機能解析**
Role of Dpcd in motile cilia of mice
Misato Tamegai[1], Mahito Kikumoto[2], Miki Kinoshita[3], Akihiro Kawamoto[3], Keiichi Namba[3], Hiroshi Hamada[4], Katsumi Imada[3], Akihiro Narita[2], Kyosuke Shinohara[1] ([1]*Tokyo University of Agriculture & Technology*, [2]*Nagoya University*, [3]*Osaka University*, [4]*RIKEN Center for Developmental Biology*)

2Pos178 **IFT（繊毛内輸送）に関与する基底小体微小管の機能に関する研究**
The function of the basal body microtubules associated with intraflagellar transport (IFT)
Yurika Koiso, Shin Yamaguchi, Mitsuhiro Sugawa, Takuya Kobayashi, Yoko Y. Toyoshima, Junichiro Yajima (*Department of Life Sciences Graduate School of Art & Sciences, The University of Tokyo*)

2Pos179 **Distinctive structured radial spoke of mouse sperm underlies wave propagation of flagella**
Kaoru Horiuchi[1], Hironori Ueno[2], Akihiro Narita[3], Hiroshi Hamada[4], Kyosuke Shinohara[1] ([1]*Tokyo University of Agriculture and Technology*, [2]*Aichi University of Education*, [3]*Nagoya University*, [4]*RIKEN Center for Developmental Biology*)

2Pos180 **クラミドモナス鞭毛から精製したラジアルスポークの特性**
Properties of the Purified Radial Spoke of Chlamydomonas Flagella
Hitoshi Sakakibara[1], Yosuke Shimizu[1], Pinfen Yang[2], Hiroaki Kojima[1] ([1]*Protein Biophys. Gr., NICT*, [2]*Dept. Biol. Sci., Marquette Univ.*)

2Pos181 **ハプトネマの微小管系急速コイリング運動メカニズムを探る**
Unveiling a mechanism for rapid microtubule coiling movement of haptonema
Mami Nomura[1], Keiko Hirose[2], Kogiku Shiba[1], Kazuo Inaba[1] ([1]*Shimoda Marine Research Center, University of Tsukuba.*, [2]*Biomedical Research Institute, National Institute of Advanced Industrial Science and Technology*)

2Pos182 **cAMP シグナルリレーにおける膜電位変化の計測と制御**
Measurement and control of membrane potential changes in cAMP signal relay
Yusuke V. Morimoto[1], Masahiro Ueda[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*)

2Pos183 **動的な場における時間空間知覚メカニズムの解析：走化性パラドクスの克服と細胞の整流作用**
Delineating temporal and spatial sensing in migrating cells: chemotactic wave paradox and rectification of the leading edge response
Akihiko Nakajima[1], Shuji Ishihara[2], Motohiko Ishida[3], Daisuke Imoto[3], Satoshi Sawai[1,3] ([1]*Res. Cent. Comp. Sys. Biol., Grad. Sch. Arts Sci., Univ. Tokyo*, [2]*Sch. Sci. Tech., Meiji Univ.*, [3]*Dept. Basic Sci., Grad. Sch. Arts Sci., Univ. Tokyo*)

2Pos184 **細胞か接着により誘導される細胞の集団運動**
Cell-Cell Adhesion guiding Collective Cell Migration
Katsuyoshi Matsushita (*Department of Biological Sciences, Graduate School of Science, Osaka University*)

2Pos185 **力測定で明かす神経幹細胞の集団遊走**
Measuring the Forces in Neural Stem Cell Monolayer
Masahito Uwamichi, Masaki Sano (*Dept. of Phys., The Univ. of Tokyo*)

2Pos186 **細胞性粘菌における集団的回転運動の定量解析**
Quantitative analysis of collective rotational motion of Dictyostelium cells
Taihei Fujimori[1], Akihiko Nakajima[1,2], Ryo Yokota[3], Ryo Nakabayashi[4], Gen Honda[1], Tetsuya Kobayashi[3], Satoshi Sawai[1,2] ([1]*Grad. Sch. of Arts & Sci., Univ. Tokyo*, [2]*Res. Ctr. Complex Syst. Biol., Univ. Tokyo*, [3]*Inst. of Ind. Sci., Univ. Tokyo*, [4]*Univ. Tokyo*)

2Pos187 **Spatial heterogeneous and transient dynamics during collective cell migration in a monolayer of MDCK epithelial cells**
Preetom Nag[1], Helal Khalifa[1,2], Hiroshi Teramoto[3], Naoya Yamaguchi[4], Chun-Biu Li[1], Hisashi Haga[2], Tamiki Komatsuzaki[1,2] ([1]*Research Institute for Electronic Science, Hokkaido University*, [2]*Graduate School of Life Science, Hokkaido University*, [3]*Hitachi, Ltd. Research & Development Group*, [4]*Skirball Institute of Biomolecular Medicine, New York University Langone Medical Center, USA*)

2Pos188 **細胞の協調運動における接着結合タンパク質の役割**
Roles of adherence junction proteins in the collective cell movement in vitro and vivo
Takeomi Mizutani, Kazushige Kawabata (*Department of Advanced Transdisciplinary Sciences, Faculty of Advanced Life Science, Hokkaido University*)

2Pos189* **筋分化 C2C12 におけるインスリン刺激時 S6K 活性のダイナミクスと不均一性**
Dynamics and Heterogeneity of S6K activity in insulin stimulated-C2C12 myotubes
Haruki Inoue[1], Katsuyuki Kunida[2], Daisuke Hoshino[2], Takumi Wada[2], Shinya Kuroda[1,2] ([1]*Grad. Frontier Sci., Univ. Tokyo*, [2]*Grad. Sch. Sci., Univ. Tokyo*)

2Pos190 **Beating rate changes of isolated cardiomyocyte clusters in different thermal environments**
Wei Wang, Tomoyuki Kaneko (*LaRC, Dept. Frontier Biosci., Hosei Univ.*)

2Pos191 **改良型蛍光 ATP センサーを用いた一細胞及び細胞内局所 ATP 濃度の測定**
Quantification of single-cell and subcellular ATP concentrations using an improved fluorescent ATP indicator in mammalian cells
Hideyuki Yaginuma, Yasushi Okada (*QBiC, RIKEN*)

2Pos192* **がん細胞の損傷回復過程の定量評価**
Quantitative evaluation of recovery from damage in cancer cells
Morito Sakuma[1,2], Kazuhito Tabata[1,2], Hiroyuki Noji[2], Hideo Higuchi[1] ([1]*Department of Physics, Graduate School of Science, The University of Tokyo*, [2]*Department of Applied Chemistry, Graduate School of Engineering, The University of Tokyo*, [3]*JSPS Research Fellow*)

2Pos193* **ナノ秒パルス電場による細胞内応答の顕微ラマン・蛍光分光法を用いたその場観測**
In situ observation of the intracellular responses to nanosecond pulsed electric fields by Raman and fluorescence spectroscopy
Yusuke Horii, Hirotsugu Hiramatsu, Takakazu Nakabayashi (*Graduate School of Pharmaceutical Sciences, Tohoku University*)

2Pos194* **ミトコンドリア輸送・膜電位・ATP と神経伸展の相関解析**
Correlation analysis of transport, membrane potential, and ATP levels of mitochondria and neurite extension
Rika Suzuki, Kohji Hotta, Kotaro Oka (*Keio Univ.*)

2Pos195* **細胞内局所発熱がストレス顆粒形成を開始する**
Intracellular local thermogenesis initiates stress granule formation
Beini Shi[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*PRESTO, JST*)

2Pos196 **Investigating the contribution of cytoskeletons on intracellular temperature variation**
Takashi Yanagi[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharmac. Sci., Univ. Tokyo*, [2]*JST, PRESTO*)

2Pos197 **Attempt to Detect Functional Interaction between FoF1-ATPase and Adenine Nucleotide Translocator**
Saki Yamashita, Takahiro Shibata, Yoshihiro Ohta (*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*)

2Pos198 **Effects of mitochondrial volume on the generation of reactive oxygen species**
Sawako Kimura[1], Satoshi Honda[2], Norihiro Umeda[2], Yoshihiro Ohta[1] ([1]*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*, [2]*Dept. of Mech. Sys. Eng., Tokyo Univ. of Agr. and Tech.*)

2Pos199 **Partial contribution of mitochondrial permeability transition to t-butyl hydroperoxide-induced cell death**
Naoko Ashida, Xiaolei Shi, Hyonjin Choi, Yoshihiro Ohta (*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*)

2Pos200 **Monitoring of mitochondrial activity during cell division**
Kyunghak Cho, Kotoe Hirusaki, Yoshihiro Ohta (*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*)

生体膜・人工膜：情報伝達 / Biological & Artificial membrane: Signal transduction

2Pos201* **パターン化人工膜を用いて脂質ラフトによる光シグナル伝達の制御機構を解明する**
Regulation of phototransduction by lipid rafts studied with a micropatterned model membrane
Yasushi Tanimoto[1], Sakiko Kojima[1], Fumio Hayashi[2], Kenichi Morigaki[1,3] ([1]*Grad. Sch. Agri, Univ. Kobe*, [2]*Grad. Sch. Scie, Univ. Kobe*, [3]*Biosignal Research Center, Univ. Kobe*)

2Pos202 **全反射照明蛍光顕微鏡を用いた RalGDS 分子の EGF 依存的な膜局在化メカニズムの解明**
Elucidation of the EGF dependent localization mechanism of RalGDS molecule to plasma membrane using TIRF microscopy
Ryo Yoshizawa[1,2], Nobuhisa Umeki[2], Masataka Yanagawa[2], Masayuki Murata[1], Yasushi Sako[2] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Wako Inst., Riken*)

生体膜・人工膜：ダイナミクス / Biological & Artificial membrane: Dynamics

2Pos203 **分子動力学シミュレーションによる膜の細孔形成自由エネルギー解析**
Free energy analysis of membrane pore formation by molecular dynamics simulations
Yusuke Miyazaki, Wataru Shinoda, Susumu Okazaki (*Grad. Eng., Univ. Nagoya*)

2Pos204* **Min 反応拡散波の油中水滴内再構成**
Reconstitution of Min Reaction-Diffusion Waves in Water-in-Oil Microdroplets
Shunshi Kohyama, Nobuhide Doi, Kei Fujiwara (*Grad. Sch. Sci. Tech., Keio Univ.*)

2Pos205 **ラクトフェリシン B フラグメントと大腸菌および GUV との相互作用**
Interactions of a fragment of lactoferricin B with *E.coli* and single GUVs
Md. Moniruzzaman[1], Hideo Dohra[2], Masahito Yamazaki[1,3,4] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Green Sci. Tech., Shizuoka University*, [3]*Res. Inst. Ele., Shizuoka Univ.*, [4]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos206 **フォトクロミック脂質を用いた蛍光脂質のダイナミクス**
Intermembrane transfer of fluorescent lipid analogs using photochromic lipid analogs as FRET acceptors
Mariko Sumi[1,2], Asami Makino[1], Takehiko Inaba[1], Fumihiro Fujimori[2], Peter Greimel[1], Toshihide Kobayashi[1,3] ([1]*Lipid Bio. Lab., RIKEN*, [2]*Grad. Sch. of Humanities and Life Sci., Tokyo Kasei Univ.*, [3]*CNRS, France*)

2Pos207 **脂質膜の力学的特性は細胞透過ペプチド・トランスポータン 10(TP10)の単一ベシクルへの侵入に影響を与える**
Mechanical Properties of Lipid Bilayers Affect the Entry of Cell-Penetrating Peptide Transportan 10 (TP10) into Single Vesicles
Md. Zahidul Islam[1], Sabrina Sharmin[1], Masahito Yamazaki[1,2,3] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Ele., Shizuoka Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos208 **一定張力が誘起する膜破壊の活性化エネルギーを用いた解析**
Analysis of Constant Tension-Induced Rupture of Lipid Membranes Using Activation Energy
Md. Abu Sayem Karal[1], Victor Levadnyy[1,3], **Masahito Yamazaki**[1,2] ([1]*Shizuoka Univ. Grad. Sch. Sci. Tech.*, [2]*Shizuoka Univ. Res. Inst. Ele.*, [3]*Rus. Acad. Sci.*)

2Pos209 **抗菌ペプチド・マガイニン２が脂質膜中に誘起するポア形成のメカニズム**
A Mechanism of Antimicrobial Peptide, Magainin 2-Induced Pore Formation in Lipid Membranes
Moynul Hasan[1], Mohammad Abu Sayem Karal[1], Victor Levadnyy[1,2], Md. Zahidul Islam[1], Masahito Yamazaki[1,3,4] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Rus. Acad. Sci.*, [3]*Res. Inst. Ele., Shizuoka Univ.*, [4]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos210 **引張下コレステロール含有リン脂質二重膜における指組み構造の形成：分子動力学シミュレーション**
Stretch-Induced Interdigitated Phase Formation in Phospholipid/Cholesterol Bilayer: Molecular Dynamics Simulation
Taiki Shigematsu[1], Kenichiro Koshiyama[2], Shigeo Wada[2] ([1]*JAMSTEC*, [2]*Grad. Sch. Eng. Sci., Osaka Univ.*)

2Pos211 **細胞サイズ液滴内における高分子溶液の拡散とその空間閉じ込めの影響**
Diffusion in polymer solutions confined in cell-sized droplets: effect of confinement size
Chiho Watanabe[1], Miho Yanagisawa[2] ([1]*Inst. Glob. Innov., Tokyo Univ. Agri. & Tech.*, [2]*Grad. Sch. Tech., Tokyo Univ. Agri. & Tech.*)

2Pos212 **Investigating interactions and dynamics of pleckstrin homology domains on a lipid membrane surface**
Eiji Yamamoto[1], Antreas C. Kalli[2], Takuma Akimoto[1], Mark S.P. Sansom[2], Kenji Yasuoka[3] ([1]*Grad. Sch. Sci. Technol., Keio Univ.*, [2]*Dept. Biochem., Univ. Oxford*, [3]*Dept. Mech. Eng., Keio Univ.*)

生体膜・人工膜：構造・物性 / Biological & Artificial membrane: Structure & Property

2Pos213* **D 体フェニルアラニンを含む抗菌ペプチド Phenylseptin の脂質膜との相互作用解析**
Analysis of interaction between antimicrobial peptide phenylseptin containing a D-phenylalanine and membrane
Yuta Matsuo, Izuru Kawamura (*Grad. Sch. Eng., Yokohama Natl. Univ.*)

2Pos214* **人工生体膜とナノ空間を利用した 1 分子計測技術の開発**
Single-molecule observation technique based on a model membrane and a nanometric gap structure
Koji Ando[1], Humio Hayashi[2], Kenichi Morigaki[1,3] ([1]*Grad. Agri., Univ. Kobe*, [2]*Grad. Sci., Univ. Kobe*, [3]*Biosignal. Univ. Kobe*)

2Pos215* **カチオン性抗菌ペプチド ボンビニン H2 および H4 のリーシュマニア原虫模倣膜との特異的な相互作用**
Specific interaction of cationic antimicrobial peptides bombinin H2 and H4 with *Leishmania* protozoa mimetic membrane
Shiho Kaneda, Akira Naito, Izuru Kawamura (*Grad. Sch. Eng., Yokohama Natl. Univ.*)

2Pos216 **水の脂質膜透過に対する膜の張力の効果**
Effect of Lateral Tension on Membrane Permeability of Water in Lipid Membranes
Sayed Ul Alam Shibly[1], Masahito Yamazaki[1,2,3] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Ele., Shizuoka Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos217 **部分フッ素化リン脂質からなる人工膜内色素分子の発光挙動**
Investigation of fluorescence emission from dye-lipid in partially fluorinated lipid bilayer
Toshinori Motegi[1], Ryugo Tero[2], Toshiyuki Takagi[3], Hiroshi Takahashi[1], Hideki Amii[1], Toshiyuki Kanamori[3], Masashi Sonoyama[1] ([1]*Gunma Univ.*, [2]*Toyohashi Univ. Tech.*, [3]*AIST*)

2Pos218 **パターン化人工膜を利用した NAP-22 の膜結合と凝集挙動解析**
Membrane binding and aggregation of neuronal acidic protein of 22kDa (NAP-22) studied with a patterned model membrane
Sakiko Kojima[1], Yasushi Tanimoto[1], Fumio Hayashi[3], Shohei Maekawa[3], Kenichi Morigaki[1,2] ([1]*Grad. Sch. Agri., Univ. Kobe*, [2]*Biosignal Research Center, Univ. Kobe*, [3]*Grad. Sch. Sci., Univ. Kobe*)

2Pos219 **Phospholipase C and D induced defects in POPC and POPC: POPG lipid bilayers: A simulation study**
M Harunur Rashid[1], M N Holme[2], M M Stevens[2], I Yarovsky[1] ([1]*School of Engineering, RMIT university, Melbourne, Australia*, [2]*Department of Materials, Department of Bioengineering and Institute of Biomedical Engineering, Imperial College, London, UK.*)

2Pos220 **固体 NMR と MD シミュレーションによる抗菌ペプチドアラメチシンとメリチンの膜結合構造と配向の解明**
Structure and orientation of antimicrobial peptides alamethicin and melittin in membrane revealed by solid-state NMR and MD simulation
Akira Naito[1], Takashi Nagao[1], Daisuke Mishima[1], Namsrai Javkhalantugs[2], Jun Wang[1], Daisuke Ishioka[1], Kiyonori Yokota[1], Kazushi Norisada[1], Izuru Kawamura[1], Kazuyoshi Ueda[1] ([1]*Grad. Schl. Eng. Yokohama Natl. Univ.*, [2]*Schl. Eng. and Appl. Sci., Natl. Univ. Mongolia*)

2Pos222 **巨大一枚膜ベシクルに内包された DNA コンピュータ基盤遺伝子発現制御システム**
Development of a DNA computer-based gene-regulatory system encapsulated in a giant unilamellar vesicle
Koh-ichiroh Shohda[1], Toru Nishikata[1], Yutetsu Kuruma[2], Akira Suyama[1] ([1]*Grad. Sch. Arts and Sciences, The University of Tokyo*, [2]*Earth-Life Science Institute, Tokyo Institute of Technology*)

2Pos223 **Local pressure tensor calculation for molecular simulations and its application to lipid membranes**
Koh Nakagawa, Hiroshi Noguchi (*ISSP, Univ. of Tokyo*)

2Pos224 **Formation of vesicles using self-reproducing oil droplet system**
Kensuke Kurihara[1,2,3] ([1]*Okazaki Institute for Integrative Bioscience*, [2]*Institute for Molecular Science*, [3]*Research Center for Complex Systems Biology, The Univ. of Tokyo*)

2Pos225* **アクチンフィラメントの細胞膜シート上への結合**
The binding of actin filament on the cell membrane flat sheet
Shun Wakamatsu[1], Kuniyuki Hatori[1], Takashi Okuno[2] ([1]*Grad. Sch. Sci. & Eng., Yamagata Univ.*, [2]*Fac. Sci., Yamagata Univ.*)

2Pos226* **アクチン線維を封入した巨大リポソームの形態変化**
Shape change of giant liposomes encapsulating actin filaments
Shunsuke Tanaka, Masahito Hayashi, Kingo Takiguchi (*Grad. Sch. Sci., Nagoya Univ.*)

光生物：視覚･光受容 / Photobiology: Vision & Photoreception

2Pos227 **全反射赤外分光法を用いたウシオプシンと匂い分子の相互作用研究**
ATR-FTIR study on the interactions between bovine opsin and odorants
Kunisato Kuroi[1], Takefumi Morizumi[2], Hisao Tsukamoto[1], Oliver P Ernst[2], Yuji Furutani[1] ([1]*Inst. for Mol. Sci.*, [2]*Univ. Toronto*)

2Pos228 **Glu381Lys 点変異体を用いたニワトリクリプトクロム 4 の光反応メカニズムの解析**
Photoreaction mechanism of chicken cryptochrome4 studied by using a Glu381Lys mutant
Hiromasa Mitsui, Kota Miura, Keiko Okano, Toshiyuki Okano (*Dept. Eng. and Biosci., Grad. Sch. Adv. Sci. and Eng., Waseda Univ.*)

2Pos229* **ニワトリクリプトクロム４の光依存的な相互作用分子とその分子メカニズム**
Identification of chicken CRY4-interacting molecules and the interaction mechanism
Ayano Orii, Shingo Kondo, Keiko Okano, Toshiyuki Okano (*Dept. Eng. and Biosci., Grad. Sch. Adv. Sci. and Eng., Waseda Univ.*)

2Pos230 **桿体視細胞に発現する視物質の熱活性化頻度**
Thermal activation rates of visual pigments expressed in rods
Keiichi Kojima[1], Yuki Matsutani[1], Masataka Yanagawa[2], Takahiro Yamashita[1], Yasushi Imamoto[1], Osamu Hisatomi[3], Yumiko Yamano[4], Akimori Wada[4], Yoshinori Shichida[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Cell. Info. Lab., Riken*, [3]*Grad. Sch. Sci., Osaka Univ.*, [4]*Kobe Pharm. Univ.*)

2Pos231 **トランスデューシンは PDE を“間接的に”活性化する**
Transducin activates cGMP phosphodiesterase indirectly
Teizo Asano, **Shuji Tachibanaki**, Satoru Kawamura (*Grad. Sch. Frontier Biosci., Osaka Univ.*)

2Pos232* **低温赤外分光法によるサル緑感受性視物質がもつ塩化物イオン結合部位の構造解析**
Structural analysis of chloride binding site of monkey green studied by light-induced difference FTIR spectroscopy
Shunta Nakamura[1], Kota Katayama[2], Hiroo Imai[3], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*Dept. Pharm CWRU, USA*, [3]*Primate Res Inst., Kyoto Univ.*)

2Pos233 新口動物の光受容タンパク質 Opn5 の多様性

Diversity of the photoreceptor protein Opn5 found in deuterostomes

Takahiro Yamashita[1], Ikutaro Sawada[1], Keita Sato[2], Naoaki Sakamoto[3], Keisuke Takahashi[1], Naoyuki Iwabe[1], Hideyo Ohuchi[2], Takashi Yamamoto[3], Yoshinori Shichida[1] ([1]*Grad. Scl. of Sci., Kyoto Univ.*, [2]*Okayama Univ. Grad. Sch. of Med.*, [3]*Grad. Scl. of Sci., Hiroshima Univ.*)

2Pos234 Light-dependent association and dissociation of arrestin with bistable opsins

Takashi Nagata[1], Mitsumasa Koyanagi[1, 2], Emi Yamashita-Kawano[1], Robert Lucas[3], Akihisa Terakita[1] ([1]*Graduate School of Science, Osaka City University*, [2]*JST PRESTO*, [3]*Faculty of Life Sciences, The Unversity of Manchester*)

2Pos235 桿体アレスチンのスプライスバリアント・p44 の自己会合の解析

Self-association of p44, a splice variant of visual rod arrestin

Yasushi Imamoto[1], Keiichi Kojima[1], Toshihiko Oka[2], Takahiro Yamashita[1], Yoshinori Shichida[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos236 疾患に関わるロドプシン変異体の FTIR 研究

FTIR study of disease-causing mutations of rhodopsin

Akiko Enomoto[1], Shunta Nakamura[1], Kota Katayama[2], Hiroo Imai[3], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*Dept. Pharm., CWRU, USA*, [3]*Primate Res. Inst., Kyoto Univ.*)

2Pos237 N-terminal region of modified Volvox channel rhodopsin-1(mVChR1) enchances Na+ Influx by drowing hydrogen ion

Yuko Sakajiri[1], Kanako Hara[2], Yoshito Watanabe[2], Tetsuya Sakajiri[3], Eriko Sugano[2], Hiroshi Tomita[1,2] ([1]*Ugas. Agr. Iwate Univ.*, [2]*Se. Iwate Univ.*, [3]*Fac. of Nutr. Sci., Morioka Univ.*)

2Pos238 フグ眼球由来の細胞株における広範囲な光波長応答性

A wide-range spectral photosensitivity in the puffer fish ocular cells

Keiko Okano[1], Shoichi Ozawa[1], Hayao Sato[1], Sawa Kodachi[1], Masaharu Ito[1], Toshiaki Miyadai[2], Akihiro Takemura[3], Toshiyuki Okano[1] ([1]*Dept. Eng Biosci, Grad. Sch. Adv Sci. and Eng, Waseda Univ.*, [2]*Fac Marine Biosci, Fukui Pref. Univ.*, [3]*Dept. Chem Biol. & Marine Sci. Fac Sci., Univ. Ryukyus*)

2Pos239 *Guillardia theta* 由来ロドプシン様タンパク質の分子機能解明

Molecular functions of rhodopsin-like proteins from *Guillardia theta*

Yumeka Yamauchi[1], Masae Konno[1], Keiichi Inoue[1,2], Satoshi Tsunoda[1], Hideki Kandori[1] ([1]*Nagoya Inst. tech.*, [2]*JST. PRESTO*)

2Pos240* 海洋性細菌のもつ光駆動イオンポンプ

Light-driven ion-pump activity of native marine bacteria

Yuichi Hashimoto[1], Rei Abe-Yoshizumi[1], Yoshitaka Kato[1], Keiichi Inoue[1,2], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*JST PRESTO*)

光生物：光合成 / Photobiology: Photosynthesis

2Pos241 好熱性紅色光合成細菌 *Alc. tepidum* 由来の光捕集複合体の単離精製と分光学的特性評価

Purification and spectroscopic study of the light-harvesting complexes from thermophilic purple bacterium *Allochromatium tepidum*

N. Nakamura[1], S.-W. Lu[2], A. Ohkoshi[1], K. Okazaki[1], T. Kawakami[1], M. T. Madigan[3], Y. Kimura[2], **S. Otomo**[1] ([1]*Ibaraki Univ.*, [2]*Grad. Sch. Agri. Sci., Kobe Univ.*, [3]*Southern Illinois Univ.*)

2Pos242 Thermochromatium tepidum 由来光捕集１複合体における部位特異的変異体の分光学的解析

Spectroscopic characterization of site-directed mutants in Light-Harvesting 1 complex from Thermochromatium tepidum

Kanako Hashimoto[1], Seiji Akimoto[2], Kenji Nagashima[3], Takashi Ohno[1], Sheiu Otomo[4], Yukihiro Kimura[1] ([1]*Graduate school of Agriculture, Kobe University*, [2]*Graduate school of Science, Kobe University*, [3]*Research Institute for Photobiological Hydrogen Production, Kanagawa University*, [4]*Faculity of Science, Ibaraki University*)

2Pos243 光合成光捕集複合体における金属イオン認識の構造基盤

Structural basis for the metal-ion recognition of the bacterial core light-harvesting complex

T. Kawakami[1], L.-J. Yu[2,3], Y. Kimura[4], S. Otomo[2] ([1]*Grad. Sch. Sci. Eng., Ibaraki Univ.*, [2]*Ibaraki Univ.*, [3]*Present address: Grad. sch. Boi., Okayama Univ.*, [4]*Grad. Sch. Agri. Sci., Kobe Univ.*)

2Pos244 NMR study of the interaction on the two ferredoxin isoforms with ferredoxin-NADP$^+$ reductase

Risa Mutoh[1,2], Akane Furuya[2,3], Takahisa Ikegami[4], Michael Hippler[5] ([1]*Faculty of Sci., Fukuoka Univ.*, [2]*Inst. for Protein Research, Osaka Univ.*, [3]*Grad. Sch. of Sci., Osaka Univ.*, [4]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*, [5]*Inst. for Biology and Biotechnology, Univ. of Munster*)

2Pos245* 同一メナキノン分子の酸化還元電位が２種の光合成反応中心蛋白質で 500mV も異なる理由

Redox potential difference of 500 mV for menaquinones in two types of photosynthetic reaction centers

Keisuke Kawashima[1], Hiroshi Ishikita[1,2] ([1]*Grad. Sch. Eng., Univ. of Tokyo*, [2]*RCAST, Univ. of Tokyo*)

2Pos246 ヘリオバクテリア光合成反応中心の過渡吸収変化と低温蛍光解析

Analyses of transient absorption changes and low-temperature fluorescence in the photosynthetic reaction center of heliobacteria

Hirozo Oh-oka[1], Risa Kojima[1], Chihiro Azai[2], Risa Mutoh[3], Genji Kurisu[3], Shigeru Itoh[4] ([1]*Graduate School of Science, Osaka University*, [2]*College of Life Science, Ritsumeikan University*, [3]*Institute for Protein Research, Osaka University*, [4]*Center for Gene Research, Nagoya University*)

2Pos247 Magnetic structure of reduced [2Fe-2S] Rieske cluster from green sulfur bacteria Chlorobaculum tepidum studied by ESEEM

Naotaka Terashima (*photobioenergetics lab, graduate school of science, Nagoya university*)

2Pos248 **好熱性紅色光合成細菌 Thermochromatium tepidum 由来反応中心複合体におけるカルシウムイオンの機能的、構造的役割**
Functional and structural roles of calcium ion in the reaction center from thermophilic purple bacterium, Thermochromatium tepidum
Michie Imanishi[1], Masayuki Kobayashi[2], Manami Kobayashi[1], Mari Matsuzaki[3], Yuki Yura[3], Takashi Ohno[3], Seiu Otomo[4], Yukihiro Kimura[3] ([1]*Faculty of Agriculture, Kobe University*, [2]*Ariake National College of Technology*, [3]*Guraduate school of Agriculture, Kobe university*, [4]*Faculty of Science, Ibaraki University*)

2Pos249 **Initial formation of the radical pair in reaction center complex of *Heliobacterium modesticaldum* detected by transient ESR**
Hiroyuki Tsukuno[1], Risa Mutoh[3], Genji Kurisu[2,4], Hirozo Oh-oka[2], Hiroyuki Mino[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Dept. Applied Phys. Fac. Sci., Fukuoka Univ.*, [4]*Ins. Pro. Res., Osaka Univ.*)

2Pos250* **フラビンタンパク質で目指す人工光合成**
Artificial photosynthesis based on the engineered flavoprotein LOV
Nozomi Ueda, Yukiko Ono, Tatsuya Iwata, Masayo Iwaki, Hideki Kandori (*Nagoya Institute of Technology*)

2Pos251* **分子動力学シミュレーションによる光捕集複合体の自己組織化過程に関する理論的研究**
Theoretical study on the self-organization process of the light-harvesting complexes with molecular dynamics simulation
Marie Yamauchi[1], Shinji Saito[2,3], Masahiro Higashi[4] ([1]*Graduate School of Engineering and Science, University of the Ryukyus*, [2]*Department of Theoretical and Computational Molecular Science, Institute for Molecular Science*, [3]*The Graduate University for Advanced Studies (SOKENDAI)*, [4]*Faculty of Science, University of the Ryukyus*)

光生物：光遺伝学･光制御 / Photobiology: Optogenetics & Optical Control

2Pos252 **クリプト藻由来のカチオンチャネルロドプシンのイオン透過メカニズムの電気生理学による研究**
Electrophysiological study of cation channelrhodopsins from cryptophyte algae
Satoshi Tsunoda[1], Yumeka Yamauchi[1], Masae Konno[1], Keiichi Inoue[1,2], Hideki Kandori[1] ([1]*Grad. Sch. Sci., Nagoya Inst. of Tech.*, [2]*JST, PREST*)

2Pos253 **アニオンチャネルロドプシン 2 の光開閉型 Cl^-/H^+ 対向輸送活性**
A light-dependent Cl^-/H^+ antiport activity in anion channelrhodopsin-2
Satoko Doi[1], Takashi Tsukamoto[1], Srikanta Chowdhury[2], Susumu Yoshizawa[3], Akihiro Yamanaka[2], Yuki Sudo[1] ([1]*Grad. Sch. of Med. Dent. & Pharm. Sci., Okayama Univ.*, [2]*RIEM, Nagoya Univ.*, [3]*AORI, Univ. of Tokyo*)

2Pos254 **サーモフィリックロドプシンの耐熱性・高光遺伝学活性の構造基盤**
Structural basis for high thermal stability and efficient optogenetic function of thermophilic rhodopsin
Takashi Tsukamoto[1], Kenji Mizutani[2], Taisuke Hasegawa[3], Megumi Takahashi[4], Naoya Honda[1], Naoki Hashimoto[2], Kazumi Shimono[5], Seiji Miyauchi[5], Shin Takagi[4], Shigehiko Hayashi[3], Takeshi Murata[2], **Yuki Sudo**[1] ([1]*Okayama Univ.*, [2]*Chiba Univ.*, [3]*Kyoto Univ.*, [4]*Nagoya Univ.*, [5]*Toho Univ.*)

2Pos255 **新規酵素ロドプシンの機能解析**
Characterization of a novel enzyme rhodopsin
Kazuho Yoshida[1], Satoshi Tsunoda[1], Leonid S. Brown[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*Univ. Guelph*)

2Pos256 **ユニークな光反応を示す微生物型ロドプシンの研究**
Microbial rhodopsins with unique photoreaction
Yoshitaka Kato, Keiichi Inoue, Shota Ito, Satoshi Tsunoda, Yurika Nomura, Hideki Kandori (*Grad. Sch. Eng., Nagoya Inst. Tech.*)

生命の起源･進化 / Origin of life & Evolution

2Pos257* **マイクロデバイスと大腸菌の融合を基とした人工細胞の開発**
A step towards creating life: Development of the hybrid cell based on the fusion of micron-scaled device and *E. coli*
Yoshiki Moriizumi[1,2], Kazuhito V. Tabata[1,2,3], Rikiya Watanabe[1,3], Tomohiro Doura[4], Mako Kamiya[3,4], Yasuteru Urano[4,5,6], Hiroyuki Noji[1,2] ([1]*Dept. Appl. Chem., Grad. Sch. Eng, Univ. Tokyo*, [2]*ImPACT, Cab. Office, Gov. Japan*, [3]*PRESTO, JST*, [4]*Grad. Sch. Med., Univ. Tokyo*, [5]*Grad. Sch. Pharm, Univ. Tokyo*, [6]*CREST, AMED*)

2Pos258* **翻訳と共役した再帰的 DNA 複製システムの確立**
The establishment of translation-coupled recursive DNA replication system
Yoshihiro Sakatani[1], Norikazu Ichihashi[1,2] ([1]*Grad. Sch. Info., Osaka Univ.*, [2]*Grad. Sch. Bio., Osaka Univ.*)

2Pos259 **Characterization on *Escherichia coli* L-Form**
Shino Toe[1], Kazuhito V. Tabata[2,3,4], Yoshiki Moriizumi[2,3], Hiroyuki Noji[2,3] ([1]*Dept. Appl. Chem., UG. Sch. Eng, Univ. Tokyo*, [2]*Dept. Appl. Chem., Grad. Sch. Eng, Univ. Tokyo*, [3]*ImPACT, Cab. Office, Gov. Japan*, [4]*PRESTO, JST*)

2Pos260 **対称性の自発的破れによる遺伝子の起源**
The origin of genes through spontaneous symmetry breaking
Nobuto Takeuchi (*Univ. Tokyo, Grad. Sch. of Arts and Sciences*)

2Pos261* **円偏光による L 型アミノ酸過剰生成機構の理論的探求**
Theoretical investigation of the generation of L-form amino acid excess by the CPL irradiation
Akimasa Sato[1], Mitsuo Shoji[2], Katsumasa Kamiya[3], Kenji Shiraishi[4], Kazuhiro Yabana[2], Yasuteru Shigeta[2], Masayuki Umemura[2] ([1]*Grad. Sch. Pure. App. Sci., Univ. Tsukuba*, [2]*Center Comp. Sci., Univ. Tsukuba*, [3]*Center Basic Edu. Integ. Learn., Kanagawa Inst. Tech.*, [4]*Inst. Mat. Sys. Sust., Nagoya Univ.*)

ゲノム生物学：ゲノム構造 / Genome biology: Genome structure

2Pos262 **3D ゲノム構造の集団ベースモデリング**
Population-based framework of 3D genome modeling
Takeshi Sugawara (*RcMcD, Hiroshima University*)

2Pos263 **Dynamic chromatin domains revealed by super-resolution live-cell imaging**
Tadasu Nozaki[1], Sachiko Tamura[1], Ryosuke Imai[1], Tomomi Tani[2], Masaru Tomita[3], Takeharu Nagai[4], Yasushi Okada[5], Kazuhiro Maeshima[1] ([1]*Natl. Inst. Genet.*, [2]*MBL*, [3]*Inst. Adv. Biosci., Keio Univ.*, [4]*ISIR, Osaka Univ.*, [5]*QBiC, RIKEN*)

2Pos264 **分裂酵母クロマチン動態の網羅的解析**
Comprehensive Analysis of Chromatin Dynamics in Fission Yeast
Toshinori Namba[1], Sayaka Suzuki[2], Takeshi Sugawara[1], Da-Qiao Ding[3], Yasushi Hiraoka[4], Yuichi Togashi[1], Masaru Ueno[5], Shin-ichi Tate[1] ([1]*RcMcD, Hiroshima Univ.*, [2]*Dept. of Math. and Life Sci., Hiroshima Univ.*, [3]*Adv. ICT Res. Inst., NICT*, [4]*Dept. of Biol. Sci., Osaka Univ.*, [5]*Grad. Sch. of Adv. Sci. of Matter, Hiroshima Univ.*)

バイオインフォマティクス：構造ゲノミクス / Bioinformatics: Structural genomics

2Pos265 **長さの異なる塩基配列組み合わせの頻度・分布を用いたヌクレオソーム配置推定**
Predict nucleosome positioning by incorporating the frequencies and distributions of three length-different nucleotide segments
Akinori Awazu[1,2] ([1]*Dept. of Math. and Life Sciences, Hiroshima Univ.*, [2]*RcMcD , Hiroshima Univ.*)

2Pos266 **New rules of protein structures**
Shunsuke Nishiyama[1], Shintaro Minami[2], **George Chikenji**[1] ([1]*Dept. of Comp. Sci. & Eng., Nagoya Univ.*, [2]*Grad. Sch. of Inf. Sci., Nagoya Univ.*)

2Pos267 **全原子 Motion Tree による構造変化の解析**
Description of protein structural changes by full-atom Motion Tree
Ryotaro Koike (*Grad. Sch. of Info. Sci., Nagoya Univ.*)

2Pos268 **分子動力学法を用いた、ポリグルタミン酸のアンフォールドダイナミクス**
Unfolding dynamics of poly-glutamic acid in using molecular dynamics method
Naoki Ogasawara[1], Ryosuke Iwai[1], Kota Kasahara[2], Tetsuro Nagai[3], Takuya Takahashi[2] ([1]*Grad. Sch. Life. Sci., Ritsumei. Univ.*, [2]*Col. Life. Sci., Ritsumei. Univ.*, [3]*Col. Sci., Univ. Nagoya*)

2Pos269 **MEGADOCK-Azure: Microsoft Azure クラウド環境での並列タンパク質間相互作用予測計算**
MEGADOCK-Azure: High-performance protein-protein interaction predictions on Microsoft Azure HPC
Masahito Ohue[1], Yuki Yamamoto[1,2], Hiroyuki Sato[3], Takashi Matsushita[3], Yutaka Akiyama[1,2] ([1]*Sch. of Computing, Tokyo Tech.*, [2]*ACLS, Tokyo Tech.*, [3]*IMSBIO Co., Ltd.*)

2Pos270 **非エバルト静電ポテンシャル計算法“零多重極子和法”の開発と検証**
Development and Evaluations of a Fast and Accurate Non-Ewald Electrostatic Potential Scheme, the Zero-Multipole Summation Method
Kota Kasahara[1], Shun Sakuraba[2], Ikuo Fukuda[3], Jinzen Ikebe[4], Ryuhei Harada[5] ([1]*Col. Life Sci., Ritsumeikan Univ.*, [2]*Grad. Sch. Frontier Sci., Univ. Tokyo*, [3]*IPR, Osaka Univ.*, [4]*QST, MMS*, [5]*CCS, Univ. Tsukuba*)

2Pos271 **Local structures around protein phosphorylation sites**
Hafumi Nishi, Kengo Kinoshita (*Grad. Sch. Info. Sci., Tohoku Univ.*)

2Pos272 **膜タンパク質の構造分類：93 フォールドの同定**
We found at least 93 membrane protein folds in structure classification
Tsukasa Ueno[1], Masato Sakai[1], Masami Ikeda[2], Makiko Suwa[1,2] ([1]*Biol. Sci., Grad. Sci. Eng., Aoyama Gakuin Univ.*, [2]*Chem. Biol. Sci., Sci. Eng., Aoyama Gakuin Univ.*)

バイオインフォマティクス：分子進化 / Bioinformatics: Molecular evolution

2Pos273 **Culture-independent identification of genes encoding agarase from environmental bacteria using agarose gel microdroplets**
Eiji Shigihara[1], Ryo Iizuka[1], Takashi Sakurai[1], Yuji Hatada[2], Dong Hyun Yoon[3], Tetsushi Sekiguchi[4], Shuichi Shoji[3], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., Univ. Tokyo.*, [2]*Dept. of Life Sci. and Green Chem., Saitama Inst. of Technol.*, [3]*Dept. of Nanosci. and Nanoeng., Waseda Univ.*, [4]*Res. Org. for Nano&Life Innov., Waseda Univ.*)

2Pos274 **人工平面脂質二重膜を用いた抗菌性ペプチドの分子進化研究**
Molecular evolution of antimicrobial peptides using artificial planar lipid bilayers
Naoki Saigo[1], Yusuke Sekiya[2], Hirokazu Watanabe[2], Ryuji Kawano[3] ([1]*Tokyo Univ. of Agri. & Tech. Dept. of Biotech. Life Sci.*, [2]*Tokyo Univ. of Agri. & Tech. Dept. of Biotech. Life Sci.*, [3]*Tokyo Univ. of Agri. & Tech. Dept. of Biotech. Life Sci.*)

2Pos275 **タンパク質コーパスによる分散表現：ランダム配列の意味空間マッピングによる偽タンパク質の探索**
Distributed representation analysis of a protein corpus: Can we identify fake proteins by mapping random sequences on a semantic space?
Hiroshi Imamura, Shinya Honda (*AIST*)

2Pos276 **蛋白質構造安定性の平衡淘汰**
Selection maintaining protein stability at equilibrium
Sanzo Miyazawa

計測 / Measurements

2Pos277* **原子間力顕微鏡による初期発生胚の弾性率のタイムラプスイメージング**
Time-lapse imaging of elastic modulus of ascidian embryo during early development by atomic force microscopy
Yuki Fujii[1], Wataru Koizumi[2], Taichi Imai[2], Kohji Hotta[2], Kotaro Oka[2], Takaharu Okajima[1] ([1]*Grad. Schl. Inform. Sci. and Tech. Hokkaido Univ.*, [2]*Grad. Schl. Biosci. and Bioinfo. Keio Univ.*)

2Pos278 **信号処理蛋白質 Raf の生細胞内 ALEX 計測**
In-cell ALEX measurement of cytosolic signaling protein Raf
Kenji Okamoto[1], Kayo Hibino[2], Yasushi Sako[1] ([1]*RIKEN*, [2]*NIG*)

2Pos279 **細胞内環境におけるアンチセンス分子自己相補形成の mRNA に対する親和性への寄与**
Contribution of self-complementarity of antisense molecule to the affinity for mRNA in intracellular environment
Shunsuke Takeda[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*PRESTO, JST*)

2Pos280 **顕微ラマン分光法によるバクテリア一細胞の代謝活性測定**
Measuring metabolic activities in single bacterial cells by Raman microspectroscopy
Yota Kato[1], Hiroshi Ueno[1], Hiroyuki Noji[1,2] ([1]*Grad. Sch. Eng., Univ. Tokyo*, [2]*ImPACT, JST*)

2Pos281 **上皮細胞シートの頂端膜揺らぎ：走査型イオンコンダクタンス顕微鏡**
Scanning ion conductance microscopy (SICM) measurement of apical membrane fluctuation in epithelial cell monolayer
Kenta Aoki[1], Ryosuke Tanaka[1], Cho Nam-Joon[2], Takaharu Okajima[1] ([1]*Grad. Schl. Inform. Sci. and Technol., Hokkaido Univ.*, [2]*NanyangTechnol Univ.*)

2Pos282* **蛍光偏光相関分光法により明らかになった生細胞内での分子混雑と回転拡散の関係**
The relationship between rotational diffusion and crowding in living cell revealed by polarized fluorescence correlation spectroscopy
Makoto Oura[1], Johtaro Yamamoto[2], Takahiro Matsuda[1], Jian Ping Gong[2], Masataka Kinjo[2] ([1]*Hokkaido Univ. Grad. Sch. Life Sci.*, [2]*Hokkaido Univ. Fac. Adv. Life Sci.*)

2Pos283 **細胞イメージングシステムを用いたナノバイオプローブの生体適合性評価**
Evaluation of biocompatibility of nano-bio probes by using Cell imaging system
Yuko Nakane[1,2], Takashi Jin[2] ([1]*Tomy Digital Biology Co., Ltd.*, [2]*RIKEN QBiC*)

2Pos284 **金ナノ粒子を用いた加熱による細胞内局所温度の制御**
Manipulating the local temperature in a single cell with gold nanoparticles
Takaaki Honda[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharma., Univ. Tokyo*, [2]*JST PRESTO*)

2Pos285 **人工細胞開発に向けた細胞内 ATP 濃度の定量計測系開発**
Development of Quantitative ATP Concentration Measurement Method in Single Cells for Artificial Cell System
Hiroki Ashikawa[1], Kazuhito V. Tabata[1,2,3], Hiromi Imamura[4], Rikiya Watanabe[1,3], Hideyuki Yaginuma[5], Hiroyuki Noji[1,2] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Toyko*, [2]*ImPACT, CAO, Govt. Japan*, [3]*PRESTO, JST*, [4]*Grad. Sch. Bio., Univ. Kyoto*, [5]*QBiC, RIKEN*)

2Pos286 **バイオセンサシステムのための水素化アモルファスシリコン薄膜上のアミノ酸含有ゲルの電圧電流特性解析および蛍光性分子薄膜に関する研究**
Voltage current property of amino acid containing hydrogel and molecular film on hydrogenated amorphous silicon film for biosensor system
Makoto Horigane[1], Shotaro Minato[1], Hiroshi Masumoto[2], Takashi Goto[3], **Yutaka Tsujiuchi**[1] ([1]*Mat. Sci. & Eng., Akita Univ.*, [2]*Front. Res. Inst., Tohoku Univ.*, [3]*Inst. Mat. Res., Tohoku Univ.*)

2Pos287 **オンチップ１細胞計測系によるマクロファージの複数貪食の制御解析**
Studies on regulation mechanism of multiple phagocytosis of macrophage by single cell on-chip measurement assay
Yoshiki Nakata[1], Hideyuki Terazono[2], Masao Odaka[2], Kenji Matsuura[2], Akihiro Hattori[2], Kenji Yasuda[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*)

2Pos288 **Development of Novel Scanning Microscope for Measurement of Emission and Excitation Spectra Simultaneously**
Sankar Jana, Yutaka Shibata (*Tohoku University*)

バイオイメージング / Bioimaging

2Pos289 **高速 AFM によるアクチン様細胞骨格タンパク質 MamK 繊維の直接観察**
Direct observation of actin-like MamK cytoskeletal filaments by high-speed AFM
Yousuke Kikuchi[1], Marina Inagawa[2], Zachery Oestreicher[1], Azuma Taoka[1,3], Yoshihiro Fukumori[1] ([1]*Sch. of Nat. Sys., Col. of Sci. and Eng., Kanazawa Univ.*, [2]*Grad. Sch. of Nat. Sci. and Tech., Kanazawa Univ.*, [3]*Bio-AFM Center, Col. of Sci. and Eng., Kanazawa Univ.*)

2Pos290 **高速 AFM による古細菌 *S. solfataricus* 由来ミニ染色体維持(ssoMCM)タンパク質複合体の観察**
Observation of *S. solfataricus* archaeal minichromosome maintenance (ssoMCM) protein complex by high-speed AFM
Daisuke Noshiro[1], Noriyuki Kodera[1,2], Toshio Ando[1,3] ([1]*Bio-AFM FRC, Inst. of Sci. & Eng., Kanazawa Univ.*, [2]*PRESTO, JST*, [3]*CREST, JST*)

2Pos291 **ストレプトリジン O による膜孔形成の高速 AFM 観察**
High-speed AFM Observation of Membrane Pore Formation by Streptolysin O
Hirotaka Ariyama[1], Noriyuki Kodera[1], Toshio Ando[1,2] ([1]*Bio-AFM Frontier Research Center, Kanazawa Univ.*, [2]*Dept. Phys., Kanazawa Univ.*)

2Pos292 **ストレス顆粒内内在性 mRNA のナノスケール蛍光イメージング**
Nanoscale Fluorescence Imaging of Endogenous mRNAs in Stress Granules
Ko Sugawara[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*JST, PRESTO*)

2Pos293 **人知を超える超高速・高精度蛍光形態サイトメトリー**
Ghost Cytometry: fluorescence “imaging” cytometry beyond human's limit
Sadao Ota[1,2], Hiroyuki Noji[1,3] ([1]*Sch. Eng., Univ. Tokyo*, [2]*JST, PRESTO*, [3]*JST, ImPACT*)

2Pos294 **線形ゼロモード導波路を用いたアクチン重合メカニズムの 1 分子解析**
Single molecule observation of actin polymerization using linear zero-mode waveguides
Soichiro Fujii[1], Ryo Iizuka[1], Masamichi Yamamoto[1], Makoto Tsunoda[1], Takashi Tanii[2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*Fac. Sci. Eng., Waseda Univ.*)

2Pos295 **T 細胞活性化における微小管動態の超解像解析**
Super-resolution analysis of microtubule dynamics on T cell activation
Hengyu Shi, Yuma Ito, Wei Ming Lim, Kumiko Sakata-Sogawa, Makio Tokunaga (*Sch. Life Sci. Tech., Tokyo Inst. Tech.*)

2Pos296 **自発的光スイッチング蛍光タンパク質による簡便超解像イメージング**
Simple and easy way for superresolution imaging by spontaneously switching-on fluorescent protein
Yoshiyuki Arai, Hiroki Takauchi, Takeharu Nagai (*ISIR, Osaka Univ.*)

2Pos297* **細胞内グルタチオンの求核付加・解離平衡に基づく超解像蛍光イメージングプローブの開発**
Development of spontaneously blinking fluorophores based on nucleophilic addition of intracellular glutathione for superresolution imaging
Akihico Morozumi[1,4], Mako Kamiya[2,5], Shinnosuke Uno[1], Keitaro Umezawa[1], Toshitada Yoshihara[3], Seiji Tobita[3], Yasuteru Urano[1,2,4] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Grad. Sch. of Med., The Univ. of Tokyo*, [3]*Grad. Sch. Sci. Tech., Gunma Univ.*, [4]*AMED CREST*, [5]*JST PRESTO*)

2Pos298* **自由行動マウスの脳活動計測を可能にする化学発光膜電位センサーの開発**
Development of a chemiluminescent voltage indicator applicable to brain activity recording in freely moving mice
Shigenori Inagaki[1], Masakazu Agetsuma[2], Hidekazu Tsutsui[3,4], Yoshiyuki Arai[2], Kazushi Suzuki[5], Yuka Jinno[4], Yasushi Okamura[1,4], Tomoki Matsuda[2], Takeharu Nagai[1,2,5] ([1]*FBS., Univ. Osaka*, [2]*ISIR., Univ. Osaka*, [3]*Sch. Mat. Sci., JAIST*, [4]*Grad. Sch. Med., Univ. Osaka*, [5]*Dep of Biotech, Univ. Osaka*)

2Pos299 **自動 1 分子イメージング装置の開発**
Development of automatic single molecular imaging system
Masato Yasui[1], Jun Kozuka[1], Michio Hiroshima[1], Taku Tsuzuki[2], Yasushi Sao[3] ([1]*RIKEN QBiC*, [2]*Osaka University*, [3]*RIKEN Cellular Informatics Laboratory*)

2Pos300 **Fluorescence recovery after photobleaching (FRAP) analysis of INO80 chromatin remodeling complex**
Tsubasa Isogaki[1], Yuma Ito[1], Shota Ichikawa[1], Hiroshi Kimura[2], Masahiko Harata[3], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*Inst. Innov. Res., Tokyo Inst. Tech.*, [3]*Grad. Agr. Sci., Tohoku Univ.*)

2Pos301* **The Correspondence between Raman Microspectroscopy and Omics Data**
Koseki Kobayashi-Kirschvink[1], Hidenori Nakaoka[1], Arisa Oda[2], Kunihiro Ohta[2,3], Yuichi Wakamoto[1] ([1]*Dep. Bas. Sci., Univ. Tokyo*, [2]*Dep. Life. Sci., Univ. Tokyo*, [3]*Dep. Bio. Sci., Univ. Tokyo*)

2Pos302 **電子顕微鏡法のための画像処理パッケージ Eos/PIONE の更なる発展**
Further progress of Eos/PIONE for image analysis packages for electron microscopy
Takuo Yasunaga, Takafumi Tsukamoto, Ayaka Iwasaki (*Dept. of Biosci. Bioinfo., School of Comp. Sci. Systems, Eng., Kyushu Inst. Tech.*)

2Pos303 **X 線自由電子レーザーを用いたコヒーレント回折イメージング実験におけるデータ解析の自動化と酵母細胞核の三次元構造解析への応用**
Automated data analyses for 3D structural reconstruction of yeast nuclei in coherent diffraction imaging using X-ray free-electron laser
Yuki Sekiguchi[1,2], Amane Kobayashi[1,2], Koji Okajima[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Masaki Yamamoto[2] ([1]*Grad. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*)

バイオエンジニアリング / Bioengineering

2Pos304 **フォトクロミック分子を利用した低分子量 G タンパク質の光可逆的制御**
Photo-regulation of Small G protein RhoA using Photochromic Molecules
Kaori Masuhara[1], Masahiro Kuboyama[1], Nobuyuki Nishibe[2], Shinsaku Maruta[1,2] ([1]*Grad. Sch. Bioinfo., Univ. SOKA*, [2]*Dept. Bioinfo., Fac. Engineer., Univ. SOKA*)

2Pos305 **Structure and mechanism of the multimerization of small GTPase protein Ras induced by chamical modification at HVR domain**
Takashi Hashimoto[1], Shinsaku Maruta[1], Yasunobu Sugimoto[2] ([1]*Soka University*, [2]*Nagoya University*)

2Pos306 **環状型サイトカインの安定性を向上させる結合末端ループ長の選択**
Selection of the loop length about circularized cytokines
Risa Shibuya[1], Takamitsu Miyafusa[2], Wataru Nishima[2], Shinya Honda[2] ([1]*Front. Sci. , Univ. of Tokyo*, [2]*BMRI, AIST*)

2Pos307 **Interaction between ring or linear DNA vs. nanopore/nanoslit**
Takayuki Nakayama[1], Yoshiaki Iitsuka[1], Seiya Minato[1], Surat Wangwarunyoo[2], Naoto Sakashita[1], Kentaro Ishida[1], Toshiyuki Mitsui[1] ([1]*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*, [2]*Chulalongkorn Univ.*)

2Pos308 **Photo-regulation of Small G-proteins Ras Using Photochromic Molecules**
Masahiro Kuboyama, Kaori Masuhara, **Shinsaku Maruta** (*Soka University*)

2Pos309 **脂質膜上チャネル形成のための 10 ナノメートルスケールのポアを持つ DNA オリガミナノ構造**
DNA origami nanostruture with pore of ten nanometer scale for forming channel on lipid membrane
Koichiro Katayama[1], Ibuki Kawamata[1], Yuki Suzuki[1,2], Satoshi Murata[1], Shin-ichiro Nomura[1] ([1]*Graduate school of Engineering, Tohoku University*, [2]*Frontier Research Institute for Interdisciplinary Sciences, Tohoku University*)

2Pos310* **撹拌操作が引き起こすゲノム DNA の二本鎖切断：新規実験手法の提案**
How to keep genome-sized DNA safe against stirring stress: Quantitative analysis through single DNA observation
Hayato Kikuchi, Yuko Yoshikawa, Rinko Kubota, Kenichi Yoshikawa (*Lab. Biol. Phys., Facul. Life Med. Sci., Doshisha Univ.*)

2Pos311* **Four-way junction DNA 形成による癌特異的 microRNA 発現パターンの自律的検出**
Programmable system for recognition of microRNA expression pattern using four-way junction DNA formation
Moe Hiratani, Masayuki Ohara, Ryuji Kawano (*The Dep. of Biotech. and Life Sci., Tokyo Univ. of Agr. and Tech.*)

2Pos312 **光による DNA ハイドロゲルのパターン形成**
Patterning of DNA hydrogel using light
Suguru Shimomura, Takahiro Nishimura, Yusuke Ogura, Jun Tanida (*Grad. Sch. Info. Sci. & Tech., Osaka Univ.*)

2Pos313 **キメラ受容体によるバクテリアバイオセンサーの特異性改変**
Modification of ligand specificity in bacterial biosensor with hybrid chemoreceptors
Hana Satou[2], Nao Fujii[2], Takashi Sagawa[1], Hiroto Tanaka[1], Kazuhiro Oiwa[1,2], **Hiroaki Kojima**[1] ([1]*Frontier Lab.,KARC,NICT*, [2]*Sch. Sci., Univ. Hyogo*)

2Pos314* **金ナノ粒子のデジタル計数法による標的 DNA の高感度検出**
High-sensitivity Homogeneous DNA hybridization assay by Digital Counting of Gold Nanoparticle Dimers
Takaha Mizuguchi, Keiko Esashika, Toshiharu Saiki (*Grad. Sch. Sci. Tech., Keio Univ.*)

2Pos315* **A single integrated gene nano-chip functioning in an artificial cell**
Takeya Masubuchi[1], Masayuki Endo[2], Ryo Iizuka[3], Ayaka Iguchi[4], Yoon Doung Hyun[4], Tetsushi Sekiguchi[5], Hao Qi[1,6], Ryosuke Iinuma[1], Yuya Miyazono[1], Shuichi Shoji[4], Takashi Funatsu[3], Hiroshi Sugiyama[2,7], Yoshie Harada[2], Takuya Ueda[1], Hisashi Tadakuma[1,2] ([1]*Grad. Sch. of Frontier Sci., The Univ. of Tokyo*, [2]*iCeMS, Kyoto Univ.*, [3]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [4]*Grad. Sch. of Adv. Sci. and Eng., Waseda Univ.*, [5]*Research Organization for Nano & Life Innovation, Waseda Univ.*, [6]*Dept. of Chem. Eng. and Tech., Tianjin Univ.*, [7]*Grad. Sch. of Sci., Kyoto Univ.*)

2Pos316* **アメーバ型分子ロボット：モータータンパク質と DNA デバイスを内包した巨大リポソームの形状変化とその制御**
Amoeba type molecular robot: controlling shape change of giant liposome entrapping molecular motors and DNA circuits
Yusuke Sato[1], Yuichi Hiratsuka[2], Ibuki Kawamata[1], Satoshi Murata[1], Shin-ichiro Nomura[1] ([1]*Grad. Sho. Eng., Tohoku Univ.*, [2]*Sch. Mat. Sci., JAIST*)

2Pos317 **心筋細胞集団同士を繋ぐ線維芽細胞の距離に対する同期**
Synchronization of large clusters of cardiomyocytes connected with fibroblasts and its distance change
Shota Miyakoshi[1], Toshiyuki Mitsui[2], Tomoyuki Kaneko[1] ([1]*LaRC, Grad. Sci. Eng., Hosei Univ.*, [2]*Dept. Math. Phys. Col. Sci. Eng,. Aoyama Univ.*)

2Pos318 **目的細胞の回収を目指した微小液滴内培養法の開発**
Development of a single cell cultivating method using a microdroplets forming technique for sorting specific cells
Hideyuki Terazono[1], Masao Odaka[1], Akihiro Hattori[1], Kenji Matsuura[1], Kenji Yasuda[1,2] ([1]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*, [2]*Dept. Physics, Waseda Univ.*)

第3日目（11月27日（日））／Day 3 (Nov. 27 Sun.) 大会議室 101 ＋102、多目的ホール／Conference Room 101+102, Multi-Purpose Hall

蛋白質：構造 / Protein: Structure

3Pos001 **Single Particle Analysis of *Eh*V-ATPase by Phase-contrast cryo-Electron Microscopy**
Jun Tsunoda[1,2], Chihong Song[2], Fabiana Lica Yakushiji[3], Takeshi Murata[3], Hiroshi Ueno[4], Junichi Takagi[6], Ryota Iino[1,5], Kazuyoshi Murata[1,2] ([1]*SOKENDAI*, [2]*NIPS*, [3]*Dept. Chem., Chiba Univ.*, [4]*Dept. Appl. Chem., Sch. Eng., Univ. Tokyo*, [5]*OIIB/IMS*, [6]*Osaka Univ. IPR*)

3Pos002 **TEM と ASEM を用いたタンパク質複合体・細胞組織の親水環境での観察**
Electromicroscopy of protein complexes, cells and tissues in hydrophilic environment
Chikara Sato[1], Nassirhadjy Memtily[1], Mari Sato[1], Toshiko Yamazawa[2], Masaaki Kawata[1] ([1]*AIST*, [2]*Dept. Mol Physiol, Jikei Univ. Sch. Med.*)

3Pos003 **クライオ電子顕微鏡による *Thermus thermophilus* V-ATPase の単粒子解析**
Single-particle analysis of *Thermus thermophilus* V-ATPase by Cryo-EM
Atsuko Nakanishi[1], Jun-ichi Kishikawa[1], Kaoru Mitsuoka[2], Ken Yokoyama[1] ([1]*Dept. LifeSci. Kyoto Sangyo Univ.*, [2]*Res. Ctr. UVHEM. Univ. Osaka.*)

3Pos004 **クライオ電子顕微鏡単粒子解析によるマウスノロウイルス VLP の構造解析**
Structural Analyses of Murine Norovirus VLPs by Cryo-Electron Microscopy Single Particle Analysis
Chihong Song[1], Motohiro Miki[2,3], Reiko Todaka[2], Kei Haga[2], Akira Fujimoto[2], Kazuhiko Katayama[2], Kazuyoshi Murata[1] ([1]*NIPS*, [2]*NIID*, [3]*Denka Seiken*)

3Pos005 2D ハイブリッド解析による電子顕微鏡平均画像の成分解析

Component analysis of averaged EM images by 2D hybrid analysis

Atsushi Matsumoto[1], Junichi Takagi[2], Atsushi Kawaguchi[3], Kenji Iwasaki[2] ([1]*National Institutes for Quantum and Radiological Science and Technology*, [2]*Osaka University*, [3]*Tsukuba University*)

3Pos006 Structural analysis for V1-ATPase from a variety of prokaryotes

Nao Takeuchi[1], Atsuko Nakanishi[1], Jun-ichi Kishikawa[1], Kaoru Mitsuoka[2], Ken Yokoyama[1] ([1]*Kyoto Sangyo Univ. LifeSci.*, [2]*Osaka Univ. Res. Ctr. UHVEM*)

3Pos007 最新低温電子顕微鏡“CryoARM”の性能

Performance of State-of-the-art CryoEM, named “CryoARM”

Takayuki Kato[1], Naoki Hosogi[2], Takeshi Kaneko[2], Isamu Ishikawa[2], Keiichi Namba[1,3] ([1]*Grad. Sch. of Front. Biosci., Osaka Univ.*, [2]*JEOL*, [3]*RIKEN, QBiC*)

3Pos008 電顕３次元密度マップからαヘリックスを認識する混合正規分布モデルの開発

Detection of alpha-helices from the 3D EM density map using Gaussian mixture model

Takeshi Kawabata, Haruki Nakamura (*IPR, Osaka U.*)

3Pos009 Towards Understanding the Molecular Architecture of Human DNA Polymerase δ using Electron Microscopy and Computational Modeling

Ashutosh Srivastava[1], Yuji Masuda[2], Jiro Usukura[3], Motoshi Suzuki[4], Florence Tama[1,5] ([1]*ITbM, Nagoya Univ.*, [2]*Res. Inst. Env. Med., Nagoya Univ.*, [3]*Str. Bio. Cen., Grad. Sch. Sci., Nagoya Univ.*, [4]*Div. Mol. Carc., Grad. Sch. Med., Nagoya Univ.*, [5]*Dept. Phys., Sch. Sci., Nagoya Univ.*)

3Pos010 GPI アタッチメントシグナル領域の二次構造解析

Secondary structural analysis of GPI attachment regions

Keiya Inoue[1], Daiki Takahashi[2], Tatsuki Kikegawa[2], Kenji Etchuya[2], Yuri Mukai[1,2] ([1]*Sch. Sci. & Tech., Meiji Univ.*, [2]*Grad. Sch. Sci. & Tech., Meiji Univ.*)

3Pos011 インターセクチン２のコンホメーション解析

Conformational Analysis of Multidomein Protein Intersectin 2

Kazutaka Murayama[1,2], Miyuki Murayama-Kato[2], Ryogo Akasaka[2], Daisuke Sugimori[3], Mikako Shirouzu[2] ([1]*Tohoku Univ. Biomed. Eng.*, [2]*RIKEN, CLST*, [3]*Fukushima Univ. Symbio. Sys. Sci.*)

3Pos012 タンパク質の構造変性と回転拡散係数

Rotational diffusion coefficients of proteins along denaturation curve

Yoshitake Tomoyuki, Terazima Masahide (*Graduate School of Science, Kyoto University*)

3Pos013 シアノバクテリア時計タンパク質 KaiC の AFM 観察

AFM observation of a ring-shaped structure of KaiC

Jun Abe, Atsushi Mukaiyama, Yoshihiko Furuike, Shuji Akiyama (*Division of Trans-Hierarchical Molecular Systems, Research Center of Integrative Molecular Systems (CIMoS), Institute for Molecular Science (IMS)*)

3Pos014 高速 AFM による 20S プロテアソーム関連タンパク質の動態観察

Dynamics observation of the 20S proteasome-related proteins using High-Speed AFM

Toshiya Kozai[1], Tadashi Satoh[2], Arunima Sikdar[3,4], Hirokazu Yagi[2], Maho Yagi-Utsumi[3], Takayuki Uchihashi[1], Toshio Ando[1], Koichi Kato[3,4] ([1]*Dept. of phys., Kanazawa Univ.*, [2]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [3]*Okazaki Inst. Integ. Biosci.*, [4]*Nat. Univ. SOKENDAI*)

3Pos015 立体構造予測において疎水効果を評価するための新しい指標： 仮想原子の周りのコンタクト数

A new measure for hydrophobicity: Contact number around an imaginary atom

Yota Masuyama, George Chikenji (*Grad. Sch. Eng., Nagoya Univ.*)

3Pos016 統合失調症疾患感受性遺伝子産物 G72 タンパク質の構造機能予測

Structure and function prediction of the G72 protein, the product of a susceptible gene for schizophrenia

Yusuke Kato, Kiyoshi Fukui (*Institute for Enzyme Research, Tokushima University*)

3Pos017 残基間平均距離統計に基づくコンタクトマップによる天然変性領域の予測

Prediction of IDRs by a contact map based on inter residue average distance statistics

Takumi Shimomura, Takeshi Kikuchi (*Univ. Ritsumei*)

3Pos018 A new threading method based on the physical characteristics of sequence-structure compatibility

Kyosuke Tomoda, Yota Masuyama, George Chikenji (*Grad. Sch. Eng., Nagoya Univ.*)

3Pos019 EMDB, PDB, SASBDB 中の多階層構造データを対象としたウェブベースのサービス

Web based services for multiscale structure data in EMDB, PDB and SASBDB

Hirofumi Suzuki[1,2], Takeshi Kawabata[1], Gert-Jan Bekker[1,2,3], Haruki Nakamura[1,2] ([1]*IPR, Osaka Univ.*, [2]*PDBj*, [3]*FBS, Osaka Univ.*)

3Pos020 単独で構造を維持するドメインデータベース「IS-Dom」の他のデータベースに依存しない拡張

Standalone definition of putatively independent structural domain: IS-Dom

Soichiro Ide[1], Teppei Ebina[2], Richa Tanbi[1], Yutaka Kuroda[1] ([1]*Tokyo University of Agriculture and Technology*, [2]*Department of Physiology, Graduate School of Medicine, The University of Tokyo*)

3Pos021 Attempts at CA-type formal analysis of fibrous assembly of particles

Takashi Konno (*Mol. Physiol., Med., Univ. Fukui*)

3Pos022 生物の低温適応と蛋白質配列の進化

Cold adaptation of organisms and the evolution of protein sequences

Matsuyuki Shirota[1,2,3] ([1]*Grad. Sch. Med., Tohoku Univ.*, [2]*ToMMo, Tohoku Univ.*, [3]*Grad. Sch. Inform Sci., Tohoku Univ.*)

3Pos023 **スライディングとストランド間移動を用いたヒト抗ウイルス因子 APOBEC3G の高効率な DNA 配列探索：実時間 NMR による新知見**
Sliding and intersegmental transfer on DNA enhance target search of human anti-viral factor APOBEC3G: insight by the real-time NMR study
Keisuke Kamba[1], Takashi Nagata[1,2], Masato Katahira[1,2] ([1]*Inst. of Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. of Energy Sci., Kyoto Univ.*)

3Pos024 **分子動力学ドッキング・シミュレーションによるスーパーコイル DNA 結合（SDR）ペプチドとクロスオーバー DNA の選択的結合メカニズムの解析**
Molecular dynamics docking study on selective binding mechanisms of supercoiled-DNA recognition (SDR) peptide and spatially-crossover DNA
Hiroshi Nishigami[1], Kakeru Sakabe[1], Jiyoung Kang[1], Kuniaki Sano[2], Kimiko Tsutsui[2], Ken Tsutsui[2], Kazuhiko Yamasaki[3], Masaru Tateno[1] ([1]*Grad. Sch. Life Sci., Univ. Hyogo*, [2]*Dept. Neurogenomics, Grad. Sch. Med. Dent. Pharm. Sci., Okayama Univ.*, [3]*Biomedical Res. Inst., AIST*)

3Pos025 **Unravelling the mechanism of (6-4) photolyase enzyme**
Hisham Dokainish, AKio Kitao (*The University of Tokyo*)

3Pos026 **暗視野顕微鏡を用いたタンパク質過飽和溶液中ナノスケールダイナミクス構造観察**
Nano-scale Observations of Supersaturated Protein Dynamics using Dark-Field Microscopy
Kazuki Yoshimura[1], Yufuku Matsushita[1], Keigo Ikezaki[1], Hiroshi Sekiguchi[2], Yuji Goto[3], Yuji Sasaki[1,2] ([1]*Grad. Sch. Front. Sci., Univ. Tokyo*, [2]*JASRI/SPring-8*, [3]*IPR, Osaka Univ.*)

3Pos027 **Analysis and control of protein crystallization using short peptide tags without affecting structure, thermal stability and function**
MM. Islam[1], N. Shigeyoshi[2], K. Noguchi[3], M. Yohda[3], SI. Kidokoro[2], Y Kuroda[3] ([1]*CU*, [2]*NUT*, [3]*TUAT*)

3Pos028 **滴定 X 線溶液散乱測定を用いたアダプター蛋白質 GGA-ユビキチン相互作用の解析**
An analysis of the interaction of GGA with ubiquitin by using titration SAXS measurement
Yugo Hayashi[1], Miho Shinohara[1], Keito Yoshida[1], Yoichi Yamazaki[1], Kazuhisa Nakayama[2], Soichi Wakatsuki[3], Hironari Kamikubo[1] ([1]*Grad. Sch. Mat. Sci., NAIST*, [2]*Grad. Sch. Pharm., Kyoto Univ.*, [3]*Stanford Univ.*)

3Pos029 **K63 ジユビキチンと TAB2 複合体の拡張サンプリング**
Large-scale configurational sampling of K63-linked di-ubiquitin complexed with TAB2
Keiichi Inariyama[1], Hafumi Nishi[2], Kei Moritsugu[1], Akinori Kidera[1] ([1]*Grad. Sch. of Med. Life Sci., Yokohama City University*, [2]*Grad. Sch. Info. Sci., Tohoku University*)

3Pos030 **Periodic Formation of the Cyanobacterial Circadian Clock Protein Complexes**
Shun Terauchi[1], Takahiro Iida[1,2], Kentaro Ishii[2], Masahiro Ishiura[2], Kosuke Maki[1] ([1]*Sch. of Sci., Nagoya Univ.*, [2]*Center for Gene Res., Nagoya Univ.*)

3Pos031 **広角溶液散乱測定のための環境整備と時計タンパク質への応用**
Wide-angle x-ray scattering studies on circadian clock systems
Shuji Akiyama[1,2], Takaaki Hikima[2], Atsushi Mukaiyama[1,2], Jun Abe[1,2], Yoshihiko Furuike[1,2] ([1]*CIMoS, IMS*, [2]*RIKEN SPring-8 Center*)

3Pos032 **X 線小角散乱法を用いた神経軸索伸長系関連蛋白質 shootin1 の動的な構造変化の解析**
Structural alteration of shootin1 upon phosphorylation revealed by using small angle x-ray scattering
Shoki Nakata[1], Keito Yoshida[1], Kentarou Baba[2], Yohei Shibata[1], Yoichi Yamazaki[1], Naoyuki Inagaki[2], Hironari Kamikubo[1] ([1]*Grad. Sch. of Mater. Sci., Nara Inst. of Sci. & Tech.*, [2]*Grad. Sch. of Biol. Sci., Nara Inst. of Sci. & Tech.*)

3Pos033 **Dynamical system of alpha-crystallin oligomers**
Rintaro Inoue, Takumi Takata, Noriko Fujii, Masaaki Sugiyama, Nobuhiro Sato, Yojiro Oba (*Research Reactor Institute,Kyoto University*)

3Pos034 **Refinement of Cryo-EM Structures Using Scattering Factors of Charged Atoms**
Koji Yonekura, Saori Maki-Yonekura, Rei Matsuoka, Yoshiki Yamashita, Fumie Iwabuki, Maiko Tanaka (*RIKEN SPring-8 Center*)

3Pos035 **Visualization of 11- and 34-fold rotational symmetries in the MS ring of the bacterial flagellum by electron cryomicroscopy**
Akihiro Kawamoto[1], Ayana Kaido[2], Miki Kinoshita[1], Tomoko Miyata[1], Tohru Minamino[1], Takayuki Kato[1], Keiichi Namba[1,3] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Dept. Food Science and Nutrition., Doshisha Women's College of Liberal Arts*, [3]*QBiC, RIKEN*)

3Pos036 **単粒子コヒーレント X 線回折像の類似積判定のためのマルチステップアルゴリズム**
Multistep similarity detection algorithm for single particle X-ray coherent diffractions
Atsushi Tokuhisa[1], Osamu Miyashita[1], Florence Tama[1,2] ([1]*AICS, RIKEN*, [2]*Department of Physics, Nagoya University*)

3Pos037 **Molecular determinants of the ATP binding properties of the ε subunit from bacterial ATP synthases**
Alexander Krah[1,2], Yasuyuki Kato-Yamada[3], Changbong Hyeon[2], Shoji Takada[1] ([1]*Dept. Biophys., Kyoto Uni.*, [2]*KIAS*, [3]*Dept. Life Sci., Rikkyo Uni.*)

3Pos038 **Microtubule stability and the tubulin molecule interactions within the microtubule lattice**
Kenta Hirasada, Daisuke Yamamoto, **Miho Katsuki** (*Fukuoka Univ., Faculty of Science*)

3Pos039 **隣り合うチューブリン存在下と非存在下における α/β チューブリン C 末端の異なる三次元空間分布**
Three-dimensional distributions of α/β-tubulin C-terminal tails and the influence of neighboring tubulins
Koji Umezawa[1,2], Yukinobu Mizuhara[3], Jun Ohnuki[3], Mitsunori Takano[3] ([1]*Grad. Sch. of Sci. & Tech., Shinshu Univ.*, [2]*IBS, Shinshu Univ.*, [3]*Grad. Scl. of Adv. Sci. & Eng., Waseda Univ.*)

3Pos040 **F アクチンの水和状態は Mg^{2+}/Ca^{2+} イオンに強く依存する**
Strong Mg/Ca Ion Dependence of Hydration State of F-actin
Makoto Suzuki[1], Asato Imao[1], George Mogami[1], Ryotaro Chishima[1], Takahiro Watanabe[1], Takaya Yamaguchi[1], Nobuyuki Morimoto[1], Tetsuichi Wazawa[2] ([1]*Grad. School of Eng. Tohoku Univ.*, [2]*The Institute of Scientific and Industrial Research, Osaka University*)

3Pos041 アクチンフィラメントに結合したローダミンファロイジン蛍光のゆらぎ
Fluctuation of rhodamine-phalloidin fluorescence along actin filaments
Taro Ueda[1,3], Saku Kijima[2,3], **Takahiro Suzuki**[1] ([1]*Dept. of Physics, Waseda Univ.*, [2]*Biomed. Res. Inst., AIST*, [3]*Grad. Sch. Life Environ. Sci., Univ. of Tsukuba*)

3Pos042 OH 伸縮振動のラマン分光によるミオシン S1 および他のいくつかのタンパク質の水和状態の測定
Hydration study on myosin subfragment-1 (S1) and some other proteins by Raman OH-atretching spectroscopy
Yuki Ochiai[1], George Mogami[1], Tetsuo Taniuchi[2], Makoto Suzuki[1] ([1]*Grad. Sch. Eng., Univ. Tohoku*, [2]*IMRAM, Univ. Tohoku*)

3Pos043 ウルトラファインバブル水中のタンパク質構造の研究
Study of protein structure in ultra-fine bubble water
Mitsuhiro Hirai[1], Satoshi Ajito[1], Kosuke Takahashi[1], Noboru Ohta[2], Tatsuo Iwasa[3] ([1]*Grad. Sch. Sci. Tech., Gunma Univ.*, [2]*Jpn. Syn. Rad. Res. Inst.*, [3]*Muroran Inst. Tech.*)

3Pos044 3D−RISM 理論を応用した溶液中における Met−enkephalin の構造揺らぎの解析
Analysis of structural fluctuations of Met-encephalin in the solution phase by means of 3D-RISM theory
Masatake Sugita[1], Fumio Hirata[2] ([1]*Dept. of Bioinfo., Col. of Life Sci., Ritsumeikan Univ.*, [2]*Toyota Phys. & Chem. Res. Inst.*)

3Pos045 分子動力学シミュレーションとエネルギー表示理論を用いた共溶媒変性効果の自由エネルギー解析
Free energy analysis of cosolvent effect through molecular dynamics simulation and energy-representation method
Yu Yamamori, Nobuyuki Matsubayasi (*Grad. Sch. of Eng. Sci., Osaka Univ.*)

3Pos046 タンパク分子内情報伝達を実現する構造基盤の探索―既知の構造から
Search for Common Structural Basis of Mechanical Communication in Proteins: from Known Structures
Yuichi Togashi (*RcMcD, Hiroshima Univ.*)

3Pos047 CAPAXIS と PyMOL を用いたウイルス粒子脱殻の描画・操作
Modeling of uncoating of virus capsid by using CAPAXIS and PyMOL
Shunsuke Sato[1], Aya Kosugi[1], Go Wabanabe[2], Shigetaka Yoneda[2] ([1]*Grad. Sch. Sci., Kitasato Univ.*, [2]*Sch. Sci., Kitasato Univ.*)

蛋白質：物性 / Protein: Property

3Pos048 生体分子の分子動力学計算を取り扱う高速な QM/MM 理論の開発
Development of rapid QM/MM approach for biomolecular simulations
Hiroaki Nishizawa[1], Hisashi Okumura[1,2] ([1]*IMS*, [2]*Sokendai*)

3Pos049 タンパク質の基準振動モードのネットワーク解析：中心性指標の計算
Network analysis of normal modes of proteins: calculations of various centrality measures
Hiroshi Wako[1], Shigeru Endo[2] ([1]*Sch. of Soc. Sci., Waseda Univ.*, [2]*Sch. of Sci., Kitasato Univ.*)

3Pos050 タンパク質構造変化における経路の多様性：分子動力学シミュレーションによる解析
A variety of pathways for a conformational change of a protein investigated by molecular dynamics simulation
Sotaro Fuchigami (*Grad. Sch. of Medical Life Science, Yokohama City Univ.*)

3Pos051 カメレオンモデルによる NtrC の構造転移機構の研究
Mechanism of conformational transition of NtrC studied by using chameleon model
Shinya Abe[1], Atsushi Mizuno[2], Masaki Sasai[1], Tomoki P. Terada[1] ([1]*Dept. Comput. Sci. Eng., Grad. Sch. Eng., Nagoya Univ.*, [2]*Dept. Appl. Phys., Sch. Eng., Nagoya Univ.*)

3Pos052 Oct4 の 2 つの DNA 結合サブドメインを結ぶ柔軟な linker 領域の自由エネルギー地形
Free-energy landscape of the flexible linker connecting two DNA-binding subdomains of Oct4
Tomonori Hayami[1,2], Shoji Takada[3], Haruki Nakamura[1], Junichi Higo[1] ([1]*Inst. Protein Res., Osaka Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Dept. Biophys., Grad. Sch. Sci., Kyoto Univ.*)

3Pos053 タンパク質の協同的な折れたたみとループのつながり方の関係
Relation between cooperative protein folding and loop connections
Nobu C. Shirai[1], Shintaro Minami[2] ([1]*Center for Information Technologies and Networks, Mie University*, [2]*Graduate School of Information Science, Nagoya University*)

3Pos054 酸曝露後中和による抗体のフォールディングと凝集: 二種の光子相関分光法による追跡
Folding versus aggregation of an antibody initiated by pH-shift stress: Double tracking by photon correlation spectroscopies
Hiroshi Imamura, Akira Sasaki, **Shinya Honda** (*Natl Inst Adv Indust Sci. Tech.*)

3Pos055 NMR を用いた血清環境での相互作用解析
NMR approach for understanding protein interactions in serum environments
Saeko Yanaka[1,2], Rina Yogo[1,2], Hirokazu Yagi[1,2], Koichi Kato[1,2] ([1]*Department of Bioorganization Research, Okazaki Institute for Integrative Bioscience and Institute for Molecular Science, National Institutes of Natural Sciences*, [2]*Department of Structural Biology and Biomolecular Engineering, Graduate School of Pharmaceutical Sciences, Nagoya City University*)

3Pos056 酸化と酵素切断が LDL の物性に与える影響
Physical properties of low-density lipoprotein after oxidation or proteolytic enzyme treatment
Seiji Takeda[1], Agus Subagyo[2], Shu-Ping Hui[1], Hirotoshi Fuda[1], Kazuhisa Sueoka[2], Hitoshi Chiba[1] ([1]*Fac. Health Sci., Univ. Hokkaido*, [2]*Grad. Sch. Inf. Sci. Tech., Univ. Hokkaido*)

3Pos057 **イムノグロブリン G のマルチドメイン構造形成におけるエントロピー効果**
Entropic stabilization of the multi-domain architecture in immunoglobulin G
Seiki Yageta, Hiroshi Imamura, Shinya Honda (*Biomedical Research Institute, National Institute of Advanced Industrial Science and Technology*)

3Pos058 **統計熱力学に基づいたサーモフィリックロドプシンの熱安定化変異体の作製**
Identification of thermostabilizing mutations for thermophilic rhodopsin based on statistical thermodynamics
Sayaka Nemoto[1], Satoshi Yasuda[1,2], Kenji Mizutani[1], Takashi Tsukamoto[3], Yuki Sudo[3], Masahiro Kinoshita[2], Takeshi Murata[1,4] ([1]*Grad. Sch. Sci., Univ. Chiba*, [2]*Inst. Advanced Energy, Univ. Kyoto*, [3]*Grad. Sch. Medicine, Dentistry, and Pharmaceutical Sciences*, [4]*JST, PRESTO*)

3Pos059 **Amorphous aggregation of cytochrome c with inherently low amyloidogenicity is characterized by the phase diagram**
Yuxi Lin[1], Jozsef Kardos[2], Misaki Kinoshita[1], Toshihiko Sugiki[1], Koichiro Ishimori[3], Yuji Goto[1], Young-Ho Lee[1] ([1]*Institute for Protein Research, Osaka University*, [2]*Department of Biochemistry, Eotvos Lorand University*, [3]*Department of Chemistry, Faculty of Science, Hokkaido University*)

3Pos060 **The virial coefficients based on the rotaitional diffusion as a criterion of the protein crystallizaton**
Yudai Katsuki[1], Akane Kato[2], Etsuko Nishimoto[3] ([1]*sch. Agr., Kyushu Univ.*, [2]*Grad. Sch. Bioresour. Bioenviron. Sci., Kyushu Univ.*, [3]*Fac. Agr., Kyushu Univ.*)

3Pos061 **The hydration state near the binding site of human Serum Albumin revealed by the time-resolved fluorescence spectrum of Trp214**
Shoutaro Kubo[1], Etsuko Nishimoto[2] ([1]*Sch. Agr., Kyushu Univ.*, [2]*Fac. Agr., Kyushu Univ.*)

3Pos062 **アクチンフィラメントの圧電特性 III**
Piezoelectric property of an actin filament III
Jun Ohnuki, Takato Sato, Hideyo Okamura, Taro Q.P. Uyeda, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

3Pos063 **テトラヒメナ外腕ダイニン重鎖（Dyh3p)における運動系の開発と運動特性**
Motor domain-based motility system and motile properties of alpha heavy chain in Tetrahymena outer arm dynein
Masaki Edamatsu (*Department of Life Sciences, Graduate School of Arts and Sciences, The University of Tokyo*)

蛋白質：計測·解析 / Protein: Measurement & Analysis

3Pos064 **タンパク質の NMR 解析が困難な系にも有効なシグナル帰属法**
Signal assignment strategy for protein NMR under challenging conditions
Takuma Kasai[1,2], Kae Higuchi[1], Kohsuke Inomata[1,3], Takanori Kigawa[1,2,4] ([1]*RIKEN QBiC*, [2]*CREST, JST*, [3]*PRESTO, JST*, [4]*Sch. Comput., Tokyo Inst. of Tech.*)

3Pos065 **二量子遷移 EPR 距離測定における短距離成分の影響**
Effects of Short Distance Components on Double Quantum Coherence EPR Distance Measurements
Yasunori Ohba[1], Syouji Ueki[2], Toshiaki Arata[3] ([1]*IMRAM、Tohoku Univ.*, [2]*Fac. Pharm. Sci., Tokushima Bunri Univ.*, [3]*Dep. Biol. Sci., Osaka Univ.*)

3Pos066 **天然変性タンパク質の SAXS プロフィル評価法を開発するための新たな枠組み**
A novel framework for developing the evaluation method of SAXS profile of IDP
Yasutaka Seki[1], Shigeyoshi Nakamura[2] ([1]*Kochi Med. Sch.*, [2]*Kitakushu Nat. Coll. of Tech.*)

3Pos067 **SAXS とアミノ酸残基レベル二次構造情報からのタンパク質立体構造の構築**
Protein structure constructed with SAXS and secondary structures at amino acid residue level
Yasumasa Morimoto[1], Takayuki Ichioka[1], Toru Terada[2], Kentaro Shimizu[2], Yoshitaka Matsumura[1], Masaki Kojima[1] ([1]*Sch. of Life Sci., Tokyo Univ. of Pharm. and Life Sci.*, [2]*Dept. of Appl. Biol. Chem., Grad. Sch. of Agr. and Life Sci., The Univ. of Tokyo*)

3Pos068 **タンパク質複合体の解離過程の分子動力学**
A Steered Molecular Dynamics to Understand the Dissociation Process of Protein Complex
Yutaka Ueno, Yuki Mochizuki (*AIST Kansai, Biomedical Research*)

3Pos069 **脂質ナノディスクと ZMW 法を用いた高濃度リガンドでの計測が可能な膜タンパク 1 分子計測系の構築**
Nanodiscs platform on ZMWs for single-molecule imaging of membrane proteins at high ligand concentration
Keisuke Tsukada, Kazushi Isomura, Tomotaka Komori, Sotaro Uemura (*Dep. Bio. Sci., Grad. Sch. Sci., Univ. Tokyo*)

3Pos070 **X 線 1 分子追跡法による TRPV1 チャネルの分子運動解析**
3D Motion Maps of TRPV1 cation channel depicted by Diffracted X-Ray Tracking Method
Kazuhiro Mio[1], Keigo Ikezaki[2], Hiroshi Sekiguchi[3], Yuhuku Matsushita[2], Tai Kubo[1], Yuji C. Sasaki[2] ([1]*Molprof, AIST*, [2]*Frontier Science, Adv. Material Sci., Univ. of Tokyo*, [3]*JASRI*)

3Pos071 **細胞性粘菌の生きた細胞での膜タンパク質の拡散の網羅的解析**
Comprehensive Diffusion Analysis of Membrane Proteins in Living Dictyostelium Cells
Kazutoshi Takebayashi[1], Yukihiro Miyanaga[2], Masahiro Ueda[2,3] ([1]*Grad. Sch. Sci., Univ. Osaka*, [2]*Grad. Sch. FBS., Univ. Osaka*, [3]*QBiC., Riken*)

3Pos072 **細菌Ⅲ型分泌装置の回転運動によるエフェクター輸送の制御**
Rotation of needle-like type III secretion apparatus directly regulates its effector transport
Takashi Ohgita, Kohei Fukuda, Kyoko Momiyama, Naoki Hayashi, Naomasa Gotoh, Hiroyuki Saito (*Kyoto Pharm. Univ.*)

3Pos073 **抗体修飾ナノニードルと AFM を用いた引っ張り試験による細胞骨格の機械的特性の解析**
Analysis of mechanical property of cytoskeleton by tensile test for intermediate filament using antibody-modified nanoneedle and AFM
Moe Susaki[1], Itaru Takeda[2], Ayana Yamagishi[3], Yuta Takano[2], Tomoko Okada[3], Chikashi Nakamura[1,2,3] ([1]*Fac. Eng., Tokyo Univ. Agric. Technol.*, [2]*Grad. Sch. Eng., Tokyo Univ. Agric. Technol.*, [3]*Biomed. Res. Inst., AIST*)

3Pos074 **テンダム遺伝子を用いた無細胞タンパク質発現ノイズの解析**
Stochastic gene expression in cell-free system by tandem genes
Shiori Fujimoto, Yi Zhang, Kazuhito Tabata, Hiroyuki Noji (*Grad. Sch. Eng., Univ. Tokyo*)

ヘム蛋白質 / Heme proteins

3Pos075 **凍結トラップ結晶構造解析と時間分解分光を用いたP450norの反応中間体の解析**
Reaction Intermediate Analysis of P450nor Using Freeze-Trap X-ray Crystallography and Time-Resolved Spectroscopy
Takashi Nomura[1], Takuma Nishida[2], Takehiko Tosha[1], Hiroshi Sugimoto[1], Yoshitsugu Shiro[1,2], Minoru Kubo[1,3] ([1]*Harima Inst., Riken*, [2]*Grad. Sch. Sci., Univ. Hyogo*, [3]*JST PRESTO*)

3Pos076 **結晶状態ヘモグロビンの大規模四次構造変化の直接観測**
Direct observation of large-scale quaternary motions of hemoglobin in a crystalline state
Naoya Shibayama[1], Mio Ohki[2], Sam-Yong Park[2] ([1]*Jichi Med. Univ., Div. of Biophys.*, [2]*Yokohama City Univ., Drug Design Lab.*)

3Pos077 **構造状態と関係したヘモグロビンのピコ秒ダイナミクスの変化**
Changes in the picosecond dynamics of hemoglobin related to the structural states
Satoru Fujiwara[1], Toshiyuki Chatake[2], Tatsuhito Matsuo[1], Fumiaki Kono[1], Taiki Tominaga[3], Kaoru Shibata[4], Ayana Sato[5], Naoya Shibayama[5] ([1]*QuBS, QST*, [2]*RRI, Kyoto Univ.*, [3]*CROSS-Tokai*, [4]*J-PARC Center*, [5]*Div. Biophysics, Jichi Med. Univ.*)

3Pos078 **四量体ヒトヘモグロビンにおける2つのα鎖に特有のFe-His結合と四次構造との関連**
Distinct Fe-His bond of two α subunits in human $\alpha_2\beta_2$ tetramer hemoglobins and their quaternary structures
Shigenori Nagatomo[1], Kazuya Saito[1], Masako Nagai[2], Takashi Ogura[3], Teizo Kitagawa[3] ([1]*Dept. Chem., Univ. Tsukuba*, [2]*Res. Center Micro-Nano Tech., Hosei Univ.*, [3]*Grad. Sch. Life Sci., Univ. Hyogo*)

3Pos079 **神経保護作用を持つヒトニューログロビンとヘテロ三量体 G_i 蛋白質αサブユニットとの相互作用に重要なアミノ酸残基の特定**
Identification of residues crucial for the interaction between human neuroprotective protein “neuroglobin” and $G\alpha_i$
Nozomu Takahashi, Keisuke Wakasugi (*Dep. of Life Sci., Grad. Sch. of Arts and Sci., Univ. of Tokyo*)

3Pos080 **ドメインスワッピングによるミオグロビン二量体の形成**
Formation of myoglobin dimer by domain swapping
Satoshi Nagao[1], Ayaka Suda[1], Hisashi Kobayashi[1], Naoki Shibata[2], Yoshiki Higuchi[2], Shun Hirota[1] ([1]*Graduate School of Materials Science, Nara Institute of Science and Technology*, [2]*Graduate School of Life Science, University of Hyogo*)

3Pos082 **共鳴ラマン分光法による2価コバラミンの軸配位子に依存した構造変化の検出**
Resonance Raman Study of Cobalamin (II): Axial Ligands-Dependent Structural Change
Kaoru Mieda[1], Abdullah Al Mamun[2], Pawel M. Kozlowski[2], Takashi Ogura[1] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Dept. of Chem., Univ. of Louisville*)

3Pos083 **タンパク質中のヘムの歪みの統計的解析**
Statistical analysis of heme distortion in protein
Yasuhiro Imada[1], **Yusuke Kanematsu**[2], Hiroko X. Kondo[2], Yu Takano[2] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Info. Sci., Hiroshima City Univ.*)

3Pos084 **アンサンブルドッキングを用いたCYP1A2化合物の代謝部位予測**
Prediction of site of metabolism of compounds for CYP1A2 by ensemble docking simulation
Hiroaki Saito[1], Taku Mizukami[2], Yoshinori Hirano[1], Takao Otsuka[1], Noriaki Okimoto[1], Makoto Taiji[1] ([1]*RIKEN Quantitative Biology Center (QBiC)*, [2]*Japan Institute of Science and Technology (JAIST)*)

膜蛋白質 / Membrane proteins

3Pos085 **本来の構造と機能を保持したウシミトコンドリア呼吸鎖複合体の精製**
Purification of native mitochondrial respiratory complexes from bovine heart
Satoru Shimada, Shigefumi Uene, Marika Oosaki, Ryoko Takahashi, Harunobu Shimomura, Kaoru Mieda, Shintaro Maeda, Masahide Hikita, **Kyoko Shinzawa-Itoh** (*Grad. Sch. Life Sci., Univ. Hyogo*)

3Pos086 **部分フッ素化リン脂質二分子膜中膜タンパク質バクテリオロドプシンの構造と安定性に対するフッ化アルキル鎖長依存性**
Structural stability of bacteriorhodopsin in partially fluorinated analogs of DMPC with different perfluoroalkyl chain lengths
Mami Hashimoto[1], Yuka Murai[1], Masaru Yoshino[1], Toshinori Motegi[1], Takashi Kikukawa[2], Toshiyuki Takagi[3], Hiroshi Takahashi[1], Hideki Amii[1], Toshiyuki Kanamori[3], Masashi Sonoyama[1] ([1]*Div. Mol. Sci., Gunma Univ.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*, [3]*AIST*)

3Pos087 **高圧下で界面活性剤を用いて昆虫細胞膜から可溶化したPBANR（クラス-A GPCR）はリガンド結合能を保持する**
PBANR, a class-A GPCR, solubilized under high hydrostatic pressure retains its ligand binding ability
Yukie Katayama[1], Tatsuya Suzuki[1], Tatsuki Ebisawa[1], Jun Ohtsuka[1], Ryo Natsume[2], Yu-Hua Lo[3], Toshiya Senda[3], Toshihiro Nagamine[4], J.Joe Hull[5], Shogo Matsumoto[4], Hiromichi Nagasawa[1,6], Koji Nagata[1], Masaru Tanokura[1] ([1]*UTokyo*, [2]*TDU*, [3]*KEK-PF*, [4]*RIKEN*, [5]*USDA-ARS*, [6]*ZJU*)

3Pos088 **GraDeR：単粒子解析等の膜タンパク質資料調整**
GraDeR: membrane protein preparation for single particle cryoEM & more
Christoph Gerle[1,5], Florian Hauer[4], Niels Fischer[4], Kyoko Shinzawa-Itoh[1], Satoru Shimada[1,5], Ken Yokoyama[2], Atsunori Oshima[3], Yoshinori Fujiyoshi[3], Holger Stark[4] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Kyoto Sangyo Univ.*, [3]*CeSPI, Nagoya Univ.*, [4]*Max Planck Society*, [5]*CREST, JST*)

3Pos089 **エーテル型部分フッ素化リン脂質膜中の膜タンパク質バクテリオロドプシンのサーモクロミズム**
Thermochromism of bacteriorhodopsin in partially fluorinated di-o-tetradecylphosphocholine vesicles
Masaya Miyazaki[1], Naoyuki Tuchida[1], Toshinori Motegi[1], Takashi Kikukawa[2], Toshiyuki Takagi[3], Hiroshi Takahashi[1], Hideki Amii[1], Toshiyuki Kanamori[3], Masashi Sonoyama[1] ([1]*Fac. Sci. Tech., Gunma. Univ.*, [2]*Grad. Sch. Sci., Hokkaido. Univ.*, [3]*AIST*)

3Pos090 **GXXXGモチーフによる膜貫通ヘリックスの二量体形成：会合トポロジー制御下での一分子FRET研究**
GXXXG-mediated dimerization of transmembrane helices: single-molecule FRET detection with controlled association topology
Yoshiaki Yano, Yuta Watanabe, Katsumi Matsuzaki (*Grad. Sch. Pharm Sci., Kyoto Univ.*)

3Pos091 **脂質膜分子による上皮成長因子受容体の膜近傍ドメイン構造制御機構**
Conformational regulation of the juxtamembrane domain of epidermal growth factor receptor by membrane lipid molecules
Ryo Maeda[1], Takeshi Sato[2], Yasushi Sako[1] ([1]*Cellular Informatics Lab., RIKEN*, [2]*Inst. for Protein Research, Osaka Univ.*)

3Pos092 **ESR による銅ポンプ P 型 ATPase における 金属イオン配位子の動的構造：ATP 効果の研究**
Structural dynamics in metal ion coordination of copper pump P-type ATPase as studied by EPR spectroscopy: Effect of ATP
Satoshi Yasuda[1,2], Naoyuki Kuwabara[3], Shoji Ueki[4], **Toshiaki Arata**[1,5] ([1]*Dept. Biol. Sci., Grad. Sch. Sci. Osaka Univ.*, [2]*Asahikawa Med. Univ.*, [3]*PF, KEK*, [4]*Tokushima-Bunri Univ.*, [5]*Ctr. Adv. High Mag. Field Sci., Grad. Sci. Osaka Univ.*)

3Pos093 **高速 AFM による ABC タンパク質の動態観察**
High-speed atomic force microscopy shows conformational changes of nucleic binding domains of ABC protein
Shohei Takigaura[1], Mikihiro Shibata[1,2], Kazuaki Yamahara[3], Yasuhisa Kimura[3], Kazumitsu Ueda[3,4], Takayuki Uchihashi[1,2], Toshio Ando[2] ([1]*Dep. Phys. Kanazawa Univ.*, [2]*Bio-AFM FRC*, [3]*Div. App. Lif. Sci. Kyoto Univ.*, [4]*iCeMs*)

3Pos094 **高速原子間力顕微鏡による電位依存性プロトンチャネルの直接観察**
Direct observation of voltage-gated proton channels by high speed AFM
Hayato Yamashita[1,2], Akira Kawanabe[3,4], Yasushi Okamura[3,4], Masayuki Abe[1] ([1]*Grad. Sch. of Eng. Sci., Osaka Univ.*, [2]*PRESTO, JST*, [3]*Grad. Sch. of med., Osaka Univ.*, [4]*CREST, JST*)

3Pos095 **多剤輸送担体 EmrE の基質結合エントロピー利得に対する水分子の寄与**
Contribution of water molecules for the gain in the substrate binding entropy to multidrug resistance transporter, EmrE
Kazumi Shimono[1], Keisuke Matsuda[1], Shoko Suzuki[1], Kaho Yajima[1], Sakiyo Yamamoto[1], Tomomi Kimura-Someya[2,3], Mikako Shirouzu[2,3], Shigeyuki Yokoyama[2,4], Seiji Miyauchi[1] ([1]*Fac. Pharm. Sci., Toho Univ.*, [2]*RIKEN SSBC*, [3]*RIKEN CLST*, [4]*RIKEN Struct. Biol. Lab.*)

3Pos096 **cd1NiR:cNOR 複合体構造を安定化する相互作用の理論解析**
Theoretical analysis of interaction that stabilizes cd1NiR:cNOR complex structure
Kenta Yamada[1], Takaharu Mori[1], Kiyoshi Yagi[1], Takehiko Tosha[2], Yoshitsugu Shiro[2], Yuji Sugita[1,3,4,5] ([1]*RIKEN TMS*, [2]*RIKEN BSL*, [3]*RIKEN AICS*, [4]*RIKEN QBiC*, [5]*RIKEN iTHES*)

3Pos097 **3 次元立体構造が不明の膜タンパク質に対する耐熱化置換体の特定**
Identification of Thermostabilizing Mutations for a Membrane Protein Whose Three-Dimensional Structure is Unknown
Yuta Kajiwara[1], Satoshi Yasuda[2,3], Yuuki Takamuku[2], Takeshi Murata[2,3,5], Masahiro Kinoshita[4] ([1]*Graduate School of Energy Science, Kyoto University*, [2]*Graduate School of Science, Chiba University*, [3]*Molecular Chirality Research Center, Chiba University*, [4]*Institute of Advanced Energy, Kyoto University*, [5]*JST, PRESTO*)

3Pos098 **In silico screening of novel stress response factors regulated by mitochondrial inner membrane proteases**
Kenichiro Imai, Yoshinori Fukasawa, Kentaro Tomii, Paul Horton (*AIRC, AIST*)

核酸：相互作用・複合体形成 / Nucleic acid: Interaction & Complex formation

3Pos099 **MD シミュレーションを用いた RNA 二重らせん構造の熱安定性予測**
Predicting RNA Duplex Dimerization Free-Energy Changes upon Mutations Using Molecular Dynamics Simulations
Shun Sakuraba[1], Kiyoshi Asai[1,2], **Tomoshi Kameda**[2] ([1]*Grad. Sch. Frontier Sci., Tokyo Univ.*, [2]*AI center, AIST*)

3Pos100 **単層および多層カーボンナノチューブ上における蛍光 DNA の蛍光強度の塩基配列依存性**
Base sequence dependence for fluorescence intensity of fluorescent dye-labeled DNA on single- and multi-walled carbon nanotubes
Shusuke Oura, Kazuo Umemura (*Tokyo Univ. of Sci.*)

3Pos101 **メタダイナミクスとアルケミカル変換法を用いた定量的結合活性予測**
Approach to the quantitative prediction of the binding affinity using metadynamics and alchemical transformation
Yoshiaki Tanida, Azuma Matsuura (*FUJITSU LABORATORIES LTD.*)

3Pos102 **紫外線損傷 DNA における Flippling 機構**
On the Flipping-out mechanism of the UV-induced DNA damage
Ryuma Sato, Ryuhei Harada, Yasuteru Shigeta (*Center of comp. Sci., Univ. Tsukuba*)

3Pos103 **Comparison of Multi-Dyes Quenching by Single-Walled Carbon Nanotube Dispersion with Single Stranded DNA**
Ying Tan, Katsuki Izumi, Kazuo Umemura (*Tokyo University of Science*)

電子状態 / Electronic state

3Pos104 **サイトキニン脱水素酵素における独特なフラビン-基質配置に関する量子化学的研究**
Quantum chemical study on unusual flavin-substrate alignment in cytokinin dehydrogenase
Kyosuke Sato (*Dept. Mol. Physiol., Facult. Life Sci., Kumamoto Univ.*)

3Pos105 **生体分子の電子状態解析のための大規模第一原理 DFT 計算手法の開発**
Large-scale DFT calculation method for electronic-structure analysis of biomolecules
Ayako Nakata[1], Takao Ostuka[2], David R. Bowler[3], Tsuyoshi Miyazaki[1] ([1]*NIMS*, [2]*RIKEN*, [3]*UCL*)

3Pos106 **生体分子系における定温オーダーＮ法第一原理分子動力学計算**
Constant temperature order-N first-principles molecular dynamics calculations of biomolecular system and short-time behavior
Takao Otsuka[1], Makoto Taiji[1], David R. Bowler[2], Tsuyoshi Miyazaki[3] ([1]*RIKEN QBiC*, [2]*UCL*, [3]*NIMS*)

水·水和·電解質 / Water & Hydration & Electrolyte

3Pos107 **両親媒性抽出剤を用いた水相から有機相へのリン酸化合物の抽出**
Extraction of phosphoric compounds from aqueous phase into an organic phase with an amphiphilic extractant
Hideyuki Komatsu (*Bioinfo. & Biosci., Kyushu Inst. Tech.*)

3Pos108 **リン酸イオンの水和エネルギー空間分割解析**
Spatial-Decomposition Analysis of Hydration Energy of Phosphate Ions
George Mogami[1], Nobuyuki Matubayasi[2], Makoto Suzuki[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Eng. Sci., Osaka Univ.*)

3Pos109 **実効相互作用を用いた電解質中のマクロアニオンの分子シミュレーション**
Molecular simulation of macroanions in an electrolyte solution based on the effective potential
Ayumi Suematsu, **Ryo Akiyama** (*Dept. Chem., Kyushu Univ.*)

3Pos110 **MM/3D-RISM 法を用いたシクロデキストリン誘導体とロクロニウム臭化物の結合自由エネルギーの予測**
Estimation of binding free energies for inclusion processes of Rocuronium bromide by cyclodextrin derivatives using MM/3D-RISM method
Yuji Hayashino[1], Masatake Sugita[1], Fumio Hirata[2], Takeshi Kikuchi[1] ([1]*Dept. of Bioinfo., Col. Life Sci., Ritsumeikan Univ.*, [2]*Toyota Phys. & Chem. Res. Inst.*)

3Pos111 **ボルンエネルギーとクーロンエネルギー間のバランスの物理的理解と計算指針**
Physical understanding and computational guideline for the balance between Born and Coulomb energies
Dan Parkin, Yukinobu Mizuhara, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

分子遺伝学·遺伝子発現 / Molecular genetics & Gene expression

3Pos112 **何故無細胞タンパク質合成系においてはそんなに多くのリボソームが必要なのか？**
Why we need so many ribosomes in cell-free protein synthesis?
Yue Xu, Yi Zhang, Hiroyuki Noji (*Grad. Sch. Eng., Univ. Tokyo*)

3Pos113 **一細胞トランスクリプトーム解析へ向けた、PCR を含まないライブラリー調製**
Amplification-free library construction for single-cell transcriptome analysis
Tetsuo Fujinami, Yusuke Oguchi, Mai Yamagishi, Yoshitaka Shirasaki, Sotaro Uemura (*Grad. Sch. Sci. Univ. Tokyo*)

3Pos114 **無細胞タンパク質合成系に向けた最良 T7 プロモーター配列の探索**
Improvement of T7 promoter sequence for cell-free protein synthesis
Tomoya Nishimura[1], Yi Zhang[2], Hiroyuki Noji[2] ([1]*Undergrad. Sch. Eng., Univ. Tokyo*, [2]*Sch. Eng., Univ. Tokyo*)

3Pos115 **細胞集積度によるパラクラインシグナリングの制御により遺伝子発現の安定性が変化した**
Paracrine signaling modulated by the accumulation of cells altered the stability of gene expression
Mai Yamagishi[1,3], Yoshitaka Shirasaki[1,3], Yutaka Hori[2], Nobutake Suzuki[1], Osamu Ohara[3], Sotaro Uemura[1] ([1]*Grad. Sci. Sci., The Univ. of Tokyo*, [2]*Fac. Sci. Technol., Keio Univ.*, [3]*IMS, RIKEN*)

3Pos116 **Causal role of DNA methylation ?: A computational model**
Ashwin S.S, Masaki Sasai (*Dept. of Computational Sciences and Engineering & Dept. of Applied Physics, Nagoya Univ., Nagoya*)

3Pos117 **An in silico Approach to Investigating Gene Variants of Unknown Significance in a Clinical Context**
Stefanie S. Portelli[1], Elizabeth N. Robertson[1,2], Yaxin Lu[1], Murat Kekic[1], Brett D. Hambly[1], Richmond Jeremy[1,2,3] ([1]*Univ. Sydney*, [2]*Royal Prince Alfred Hospital*, [3]*Baird Institute*)

3Pos118 **遺伝子発現におけるポリアミンの DNA 高次構造との関係性**
Relationship between DNA higher order structure and Gene-Expression with Polyamines
Ai Kanemura[1], Yuko Yoshikawa[1], Takahiro Kenmotsu[1], Wakao Hukuda[2], Kenichi Yoshikawa[1] ([1]*Grad. Sch. Life Med. Sci., Univ. Doshisha*, [2]*Coll. Life Sci., Univ. Ritsumeikan*)

3Pos119 ***Guillardia theta*** **におけるロドプシン様遺伝子群の発現解析**
Expression analysis of microbial rhodopsin-like genes in *Guillardia theta*
Masae Konno[1,3], Keiichi Inoue[1,2], Hideki Kandori[1,3] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*PRESTO, JST*, [3]*OPTRC, Nagoya Inst. Tech.*)

筋肉 / Muscle

3Pos121 **細胞性粘菌の Tyr143 変異アクチンのカルボキシ末端領域にある Phe352、Met355 と Trp356 の側鎖の二形性**
Dimorphism of the side-chains of Phe352, Met355, and Trp356 in the carboxyl-terminal region of Dictyostelium actin mutants
Yuki Gomibuchi[1], Taro Q.P. Uyeda[2], Takeyuki Wakabayashi[1] ([1]*Teikyo Univ.*, [2]*Waseda Univ.*)

3Pos122 **筋収縮制御メカニズムの解明を目指した細いフィラメントの立体構造解析**
CryoEM structural analysis of muscle thin filament composed of actin filament, tropomyosin and troponin
Yurika Yamada[1], Keichi Namba[1,2], Takashi Fujii[1] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*RIKEN QBiC*)

3Pos123 **中性子準弾性散乱により明らかとなった心筋症原因変異がもたらすトロポニンの動力学異常**
Effects of a cardiomyopathy-causing mutation on the internal dynamics of troponin revealed by quasielastic neutron scattering
Tatsuhito Matsuo[1], Taiki Tominaga[2], Kaoru Shibata[3], Satoru Fujiwara[1] ([1]*QST/J-PARC*, [2]*CROSS-Tokai*, [3]*J-PARC*)

3Pos124 **ウニのコネクチン様タンパク質の構造解析**
Sequential analysis of connectin-like protein in sea urchin
Sumiko Kimura[1], Akira Hanashima[2], Maki Yamaguchi[1], Toshiko Yamazawa[1], Tetsuo Ohno[1], Naoya Nakahara[1], Mika Taguchi[1], Shigeru Takemori[1] ([1]*Dept. Mol. Physiol., Jikei Univ. Sch. Med.*, [2]*First Dept. Physiol., Kawasaki Med. Sch.*)

3Pos125 **Mg ポリマー再考**
Revisiting "Mg-Polymer"
Mahito Kikumoto, Shuuichi Takeda, Yuichiro Maeda (*Structural Biology Research Center, Nagoya-Univ.*)

3Pos126 **細いフィラメント上のトロポミオシンのモデル：スピンラベル ESR 距離マップ**
Modeling for tropomyosin position in the thin filament by distance measurements using spin-labeling dipolar EPR spectroscopy
Keisuke Ueda[1,2], Yoshiki Tsujimoto[2], Hiroaki Yamashita[2], Kouichi Sakai[2], Shoji Ueki[4], Masao Miki[3], Toshiaki Arata[2,5] ([1]*CLIST, Riken-Yokohama*, [2]*Dept. Biol. Sci. Grad. Sch. Osaka Univ.*, [3]*Univ. Fukui*, [4]*Tokushima-Bunri Univ.*, [5]*Ctr. Adv. High Mag. Field Sci., Grad. Sci. Osaka Univ.*)

3Pos127 **筋原線維懸濁液の ATP 分解素過程中のプロトン NMR 緩和経過**
Spin-spin relaxation of 1H NMR signals from myofibril suspension during cross-bridge cycling
Tetsuo Ohno, Hitomi Sano (*Dept. Physiol., The Jikei Univ. Sch. Med.*)

3Pos128 **骨格筋タンパク質と水の相互作用を融点から探る**
Interaction between water and myoproteins revealed by melting points
Naoya Nakahara[1], Tetsuo Ohno[1], Masako Kimura[2], Sumiko Kimura[1], Shigeru Takemori[1] ([1]*Jikei Univ. Sch. Med.*, [2]*Kagawa Nutri. Univ.*)

3Pos129 **T-plastin の 2 つのアクチン結合ドメインとアクチンフィラメントとの結合性の比較**
Comparison of binding affinities of two actin-binding domains of T-plastin to actin filament
Taiki Hirate, Atsusi Ooi, Tsuyoshi Okagaki (*Dept., Bioresources, Mie Univ.*)

3Pos130 **心筋細胞集団の伝搬のゆらぎの局所・全体相関の解明のためのオンチップ心筋細胞ネットワーク解析技術の開発**
Development of On-chip Cardiomyocyte Network Analysis Assay for Understanding of Fluctuation Correlation in Cell-to-cell Conduction
Naoki Takahashi[1], Hideyuki Terazono[2], Masao Odaka[2], Kenji Matsuura[2], Akihiro Hattori[2], Kenji Yasuda[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*WASEDA Biosci. Res. Inst. Singapore(WABIOS), Waseda Univ.*)

3Pos131 **肥大型心筋症特異的なトロポミオシン変異体(V95A,D175N)のアクトミオシン収縮速度・収縮力への異なる影響**
Tropomyosin's HCM mutants (V95A, D175N) differently affect the actomyosin sliding velocity and force
Shuya Ishii[1], Shin'ichi Ishiwata[2], Masataka Kawai[3] ([1]*Sch. Adv. Sci. Eng., Waseda Univ.*, [2]*Fac. Sci. Engn., Waseda Univ.*, [3]*Coll. Med., Univ. Iowa*)

3Pos132 **温めた心筋細胞に備わった収縮振動は遅い Ca^{2+}変動に対して周期を一定に保つ**
Contractive oscillations intrinsic to heating cardiomyocytes maintain the period against late Ca^{2+} variations
Seine A. Shintani[1,2], Takumi Washio[3], Hideo Higuchi[1] ([1]*Dept. Physics, Univ. Tokyo*, [2]*JSPS Research Fellow*, [3]*Dept. Human and Engineered Enviromental, Univ. Tokyo*)

3Pos133 **周期性伸展刺激における伸展周期と心筋細胞の応答の関係**
Relation between stretch cycles and response of cardiomyocytes in cyclic stretch stimulation
Chiho Nihei, Tomoyuki Kaneko (*LaRC, Grad. Sci. Eng., Hosei Univ.*)

3Pos134 **Mg^{2+}/Ca^{2+} 交換及び温度変化による F-アクチンの三次構造変化**
Tertiary structure of F-actin affected by Mg^{2+}/Ca^{2+} and temperature
Takaya Yamaguchi, George Mogami, Makoto Suzuki (*Grad. Sch. Eng., Univ. Tohoku*)

分子モーター / Molecular motor

3Pos135 **Does homo hexamer function as a stator of rotary motor?**
Aiko Endo[1], Junichi Kishikawa[2], Ken Yokoyama[2] ([1]*Grad. Sch. Biochem., Kyoto sangyo Univ.*, [2]*Dept. Mol. Biosci., Kyoto sangyo Univ.*)

3Pos136 **全原子 MD と粗視化 MD を組み合わせたマルチスケール MD 解析による V1-ATPase の回転機構の解明**
Rotation mechanism of V1-ATPase elucidated by multi-scale MD analysis
Yuta Isaka[1], Toru Ekimoto[1], Yuichi Kokabu[1], Takeshi Murata[2,3] ([1]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*, [2]*Fac. of Sci., Chiba Univ.*, [3]*JST, PRESTO*)

3Pos137 **Probing the biophysical properties of a Thermoalkaliphilic F1 ATPase gives insight into adaptation and regulation**
Duncan G. G. McMillan[1], Rikiya Watanabe[1], Hiroshi Ueno[1], Gregory M. Cook[2], Hiroyuki Noji[1] ([1]*Dept. of Applied Chemistry, The Univ. of Tokyo*, [2]*Dept. of Microbiology and Immunology, Univ. of Otago, Dunedin, New Zealand*)

3Pos138 **高速 AFM で明らかにする回転軸の無い腸内連鎖球菌由来 V_1-ATPase の一方向的協同性度合**
The Extent of Unidirectional Cooperativity in Rotorless *Enterococcus hirae* V_1-ATPase Revealed by High-speed AFM
Motonori Imamura[1], Kazuya Nakamoto[2], Shintaro Maruyama[2], Fumihiro Kawai[3], Ryota Iino[3], Takayuki Uchihashi[1,4,5], Takeshi Murata[2,6], Toshio Ando[1,4,5] ([1]*Bio-AFM FRC*, [2]*Grad. Sch. Sci., Chiba Univ.*, [3]*Okazaki Inst. Integ. BioSci., IMS, NINS*, [4]*Dept. Phys., Kanazawa Univ.*, [5]*CREST, JST*, [6]*PRESTO, JST*)

3Pos139 **リン酸結合蛋白を封入したフェムトリットル体積のドロップレットアレイによる無機リン酸検出**
Detection of inorganic phosphate by phosphate binding protein encapsulated in femtoliter droplet arrays
Masayuki Higuchi[1], Kazuhito V. Tabata[2,4], Hiroyuki Noji[2], Tomoko Masaike[1,3,4] ([1]*Dept. Appl. Biol. Sci., Tokyo Univ. of Sci.*, [2]*Dept. Appl. Chem., Sch. of Eng., Univ. of Tokyo*, [3]*Res. Inst. for Sci. and Tech., Tokyo Univ. of Sci.*, [4]*PRESTO, JST*)

3Pos140 **Unveiling the chemomechanical coupling of F1 ATPase of Paracoccus denitrificans**

Mariel Zarco - Zavala[1], Duncan G G McMillan[1], Toshiharu Suzuki[1], Hiroshi Ueno[1], Francisco Mendoza-Hoffmann[2], Jose J. Garcia-Trejo[2], Hiroyuki Noji[1] ([1]*Department of Applied Chemistry, Graduate School of Engineering, The University of Tokyo*, [2]*Depbest of Biology, Chemistry Faculty, National Autonomous University of Mexico*)

3Pos141 **腸球菌由来 ADP 結合型 V_1-ATPase の X 線結晶構造解析**

Crystal structures of the ADP-bound V_1-ATPase from *Enterococcus hirae*

Kano Suzuki[1], Kenji Mizutani[1,2,3], Shintaro Maruyama[1], Kazumi Shimono[4], Fabiana L. Yakushiji[1], Eiro Muneyuki[5], Yoshimi Kakinuma[6], Yoshiko Ishizuka-Katsura[7], Mikako Shirouzu[7], Shigeyuki Yokoyama[8], Ichiro Yamato[3], Takeshi Murata[1,2,7,9] ([1]*Chiba Univ.*, [2]*Mol. Chirality Res. Center, Chiba Univ.*, [3]*Dept. Biol. Sei. Tech., Tokyo Univ. of Science*, [4]*Faculty of Pharm. Sci., Toho Univ.*, [5]*Faculty of Sci. and Eng., Chuo Univ.*, [6]*Faculty of Agri., Ehime Univ.*, [7]*DSSB, RIKEN*, [8]*Struct. Biol. Lab., RIKEN*, [9]*JST, PRESTO*)

3Pos142 ***Enterococcus hirae* 由来 V_1-ATPase アルギニンフィンガー変異体が示す特異な回転特性**

Arginine finger mutant of *Enterococcus hirae* V_1-ATPase shows unusual rotational behaviors

Tatsuya Iida[1], Yoshihiro Minagawa[2], Hiroshi Ueno[2], Takeshi Murata[3], Ryota Iino[1,4,5] ([1]*SOKENDAI (The Grad. Univ. for Adv. Stud.)*, [2]*The Univ. Tokyo*, [3]*Chiba Univ.*, [4]*Inst. for Mol. Sci.*, [5]*Okazaki Inst. for Integr. Biosci.*)

3Pos143 **Structural analysis by NMR on C-terminal region of FliG, an essential motor component of Vibrio Na^+-driven flagella**

Yohei Miyanoiri[1], Yuuki Nishino[2], Mizuki Gohara[2], Atsushi Hijikata[3], Yasuhiro Onoue[2], Seiji Kojima[2], Tsuyoshi Shirai[3], Masatsune Kainosho[1], Michio Homma[2] ([1]*SBRC. Grad. Sch. Sci., Nagoya Univ.*, [2]*Biosci. Grad. Sch. Sci., Nagoya Univ.*, [3]*Biosci. Nagahama Inst. Biosci. Tech.*)

3Pos144 **サルモネラ属菌 FliFG 融合変異型べん毛モーターの動態機能計測**

Functional analysis of a FliFG deletion-fusion mutant flagellar motor

Tomofumi Sakai[1], Koichiro Mori[1], Yumi Inoue[1], Tomoko Miyata[1], Naoya Terahara[1], Yusuke Morimoto[2], Takayuki Kato[1], Keiichi Namba[1,2], Tohru Minamino[1] ([1]*Grad. Sch. Frontier BioSci., Osaka Univ.*, [2]*RIKEN QBiC*)

3Pos145 **光ピンセットを用いたバクテリアべん毛モーターの最大トルクの計測**

Maximum torque generated by the bacterial flagellar motor measured by optical tweezers

Taishi Kasai[1], Yoshiyuki Sowa[1,2] ([1]*Reserch center for Micro-Nano Tech. Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*)

3Pos146 **べん毛モーターのトルク特性とその個体差の精密測定**

Precise measurement of torque characteristics and individual variability of bacterial flagellar motor

Kento Sato, Shuichi Nakamura, Seishi Kudo, Shoichi Toyabe (*Grad. Sch. Eng., Tohoku Univ.*)

3Pos147 **The mechanism of Vibrio alginolyticus polar flagellum growth**

Chien-Jung Lo, Meiting Chen (*Department of Physics, National Central University*)

3Pos148 **Functional Analysis of Slow-Motile Mutations in Flagellar Stator MotA/B of Salmonella**

Seyedehnoorolhoda Shajaripourjaberi, Naoya Terhara, Tohru Minamino (*frontier biosciences*)

3Pos149 **Rng2 による F-アクチンとミオシン間の協同的相互作用の制御**

Regulation of cooperative interaction between myosin and F-actin by Rng2

Taiga Imai[1], Masak Takaine[2], Kentaro Nakano[2], Osamu Numata[2], Taro Uyeda[3], Kiyotaka Tokuraku[1] ([1]*Muroran institute of technology*, [2]*University of Tsukuba*, [3]*Waseda University*)

3Pos150 **SH1 ヘリックス内に変異をもつミオシン II はアクチンフィラメントの滑りの活性化エネルギーを減少させる**

Myosin II SH1 helix mutant lowers the activation energy for sliding of Factin

Shigeru Chaen[1], Kotomi Shibata[1], Tsubasa Koyama[1], Atsushi Suenaga[1], Sosuke Iwai[2] ([1]*Dept. Biosci. Nihon Univ.*, [2]*Dept. Biol. Hirosaki Univ.*)

3Pos151 **プログラム可能なミオシンアセンブリの設計と高解像 1 分子イメージング**

Design of a programmable myosin motor assembly and nanometer-precision single-molecule imaging

Masashi Ohmachi[1], Keisuke Fujita[1], Keigo Ikezaki[2], Toshio Yanagida[1,3], **Mitsuhiro Iwaki**[1,3] ([1]*QBiC, RIKEN*, [2]*Univ. of Tokyo*, [3]*Osaka Univ.*)

3Pos152 **単純化された筋繊維の計算モデルを用いた筋収縮におけるバイアスブラウン運動の寄与の研究**

Contribution of biased Brownian motion in muscle contraction studied by a simplified computational model of muscle fiber

Daisuke Watanabe, Masaki Sasai, Tomoki P. Terada (*Dept. Comput. Sci. Eng., Grad. Sch. Eng., Nagoya Univ.*)

3Pos153 **Rng2 によるアクチンフィラメントの構造変化と、HMM で駆動されるアクチン運動の協同的阻害**

Structural changes of actin filaments induced by Rng2, and the resultant inhibition of actin movement on HMM

Yuki Hayakawa[1], Keiko Hirose[2], Masafumi Yamada[2], Kien X. Ngo[1], Noriyuki Kodera[4], Masak Takaine[3], Kentaro Nakano[3], Osamu Numata[3], Taro Uyeda[1,2] ([1]*dep physics, Waseda Univ.*, [2]*Biomed Res Inst, AIST*, [3]*Grad. School of Life and Environ Sci., Univ. Tsukuba*, [4]*Bio-AFM Res Ctr, Kanazawa Univ.*)

3Pos154 **Effect of external mechanical stress on collective motion of microtubules**

Tamanna Ishrat Farhana[1], Arif Md. Rashedul Kabir[2], Daisuke Inoue[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. Chem. Sci. & Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*)

3Pos155 **微小管内 GDP-チューブリンの精密な周期決定**

Determination of accurate axial tubulin repeat in GDP-microtubules

Shinji Kamimura[1], Hiroshi Imai[1], Toshiki Yagi[2], Tomohiro Shima[3,4], Yasushi Okada[4,5], Hiroyuki Iwamoto[6] ([1]*Dept. Biol. Sci., Chuo University*, [2]*Dept. Life Sci., Pref. Univ. Hiroshima*, [3]*Dept. Biol. Sci., Univ. Tokyo*, [4]*Lab. Cell Polarity Regulation., Qbic, RIKEN*, [5]*Dept. Phys., Univ. Tokyo*, [6]*Life & Environmental Div., SPring-8*)

3Pos156 高精度な微小管分離に向けた微小管の持続長設計

Design of Microtubule Persistence Length Toward High-precision Microtubule Sorting

Naoto Isozaki[1], Hirofumi Shintaku[1], Hidetoshi Kotera[1], Taviare L. Hawkins[2], Jennifer L. Ross[3], Ryuji Yokokawa[1] ([1]*Kyoto University*, [2]*University of Wisconsin - La Crosse*, [3]*University of Massachusetts - Amherst*)

3Pos157 Regulated swarming of molecular robots prepared from a DNA programmed biomolecular motor system

Jakia Jannat Keya[1], Ryuhei Suzuki[1], Arif Md. Rashedul Kabir[2], Daisuke Inoue[2], Kazuki Sada[1,2], Akinori Kuzuya[3], Akira Kakugo[1,2] ([1]*Graduate School of Chemical Sciences and Engineering, Hokkaido University*, [2]*Faculty of Science, Hokkaido University*, [3]*Faculty of Chemistry and Bioengineering, Kansai University*)

3Pos158 構成論的手法を用いた鞭毛の機能的再構築

Functional reconstitution of flagellar axonemes by self-organization of microtubules, dynein docking complex and outer arm dyneins

Misaki Shiraga[1,2], Jyunya Kirima[1,2], Hiroaki Kojima[1], Kazuhiro Ooiwa[1,2] ([1]*Adv. ICT Res. Inst. NICT*, [2]*Grad. Sch. Sci., Univ. Hyogo*)

3Pos159 電子顕微鏡トモグラフィーによって明らかになった細胞質ダイニンのモータードメインの配置

Orientation of two motor domains of cytoplasmic dynein characterized using electron computed tomography

Kotaro Koyasako[1], Shiori Toba[2], Shinji Hirotsune[2], Takuo Yasunaga[1] ([1]*Department of Bioscience and Bioinformatics, Faculty of Computer Science and Systems Engineering, Kyushu Institute of Technology*, [2]*Department of Genetic Disease Research, Osaka City University Graduate School of Medicine*)

3Pos160 ポリグルタミン酸化酵素欠損マウスの気管繊毛が示す異常な跳ね上がり運動

Abnormal "hopping" of mouse tracheal cilia deficient in tubulin polyglutamylation

Masayuki Shiina[1], Toshihito Iwase[1], Masaaki Suegara[1], Haruka Kanno[1], Takanobu A Katoh[2], Mitsutoshi Setou[3], Takayuki Nishizaka[2], Koji Ikegami[3], Tomoko Masaike[1,4] ([1]*Dept. Appl. Biol. Sci., Tokyo Univ. of Sci.*, [2]*Dept. Phys., Gakushuin Univ.*, [3]*Hamamatsu Univ. Sch. Med.*, [4]*PRESTO, JST*)

3Pos161 クラミドモナス鞭毛の波形変化に関わるタンパク質の探索

Searching a putative protein responsible for switching waveform of Chlamydomonas flagella

Junya Kirima[1], Misaki Shiraga[1], Hiroaki Kojima[2], Kazuhiro Oiwa[1,2] ([1]*Grad. Sch. Life Sci., Univ. Hyogo*, [2]*Adv. ICT Res. Inst., NICT*)

3Pos162 バクテリア・カビ由来セロビオヒドロラーゼの結合、解離、プロセッシブ運動の一分子蛍光観察

Single-molecule fluorescence analysis of binding, dissociation, and processive movement of bacterial and fungal cellobiohydrolases

Daiki Ishiwata[1], Akihiko Nakamura[1,2], Tomoyuki Tasaki[3], Akasit Visootsat[4], Maximilien Morice[5], Ryota Iino[1,2,6] ([1]*Sch. phys. sci., SOKENDAI (The Graduate University for Advanced Studies)*, [2]*Okazaki Inst. for Integr. Biosci.*, [3]*Sch. of Engi., Univ. Tokyo*, [4]*Fac. Sci., Univ. Kasetsart*, [5]*Chimie ParisTech.*, [6]*Inst. for Mol. Sci.,*)

3Pos163 分子モーター RecBCD による混雑環境下でのタンパク質-DNA 複合体の除去

Sequential eviction of crowded nucleoprotein complexes by the RecBCD molecular motor

Tsuyoshi Terakawa[1], Redding Sy[1,2], Silverstein Timothy[1], Greene Eric[1] ([1]*Columbia Univ.*, [2]*Univ. of California*)

3Pos164 RHAU ヘリカーゼがグアニン４重鎖構造の安定性を調節するメカニズムの一分子研究

RHAU helicases regulate G4 stability during the ATPase cycle revealed from single-molecule analysis

Huijuan You[1], Jie Yan[1,2] ([1]*MBI, National University of Singapore*, [2]*Dep. of Physics, National University of Singapore*)

3Pos165 転写バーストは DNA 上の RNA ポリメラーゼの相互作用によって内因的に引き起こされる

Transcriptional bursting is intrinsically caused by interplay between RNA polymerases on DNA

Keisuke Fujita[1,2], Mitsuhiro Iwaki[1,2], Toshio Yanagida[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. of Front. Biosci., Osaka Univ.*)

3Pos166 金ナノプローブで明らかにされたリニア分子モーター霊菌 *Serratia marcescens* 由来キチナーゼ A の 1 nm ステップ運動

One nanometer steps in the motion of a linear molecular motor *Serratia marcescens* chitinase A resolved by gold nanoprobe

Akihiko Nakamura[1,2], Ryota Iino[1,2,3] ([1]*Okazaki inst. for Integrative Bioscience*, [2]*The Graduate University for Advanced Studies (SOKENDAI)*, [3]*Institute for Molecular Science*)

細胞生物学 / Cell biology

3Pos167 海洋性ビブリオ菌のべん毛形成に関わる DnaJ ファミリータンパク質 SflA のペリプラズム領域の構造特性

Structural property of the periplasmic TPR domain of SflA, a DnaJ family protein involved in flagellation of *Vibrio alginolyticus*

Mayuko Sakuma[1,2], Satoshi Inaba[2], Shoji Nishikawa[3], Takehiko Nishigaki[2], Seiji Kojima[2], Katsumi Imada[3], Michio Homma[1,2] ([1]*Radioisotpe Res. Cent., Nagoya Univ.*, [2]*Div. Biol. Sci. Grad. Sch. Sci., Nagoya Univ.*, [3]*Dept. of Macromol. Sci., Grad. Sch. Sci., Osaka Univ.*)

3Pos168 FlhF がもつ GTPase モチーフへの変異によるビブリオ菌極べん毛数と位置への影響

Effect of mutations in the GTPase motif of FlhF on the number and location of the polar flagellum of Vibrio alginolyticus

Shota Kondo, Michio Homma, Seiji Kojima (*Division of Biological Science, Graduate School of Science, Nagoya University*)

3Pos169 細菌 Rhomboid プロテアーゼ GlpG の生理的基質の探索：GlpG のべん毛 III 型分泌装置機能への関与の可能性

Screening of physiological substrates of *E. coli* rhomboid protease GlpG: possible involvement of GlpG in the flagellar function

Yohei Hizukuri, Kosuke Terushima, Yoshinori Akiyama (*Inst. Virus Res., Kyoto Univ.*)

3Pos170 集合に共役したべん毛モーター固定子 MotB のペリプラズム領域における構造変化の変異体解析

Mutational studies of the assembly-coupled conformational change in the periplasmic region of a flagellar stator protein MotB

Seiji Kojima[1], Masato Takao[2], Gaby Almira[3], Ikumi Kawahara[3], Mayuko Sakuma[1], Michio Homma[1], Chojiro Kojima[3,4], Katsumi Imada[2] ([1]*Div. of Biol. Sci., Grad. Sch. of Sci., Nagoya Univ.*, [2]*Dept. of Macromol. Sci., Grad. Sch. of Sci., Osaka Univ.*, [3]*Inst. for Prot. Res., Osaka Univ.*, [4]*Grad. Sch. of Eng., Yokohama National Univ.*)

3Pos171 **細菌べん毛モーター固定子蛋白質 MotA および回転子蛋白質 FliF の構造解析のための条件検討**

Screening for the structural analysis of the stator and the rotor proteins MotA and FliF in the bacterial flagellar motor

Norihiro Takekawa[1], Mayuko Sakuma[2,3], Erika Yamaguchi[2], Seiji Kojima[2], Michio Homma[2], Katsumi Imada[1] ([1]*Dept. of Macromol. Sci., Grad. Sch. of Sci., Osaka Univ.*, [2]*Div. of Biol. Sci., Grad. Sch. of Sci., Nagoya Univ.*, [3]*Radioisotope Res. Cent., Nagoya Univ.*)

3Pos172 **FlhA の構造変換がべん毛蛋白質輸送順序の決定に重要である**

Conformational rearrangements of FlhA is critical for ordered protein export during flagellar assembly

Yumi Inoue[1], Yuya Ogawa[2], Miki Kinoshita[1], Katsumi Imada[2], **Keiichi Namba**[1,3], Tohru Minamino[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*QBiC, RIKEN*)

3Pos173 **サルモネラべん毛モーターにおける MotA Met-206 の役割**

Roles of MotA Met-206 in rotation and assembly of the Salmonella flagellar motor

Yuya Suzuki[1], Kodai Oono[1], Shuichi Nakamura[1], Fumio Hayashi[2], Kenji Oosawa[3], Yusuke V. Morimoto[4], Seishi Kudo[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Center for Inst. Anal.*, [3]*Div. Mol. Sci., Fac. Sci. and Tech., Gunma Univ.*, [4]*QBiC, Riken*)

3Pos174 **細菌べん毛 III 型タンパク質輸送の *in vitro* 再構築**

***In vitro* reconstitution of the bacterial flagellar type III protein export**

Hiroyuki Terashima[1], Akihiro Kawamoto[2], Chinatsu Tatsumi[1], Keiichi Namba[2,3], Tohru Minamino[2], Katsumi Imada[1] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*, [3]*Quant. Bio. Cent., Riken.*)

3Pos175 **反転膜を用いたべん毛 III 型蛋白質輸送の蛍光による検出**

Fluorescence detection of the flagellar type III protein export using the inverted membrane vesicles

Tsuyoshi Tono[1], Hiroyuki Terashima[1], Kazuhito Tabata[2], Hiroshi Ueno[2], Tomoki Matsuda[3], Takeharu Nagai[3], Hiroyuki Noji[2], Katsumi Imada[1] ([1]*Grad. Sch. of Sci., Univ. Osaka.*, [2]*Sch. of Eng., Univ. of Tokyo*, [3]*ISIR, Univ. Osaka.*)

3Pos176 **バクテリアべん毛輸送ゲート複合体の発現系の構築と精製**

Expression and purification of the bacterial flagellar type III export gate complex

Takuma Fukumura[1], **Miki Kinoshita**[1], Keiichi Namba[1,2], Tohru Minamino[1] ([1]*Grad. Sch. Frontier Biosci, Osaka Univ.*, [2]*QBiC, RIKEN*)

3Pos177 **三次元的形状解析を用いたらせん形細菌の構造的理解**

3D microscopic observation of the cell shape of spiral shaped bacteria

Hajime Tahara, Shuichi Nakamura (*Grad. Sch. Eng. Univ. Tohoku*)

3Pos178 **Coordinated cell-body rotation in spirochete motion**

Kyosuke Takabe, Seishi Kudo, Shuichi Nakamura (*Grad. Sch. Eng., Univ. Tohoku*)

3Pos179 **局所的な照明によって誘起されたスピロプラズマの遊泳方向の反転**

Reversal motion of *Spiroplasma* induced by partial illumination

Tatsuro Itou[1], Daisuke Nakane[1], Wen Wang[2], Takayuki Nishizaka[1] ([1]*Department of Physics, Gakushuin Univ.*, [2]*College of Life Sciences, Nanjing Normal Univ.*)

3Pos180 **ロッド様に直線状で固く，野生型より長い変異型フックの金ナノ粒子標識によるべん毛モーターの高分解能回転計測**

High resolution measurements of flagellar motor rotation by nanogold attached to a straight, solid hook mutant longer than the wild-type

Shuichi Nakamura[1,2], Yusuke V. Morimoto[2,3], Tohru Minamino[2], Keiichi Namba[2] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Frontier BioSci., Osaka Univ.*, [3]*QBiC, Riken*)

3Pos181 **シュードモナス属べん毛モーターの回転計測**

Rotation assay of the Pseudomonas flagellar motor

Taro Hariu, Takuto Tensaka, Seisi Kudo, Shuichi Nakamura (*Grad. Sch. Eng., Tohoku Univ.*)

3Pos182 **ガラスに付着したビブリオ菌を用いたべん毛フリッキングの解析**

Analysis of flagellar flicks of Vibrio cells stuck to a glass slide

Taichi Ohnuki, Shuichi Nakamura, Shoichi Toyabe, Seishi Kudo (*Grad. Sch. Eng., Univ. Tohoku*)

3Pos183 **大腸菌の忌避刺激で見られた時間遅れは FliM の共同的な振る舞いにより説明される**

Simulation of delays in repellent responses of *Escherichia coli* using a conformational spread model

Takashi Sagawa[1], Yoshiyuki Sowa[2], Ikuro Kawagishi[2], Kazuhiro Oiwa[1], Hiroaki Kojima[1] ([1]*Adv. ICT Res. Inst., NICT*, [2]*Dept. of Front. Biosci., Hosei Univ.*)

3Pos184 **大腸菌二成分制御系 AtoS，AtoC の相互依存的細胞内局在**

Interdependent co-localization of the histidine kinase AtoS and the response regulator AtoC of *Escherichia coli*

Takahide Endo[1], Yukiko Miyao[1], Kentaro Yamamoto[1], Masatoshi Nishikawa[1], Yoshiyuki Sowa[1,2], Ikuro Kawagishi[1,2] ([1]*Dept. Frontier Biosci., Hosei Univ.*, [2]*Research Center for Micro-Nano Technology, Hosei Univ.*)

3Pos185 **大腸菌は、アミノ酸種を識別する：データ駆動的アプローチにより明らかにする単細胞生物の化学知覚**

Escherichia coli identify amino-acid species : unicellular organism's chemical perception revealed by using a data-driven approach

Hiroto Tanaka[1], Yasuaki Kazuta[1], Tadashi Matsukawa[1], Yasushi Naruse[2], Yukihiro Tominari[1], Masato Okada[3], Yoshiyuki Sowa[4], Ikuro Kawagishi[4], Kazuhiro Oiwa[1], Hiroaki Kojima[1] ([1]*Adv ICT Res Inst, NICT*, [2]*CiNet, NICT*, [3]*Tokyo Univ.*, [4]*Housei Univ.*)

3Pos186 **コレラ菌走化性受容体 Mlp24 のアミノ酸受容能はカルシウムイオンで増強される**

Ca^{2+} potentiates attractant responses to amino acids mediated by the chemoreceptor Mlp24 of *Vibrio cholerae*

So-ichiro Nishiyama[1,2], Yohei Takahashi[3], Katsumi Imada[3], Ikuro Kawagishi[1] ([1]*Dept. Frontier Biosci., Hosei Univ.*, [2]*Res. Cen. Micro-Nano Tech., Hosei Univ.*, [3]*Dep. Macromol. Sci., Grad. Sch. Sci., Osaka Univ.*)

3Pos187 バクテリアのマグネットコンパスの人為的反転

Artificial polarity-reversal of bacterial magnetic compass

Seiji Iwata[1], Shouhei Kuzoo[1], Daisuke Nakane[1], Azuma Taoka[2,3] ([1]*Dept. of Phys., Gakushuin Univ.*, [2]*Col. of Sci. Eng., Kanazawa Univ.*, [3]*Bio-AFM Front. Res. Cent., Kanazawa Univ.*)

3Pos188 **Importance of receptor cooperativity on the switching coordination of flagellar motors on a single *Escherichia coli* cell**

Yong-Suk Che[1], Hiroto Takahashi[2], Akihiko Ishijima[1], Hajime Fukuoka[1] ([1]*Dept. Frontier Biosci., Osaka Univ.*, [2]*IMRAM, Tohoku Univ.*)

3Pos189 変性蛋白質センサーとしての大腸菌ヒスチジンキナーゼ BaeS

The histidine kinase BaeS of Escherichia coli may sense denatured proteins

Tohru Umemura[1], Yoshiyuki Sowa[1,2], Ikuro Kawagishi[1,2] ([1]*Dept. Frontier Bio., Hosei Univ.*, [2]*Research Center for Micro-Nano Technology, Hosei Univ.*)

3Pos190 G タンパク質共役型受容体の動的なホモ・ヘテロダイマー形成：二色同時蛍光１分子観察による解析

Dynamic homo- and hetero-dimerizations of G-protein coupled receptors: An analysis by dual-channel single fluorescent molecule observation

Rinshi Kasai[1], Akihiro Kusumi[1,2] ([1]*Inst. Front. Med. Sci., Kyoto Univ.*, [2]*Membrane Cooperativity Unit, OIST*)

3Pos191 共焦点画像解析と 1 分子計測を用いた ERK シグナル伝達系のボトルネックの解明

Unraveling origins of bottleneck effects for ERK signal transduction using confocal image analyses and single molecule imaging

Kazunari Mouri, Yasushi Okada (*RIKEN QBiC*)

3Pos192 三量体 G タンパク質の制御を介した走化性レンジの拡張機構

Dynamic range extension of eukaryotic chemotaxis via regulation of heterotrimeric G protein dynamics

Yukihiro Miyanaga[1,2], Yoichiro Kamimura[1,2], Masahiro Ueda[1,2] ([1]*Frontier Biosciences, Osaka Univ.*, [2]*QBiC, Riken*)

3Pos193 走化性タンパク質の細胞内動態と細胞応答の同時計測

Simultaneous observation of the intracellular chemotactic proteins and the cellular behavior

Hajime Fukuoka[1], Hiroto Takahashi[2], Akihiko Ishijima[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*IMRAM, Tohoku Univ.*)

3Pos194 ライブセルイメージングと薬剤実験に基づく動的な誘引物質勾配場における HL60 細胞の走化性運動の解析

Pharmacological and live-cell imaging analysis of chemotactic HL60 cells under dynamically changing chemoattractant gradient

Motohiko Ishida[1], Akihiko Nakajima[2], Satoshi Sawai[1,2] ([1]*Dept. Basic Sci., Grad. Sch. of Arts & Sci., Univ. of Tokyo*, [2]*Research Center for Complex Systems Biology, Grad. Sch. of Arts & Sci., Univ. of Tokyo*)

3Pos195 マクロファージの走熱性メカニズムの解明

Thermotaxis mechanism of mouse macrophage

Hideo Saitou, Seine Shintani, Hideo Higuchi (*Grad. Sch. Sci., The university of Tokyo*)

3Pos196 免疫受容体シグナルを担う足場分子 LAT は細胞膜に繋留された小胞で機能する：1 分子イメジングによる解明

The immune signal adaptor molecule LAT works on cytoplasmic vesicles tethered to the plasma membrane: a single-molecule imaging study

Koichiro M. Hirosawa[1], Bo Tang[2], Nao Hiramoto-Yamaki[1,3], Kenta J. Yoshida[1], Shohei Nozaki[4], Takaaki Tsunoyama[5], Kenichi G.N. Suzuki[6], Kazuhisa Nakayama[4], Takahiro K. Fujiwara[1], Akihiro Kusumi[5] ([1]*WPI-iCeMS, Kyoto Univ.*, [2]*College of Chemistry and Molecular Sciences, Wuhan Univ.*, [3]*JSPS Research Fellow*, [4]*Grad. Sch. Pharm., Kyoto Univ.*, [5]*OIST*, [6]*NCBS-inStem, Bangalore, India.*)

3Pos197 PIP3 と Ras の自己組織的な局在形成過程とその制御メカニズムの解析

Analysis of Self-organized Domain Formation and Regulation Mechanism of PIP3 and Ras

Seiya Fukushima[1,2], Satomi Matsuoka[2,3], Masahiro Ueda[1,2,3] ([1]*Grad. Sch. Sci., Univ. Osaka*, [2]*QBiC, RIKEN*, [3]*Grad. Sch. Frontier Biosciences., Univ. Osaka*)

3Pos198 **Controlling contractile instabilities in the actomyosin cortex**

Masatoshi Nishikawa[1,2,3,4], Sundar Naganathan[2,3,4], Frank Julicher[4], Stephan Grill[2,3,4] ([1]*Hosei University*, [2]*TU-Dresden*, [3]*MPI-CBG*, [4]*MPI-PKS*)

3Pos199 増殖中の培養細胞における遊離コレステロールの分布

Distribution of free cholesterol in MDCK cells during the migration on culture dish

Yoshikatsu Ogawa (*AIST BMD*)

3Pos200 獲得免疫応答における T リンパ球の単一細胞測定系の開発

Development of a single cell assay system for T lymphocytes in adaptive immunity

Hiroaki Machiyama[1], Tomoyuki Yamaguchi[1], Tomonobu Watanabe[1,2], Hideaki Fujita[1,2] ([1]*IFReC, Osaka U.*, [2]*QBiC, RIKEN*)

3Pos201 **Drug response of lymphocytic leukemia cells to anticancer drug is affected by experience of cell division prior to treatment**

Akihisa Seita, Takahiro Yamada, Yuichi Wakamoto (*Department of Basic Science, Graduate School of Arts and Science, University of Tokyo*)

3Pos202 ErbB レセプターの相互リン酸化ネットワーク

Cell and signal specific phosphorylation networks of the ErbB receptor family

Hiraku Miyagi, Michio Hiroshima, Atsushi Mochizuki, Yasushi Sako (*RIKEN*)

生体膜・人工膜：構造・物性 / Biological & Artificial membrane: Structure & Property

3Pos203 膜活性ポリマーによる脂質二分子膜ナノディスクの形成

Lipid bilayer nanodiscs formed by designed membrane-active polymers

Kazuma Yasuhara, Jin Arakida, Masaya Inoue, Jun-ichi Kikuchi (*Grad. Sch. Mat. Sci., Nara Inst. Sci. Tech.*)

3Pos204 可溶性高分子で生成された水性相分離のマイクロ液滴内への生細胞の含有

Entrapment of Living Cells inside Micro-Droplet under Aqueous/Aqueous Segregation with Solvable Polymers

Tadashi Fujimoto[1], Naoki Nakatani[1], Kanta Tsumoto[2], Chwen-Yang Shew[3], Kenichi Yoshikawa[1] ([1]*Fac. Life Medical Sciences, Univ. Doshisha*, [2]*Grad. Sch. Eng, Univ. Mie*, [3]*Chemistry, City. Univ. New York*)

3Pos205 粗視化モデルによる脂質分子の集合体の形成過程に関する理論的研究

Theoretical study on the process of the formation of lipid molecule cluster by coarse-grained model

Shogo Kinoshita, Satoshi Nakagawa, Makoto Wada, Seiichiro Ito, Kazutomo Kawaguchi, Hidemi Nagao (*Nat. Sci. Kanazawa Univ.*)

3Pos206 ベシクルに大小２種のコロイド粒子を内包させた系の相分離ー朝倉・大沢理論の拡張―

Phase Separation of Two Kinds of Colloidal Particles in Giant Vesicles -Extension of Asakura-Osawa Theory-

Yuno Natsume[1], Kazumi Itoh[1], **Yuhei Natsume**[2], Kensuke Kurihara[3,4,5] ([1]*Japan Women's Univ.*, [2]*Chiba Univ.*, [3]*Okazaki Institute for Integrative Bioscience*, [4]*Institute for Molecular Science*, [5]*Research Center for Complex Systems Biology, The Univ. of Tokyo*)

3Pos207 種々の脂質組成によるタンパク質内包リポソームの構造学的研究

Structural study of liposomes encapsulating proteins depending on lipid composition and species

Kosuke Takahashi, Mitsuhiro Hirai (*Grad. Sch. Eng., Gunma Univ.*)

3Pos208 アシル鎖長の異なるホスファチジルエタノールアミン二重膜の熱および圧力相転移

Thermotropic and barotropic phase transitions in bilayer membranes of phosphatidylethanolamines with varying acyl chain lengths

Masaki Goto[1], Shigeru Endo[2], Nobutake Tamai[1], Hitoshi Matsuki[1] ([1]*Grad. Sch. of Biosci. and Bioindus., Tokushima Univ.*, [2]*Fac. of Engin., Tokushima Univ.*)

3Pos209 親水性高分子により基板との距離を制御した人工膜への膜タンパク質再構成

Patterned model membrane with hydrophilic polymer brushes for the functional incorporation of membrane proteins

Fuyuko Tamura[1], Yasushi Tanimoto[1], Yasuhiko Iwasaki[2], Fumio Hayashi[3], Yuki Sudo[4], Kenichi Morigaki[5] ([1]*Grad. Agri., Univ. Kobe*, [2]*Chem. Bioeng., Univ. Kansai*, [3]*Sci., Univ. Kobe*, [4]*Grad. Med. Dent. Pharm., Univ. Okayama*, [5]*Biosignal., Univ. Kobe*)

3Pos210 バクテリオロドプシン球殻構造体の形成機構

A mechanistic insight into the formation of bacteriorhodopsin vesicle

Daisuke Yamamoto, Risa Mutoh (*Fac. Sci. Fukuoka Univ.*)

3Pos211 ホスホリパーゼ Cβ １の C 末端には脂質膜のチューブ形成能がある

Membrane tubulation ability of phospholipase Cβ1 C terminal domains

Takehiko Inaba[1], Takuma Kishimoto[2], Motoshide Murate[1], Takuya Tajima[1,3], Mitsuhiro Abe[1], Asami Makino[1], Nario Tomishige[1], Reiko Ishitsuka[1], Yasuo Ikeda[3], Shinji Takeoka[3], Toshihide Kobayashi[1,4] ([1]*RIKEN*, [2]*Kyorin Univ. Sch. Medicine*, [3]*Waseda Univ.*, [4]*UMR 7213 CNRS, University of Strasbourg*)

3Pos212 巨大分子系シミュレーションに向けた粗視化脂質モデルの開発

Developing a coarse-grained model of lipid for large molecular simulations

Suguru Kato, Shoji Takada (*Kyoto University*)

3Pos213 アルギニンペプチドの膜透過を促進する両親媒性ペプチドによる脂質パッキングの変化

Alteration of Lipid Packing State by Amphipathic Peptides Promoting Membrane Penetration of Octaarginine

Tomo Murayama, Shiroh Futaki (*ICR, Kyoto Univ.*)

3Pos214 FTIR-ATR のプリズム上に作製した皮膚角層モデル膜への物質透過解析

Permeation of substances into stratum corneum model membranes prepared directly on FTIR-ATR prism

Kohei Oka, Satoru Kato (*Grad. Sch. Sci. & Tech., Univ. Kwansei Gakuin*)

神経回路･情報処理 / Neuronal circuit & Information processing

3Pos227 神経振動活動のシナプス入力による位相依存的な調節

Phase-dependent modulation of neural oscillations by synaptic inputs

Satoshi Watanabe[1], Moritoshi Hirono[2] ([1]*NCNP*, [2]*Grad. Sch. Brain Sci., Doshisha Univ.*)

3Pos228 線虫の whole-brain イメージングデータに関する位相同期解析

Phase synchronization analysis of whole-brain imaging data of C. elegans

Yuishi Iwasaki[1,7], Takayuki Teramoto[2,7], Terumasa Tokunaga[3,7], Osamu Hirose[4,7], Yu Toyoshima[5,7], Ryo Yoshida[6,7], Yuichi Iino[5,7], Takeshi Ishihara[2,7] ([1]*Fac. Eng., Ibaraki Univ.*, [2]*Grad. Sch. Sci., Kyushu Univ.*, [3]*Grad. Sch. Comp. Sci. and Sys. Eng., Kyushu Institute Tech.*, [4]*Institute. Sci. and Eng., Kanazawa Univ.*, [5]*Grad. Sch. Sci., Univ. Tokyo*, [6]*Institute Stat. Math.*, [7]*JST, CREST*)

3Pos229 神経細胞から伸長する神経突起の特性のオンチップ１細胞解析

Minimum Requirements of Microchannel Patterns for Building of Stable Neuronal Circuits in On-chip Cell Network Assay

Takahito Kikuchi[1], Hideyuki Terazono[2], Kenji Matsuura[2], Akihiro Hattori[2], Masao Odaka[2], Kenji Yasuda[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*)

3Pos230 フェムト秒レーザー光刺激による神経回路網の誘発応答特性

Spatio-temporal activity pattern in neuronal network evaluated by femtosecond laser-induced stimulation

Yuji Fujioka[1,2], Yuta Nakagawa[1,2], Suguru N. Kudoh[2], Takahisa Taguchi[3], Chie Hosokawa[1] ([1]*Biomed. Res. Inst., AIST*, [2]*Sch. Sci. & Tech., Kwansei Gakuin Univ.*, [3]*CiNet, NICT*)

3Pos231 **培養神経回路網におけるネットワークグラフ構造の培養日数依存的変化**
Developmental changes of graph structures in cultured neurons -the analysis with functional-connection map
Nanami Hirata, Wataru Minoshima, Hidekatsu Ito, Suguru Kudoh (*Department of Human System Interaction, School of Science and Technology, Kwansei Gakuin University*)

3Pos232 **エピカテキンは、ヨーロッパモノアラガイの呼吸行動の中枢リズム発生器であるRPeD1ニューロンの興奮性を変化させる**
Epicatechin alters the electrophysiological activity of RPeD1 in the pond snail, Lymnaea
Yoshimasa Komatsuzaki[1], Minoru Saito[2], Ken Lukowiak[3] ([1]*Dept. Phys., CST, Nihon Univ.*, [2]*Grad. Sch. Integ. Basic Sci., Nihon Univ.*, [3]*Hotchkiss Brain Inst., Cumming Sch. of Med., Univ. of Calgary*)

3Pos233 **Increasing reproducibility of activity patterns in nenural network during culture days**
Takumi Okada, Wataru Minoshima, Hidekatsu Ito, Suguru Kudoh (*Dept. of Human System Interaction, School of Sci. and Tech., Kwansei Gakuin University,*)

3Pos234 **超高感度な匂い識別は、受容体 - G 蛋白質の初期一過性相互作用と前梨状皮質振動性応答に部分的に支配される**
Supersensitive odor discrimination is controlled in part by initial interactions of receptor-G-protein and cortical oscillatory responses
Takaaki Sato[1], Reiko Kobayakawa[2], Ko Kobayakawa[2], Makoto Emura[3], Shigeyoshi Itohara[4], Takashi Kawasaki[1], Riichi Kajiwara[5], Ichiro Takashima[1], Toshio Iijima[6], Akio Tsuboi[7], Hiroyoshi Matsumura[8] ([1]*AIST*, [2]*Kansai Med. Univ.*, [3]*Takasago Internat'l. Corp.*, [4]*BSI., RIKEN*, [5]*Sch. Sci. & Technol., Meiji Univ.*, [6]*Grad. Sch. Life Sci., Tohoku Univ.*, [7]*Nara Med. Univ.*, [8]*Ritsumeikan Univ.*)

3Pos235 **定量的マンガン造影 MRI による全脳神経活動計測**
Whole brain activity mapping using quantitative activation-induced manganese-enhanced MRI
Makoto Osanai[1,2], Satomi Kikuta[1,3], Hajime Tamura[1], Noriyasu Homma[1,2] ([1]*Tohoku Univ. Grad. Sch. Med.*, [2]*Grad. Sch. Bio Med. Eng, Tohoku Univ.*, [3]*Research Fellow, JSPS*)

行動 / Behavior

3Pos236 **Foraging behavior of Caenorhabditis elegans**
Chien Jung Lo, **Mao Ting Cheng** (*Dept. of Physics, NCU, Taiwan*)

3Pos237 **2 次元系および 3 次元系における細胞の自発運動動態**
Spontaneous cell migration dynamics in 2D and 3D environment
Hiroaki Takagi (*Dept. of Phys., Nara Med. Univ.*)

3Pos238 **活動量と睡眠との関連解析**
Association between physical activity and sleep efficiency
Ikuko Motoike[1,2], Atsushi Koike[1], Akihiro Karashima[1,3], Mitsuyuki Nakao[1] ([1]*Grad. Sch. Info. Sci., Tohoku Univ.*, [2]*Medical Megabank Org., Tohoku Univ.*, [3]*Grad. Sch. Eng., Tohoku Inst. Tech.*)

光生物：視覚･光受容 / Photobiology: Vision & Photoreception

3Pos239 **光誘起チャネルロドプシンに関する理論研究**
Theoretical study on molecular mechanism of photo-induced gate opening of channelrhodopsin
Cheng Cheng[1], Motoshi Kamiya[1], Norio Yoshida[2], Shigehiko Hayashi[1] ([1]*Kyoto Univ.*, [2]*Kyushu Univ.*)

3Pos240 **古細菌型 TSA モチーフ配列をもつ真正細菌由来ハライドイオンポンププロドプシンの光反応解析**
Cl^--pumping Photoreaction of a Bacterial Halide-ion Pumping Rhodopsin with an Archaeal-type TSA motif
Takashi Tsukamoto[1], Susumu Yoshizawa[2], Takashi Kikukawa[3], Makoto Demura[3], Yuki Sudo[1] ([1]*Grad. Sch. Med. Dent. & Pharm. Sci., Okayama Univ.*, [2]*AORI, Univ. of Tokyo*, [3]*Fac. Adv. Life Sci., Hokkaido Univ.*)

3Pos241 ***Mastigocladopsis repens* halorhodopsin のフォトサイクルの解析と TSD モチーフの役割の解明**
Photocycle of *Mastigocladopsis repens* halorhodopsin and the role of its TSD motif
Takatoshi Hasemi, Takashi Kikukawa, Tomoyasu Aizawa, Naoki Kamo, Makoto Demura (*Grad. Sch. Life Sci., Hokkaido Univ.*)

3Pos242 **ハロロドプシンの陰イオン輸送サイクルにおけるレチナール色素の異性化**
Isomerization of the retinal chromophore during the anion pumping cycle of halorhodopsin
Tsutomu Kouyama, Hiroki Kubo, Siu Kit Chan, Kousuke Maki (*Dept. Physics, Graduate School of Science, Nagoya University*)

3Pos243 **X 線小角散乱法を用いたシロイヌナズナ phototropin1 と変異体の構造及び機能研究**
Structural and functional study of Arabidopsis phototropin1 and its mutants by using small-angle X-ray scattering
Mao Oide[1,2], Koji Okajima[1,2], Sachiko Kashojiya[2,3], Yuki Takayama[1,2], Tomotaka Oroguchi[1,2], Takaaki Hikima[2], Masaki Yamamoto[2], Masayoshi Nakasako[1,2] ([1]*Grad. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*, [3]*Dept. of Biol. Sci., Osaka Pref. Univ.*)

3Pos244 **Chromophore conformation in active site of orange carotenoid protein studied by Raman optical activity spectroscopy**
Tomotsumi Fujisawa[1], Masashi Unno[1], Ryan Leverenz[2], Cheryl Kerfeld[2] ([1]*Saga Univ.*, [2]*Michigan State Univ.*)

3Pos245 **Functional characterization of a microbial rhodopsin from the marine eubacterium *Rubricoccus marinus* SG-29**
Saki Inoue[1], Susumu Yoshizawa[2], Takashi Tsukamoto[1,3], Yuki Sudo[1,3] ([1]*Fac. Pharm. Sci., Okayama Univ.*, [2]*AORI, Univ. of Tokyo*, [3]*Grad. Sch. Med. Dent. & Pharm. Sci., Okayama Univ.*)

3Pos246 **Signaling kinetics of Cyanabacterial phytochrome (Cph1) studied by the transient grating method**
Kimitoshi Takeda, Masahide Terazima (*Department of chemistry, Kyoto University*)

3Pos247 **In situ 光照射固体 NMR によるセンサリーロドプシン II の光中間体の解析**

Characterization of photo intermediates in sensory rhodopsin II as revealed by in-situ photo-irradiation solid-state NMR

Yoshiteru Makino[1], Izuru Kawamura[1], Takashi Okitsu[2], Akimori Wada[2], Yuki Sudo[3], Naoki Kamo[4], Akira Naito[1], Kazuyoshi Ueda[1] ([1]*Grad. Sch. Eng., Yokohama Natl. Univ.*, [2]*Kobe Pharm. Univ.*, [3]*Grad. Sch. Med. Dent. Pharm., Okayama Univ.*, [4]*Grad. Sch. Life Sci., Hokkaido Univ.*)

3Pos248 **異なる生物種におけるフォトトロピン光反応の多様性**

Diversity of photoreaction of phototropins among different organisms

Yusuke Nakasone[1], Koji Okajima[4], Kenichi Hitomi[3], John Christie[3], Satoru Tokutomi[2], Masahide Terazima[1] ([1]*Kyoto University*, [2]*Osaka Prefecture University*, [3]*Scripps research institute*, [4]*Keio University*)

3Pos249 **EPR 法による Photozipper 反応過程の解析**

Reaction mechanism in Photozipper monitored by Electron Paramagntic Resonance

Kouhei Ozeki[1], Hiroki Nagashima[1], Osamu Hisatomi[2], Hiroyuki Mino[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*)

3Pos250 **Substrate recognition of the (6-4)photolyase**

Yuma Terai[1], Takahiro Yumiba[1], Tomoko Ishikawa[2], Takeshi Todo[2], Junpei Yamamoto[1], Shigenori Iwai[1] ([1]*Grad. Sch. Eng. Sci., Osaka Univ.*, [2]*Grad. Sch. Med., Osaka Univ.*)

3Pos251 **(6-4)光回復酵素による逐次的 2 光子 DNA 修復における逆電子移動の観測**

Monitoring of the back electron transfer in the successive two-photons DNA repair by the (6-4) photolyase

Junpei Yamamoto[1], Kohei Shimizu[1], Shigenori Iwai[1], Klaus Brettel[2] ([1]*Grad. Sch. Eng. Sci., Osaka Univ.*, [2]*CEA Saclay, France*)

光生物：光合成 / Photobiology: Photosynthesis

3Pos252 **イチョウの葉の微量色素分析**

Precise pigment analysis of ginkgo leaves

Yuhta Isei[1], Katsuhiro Wada[1], Tadashi Watanabe[2], Norio Tanaka[3], Masami Kobayashi[1] ([1]*Divi. Materials Sci., Fac. Pure and Applied Sci., Univ. Tsukuba*, [2]*Res. Center Math and Sci. Edu., Org. Adv. Edu., Tokyo University of Science*, [3]*Tsukuba Botanical Garden*)

3Pos253 **新奇クロロフィルを持つシアノバクテリアより光化学系 II 標品の単離精製とエネルギー移動機構の解析**

Analysis of energy transfer system of photosystem II complexes isolated from new chlorophyll containing cyanobacterium

Toshiyuki Shinoda[1], Daisuke Nii[1], Seiji Akimoto[2,3], Tatsuya Tomo[1,4] ([1]*Grad. Sch. Sci., Tokyo Univ. of Sci.*, [2]*Molecular Photoscience Research Center, Kobe Univ.*, [3]*JST CREST*, [4]*JST PRESTO*)

3Pos254 **Aggregation of chlorophylls d and f in n-hexane**

Katsuhiro Wada[1], Terumitsu Kanjoh[1], Yuhta Isei[1], Yutaka Hanawa[2], Yoshihiro Shiraiwa[2], Masataka Nakazato[3], Hideaki Miyashita[4], Masami Kobayashi[1] ([1]*Div. Materials Sci., Fac. Pure Applied Sci., Univ. Tsukuba*, [2]*Fac. Life Environ. Sci., Univ. Tsukuba*, [3]*Chlorophyll Research Institute Co., Ltd*, [4]*Graduate School Human Environ. Studies, Kyoto Univ.*)

3Pos255 **緑藻ミル糸状体における培養時光強度に依存したカロテノイドの蓄積**

Extra accumulation of carotenoids upon intense irradiation during culture of a siphonous green algae, *Codium fragile*

Kentaro Fujiwara[1], Ritsuko Fuji[1,2] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*OCARINA, Osaka City Univ.*)

3Pos256 **チラコイド膜での光還元に対する共溶媒の効果**

Effect of co-solvents on photo-reduction in thylakoid membranes

Kuniyuki Hatori, Yuko Kokaji, Tomoyuki Toyama (*Dept. Bio-Systems, Yamagata Univ.*)

3Pos257 **Identity of chlorophyll e**

Yuhta Sorimachi[1], **Taku Kaitani**[1], Masataka Nakazato[2], Hideaki Miyashita[3], Masami Kobayashi[1] ([1]*Div. Materials Sci. Pure and Applied Sci. Univ.*, [2]*Chlorophyll Res. Inst.*, [3]*Graduate School of Human and Environ. Sci., Univ.*)

3Pos258 **Solubility and stability of chlorophylls in algal oil**

Terumitsu Kanjoh[1], Mikihide Demura[2], Masaki Yoshida[2], Makoto Watanabe[2], Masataka Nakazato[3], Masami Kobayashi[1] ([1]*Div. Materials Sci., Fac. Pure Appl. Sci., Univ. Tsukuba*, [2]*Fac. Life Environ. Sci., Univ. Tsukuba*, [3]*Chl. Tes. Inst.*)

3Pos259 **Complex formation between carbon nanomaterials and photosystem complexes**

Shota Tanaka, Tatsuya Tomo (*Grad. Sch. Sci., Tokyo Univ. of Sci.*)

3Pos260 **緑化途上トウモロコシ生葉の極低温顕微分光による光合成タンパク質前駆体の蛍光スペクトル同定**

Spectral identification of late precursors to photosynthetic proteins by cryogenic microscopy of greening etiolated Zea mays leaves

Hirotomo Nagasawa, Tomofumi Chiba, Yutaka Shibata (*Graduate School of Science, Tohoku University*)

3Pos261 **北海道で採取した紅色非硫黄細菌による酢酸塩からの光水素生成**

Phototrophic hydrogen production from acetate by purple non-sulfur bacteria from rivers in Hokkaido

Mayoka Kanoh[1], Kazuma Tazawa[1], Seigo Kumakura[2], **Masahiro Hibino**[1,2] ([1]*Div. Sustain. Enviro. Eng., Muroran Inst. Tech.*, [2]*Dept. Appl. Sci., Muroran Inst. Tech.*)

放射線生物学･活性酸素 / Radiobiology & Active oxygen

3Pos262 **低線量或いは高線量 X 線被ばく後のマウス肝臓におけるメタボローム解析**

Metabolome analyses in livers of mice exposed to low or high-dose X-ray-irradiation

Tetsuo Nakajima[1], Guillaume Vares[2], Yasuharu Ninomiya[1], Bing Wang[1], Takanori Katsube[1], Kaoru Tanaka[1], Cuihua Liu[1], Hirokazu Hirakawa[1], Kouichi Maruyama[1], Akira Fujimori[1], Mitsuru Nenoi[1] ([1]*Natl. Inst. Radiol. Sci., QST.*, [2]*OIST*)

3Pos263 **ビタミンDによって誘発される単球の分化と酸化ストレスの関係**
Relations of oxidative stress and monocytic differentiation induced by vitamin D_3
Naoya Matsunaga, Kiyotaka Murakami, Wakako Hiraoka (*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*)

3Pos264 **プリオンオクタペプチドと二価金属錯体結合によってひきおこされるレドックス不均衡**
Redox imbalance induced by coordination of divalent metals in octarepeat region of human PrP
Shinnosuke Kondo, Wakako Hiraoka (*Dept. Phys., Grad. Sch. Sci. & tech., Meiji Univ.*)

3Pos265 **超音波に誘発される CMNB-ケージ基と脂肪酸の分解**
Ultrasound-induced scission of CMNB-caged moiety and fatty acids
Kengo Takei[1], Takuya Wada[1], Asuka Kato[1], Masato Mutoh[2], Wakako Hiraoka[1] ([1]*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Mater. & Human Env. Sci., Shonan Inst. of Tech.*)

ゲノム生物学：ゲノム機能 / Genome biology: Genome function

3Pos266 **一分子観察における DNA 二重鎖の光切断に対する PEG 保護作用**
Protective effect of PEG against DNA double-strand breaks caused by photo irradiation through single molecule observation
Moe Usui, Yuko Yoshikawa, Kenichi Yoshikawa (*Facul. Life Med. Sci., Doshisha Univ.*)

バイオインフォマティクス：機能ゲノミクス / Bioinformatics: Functional genomics

3Pos267 **機械学習を用いた機能未知スプライシングアイソフォームの機能性推定**
Evaluation of functionality of uncharacterized splicing isoforms using machine learning techniques
Pramote Teerasetmanakul, Masafumi Shionyu (*Grad. Sch. Bio-Sci., Nagahama Inst. Bio-Sci. Tech.*)

3Pos268 **Predicting protein-protein interactions using sequence homology and machine learning methods**
Yifan Tang, Wei Cao, Tohru Terada, Kazuya Sumikoshi, Shugo Nakamura, Kentaro Shimizu (*Grad. Sch. Agr., Univ. Tokyo*)

3Pos269 **クロマチン構造形成における単純反復配列の機能的役割**
Functional Roles of Simple Repeat Sequences in Chromatin Conformations
Takeru Kameda[1], Atsushi Ikegaya[1], Takeshi Sugawara[2], Naoaki Sakamoto[1,2], Akinori Awazu[1,2] ([1]*Dept. of Mathematical and Life Sciences, Hiroshima University*, [2]*Research Center for the Mathematics on Chromatin Live Dynamics*)

3Pos270 **翻訳伸長因子 1A の配列情報に基づいた機能分岐に関わる重要な残基の予測**
Prediction of key residues involving functional divergence based on sequence information of translation elongation factor 1A
Yosuke Kondo, Satoru Miyazaki (*Fac. Pharm., Tokyo Univ. Sci.*)

3Pos271 **天然変性領域の機能部位：Protean Segments が効果的に相互作用できる理由**
Interface property of protean segments: intrinsically disordered regions that undergo disorder-to-order transitions upon binding
Divya Shaji, Takayuki Amemiya, Ryotaro Koike, **Motonori Ota** (*Grad. Sch. Inf. Sci., Nagoya U.*)

3Pos272 **Immuno-Navigator: a co-expression database for cell type-specific network inference in the immune system**
Alexis Vandenbon (*IFReC, Osaka University*)

バイオインフォマティクス：比較ゲノミクス / Bioinformatics: Comparative genomics

3Pos273 **光回復酵素／クリプトクロムファミリーの機能発現に重要なアミノ酸残基部位の探索**
In search for amino acid positions that determine the molecular function of photolyase/cryptochrome family
Daichi Yamada[1], Kei Yura[1,2] ([1]*Cent. Info. Biol., Ochanimizu Univ.*, [2]*NIG*)

3Pos274 **Refining the performance of k-mer count similarity prediction and examining use of different scoring matrix for better pairwise alignment**
Kazunori Yamada, Kengo Kinoshita (*Tohoku University*)

数理生物学 / Mathematical biology

3Pos275 **緑藻の走光性と多細胞性の関係**
Relation between phototaxis and multicellularity of green algae
Keisuke Yamada, Yoshihiro Murayama (*Tokyo Univ. of Agri. and Tech.*)

3Pos276 **染色体の凝縮が染色体の構築や分離に与える影響**
Effects of chromatin condensation on chromosome construction and segregation
Yuji Sakai[1,2], Masashi Tachikawa[1], Atsushi Machizuki[1], Kazuhisa Kinoshita[3], Tatsuya Hirano[3] ([1]*RIKEN, Theoretical Biology Laboratory*, [2]*RIKEN, iTHES*, [3]*RIKEN, Chromosome Dynamics Laboratory*)

3Pos277 **クロマチン動態とコンタクトマップの関係を深く理解するための数理的研究**
A mathematical study for deep understanding of relationship between chromatin dynamics and contact map
Masaki Nakagawa (*RcMcD, Hiroshima Univ.*)

3Pos278 **Enhancement of sampling space in multivariate analysis of experimental big data in various biological sciences**
Jiyoung Kang[1], Kazuhiko Yamasaki[2], Masaru Tateno[1] ([1]*Univ. of Hyogo*, [2]*Biomed. Res. Ins. AIST*)

3Pos279 血糖値調節におけるインスリン・C ペプチドの血中動態の数理モデルを用いた解析
Mathematical model analysis of blood glucose regulation with insulin and C-peptide
Kaoru Ohashi[1], Masashi Fujii[1], Shinsuke Uda[1], Hisako Komada[2], Kazuhiko Sakaguchi[2], Wataru Ogawa[2], Shinya Kuroda[1] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Grad. Sch. Med., Univ. Kobe*)

3Pos280 生命科学研究用の特化型シミュレータ群の具現化
Implementation of a group of special-purpose simulators for life science researches
Hideto Katsuma[1,2], Jun Takayama[2], Yukako Tohsato[2], Koji Kyoda[2], Shuichi Onami[1,2] ([1]*Grad. Sch. System Inform., Kobe Univ.*, [2]*Lab. Dev Dyn., RIKEN QBiC*)

3Pos281 遺伝子発現量制御メカニズムのタイプ分類と遺伝子機能の関係
Relationship between regulatory pattern of gene expression level and gene function
Masayo Inoue, Katsuhisa Horimoto (*molprof, AIST*)

3Pos282 T 細胞の胸腺における細胞数のダイナミクスのモデリングと推定
Modeling and inferring dynamics of T cell population in thymus
Kazumasa Kaneko[1], Ryo Yokota[2], Taishin Akiyama[3], Tetsuya Kobayashi[2] ([1]*Grad. Sch. Eng., Univ. Tokyo*, [2]*Inst. Ind. Sci., Univ. Tokyo*, [3]*Inst. Med. Sci., Univ. Tokyo*)

3Pos283 触媒反応系から成る細胞のトレードオフ
Trade-off in a protocell model with catalytic reactions
Atsushi Kamimura, Kunihiko Kaneko (*The University of Tokyo*)

3Pos284 Spine における Small-volume effect: Robust, Sensitive, Efficient な情報伝達のメカニズム
Small-volume effect enables robust, sensitive and efficient information transfer in the spine
Masashi Fujii[1], Kaoru Ohashi[1], Yasuaki Karasawa[2], Minori Hikichi[1], Shinya Kuroda[1] ([1]*Dept. Biol. Sci., Grad. Sch. of Sci., Univ. of Tokyo*, [2]*Dept. Neurol., Grad. Sch. of Med., Univ. of Tokyo*)

非平衡·生体リズム / Nonequilibrium state & Biological rhythm

3Pos285 **Curvature-driven splitting of a planar traveling wave**
Kazuya Horibe[1], Ken-ichi Hironaka[2,3,4], Katsuyoshi Matsushita[2], Koichi Fujimoto[2] ([1]*Grad. Sch. Info, Univ. Osaka*, [2]*Grad. Sch. Sci., Univ. Osaka*, [3]*CDB, Inst., Riken*, [4]*JSPS Research Fellow(PD)*)

3Pos286 **Quantification of dynamic mechano-response of myoblast using stimulus responsive matrix**
Marcel Hoerning[1], Masaki Nakahata[2], Akihisa Yamamoto[1], Mariam Veschgini[3], Stefan Kaufmann[3], Yoshinori Takashima[2], Akira Harada[2,4], Motomu Tanaka[1,3] ([1]*iCeMS, Kyoto University*, [2]*Osaka University*, [3]*Heidelberg University*, [4]*ImPACT*)

3Pos287 再生ヒドラにおける形状ダイナミクスと対称性の破れの定量化
Quantification of Morphological Dynamics and Symmetry Break in Regenerating Hydra Tissues
Ryo Suzuki[1], Mariam Veschgini[2], Thomas W. Holstein[3], Motomu Tanaka[1,2] ([1]*iCeMS, Kyoto University*, [2]*Institute of Physical Chemistry, University of Heidelberg*, [3]*Centre for Organismal Studies, University of Heidelberg*)

3Pos288 **2D swarming bacteria**
Chien Jung Lo, **Ching Yuan Lin** (*Dept. of Physics, NCU, Taiwan*)

3Pos289 聴覚刺激によって引き起こされる脳波の引き込み現象と確率共鳴
Auditory entrainment and stochastic resonance in EEGs
Minoru Saito[1,2], Shogo Kawamoto[2], Yuuta Hamasaki[1], Ken Saito[3], Tetsuya Yamamoto[4] ([1]*College of Humanities and Sciences, Nihon University*, [2]*Graduate School of Integrated Basic Sciences, Nihon University*, [3]*College of Science and Technology, Nihon University*, [4]*Tokyo Metropolitan College of Industrial Technology*)

バイオイメージング / Bioimaging

3Pos290 **Observation of conformational dynamics of FliI by HS-AFM**
Kei Adachi[1], Jun-ichi Kishikawa[3], Hiroyuki Terashima[2], Takayuki Uchihashi[1], Katsumi Imada[2], Ken Yokoyama[3], Toshio Ando[1] ([1]*Coll. Sci. & Eng., Kanazawa Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Facul. Biosci., Kyoto Sangyo Univ.*)

3Pos291 高速原子間力顕微鏡と光ピンセットの複合システム
Combined system of high speed atomic force microscopy(HS-AFM) and optical tweezers
Shin'nosuke Yamanaka[1], Akane Goto[1], Hiroki Watanabe[2], Takayuki Uchihashi[1,3], Toshio Ando[1,3] ([1]*Grad. Sch. Sci., Kanazawa Univ.*, [2]*RIBM Co., Ltd.*, [3]*Bio-AFM FRC., Kanazawa Univ.*)

3Pos292 脂質膜の曲率に依存したタンパク質-脂質膜の相互作用の直接観察のための高速 AFM 用基板の開発
Development of HS-AFM substrate for observation between proteins and lipid membrane depending on the physical shape of lipid membrane
Takahiro Toyoda[1], Shin'nosuke Yamanaka[1], Akane Goto[1], Hiroki Watanabe[2], Shunsuke Shozui[1], Mikihiro Shibata[1,3], Takayuki Uchihashi[1,3] ([1]*Dept. of phys., Kanazawa Univ.*, [2]*RIBM Co., Ltd.,*, [3]*Bio-AFM FRC., Kanazawa Univ.*)

3Pos293 長時間１蛍光分子追跡法による接着班分子の動的リクルートの解明
Transient recruitment of focal adhesion molecules revealed by super-long single molecule tracking
Taka-aki Tsunoyama[1], Kenichi G.N. Suzuki[2,3], Takahiro K. Fujiwara[2], Akihiro Kusumi[1,2,4] ([1]*OIST*, [2]*Inst. Integrated Cell-Material Sciences (WPI-iCeMS), Kyoto Univ.*, [3]*NCBS/inStem, India*, [4]*Inst. Frontier Medical Sciences, Kyoto Univ.*)

3Pos294 **個々の酵母細胞内 ATP 濃度の可視化により明らかになった、酸化ストレス下における細胞内 ATP 濃度の低下**
In vivo imaging of cytoplasmic ATP in living yeast cells reveals a profound effect of oxidative stress on ATP level
Masak Takaine[1,2], Hiromi Imamura[3], Satoshi Yoshida[1,2] ([1]*Gunma Univ. Initiative for Adv. Res.*, [2]*Gunma Univ. Inst. for Mol. and Cell. Regulation*, [3]*Lab. of Funct. Biol., Grad. Sch. of Biostudies, Kyoto Univ.*)

3Pos295 **PDLIM2 の相互作用タンパク質 MKRN2 は、NF-κB の p65 サブユニットに対する新規ユビキチン E3 リガーゼとして機能する**
PDLIM2-interacting protein MKRN2 functions as a novel E3 ligase for p65 subunit of NF-κB
Chanyoung Shin[1,2], Yuma Ito[1], Makio Tokunaga[1], Takashi Tanaka[2], **Kumiko Sakata-Sogawa**[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*IMS, RIKEN*)

3Pos296 **リポ多糖刺激における炎症抑制タンパク質 PDLIM2 活性化の生細胞イメージング定量解析**
Live cell imaging and quantitative analysis of anti-inflammatory protein PDLIM2 activation upon LPS stimulation
Shota Ichikawa[1], Yuma Ito[1], Takashi Tanaka[2], Makio Tokunaga[1], Kumiko Sakata-Sogawa[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*IMS-RCAI, RIKEN*)

3Pos297 **mRNA の一分子追跡によるストレス顆粒形成初期機構の調査**
Investigating initiation mechanism of stress granule formation by tracking single mRNA particles
Masamichi Imaseki[1], Ko Sugawara[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm Sci., Univ. of Tokyo*, [2]*JST, PRESTO*)

3Pos298 **高速近接場光学顕微鏡の開発**
Development of high-speed near-field optical microscopy
Takayuki Umakoshi[1], Shingo Fukuda[2], Takayuki Uchihashi[1,2,3], Toshio Ando[1,2,3] ([1]*Bio-AFM FRC, Kanazawa Univ.*, [2]*Coll. Sci. & Eng., Kanazawa Univ.*, [3]*CREST-JST*)

3Pos299 **転写伸長制御に関わるタンパク質動態のイメージング定量解析**
Quantitative image analysis of dynamics of promoter-proximal pausing related proteins
Shinnosuke Kunimi, Yuma Ito, Yuki Yamaguchi, Kumiko Sakata-Sogawa, Makio Tokunaga (*Sch. Life Sci. Tech., Tokyo Inst. Tech.*)

3Pos300 **大気圧電子顕微鏡を用いた分泌腺組織の水中観察**
Secretory glands imaged in aqueous solution by atmospheric scanning electron microscopy (ASEM)
Toshiko Yamazawa[1], Naotoshi Nakamura[2], Mari Sato[3], Chikara Sato[3] ([1]*Dept. Mol. Physiol., Jikei Univ. Sch. Med.*, [2]*Dept. Statistical Genetics, Kyoto Univ.*, [3]*Biomed. Res. Inst., AIST*)

3Pos301 **ベイズ推定を用いた透過型電子顕微鏡画像の CTF 補正の自動化**
Development of automated CTF correction of transmission electron microscopic images using the Bayesian estimation
Koji Hisanaga, Takuo Yasunaga (*Kyushu Institute of Technology Graduate School of Computer Science and System Engineering*)

3Pos302 **様々な生物種の温度測定に利用でき且つ速い温度変化を測定可能な蛍光性温度プローブタンパク質**
Genetically encoded ratiometric fluorescent thermometer with wide temperature range and rapid response
Masahiro Nakano[1], Yoshiyuki Arai[1], Ippei Kotera[2], Kohki Okabe[3,4], Yasuhiro Kamei[5], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ.*, [2]*RIES, Hokkaido Univ.*, [3]*Grad. Sch. Pharma., Univ. Tokyo*, [4]*JST, PRESTO*, [5]*NIBB*)

3Pos303 **新規誘電率顕微鏡(SE-ADM)による生きたそのままの細胞の液中ナノスケール観察**
Nanoscale observation of intact living cells in a medium with low radiation damage using scanning electron-assisted dielectric microscopy
Tomoko Okada, **Toshihiko Ogura** (*Advanced Industrial Science and Technology (AIST), Biomedical Research Institute*)

3Pos304 **Improvement of photostability of fluorescent dyes by using lanthanide ions**
Takuma Imoto[1], Shin Mizukami[2], Kazuya Kikuchi[1,3] ([1]*Grad. Sch. Eng, Osaka Univ.*, [2]*IMRAM, Tohoku Univ.*, [3]*IFReC*)

3Pos305 **Optical measurement of diffusion and pH in nanopores of protein crystals**
Kazuo Mori, Bernd Kuhn (*OIST*)

バイオエンジニアリング / Bioengineering

3Pos306 **アポフェリチンを用いた複合(MRI 造影・発光)希土類ナノ粒子の作製**
Synthesis of rare earth hybrid nanoparticles in the apoferritin cavity
Keita Kontani, Hideyuki Yoshimura (*Meiji University*)

3Pos307 **標的遺伝子高感度検出に向けた自己組織化単分子膜修飾金ナノ粒子の分散安定化**
Optimized modification of SAMs for suppression of non-specific binding gold nanoparticles for High-sensitivity Target Genetic Assay
Keiko Esashika, Takaha Mizuguchi, Toshiharu Saiki (*Sci. Rech., Keio Univ.*)

3Pos308 **血中循環腫瘍細胞を測定するためのサイズ分画機能を備えた画像認識型セルソーターの開発**
Development of Size Classifying Imaging Cell Sorter for Identifying of Circulating Tumor Cells
Moe Iwamura[1], Masao Odaka[2], Kenji Matsuura[2], Akihiro Hattori[2], Hideyuki Terazono[2], Kenji Yasuda[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*)

3Pos309 **血中循環腫瘍細胞を無染色で識別するためのオン・チップ高機能画像認識型細胞分取装置の開発**
Development of Functional On-Chip Imaging Cell Sorter for Identification of Non-Labeled Circulating Tumor Cells
Masao Odaka[1], Akihiro Hattori[1], Kenji Matsuura[1], Hideyuki Terazono[1], Moe Iwamura[2], Kenji Yasuda[2] ([1]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*, [2]*Dept. Physics, Waseda Univ.*)

3Pos310 **ゲル媒質中で反応拡散系によるパターン形成を行う DNA 論理ゲート**
DNA logic gate performs Reaction-Diffusion Pattern formation in gel medium
Keita Abe[1], Ibuki Kawamata[2], Shin-ichiro M. Nomura[2], Satoshi Murata[2] ([1]*Dpt. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Eng., Tohoku Univ.*)

3Pos311 **電極埋め込み型ナノポアのACゲート電位によるDNAの挙動制御**
DNA motion and translocation controlled by nanopore with embedded gate electrode
Naoto Sakashita, Yuta Kato, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

3Pos312 ***In vitro* selection of novel peptide agonists for human somatostatin receptor subtype-2 using water-in-oil microdroplets**
Takashi Sakurai[1], Ryo Iizuka[1], Yasuyuki Nakamura[2], Jun Ishii[3], Akihiko Kondo[2], Ayaka Iguchi[4], Dong H. Yoon[4], Tetsushi Sekiguchi[5], Shuichi Shoji[4], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Grad. Sch. of Eng., Kobe Univ.*, [3]*Org. of Adv. Sci. and Technol., Kobe Univ.*, [4]*Dept. of Nanosci. and Nanoeng., Waseda Univ.*, [5]*Res. Org. for Nano & Life Innov., Waseda Univ.*)

3Pos313 **ナノポア計測の周波数解析による複数種類のmicroRNAのパターン認識**
Pattern Recognition for MicroRNA Expressions by using Fourier Analysis on Nanopore Sensing
Akihiro Tamotsu[1], Moe Hiratani[2], Masayuki Ohara[2], Ryuji Kawano[3] ([1]*Tokyo Univ. of Agri. and Tech. Dept. Biotech. and Life Sci.*, [2]*Tokyo Univ. of Agri. and Tech. Dept. Biotech. and Life Sci.*, [3]*Tokyo Univ. of Agri. and Tech. Dept. Biotech. and Life Sci.*)

3Pos314 **DNAを用いたナノ粒子3Dプリンタの実現**
Nanoparticle 3D Printing by DNA Bonding
Yuki Sakamoto, Shoichi Toyabe (*Grad. Sch. Eng. Applied Physics, Univ. Tohoku*)

3Pos315 **DNAナノ構造のDNAハイドロゲルへの繰り返し電子解離／会合**
Repeatable electronic dissociation/association of DNA nanostructures on DNA hydrogels
Keitel Cervantes, Ibuki Kawamata, Shin-Ichiro Nomura, Satoshi Murata (*Tohoku University*)

3Pos316 **作製済みリポソームへのマイクロピペットの穿刺**
Microinjection into already made liposome
Shota Sato, Shin Yoshida, Tomoyuki Kaneko (*LaRC, FB, Hosei Univ.*)

3Pos317 **Quantification of intersample differences in T cell populations**
Ryo Yokota[1], Yuki Kaminaga[2], Tetsuya Kobayashi J.[1,2] ([1]*Institute of Industrial Science, the University of Tokyo*, [2]*School of Engineering, The University of Tokyo*)

3Pos318 **多チャンネル局所化学刺激システムの開発**
Development of the multi-channel local chemical stimulation system
Masaru Kojima[1], Tatsuoya Furusawa[1], Hajime Fukuoka[2], Yasushi Mae[1], Tatsuo Arai[1] ([1]*Grad. Sch. Eng. Sci., Osaka Univ.*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*)

3Pos319 **多電極電位計測システムを用いた心筋細胞に対するテルフェナジンの影響**
Effect of Terfenadine to cardiomyocytes on multi electrode array system
Mitsuki Maruyama, Tomoyuki Kaneko (*LaCR, Hosei Univ.*)

1SAA-01 「使える」膜電位感受性色素による神経回路解析法 "Conventional" voltage sensitive dye imaging of neural circuit activity

Takashi Tominaga, Yoko Tominaga (*Inst. Neurotic., Tokushima Bunri Univ.*)

Synthetic voltage-sensitive dyes (VSDs) were developed to acquire fast membrane potential changes in the excitable membrane using optical measurement devices as early as the 1970s. Soon, developments of large-scale imaging device enabled us to visualize the activity of neural circuits. However, simple wide field VSD imaging methods have been for a long time a technique used in only very few laboratories until the late 1990s, mostly because of its poor signal noise ratio (SNR). In this talk, I will introduce how we solved the technical difficulties so that VSD imaging became a "conventional" method. I will present some examples including an application to analyze "circuit deficits" such as autism spectrum disorder (ASDs).

1SAA-02 Tuning Genetically-Encoded Voltage Indicators to Better Resolve Different Types of Neuronal Activity

Bradley Baker (*KIST*)

The signal-to-noise ratio of Genetically-Encoded Voltage Indicators (GEVI) are still low making resolution of action potentials from subthreshold, synaptic potentials difficult when imaging populations of neurons. To better resolve these different types of neuronal activity, we have created GEVIs that vary in their voltage-dependency. Probes with a V1/2 (the voltage that corresponds to 50% of the total fluorescence change) near 0 mV exhibit better optical resolution of action potentials than probes with a V1/2 at -30 mV. Preliminary progress has also been made to restrict the optical signal to hyperpolarization which may lead to a GEVI that optically reports only neuronal inhibition.

1SAA-03 新規偏光顕微鏡を用いたマウス海馬スライスにおけるシナプス活動の非侵襲的計測 Imaging of neuronal activity in mice hippocampal slices by instantaneous polarized light microscopy

Maki Koike-Tani[1], Shalin Mehta[1], Rudolf Oldenburg[1], Takashi Tominaga[2], Tomomi Tani[1] ([1]*Marine Biological Laboratory*, [2]*Tokushima Bunri University*)

Intrinsic optical signals have been used to map patterns of brain activity in a non-invasive manner. These signals refer to changes in the transmittance/reflectance of neuronal tissue that are related to secondary results of neuronal activity in the brain. We have been developing a cutting-edge polarized light microscope system that detects changes in the structural signatures of neuronal cells that directly associate with the activity of neurons. We have monitored reproducible patterns of polarization signals in mouse hippocampal slices in response to electrical stimulation. Our results suggest that polarization signals are related to postysynaptic glutamate responses, followed by unidentified structural changes of neuronal tissue at sub-cellular level.

1SAA-04 光活性化酵素制御とイメージング技術による cAMP/cGMP の時空間的機能探索 Two-photon optogenetic control and live imaging of postsynaptic cAMP/cGMP intracellular messengers

Kenichi Okamoto (*LTRI, MSH*)

cAMP and cGMP are ubiquitous second messengers with a variety of essential physiological roles such as synaptic plasticity. Pharmacological reagents and genetic manipulations have been used to study their functions, however, these approaches are limited in spatial and temporal specificity. To study the spatiotemporal dynamics and roles of cAMP/cGMP at synapses within brain tissue, we introduced two-photon live imaging and optogenetics to monitor and locally manipulate second messenger levels by genetically encoded FRET/FLIM sensors and light-sensitive enzymes that synthesize/hydrolyze cAMP/cGMP. We applied these tools from the single synapse to the hippocampal circuit level and revealed novel rapid cAMP/cGMP functions including crosstalk in synaptic plasticity.

1SAA-05 in vivo calcium imaging with genetically encoded calcium indicators

Junichi Nakai[1,2], Keiko Gengyo-Ando[1,2], Masaaki Sato[1,2], Masamichi Ohkura[1,2] ([1]*Grad. Sch. Sci. Eng. Saitama Univ.*, [2]*BBSSI, Saitama Univ.*)

Calcium ions play important roles in neurons and glial cells. In neurons both pre- and post-synaptic cells require intracellular calcium increase for synaptic transmission and plasticity such as learning and memory. To elucidate neuronal mechanism, we have been developing genetically encoded calcium indicators (GECIs). G-CaMP is a GECI constructed from a single green fluorescent protein. When we expressed G-CaMP7 in the mouse hippocampus, we were able to image calcium signals from the cell bodies of CA1 pyramidal neurons in vivo. We also generated improved versions of red GECIs (R-CaMPs). We expressed R-CaMP and G-CaMP in the neuromuscular system in Caenorhabditis elegans and successfully performed dual color calcium imaging of freely behaving C. elegans.

1SAA-06 Exploring input-output relations of neurons in awake mice

Christopher J. Roome, **Bernd Kuhn** (*Kuhn Unit, OIST*)

Measuring input-output relations of single neurons in vivo is very important for understanding brain function. Here we combine two-photon microscopy and electrophysiology to simultaneously measure dendritic voltage and calcium signals and somatic output from Purkinje cells (PC) in awake mice. To record dendritic voltage we labelled single PCs with the synthetic, pure electrochromic voltage sensitive dye ANNINE-6plus. Dendritic calcium was recorded with the genetically encoded calcium indicator GCaMP6f. We record spatio-temporal maps of dendritic activity and correlate this activity with extracellular recordings of the PC soma. We show how coincident input to the PC dendrite by climbing fibers and background activity permits spatial modulation of dendritic spiking.

1SBA-01　全身・全脳透明化の先に見えてくるもの～生命の『時間』の謎の解明に向けて～

Toward Organism-level Systems Biology in Mammals～ Whole-body and whole-organ clearing and imaging with single-cell resolution～

Hiroki R. Ueda[1,2] ([1]*The University of Tokyo*, [2]*RIKEN (QBiC)*)

Organism-level systems biology can be accelerated by whole-body or whole-organ analysis with single-cell resolution, in order to identify, analyze, control and design cellular circuits in organisms. In this talk, I introduce the advancements in whole-body and whole-organ clearing and imaging, and discuss how combining new clearing techniques with high performing fluorescent proteins, rapid volume imaging and efficient image informatics is resulting in comprehensive and quantitative organ-wide single-cell-resolution experimental data. These technologies are starting to yield information on connectivity and dynamics in cellular circuits at unprecedented resolution, and bring us closer to system-level understanding of physiology and diseases of complex mammalian systems.

1SBA-02　4K/8K CMOS イメージングによるマルチスケール生体全細胞解析

Multi-scale in vivo 4K/8K imaging analysis

Satoshi Nishimura[1,2] ([1]*Jichi Med. Univ.*, [2]*The Univ. of Tokyo*)

Real-time, multi-colour 4K/8K XYZT imaging enabled us to visualize single cell behaviour in living animals, and elucidate “minor” and “major” populations. We also used light-manipulation technique, to reproduce thrombus or inflammation reactions, and revealed molecular mechanisms of cardiovascular disease. We revealed platelet heterogeneity from their morphology, and tight association between platelet age and activating status. Remarkable transient neutrophil accumulation was followed by spontaneous cell death and monocyte recruitment.
In sum, we developed multi-scale imaging system which can evaluate the therapeutic strategies against adult-common disease, and also found contribution of “minority” cells to major phenotype.

1SBA-03　マイノリティ細胞の同定と解析による自己免疫疾患発症制御機構の解明

Elucidation of pathomechanisms of autoimmunity by minority cell research

Taku Okazaki (*Division of Immune Regulation, Institute for Genome Research, Tokushima University*)

The activation of T cell, one of the major cell types in acquired immunity, is primarily regulated by antigen-receptor signal. Stimulatory and inhibitory co-receptors modulate the quality and quantity of antigen-receptor signal to optimize immune responses against pathogens while limiting immune responses to self. Because of the recent success of tumor immunotherapy targeting an inhibitory co-receptor PD-1, co-receptors attract high attention of both clinicians and scientists. However, the precise mechanisms how inhibitory co-receptors regulate auto-reactive or tumor-specific T cells are poorly understood. The identification of target cells for inhibitory co-receptors is expected to reveal the complex regulatory mechanisms of autoimmunity and tumor-immunity.

1SBA-04　Raman spectroscopic approaches to label-free cell characterization and finding functional minorities

Katsumasa Fujita (*Osaka University*)

Raman spectroscopy has been utilized for analysis and investigation of materials in various research fields. Since the vibration of molecules can be detected as Raman scattering spectrum, the information of sample molecules and their environments can be monitored without labelling. Recently, we have combined Raman spectroscopy and optical microscopy for label-free imaging of living cells. With rough categorization of intracellular molecules, such as proteins and lipids, by Raman spectra, we have visualized the cell activities, such as cell division, apoptosis and differentiation without labelling. Since Raman spectroscopy can provide comprehensive information of intracellular molecules, it can be a tool to characterize cells and find the functional minorities.

1SBA-05　マイノリティ細胞研究にむけた神経細胞および脳組織内在性グルタミン酸受容体の蛍光可視化

Visualization of native glutamate receptors in live neurons or neuronal tissues for minority cell study

Shigeki Kiyonaka (*Grad. Sch. Eng., Kyoto Univ.*)

Genetically engineered fluorescent proteins are conventionally utilized for visualizing neurotransmitter receptors. However, genetic manipulation is required in most case, which may induce undesirable effects on the receptor properties. Here, we report a powerful method for visualizing native AMPA-type glutamate receptors (AMPARs) using chemical labeling reagents. High penetrability of our reagents allowed to visualize native AMPARs deep in brain tissues without affecting the receptor functions. Moreover, our methods successfully visualized diffusion dynamics of endogenous AMPARs not only in cultured neurons but also in hippocampal slices. Thus, This novel method will help clarify the localization of native AMPARs, which will be utilized for minority cell study.

1SBA-06　超解像生理機能イメージング法の開発とマイノリティ細胞の可視化の試み

Development of superresolution techniques for imaging physiological functions toward visualization of minority cells

Takeharu Nagai (*ISIR, Osaka Univ.*)

If we carefully observe the cell population that at first glance looks uniform and homogeneous, we may find small number of heterogeneous cells with a different nature. Moreover, this minority cells would sometimes significantly alter the behavior of the whole cell population. In this symposium, I would like to discuss 1) possible mechanism by which variety output could be produced even in the cells with identical biomolecular reaction networks, and 2) development of superresolution techniques for imaging physiological function at nano-meter scale, which is indispensable for visualization of minority cells.

1SBA-07 Finding genomic minority cells by sequencing

Katsuyuki Shiroguchi[1,2,3] ([1]*RIKEN Quantitative Biology Center*, [2]*RIKEN Center for Integrative Medical Sciences*, [3]*JST PRESTO*)

In biological systems, there are genomic minority cells, for example, in cancer cells, T/B cells, and microbiome in the intestine. These minority cells may affect states of higher biological layers, such as cell populations, tissues, or organisms. In order to identify the minority cells, high throughput single cell analysis is required. We have been developing an automatic cell barcoding method to perform sequence-based high throughput single cell analysis which enables indentification of each cell with single base resolution of target genomic sequences, and quantification of cell-number distribution. I would like to introduce our method including computer analyses.

1SBA-08 1細胞ラマン分光イメージングから如何にして細胞の個性を定量化するか？

How can one quantify cell individuality from Single Cell Raman Imaging?

Tamiki Komatsuzaki[1,2] ([1]*Hokkaido Univ., RIES, MSC*, [2]*Hokkaido Univ., Grad. Sch. Life Sci.*)

Minority over single cells acquires distinct functions even with the same genome such as persister cells. However, how can one quantify the state or individuality of single cells? Single cell Raman imaging contain rich information about the cell as a whole about the components that comprise the cell such as organelle and its components. Due to the inherently weak signals in Raman microscopy, it is essential that the data science should take into account the existence of large noise fluctuations and yet quantify the phenotypic state of the cells. Here we present our recent information theoretic approach to identify components of the cell such as the cell membrane, nucleus, etc. from their Raman spectra, which should shed light on defining the state of cells.

1SDA-01 蛋白質―無秩序な原子の動きを"命の動き"に変えるデバイス

Proteins-converting random motions of atoms into the dynamism of life

Kazuyuki Akasaka (*Kyoto Prefectural University*)

The complexity and diversity of life has fascinated and puzzled humans throughout the history, producing philosophies and religions, which are still shaping the world today. By now however, as we have reached the molecular and atomic levels of bioscience, our view on life must have changed dramatically to a new one acceptable to all humans irrespective of their backgrounds. Here, we biophysicists have the role for initiating the change. In this context, how we reconcile between the "simple and uniform" world of atoms and the dynamic and diverse reality of all lives on earth? I find the key in proteins, in which the random, thermal motions of atoms are elegantly converted into specific motions of atoms for function by the action of life itself.

1SDA-02 分子と細胞、そして細胞と組織をつなぐメカニカルシグナル

Mechanical signals interface molecules with cells, and cells with tissues

Masahiro Sokabe (*Nagoya Univ. Grad. Sch. Med.*)

Organisms are multi-scale hierarchical systems formed of molecular to individual layers. Life is realized by interactions among physical elements within a layer and those between the layers. Dynamics of molecules creates living cells, while cells dynamics regulate molecular behaviors. Yet the underlying logic is unclear due to lack of knowledge on the robust visualizable structures realizing those interactions. Emerging mechanobiology aims at unraveling the logic of life based on force signals generated by mechanical interactions within and between the layers, being able to analyze them through visible mechanical structures; dynamics of actin cytoskeletons create cell shape, while deformation of cells caused by cell-cell mechanical interactions regulate actin dynamics.

1SDA-03 分子情報システムとしての生命

Which parameters characterize "life"?

Shigeki Mitaku (*Science writer*)

The most significant features of living things are their very large diversity together with the state of "life", universal to whole biological kingdom. 'Which parameters characterize "life"?' is a good open question but has never been answered yet. Such parameters should have several characteristics. (1) They must be written in genome DNA sequences. (2) The formation and stability of "life" are designed by the whole genome and not by small number of genes. Here, I discuss the computational methods to develop the universal parameters and show its candidate. I also discuss the relationship between the reductionism and holism in biological science.

1SDA-04 情報進化―原子といのちを結ぶ進化能的生命観―

Informational Evolution: An evolvability view point of life composed of atoms

Yuzuru Husimi (*SOKENDAI*)

The form of information and communication in a system is closely related to the evolvability of the system. Origins of life can be regarded as the first informational revolution in our universe. At that moment, an encoded digital information system was introduced in the molecular world, and the evolvability of the molecular system became drastically higher. According to the RNA World hypothesis, the main player of this event was RNA. Replacement of genetic RNA by dsDNA made the homologous recombination possible, and made the evolvability even higher. Deletion of an oxygen atom innovated the form of information. Such an informational evolution has been a front-runner throughout material, biological, human and cultural evolution, based on its effect on evolvability.

1SEA-01　多細胞性緑藻ボルボックスの走光性：5000 の細胞が協調して泳ぐには？

Phototaxis in the multicellular green alga *Volvox*: How 5000 independent cells coordinate their motion?

Noriko Ueki (*CLS, Tokyo Tech.*)

The multicellular, spheroidal green alga *Volvox* shows remarkable phototaxis, though there are no direct interactions between ~5,000 cells. How *Volvox* accomplishes phototaxis was studied by motion analysis of its flagella upon photoreception. Pulsed light was applied to an immobilized spheroid to mimic the light fluctuation perceived by each cell during rotational swimming. Upon each light stimulus, the direction of flagellar effective strokes was reversed. This response was the greater in the cells closer to the anterior pole. This suggests that flagella reversal of cells facing the light source in the anterior hemisphere causes phototactic turning. Transition of the phototaxis mechanism from unicellular *Chlamydomonas* to multicellular *Volvox* will also be discussed.

1SEA-02　繊毛の運動を支える細胞内構造の理解へ向けて

Towards understanding of cell structure that governs motion pattern of motile cilia

Kyosuke Shinohara (*Tokyo University of Agriculture and Technology*)

Motile cilia plays important role on transport of fluid in body. Disruption of function of motile cilia leads to hydrocephalus, bronchitis, and infertility. Thus, it is important to understand mechanism of motility of cilia. Here we have examine the principle that governs motion pattern of motile cilia. We show some key genes involved in cytoskeleton and axonemal structure in motile ciliated cells. To address physiological role of the genes, we have examined phenotype of knockout mice.

1SEA-03　Roles of calcium in the regulation of sperm flagellar movement

Kogiku Shiba (*SMRC, Tsukuba Univ.*)

Ca^{2+} is known to play key roles in the regulation of flagellar waveforms. However, the molecular mechanism of how symmetric-asymmetric waveforms are regulated by Ca^{2+} is still unclear. By real-time intraflagellar Ca^{2+} imaging and detailed analysis of flagellar bending, we revealed the process of flagellar waveform conversion of sperm in response to transient increase of intracellular Ca^{2+} concentration during the chemotaxis in the ascidian *Ciona intestinalis*. A protein named calaxin was shown to directly regulate a dynein motor activity in a Ca^{2+}-dependent manner. Inhibition of calaxin causes the suppression of continuous asymmetric flagellar waveform required for chemotactic turn. These results suggest that calaxin plays a role in sustaining the asymmetrical waveform.

1SEA-04　IFT81 および IFT74 の N 末端領域によるチューブリンの鞭毛内輸送

The IFT81 and IFT74 N-termini together form the main module for intraflagellar transport (IFT) of tubulin

Tomohiro Kubo[1,4], Jason Brown[1,2], Karl Bellve[1], Branch Craige[1], Julie Craft[3], Kevin Forgarty[1], Karl Lechtreck[3], George Witman[1] ([1]*UMASS Med.*, [2]*Salem State Univ.*, [3]*Univ. of Georgia*, [4]*Univ. of Yamanashi Faculty of Medicine*)

Intraflagellar transport (IFT) is a bidirectional movement of protein particles along cilia/flagella. *In vitro* experiments have suggested that both IFT81 and IFT74 N-termini are important for IFT of tubulin. To test this hypothesis *in vivo*, we made *Chlamydomonas* mutants, *IFT81(5E)* and *IFT74Δ130*, that possess version of IFT81 or IFT74 respectively in which the predicted tubulin-binding site was modified. TIRF microscopy revealed that frequencies of IFT-based tubulin transport are greatly reduced in *IFT81(5E)* and *IFT74Δ130* flagella. Importantly, the double mutant *IFT81(5E) IFT74Δ130* failed to build normal-length flagella. These results provide first *in vivo* evidence that the IFT81/IFT74 N-termini together form a main module important for IFT of tubulin.

1SEA-05　繊毛・鞭毛の中の動きを見る

Dynamics of molecules inside cilia and flagella

Daisuke Takao (*NIG*)

In addition to dynamic movement of cilia and flagella (hereafter collectively termed cilia), molecular dynamics in these organelles is important for their function. The inside space of the cilium is narrow and crowded, limiting diffusional transport of cytosolic molecules such as ATP. Besides the limitation, interestingly, there is no obvious restriction for entry of small molecules into the ciliary compartment. On the other hand, ciliary entry of large molecules is restricted and requires an active process, keeping the unique composition of the cilium. In this talk, I will show how the dynamic properties of molecules within the cilium affect physiological processes and how transport is regulated, particularly in terms of gated entry into the ciliary compartment.

1SEA-06　脊椎動物運動性繊毛における PIH タンパク質の機能解析

The function of PIH proteins in the vertebrate motile cilium

Hiroshi Yamaguchi[1,2], Yousuke Yamazaki[1], Toshiyuki Oda[1,3], Masahide Kikkawa[1], Hiroyuki Takeda[2] ([1]*Grad. Sch. Med., Univ. Tokyo*, [2]*Grad. Sch. Sci., Univ. Tokyo*, [3]*Grad. Sch. Med., Univ. Yamanashi*)

Axonemal dyneins are huge motor molecule complexes aligned in the cilium and drive ciliary movement. Their subunits are pre-assembled in the cytoplasm before transported into cilia, but the mechanism of this process remains elusive. Previous reports identified proteins with PIH (protein interacting with HSP90) domain as the regulators of this process, but not all of them are well characterized. We established zebrafish mutant lines of all four PIH genes using genome editing, and found the abnormal ciliary motilities and the structural defects of axonemal dyneins in each mutant. Interestingly, the degree of motility defects and the types of defects in axonemal dyneins vary among mutants, suggesting the distinct functions of PIH proteins in the ciliary motility.

1SFA-01 クロマチンの三次元構造と動的構造

Three dimensional structures and dynamics of chromatin

Hitoshi Kurumizaka (*Waseda University, Faculty of Science and Engineering*)

Eukaryotic genomic DNA is compacted as chromatin. Four core histones, H2A, H2B, H3, and H4, form a complex to which 150 base pairs of DNA are wrapped. Chromatin structure and its dynamics play essential roles in the epigenetic regulation of various DNA metabolisms. Nucleosomes are connected with linker DNAs, resulting in a "beads-on-a-string" appearance, which is folded into higher order chromatin architecture. However, the functional chromatin architecture has not been revealed yet. We have studied the structures and physical characters of mono- and poly-nucleosomes containing histone variants. I will show our recent results on the structural analyses of mono- and poly-nucleosomes, and discuss how the chromatin functions as an epigenetic regulator for genomic DNA.

1SFA-02 Hi-C データを用いた遺伝子発現制御の理解

Using Hi-C data to understand gene regulation

Mikita Suyama (*Medical Institute of Bioregulation*)

The Hi-C method has been successfully applied to unveil genome-wide long-range interactions such as those between distant enhancers and promoters. I will briefly introduce how the data extracted from Hi-C analyses can be applied to the study of gene regulation. Examples of long-range interactions that can be explained by 3D chromosome conformation will also be presented together with the ChromContact web server that we recently constructed to utilize the Hi-C data.

1SFA-03 Waves of chromatin remodeling in mouse dendritic cells in response to LPS stimulation

Alexis Vandenbon (*IFReC, Osaka University*)

Causal relationships between dynamics in transcription factor (TF) binding, epigenetic markers and gene expression during the response to stimuli remain unclear. Here, we found that several histone modifications became induced at regulatory regions within well-specified time frames after stimulation of dendritic cells with lipopolysaccharide. These time frames appeared to be independent of the timing of transcriptional induction of nearby genes, but coincide with activation times of stimulus-activated TFs. Our results shed some light on the question of causality between TFs and epigenetics.

1SFA-04 出芽酵母 *yku70 esc1* 変異型における遺伝子発現の変化を引き起こすメカニズム

Mechanisms for the misregulated gene expression in the *yku70 esc1* mutant of budding yeast

Naoko Tokuda, Masaki Sasai (*Nagoya University*)

To investigate the relationship between the nuclear-scale chromosomal organization and gene expression, we have developed a 3-dimensional dynamical simulation model of genome of interphase budding yeast by using the Hi-C data (Tokuda, N., and Sasai, M., 2016 in preparation). With this simulation model, we investigated the reasons for the experimentally observed misregulation of 60 genes in the *yku70 esc1* mutant (Taddei et al. Genome Res., 2009) by examining the mutational modification of spatial distribution of genes in nucleus.

1SFA-05 刺激された血管内皮細胞における核内構造のダイナミクス

Dynamics of chromatin structure in stimulated vascular endothelial cells

Youichiro Wada[1,2], Youichi Nakata[3], Yoshihiro Ohta[3], Sigeo Ihara[2,3] ([1]*Isotope Science Center, The University of Tokyo*, [2]*Research Center for Advanced Science and Technology, The University of Tokyo*, [3]*Graduate School of Mathematical Sciences, The University of Tokyo*)

Vascular endothelial cells are exposed to a variety of stimulations, and change their chromatin structures responding to bioactive mediators in order to achieve effective transcriptional regulation. We have preformed experiments using TNF alpha stimulated human endothelial cells and obtained data in time course manner. Based on chromatin interactome/transcriptome data and using mathematical calculation to minimize potential energy, we established a simulation model of active transcription.

1SGA-01 序論

Introduction

Satoshi Akanuma (*Faculty of Hum. Sci., Waseda Univ.*)

A number of protein subunits form oligomeric structures that play important roles in biological activities. Recently, some researchers successfully created artificial protein oligomers through engineering protein-protein interactions. Oligomerization of protein subunits often sequesters the otherwise exposed hydrophobic surfaces. Because exposed hydrophobic surface reduces protein's solubility and/or induces aggregate formation, forming a desired complex and forming undesired aggregates are two sides of the same coin. In this symposium, speakers will talk about recent progress of their studies focusing on protein's solubility, solubilization of recombinant proteins, designing of artificial protein complexes and controlling amyloidogenic aggregation.

1SGA-02 脂質膜のアミロイド線維形成への影響

The effects of lipid membranes on the fibrillation of amyloidogenic proteins

Mayu S. Terakawa (*Weill Cornell Medicine, Biochemistry*)

Amyloid fibrillation is related to various deleterious neurodegenerative diseases. Recent studies have focused on the interactions between amyloidogenic proteins and membranes without delineating its effect on the fibrillation. Here, I show the effect of membranes on the fibrillation of amyloidogenic proteins, especially amyloid β and α-synuclein. In the case of amyloid β, the membrane curvature plays significant roles in accelerating the fibrillation. On the other hand, in the case of α-synuclein, compositions and concentrations of lipids are of importance for promotion and inhibition of the fibrillation. Together, I propose that the protein interaction with membranes affects the fibrillation in a protein-dependent manner, which regulates their amyloidogenesis.

1SGA-03 新規タンパク質分子間結合面の創成と人工タンパク質繊維の作成

De-novo design of a protein-protein interface and creation of protein fibrils

Sota Yagi[1], Satoshi Akanuma[2], Tatsuya Uchida[3], Akihiko Yamagishi[1] ([1]*Tokyo Univ. Pharm. Life Sci., Dep. Appl. Life Sci.*, [2]*Waseda Univ., Facul. Hum. Sci.*, [3]*Tokyo Univ. Pharm. Life Sci., Dep. Mol. Life Sci.*)

We constructed an artificial interaction between two helical bundle proteins, sulerythrin and LARFH, through designing an inter-molecular helix-helix interaction. For the design of the interface, leucine residues were introduced onto the helices of these proteins. Additionally, charged residues were placed around the leucine patch to avoid aggregation. The sulerythrin and LARFH variants interact to each other. Then, we created the same interfaces to both ends of sulerythrin and LARFH. By mixing the resulting mutants, fibrous structures were observed by atomic force microscopy. Our method may be applicable to create new biomaterials.

(1) Yagi et al. (2014) BBA Proteins and Proteomics 1844, 553-560
(2) Yagi et al. (2016) BBA Proteins and Proteomics 1864, 479-487

1SGA-04 人工タンパク質をブロックに見立てた超分子ナノ構造複合体の設計構築

Design and construction of supramolecular nanostructures by using *de novo* protein nanobuilding blocks

Naoya Kobayashi, Naoya Kimura, **Ryoichi Arai** (*Appl. Biol., Tex. Sci. & Tech., Shinshu Univ.*)

Recently, we designed and created a protein nanobuilding block (PN-Block), WA20-foldon, by fusing an intermolecularly folded dimeric *de novo* protein WA20 and a trimeric foldon domain of T4 fibritin (*JACS*, 2015). The WA20-foldon formed self-assembling polyhedral nanostructures in multiples of 6-mer. In addition, we constructed *de novo* extender protein nanobuilding blocks (ePN-Blocks) by tandemly fusing two WA20s. The ePN-Blocks form several homooligomeric states, probably circular chain structures. Then, we reconstructed heteromeric complexes from extender and stopper PN-Blocks by denaturation and refolding. These results demonstrate that the PN-Block approach using the *de novo* proteins is a powerful strategy to create novel self-assembling supramolecular nanostructures.

1SGA-05 時間分解小角 X 線小角散乱法を用いたフェリチンの会合機構の解析

Ferritin assembly mechanism studied by time-resolved small-angle X-ray scattering

Daisuke Sato, Masamichi Ikeguchi (*Fac. of Sci. and Eng., Soka Univ.*)

Escherichia coli ferritin (Ftn) is a spherical shell-shaped protein consisting of 24 identical subunits arranged with 4/3/2 symmetry. Ftn dissociates into 2-mers at acidic pH and can reassemble into the native 24-mer when pH increases. To clarify Ftn assembly mechanism, we employed a time-resolved small-angle X-ray scattering (TR-SAXS) method. SAXS change during the reaction was roughly explained by a simple model in which only 4, 6 and 12-mers were considered as intermediates. To assess influence of the subunit net charge on Ftn assembly rate, we prepared mutants with different net charges. Although the subunit net charge was an important factor for determining Ftn assembly rate at low ionic strengths, it was not significant at high ionic strengths.

1SGA-06 ペプチド溶解性の全原子分子動力学シミュレーション及びその実験的検証

Large scale molecular dynamics of peptide solubility and its experimental assessment

Yutaka Kuroda (*Dept. Biotech. Life Sci., TUAT*)

We analyzed amino acids solubility using 100 ns molecular dynamics simulations of systems containing 27 tetra-peptides composed of a single amino acid type and ~3x104 water molecules. The calculations were performed for all natural amino acids except cysteines and glycines using AMBER 8 with standard force field on a special purpose MD-GRAPE 3 computer, and without introducing any “artificial” hydrophobic interactions. Tetra-peptides composed of hydrophobic amino acids formed large peptide clusters, and those containing D, E, K, and R did not aggregate at all. Overall, this very first all-atom molecular dynamics simulation of multi-peptide systems appeared to reproduce the basic properties of peptide solubility, essentially in line with experimental observations.

1SGA-07 熱測定による高温で可逆的に形成される蛋白質の会合体の検出

High-temperature reversible oligomerization of proteins detected by calorimetry

Shun-ichi Kidokoro[1], Shigeyoshi Nakamura[1,2] ([1]*Dept. Bioeng., Nagaoka Univ. Tech.*, [2]*Dept. Creat. Eeng., Natl. Inst. Tech. Kitakyushu College*)

We performed differential scanning calorimetry (DSC) and pressure perturbation calorimetry (PPC) of the thermal transition of an acidic molten globule state (MG1) of cytochrome *c* at concentrations from 0.5 to 18.2 mg/ml. DSC profiles are highly reversible and showed clear protein-concentration dependence, indicating that a reversible oligomerization process occurs during the thermal transition. These DSC and PPC data were found to be rationalized by a six-state model, including three monomeric states: MG1, MG2, and denatured state, and three oligomeric states: dimer, trimer, and tetramer. The thermodynamic properties and some other examples of the similar oligomerization will be discussed.

1SGA-08 終わりに
Concluding Remarks

Fumio Arisaka (*Nihon U. Biores. Sci.*)

Oligomerization or association of proteins can be either specific or non-specific. Many proteins form specific oligomers or huge polymers in order to attain more sophisticated functions. Proteins may aggregate upon denaturation non-specifically to form amorphous aggregates. On the other hand, proteins may associate not specifically, but not totally randomly. This kind of association may be called "pseudo"-specific, which includes crystallization and amyloid formation, both of which contain regular repeating units. The two forms of association, however, is different in that the former is reversible and the latter is irreversible. Curiously, many proteins can form amyloids as well as crystals. The mechanism of pseudo-specific association remains to be elucidated.

1SAP-01 Intercellular propagation of ERK activity orients collective cell migration

Kazuhiro Aoki (*OIIB, NIBB, Div. of Quantitative Biology*)

The collective cell migration refers to a movement of cell groups with physical and functional cell-cell connections, and involves an inherent process of embryonic development, wound healing and cancer invasion. The biophysical framework of collective cell migration has been extensively investigated in recent years; however, it remains elusive how chemical inputs from neighboring cells are integrated to coordinate the collective movement. Here, we provide evidence that propagation waves of extracellular signal-related kinase (ERK) mitogen-activated protein (MAP) kinase activation determine the direction of the collective cell migration.

1SAP-02 外力が駆動する細胞集団運動を支えるアクチン細胞骨格制御の解明
Actin interacting protein 1 and cofilin sense the extrinsic stretching force and orient cell rearrangement in Drosophila wing

Kaoru Sugimura[1,2], Keisuke Ikawa[1] ([1]*WPI-iCeMS, Kyoto Univ.*, [2]*JST PRESTO*)

Cells sense chemical signaling and generate forces underlying morphogenetic cell movement. In contrast, our previous studies demonstrate that macroscopic mechanical signals (i.e., tissue stress anisotropy generated by the extrinsic stretching force) orient cell rearrangement in Drosophila wing. However, the mechanisms by which cells sense the extrinsic forces and transmit the information to the molecular machinery for cell rearrangement remain unknown. To address these questions, we performed a screening of actin-binding proteins (ABPs). Our data indicate that AIP1 and cofilin control extrinsic force-driven cell rearrangements via F-actin regulation. In the symposium, we shall discuss how force-responsive properties of ABPs underlie collective cell movement.

1SAP-03 細胞外基質の粘弾性に応答する上皮細胞の集団運動と 3 次元形態形成
Collective Movement and 3D Morphogensis of Epithelial Cells Responding to Viscoelasticity of the Extracellular Matrix

Hisashi Haga (*Faculty of Advanced Life Sci., Hokkaido Univ.*)

Collective cell movement and 3D morphogenesis are essential events in diverse physiological processes. We found that epithelial cells (MDCK cells) move collectively along one direction on a soft collagen gel, whereas the cells migrate randomly on a rigid collagen-coated-glass. Moreover, lumen formation occurred when an epithelial sheet on a collagen gel was overlaid with another collagen gel. We also found that MDCK cells formed 3D morphology on top of the Matrigel. The appearance of the morphologies was like a tulip hat. The cells tugged at the peripheral matrix and remodeled the gel surface. In this meeting, these dynamical behaviors of the epithelial cells are discussed in terms of the cellular contractile force and viscoelasticity of the extracellular matrix.

1SAP-04 マイクロ流体デバイスを用いた細胞性粘菌の集団的細胞運動の解析
Microfluidic analysis of group cell migration in *Dictyostelium*

Taihei Fujimori[1], Akihiko Nakajima[2], Daisuke Imoto[1], Shuji Ishihara[4], **Satoshi Sawai**[1,2,3] ([1]*Dept. Basic Sci., Grad. School of Arts and Sci., Univ. of Tokyo*, [2]*Research Ctr. for Complex Systems Biology, Univ. of Tokyo*, [3]*JST PRESTO*, [4]*School of Sci. Eng., Meiji Univ.*)

In the late stage of Dicytostelium aggregation, cells chain up in head-to-tail manner in the so-called 'contact following' motion that is so far poorly characterized. Here, we carried out single-cell based quantitative imaging analysis of shape and motion of mutually attached cells migrating under quasi-2D spatial constriction in a microfluidic chamber. We will discuss the results and suggest how local signals guided by both the chemoattractant and cell-cell contact promotes cell polarity.

1SAP-05 単一ヒト表皮幹細胞からの多層上皮構造の形成原理
A mechanistic principle of multilayered epithelial formation from single human epidermal stem cells

Daisuke Nanba (*Tokyo Medical & Dental Univ., Med. Res. Inst., Dept. Stem Cell Biol.*)

Here we dissect multilayered epithelial structure formation from single human epidermal stem cells. Image analysis uncovered the spatiotemporal multicellular dynamics of human keratinocytes during the colony formation, which allowed to build a cell kinetic model. Simulation experiments then successfully reconstituted several types of multilayered keratinocyte colonies from single stem cells in silico, and indicated that the locomotive ability of cells, which results from rotational motion of keratinocytes, is required for continuous growth of multilayered human keratinocyte colonies derived from single stem cells. This study provides experimental and theoretical evidence for the importance of locomotive ability of epidermal keratinocyte stem cells in the colony growth.

1SAP-06 がん細胞の集団的浸潤：病理からの視点

Collective invasion of cancer cells: perspectives from pathology

Atsushi Enomoto (*Dept. Pathol., Nagoya Univ. Grad. Sch. Med.*)

In many types of human cancers, cancer cells are often connected and form groups of various sizes. They achieve collective invasion into surrounding stroma, rather than spreading out individually. Mechanisms underlying collective cancer invasion differ from those observed in the migration of single cells in culture. Obviously, intercellular adhesion needs to be coordinated in contrary to the hypothesis that the epithelial-mesenchymal transition program is crucial for cancer invasion. Cancer cell groups are heterogeneous, including cells that are leaders and those that are followers. The interaction of cancer cells with the stroma is also a prerequisite for collective invasion of cancer. In the symposium, these features of collective cancer invasion will be discussed.

1SBP-01 骨格筋ミオシン間における力発生の同調現象を明らかにする

Molecular mechanism of synchronous force generations among skeletal myosins

Motoshi Kaya (*University of Tokyo, Graduate School of Science*)

For more than half a century, molecular mechanism of muscle contraction has been investigated by various experimental approaches. One of main questions is whether myosins generate force cooperatively? To address the question, we have developed the experimental system, in which synthetic myofilaments interact with a single actin filament. Our findings suggest that myosins may generate force synchronously against high loads. To gain insight into the mechanism of cooperative force generations, we developed the simulation model consisting of 17 myosin molecules arranged in series and interacting with a single actin filament. The model revealed that strain-dependent kinetics and multiple steps of power strike enhance a chance of synchronous force generations among myosins.

1SBP-02 体細胞分裂期における細胞質ダイニンの操作

Manipulation of cytoplasmic dynein during mitosis

Tomomi Kiyomitsu (*Nagoya University*)

During mitosis, cytoplasmic dynein localizes at multiple sites to coordinate chromosome alignment with the spindle assembly and helps spindle positioning. However, how dynein functions spatiotemporally at each location remains unclear. Here, we present two novel approaches to study dynein functions in human cells. First, we developed a rapid protein depletion assay by combining the auxin-inducible degron with CRISPR/Cas9-mediated genome editing. Second, we developed a light-inducible system in which cortical dynein regulators were targeted to locally illuminated regions to manipulate spindle position. Our results indicated that dynein maintains the bipolar spindle structure at the spindle poles and is sufficient to control spindle position at the cell cortex.

1SBP-03 Intracellular production of synthetic RNA granules by ligand-yielded multivalent enhancers

Takanari Inoue (*Johns Hopkins University*)

Non-membrane bound, hydrogel-like granular structures in cells nucleate cellular functions through their unique physico-chemical properties. Here, we report iPOLYMER, a strategy for rapid induction of protein-based hydrogels inside living cells, taking advantage of an inducible dimerization paradigm. A series of biochemical and biophysical characterizations, in conjunction with computational modeling, revealed that the polymer network formed in the cytosol resembles a physiological hydrogel-like entity. We then functionalized the generated hydrogel with RNA binding motifs to synthetically mimic RNA granules. iPOLYMER presents a powerful approach for unraveling the properties and functionality of RNA granules, and other hydrogel-like structures in cells.

1SBP-04 単一細胞内局所加熱による細胞熱応答の原理の解明

The mechanisms of cellular response to temperature changes as revealed by local heating in single cells

Kohki Okabe[1,2], Beini Shi[1], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*PRESTO, JST*)

Temperature influences various levels of physiological functions. In contrast to observational investigations into intracellular temperature variations in time and space, the mechanism and the significance of temperature change in cells are poorly understood. Here, we utilized local heating techniques for a single living cells to understand the effects of temperature change on cell functions. Furthermore, transient quantitative heating allowed examinations into the mechanism of temperature variation inside of cells, which shows an unique property of intracellular heat dynamics. These techniques and results will reveal the novel cellular mechanism of sensing of and response to the temperature change.

1SBP-05 血管のメカニカルストレスによるフィブロネクチンピラー形成

Vascular mechanical stress organizes Fibronectin into pillars bridging tissue gap

Yuki Sato[1,2] ([1]*Grad. Sch. Med. Sci., Kyushu Univ.*, [2]*JST, PRESTO*)

Fibronectin (FN) is an important extracellular matrix component that undergoes fibrillogenesis through cell contact and creates patterned geometries. We found that FN is deposited as pillars along with long filopodia between widely separated germ layers. Loss-of-function experiments of Ena/VASP, integrinb1 and talin in the filopodia abolished the FN pillars, indicating that FN pillar formation is dependent on the filopodia through these molecules. We identified a new mechanism underlying FN pillar formation by focusing on cyclic expansion of adjacent dorsal aorta. The FN pillars are maintained dependent on dorsal aorta pulsing stress. A large tissue gap is reinforced against blood flow-associated mechanical strain through the FN pillars and filopodia interaction.

1SBP-06 チューブリンアイソタイプと微小管動態の多様性 Distinct contribution of different tubulin isotypes to microtubule dynamics

Asako Sugimoto (*Life Sciences, Tohoku Univ.*)

Microtubules (MTs) are dynamic polymers composed of α- and β-tubulin heterodimers. Most organisms have multiple tubulin isotypes encoded by different genes. Analyses in vitro demonstrated that tubulin isotypes differently affect MT dynamics and interaction with MT motors. However, contribution of tubulin isotypes to MT dynamics in vivo is still poorly understood. To address this issue, we have been systematically analyzing expression patterns and loss-of-function phenotypes of each tubulin isotype in C. elegans, using the CRISPR-Cas9 genome editing system. Live imaging analysis of MT dynamics in these strains revealed that each tubulin isotype differently affects MT dynamics. We will discuss how tubulin isotypes contribute to creating diverse MT behaviors in vivo.

1SCP-01 Computational design of catalytic triad based organophosphate capture proteins

Chu Wang (*Dept. Chem. Biol., CCME, Peking Univ.*)

Organophosphates (OPs) are a notorious class of toxic compounds that are widely used for pesticides and nerve agents. We computationally designed proteins with idealized serine-containing catalytic triads, and assess their serine nucleophilicity using activity-based probes with OP warheads. Crystal structures of the most successful designs show unprecedented agreement with the computational models with extensive hydrogen bonding networks between the catalytic residues, and knockout experiments demonstrate that these networks are critical for serine activation and OP-reactivity. Further successful experimental optimization with a crystal structure suggests the designs could provide the basis for a new class of organophosphate capture agents.

1SCP-02 Chemical Probes with Fuorogenic Switches for Visualizing Modified Protein and DNA

Yuichiro Hori[1,2] ([1]*Grad. Sch. Eng., Osaka Univ.*, [2]*IFReC, Osaka Univ.*)

Synthetic chemical probes are powerful tools for studying biomolecules in living cells. Particularly, fluorescent probes for protein labeling offer useful live information on localization and function of proteins. To date, we have developed a protein labeling technique using PYP-tag and its labeling probes. The key advantage of this technique is the availability of multicolor fluorogenic probes, which are not fluorescent in the free state but enhance fluorescence intensity upon protein labeling. The fluorogenic switch allows quick and high-contrast imaging of proteins in living cells. In this study, the fluorogenic system was applied for investigating protein glycosylation. Furthermore, evolution of this techinique enabled visualization of endogenous DNA methylation.

1SCP-03 Self-assembly of protein nanofibrils that display active enzymes

Sarah Perrett (*Inst. Biophys., CAS*)

The yeast prion protein Ure2 has a natively disordered N-terminal prion domain and a globular C-terminal domain. The C-domain shows enzymatic activity in both soluble and fibrillar forms of Ure2. We have used a variety of biophysical approaches to investigate the structure of Ure2 fibrils and their mechanism of assembly. We have also created chimeric constructs where the prion domain is genetically fused to other enzymes of different sizes and architectures. The chimeric proteins spontaneously self-assemble into nanofibrils displaying active enzymes. Combination with microfluidic techniques allows formation of enzymatically-active microgel particles. The design principles can be adopted to create countless other bioactive amyloid-based materials with diverse functions.

1SCP-04 アミロイド線維形成初期過程のタンパク質構造化メカニズムの解析 Investigating early steps in amyloid fibril formation

Eri Chatani (*Grad. Sch. Sci., Kobe Univ.*)

Amyloid fibrils are associated with human amyloidoses and neurodegenerative diseases, and elucidating the mechanism of amyloid nucleation is important for molecular understanding of the onset of such diseases. To clarify details of protein association during the nucleation, we attempted to monitor early steps in amyloid formation with several analytical methods such as thioflavin T fluorescence, FTIR, SAXS, and DLS. By using insulin and an insulin-derived peptide fragment, we have observed the formation of early aggregated species and their subsequent structural development prior to the formation of amyloid fibrils. On the basis of the result a possible mechanism describing how the amyloid nuclei generate will be discussed.

1SCP-05 Nanozyme: discovery and its application in tumor diagnosis

Xiyun Yan (*Inst. Biophys., CAS*)

Since the first evidence that ferromagnetic nanoparticles with intrinsic peroxidase-like activity was reported in 2007[1], nanozyme has been considered as next generation of artificial enzyme[2], and become an emerging field between biology and nanotechnoly[3]. At the moment, there are over 40 different kind of nanozymes have been found and their application have widely expanded to biomedicine, chemical industry, food, agriculture and environment[4]. Here I will introduce the definition of nanozyme and their application in tumor diagnosis[5, 6] and therapy[7].

References: 1) Nat Nanotech 2007, 2, 577. 2) Chem Soc Rev 2013, 42, 6060. 3) Eur J Inorg Chem 2016, 2016, 1906. 4) Inorg Chem Front 2016, 3, 41. 5) Nat Nanotech 2012, 7, 459. 6) ACS Nano 2016. 7) PNAS USA 2014, 111, 14900.

1SCP-06 蛋白質相互作用の熱力学：分子設計と創薬
Thermodynamics of protein interaction for molecular design and therapeutics

Kouhei Tsumoto (*The University of Tokyo*)

Specific recognition of ligands by proteins is a fundamental biological phenomenon. Recent advances in physical biochemistry have enabled us to describe what factors dominated the specificity and affinity of protein interactions, especially from thermodynamic viewpoints. We have focused on several antigen-antibody interactions, including those specific for model antigens, and dissected the interactions from physicochemical viewpoints. Based on the results, we could propose one strategic scheme on improvement of antibody affinity for targets, which could be applied not only to improve the specificity and affinity of an antibody, but also to screen and/or design of small molecules.

1SDP-01 Single molecule analysis of F_oF_1-ATP synthase

Rikiya Watanabe[1,2] ([1]*Department of Applied Chemistry, The University of Tokyo*, [2]*PRESTO, JST*)

F_oF_1-ATP synthase (F_oF_1) is a rotary motor protein which reversibly converts the proton motive force (*pmf*) into ATP production via mechanical rotation of the rotor complex. Technical difficulties have for long time hampered single molecule analysis of chemomechanical coupling mechanism of F_oF_1, i.e., rotary motion and proton translocation. To address this issue, novel lipid-bilayer systems have been recently developed for direct observation of the rotation and proton translocation of F_oF_1. In this presentation, I will introduce the recent developments of lipid-bilayer systems for single molecule analysis of F_oF_1, simultaneous with the findings on the chemomechanical coupling mechanisms of F_oF_1.

1SDP-02 *De novo* 設計軸の回転から明らかになったトルク発生機構
Rotation of *de novo* designed axis and the torque generation mechanism

Jun-ichi Kishikawa, Mihori Baba, Atsuko Nakanishi, Ken Yokoyama (*Dept. LifeSci, Kyoto Sangyo Univ.*)

V_1-ATPase is composed of A_3B_3 ring and a rotor protein. The conformational changes of A_3B_3 ring associated with ATP hydrolysis leads to rotation of a rotor. Previous studies demonstrated that exogenous rod-like proteins function as a rotor. In this study, we examined whether *de novo* designed axis functions as a rotor. The designed axis formed complex with A_3B_3 and the complex exhibited higher ATPase activity than that of A_3B_3 without the rotor. In addition, the rotation of the axis was observed by single molecule analysis. These results indicate that the *de novo* designed axis function as a rotor and strongly suggest that any strict and specific interactions between the A_3B_3 ring and the rotor are not essential for torque generation.

1SDP-03 滑走バクテリアと遊泳アーキアの運動超分子マシナリーの単位ステップ観察
Unitary steps of supramolecular-motility machineries in gliding bacteria and swimming archaea

Yoshiaki Kinosita[1], Daisuke Nakane[1], Nariya Uchida[2], Makoto Miyata[3], Takayuki Nishizaka[1] ([1]*Dept. Phys., Gakushuin University*, [2]*Dept. Phys., Tohoku University*, [3]*Dept. Biol., Graduate School of Science, Osaka City University*)

We here describe stepwise movements of two novel motors: gliding and rotary machineries in bacteria and archaea. *Mycoplasma mobile* glides the solid surface at 2.5 μm/s by repeating a cycle that 450 legs attach to and detach from substrates. By high-speed tracking under a designed condition that the number of active legs were reduced, 70-nm steps were detected under various [ATP]s. We next examined the rotary motor of *Halobacterium salinarum*. The tracking of tethered cell enabled us to detect intermittent pauses during rotation, of which the periodicity was consistent with the ATPase in archaeal motor. With assumption that a unitary step consuming single ATP that supplies the chemical energy of 80 pN nm, the energy efficiency is estimated as ~6-10%.

1SDP-04 A small stroke for an individual, but giant motion for a population: negative gravitaxis and bioconvection of Chlamydomonas reinhardtii

Azusa Kage (*Dept. Finemechanics, Tohoku Univ.*)

Negative gravitaxis, biased swimming against gravity, of certain microorganisms is one of the extreme mysteries in gravitational biology. Its mechanism and function have remained controversial for more than a century. One consequence of negative gravitaxis is bioconvection, a collective motion spontaneously occurring in suspensions of such microorganisms. Ascent by negative gravitaxis drives formation of visible regular patterns, which look similar to those formed by thermal convection. We have experimentally investigated bioconvection, particularly spontaneous phase transition so far exclusively observed in bioconvection of the unicellular green alga Chlamydomonas. Its swimming characteristics such as flagellar waveform is likely to trigger the phase transition.

1SDP-05 インビトロ運動アッセイ中の自走する微小管の集団運動
Collective motion of running microtubules in in vitro motility assay

Ken Nagai (*Sch. Mater. Sci., JAIST*)

The collective motion of running microtubules in a motility assay such as global nematic phase, vortices located randomly, and a hexagonal lattice of vortices emerged when the number density of microtubules was large enough. The phase of collective motion depended on the species of motor, the density of motors attached to glass surface, microtubule's length, and the density of microtubules. We found that the memory of rotation rate of isolated microtubules is one of the key properties that determine the phase of collective motion with an agent-based model. The used model can also explain the traits of collective motion of various kinds of living things. The fact indicates that the properties of rotation are crucial parameters of collective motion in general.

1SDP-06 胚発生過程における細胞集団運動を担うアクトミオシンの制御機構

Local regulation of actomyosin for the globally orchestrated collective cell movement during tissue morphogenesis

Asako Shindo[1], John Wallingford[2], Makoto Kinoshita[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*UT Austin*)

Cells move collectively to form tissues in the embryo during development. To coordinate the multiple cell behaviors in the tissue, cytoskeleton such as actomyosin has to be regulated properly in each cell. We focus on the collective cell movement called convergent extension (CE) to investigate the molecular mechanisms of coordinated cell movements using Xenopus laevis embryo. We found that actomyosin pulses beneath the specific cell-cell junctions asymmetrically, and generates contractile force to drive the cellular intercalation movements during CE. We have found that the molecular signaling, a non-canonical Wnt pathway, locally controls the asymmetric actomyosin pulses, providing a basis of coordinated collective cell movement during morphogenesis.

1SDP-07 Shape Remodeling of Active Cytoskeletal Vesicles

Andreas Bausch (*Lehrstuhl für Biophysik, TU München*)

Living cells rely on the selforganization mechanisms of cytoskeleton to adapt to their requirements. Most processes such as cell division, or cellular motility rely on the controlled selfassembly of welldefined active cytoskeletal structures interacting with membrane, which still allow a dynamic reorganization. One important and promising strategy to identify the underlying governing principles is to quantify the physical process in model systems mimicking the functional units of living cell. Here I'll present in vitro minimal model systems consisting of actin filament, crosslinking molecule and myosin II encapsulated into lipid vesicles. I will discuss how a balance of local force exertion and tension generation results in blebbing, invagination or tethering of the membrane.

1SEP-01 Elucidation of structure-function relationship of biological active sites by molecular simulation

Yu Takano[1,2], Yusuke Kanematsu[1], Yasuhiro Imada[2] ([1]*Grad. Sch. Info. Sci., Hiroshima City Univ.*, [2]*IPR, Osaka Univ.*)

Biological active sites are regulated by protein environments and show prominent functions such as high-specific molecular recognitions and high-efficient catalysis. Our objective is to elucidate structure-function relationship of biological active sites by molecular simulations, quantum chemical calculations, molecular dynamics (MD), and hybrid quantum mechanics/molecular mechanics (QM/MM) calculations.
Here, we present three issues: (1) development of a QM/MM-MD code for the analysis of protein functions, (2) investigation of the electronic structure of the active sites in phostosystem II, and (3) examination of the relationship between the molecular and electronic structures of hemes in heme proteins.

1SEP-02 Structural analysis of photosystem II to reveal the mechanism of light-induced water-splitting

Fusamichi Akita[1], Michihiro Suga[1], Keitaro Yamashita[2], Go Ueno[2], Hironori Murakami[2], Yoshiki Nakajima[1], Yasufumi Umena[1], Kunio Hirata[2], Minoru Kubo[2], Kazuya Hasegawa[2], Masaki Yamamoto[2], Hideo Ago[2], Jian-Ren Shen[1] ([1]*RIIS, Okayama Univ.*, [2]*Riken Harima*)

Photosystem II (PSII) catalyzes light-induced water-splitting, leading to the generation of electrons, protons and oxygen. PSII is a supercomplex containing a Mn cluster where the water-splitting takes place. The structure of PSII was reported at 1.9 Å resolution in 2011, which showed the Mn cluster has a "distorted chair" shape. We have used X-ray free electron laser to obtain the PSII structure at 1.95 Å resolution in 2015. However, since our structural analysis was performed for the S1-state, the mechanism is still not clear. To analyze the structures of the intermediate S-states, we prepared microcrystals and collected their diffraction images with an approach of fixed-target serial crystallography. I will discuss the S3-state structure determined by this approach.

1SEP-03 X 線 1 分子追跡法によるマルチマータンパク質・機能的運動の可視化

Active 3D Motion Visualization of Multimeric Proteins by X-ray Single Molecule Tracking

Hiroshi Sekiguchi (*JASRI/SPring-8*)

Diffracted X-ray Tracking (DXT) is one of single molecule techniques for investigating intra-molecule dynamics of functional proteins. In DXT, a gold nanocrystal, used as motion probe, is immobilized on a target protein and the trajectory of its diffracted spot is investigated as the motion of the protein. The size of gold nanocrystal used for our measurements is ranged from 20 to 80 nm in diameter, and cooperative motions of multimeric protein could be tracked by immobilizing the probe on multi-sites on the protein. In this presentation, we review cooperative active motions of multimeric proteins, such as nAChR (Sci. Rep. 2014) or group II chaperonin (PLoS ONE 2013, JMB 2014, FEBS Open 2016).

1SEP-04 蛍光 X 線ホログラフィーによるヘモグロビンの金属周辺構造の可視化

Visualization by X-ray fluorescence holography of metal environments in hemoglobin

Ayana Sato-Tomita[1], Naoya Shibayama[1], Naohisa Happo[2], Kouichi Hayashi[3], Yuji C. Sasaki[4] ([1]*Jichi Med. Univ.*, [2]*Hiroshima City Univ.*, [3]*Nagoya Inst. Tech.*, [4]*Tokyo Univ.*)

X-ray fluorescence holography (XFH) is a powerful tool that can visualize the three-dimensional (3D) atomic structures around specific elements. An important advantage of XFH is the capability of "model-free" atomic imaging by a simple Fourier transform-like procedure. Using the developed experimental setup [A. Sato-Tomita et al., Rev. Sci. Inst. 87, 063707, 2016], we carried out the XFH measurements of CO-bound human hemoglobin crystals at the beamlines BL6C (KEK-PF) and BL39XU (Spring-8), and succeeded for the first time in obtaining the hologram of a protein active-site structure. The 3D atomic images around the central iron atoms of the individual hemes are generated, which indicate a difference in the planarity of the α and β hemes.

1SEP-05　マイクロ流路デバイスを用いた時間分解分光法による膜タンパク質の活性サイトの中間体構造解析

Intermediate structures of the active site in membrane proteins revealed by time-resolved spectroscopy with micro-channel devices

Tetsunari Kimura (*Grad. Sch. Sci., Kobe Univ.*)

Time-resolved spectroscopy is powerful to follow the structural changes in the active sites along the reaction axis. Here, to investigate the enzymatic reaction of a low-yield membrane protein triggered by the sudden changes in buffer conditions, microscopic infrared/visible spectrometers were constructed by equipping the novel micro-channel flow-cells. The first application of the developed systems was nitric-oxide reductase (NOR), a membrane enzyme that catalyzes NO reduction ($2NO + 2H^+ + 2e^- \rightarrow N_2O + H_2O$). The atomic and electronic properties of the active site composed of heme b_3 and non-heme Fe_B in the transient NO-bound intermediate was determined, enabling us to understand the molecular mechanism of NO reduction of NOR.

1SEP-06　チトクロム *c* 酸化酵素の時間分解 XFEL 結晶構造解析：機能部位間の相互作用ダイナミクスの観測

Time-resolved XFEL crystallography of cytochrome *c* oxidase: Probing the interaction dynamics between two functional sites

Minoru Kubo (*RIKEN SPring-8 Center*)

Bovine heart cytochrome *c* oxidase is the terminal oxidase of cell respiration that catalyzes O_2 reduction to water at the Fe_{a3}-Cu_B binuclear center, coupled with proton pump across the mitochondrial inner membrane. The proton pump pathway is composed of a water channel and a hydrogen-bond network in tandem. The water channel is closed upon ligand binding to the binuclear center, which prevents proton back flow during proton pumping. Here, we applied time-resolved XFEL crystallography at SACLA to observe the structural dynamics following CO-photolysis from Fe_{a3}. The result reveals a small but significant motion that plays an important role in the interaction between the binuclear center and the water channel.

1SFP-01　生細胞膜上で形成される G タンパク質共役型受容体の動的ダイマー：１分子観察法を用いたアプローチ

Dynamic dimer formation of G-protein coupled receptor in the live plasma membrane: An approach by using single molecule observation

Rinshi Kasai[1], Akihiro Kusumi[1,2] ([1]*Inst. Front. Med. Sci., Kyoto Univ.*, [2]*Membrane Cooperativity Unit, OIST*)

G-protein coupled receptor, GPCR, is one of the largest and the most important receptor families. Unlike class-C GPCRs, it has been controversial whether class-A GPCRs, consisting of over 80 % of all GPCRs, work as monomers or dimers. By observing them at the single molecule level in the live plasma membrane, we found that while all GPCRs that we observed can exist as monomers before stimulations, they form tentative homo-dimers with the lifetimes of ~100 ms. In addition, tentative hetero-dimer formation was observed for Dopamine Receptor D1 and D2 before stimulations. Combining these findings, it could be concluded that dynamic monomer-dimer transition is one of common features found in class-A GPCRs that may regulate signal transduction processes.

1SFP-02　細胞膜の分子組織構造・反応カップリング

Coupling of reactions and molecular organizations in plasma membranes

Yoshihisa Kaizuka (*NIMS*)

Mechanisms for protein clustering observed in both plasma membranes and cytoplasm has been interpreted in the context of specific protein-protein interactions. However, how such molecular organizations could regulate biochemical reactions are not well understood. We took an approach of reconstituting T cell membrane signaling clusters with planar lipid bilayers and fluorescently labeled proteins, and analyzed how individual protein clusters regulate phosphorylation-dephosphorylation reactions by quantitative microscopy. Our analysis revealed that clustering of kinase Lck recreates conditions in which a phosphatase CD45 has a positive role in signal initiation via reliving of Lck autoinhibition, demonstrating a novel regulatory mechanism of protein clustering.

1SFP-03　Cytokine receptor dimerization: molecular determinants and cellular regulation

Jacob Piehler (*University of Osnabrueck*)

The role of dimerization in cytokine receptor activation has been controversially discussed as pre-formed receptor dimers or clusters have been suggested by numerous assays. To shed light into the spatiotemporal dynamics of cytokine receptor assembly, we have developed single molecule imaging techniques for quantifying interactions in the context of lipid membranes in vitro and in living cells at physiologically relevant densities. These techniques established random distribution of the receptor subunits and ligand-induced dimerization for several hetero- and homodimeric cytokine receptors. Detailed quantitative dimerization assays suggest an intricate interplay of interactions between different components of cytokine receptors.

1SFP-04　マイクロクラスターは T 細胞受容体のエンドサイトーシスのシグナルユニットとして機能する

Microclusters as a signaling unit for T cell receptor endocytosis

Tadashi Yokosuka (*Dept. Immunol., Tokyo Medical Univ.*)

T cell activation is regulated by the signaling units, T cell receptor (TCR) microclusters, that are continuously constructed by few decades of TCRs and their downstream signaling molecules at an immunological synapse. By imaging Cbl-b, an essential E3 ubiquitin ligase for T cell quiescence, we found that ubiquitin itself translocates into TCR microclusters in a Cbl-b-dependent machinery. Clathrin light chain, dynamin, and epsin are also together accumulated at TCR microclusters bearing active signaling. We further real-time imaged transient clustering of clathrin at the outer region of the immunological synapse, where TCR microclusters are freshly and continuously generated. We here suggest a novel function of a microcluster as a signaling unit for TCR endocytosis.

1SFP-05 **BAR タンパク質による細胞膜の形態形成とファゴサイトーシスの関連**

Plasma membrane morphogenesis by the BAR domain superfamily proteins for phagocytic cup formation

Shiro Suetsugu (*NAIST*)

The BAR domain superfamily proteins are characterized by their BAR domains, which senses and deforms the membrane by their curvature of the BAR domains. Plasma membrane contains various sub-micron structures, such as clathrin-coated pits, caveolae, filopodia, lamellipodia, phagocytic cup, and so on. Most of these membrane structures are supposed to be under regulation of members of the BAR domain superfamily proteins. However, it is unclear how the BAR domain superfamily protein is involved in phagocytic cup formation. Here we will discuss the structure-function relationships of the BAR domain and phagocytic cup, with a newly solved structure of the BAR domain members.

1SFP-06 **視細胞円板膜上のロドプシン多量体クラスターがつくる一過的メゾ領域**

Transient meso-domains formed by oligomeric clusters of rhodopsin in retinal disk membrane

Fumio Hayashi[1], Natsumi Saito[1], Yasushi Tanimoto[2], Kenich Morigaki[2,3], Keiji Seno[4] ([1]*Grad. Sch. Sci. Kobe Univ.*, [2]*Grad. Sch. Agri. Kobe Univ.*, [3]*Biosig. Res. Cent. Kobe Univ.*, [4]*Hamamatsu Univ. Med.*)

A prototypical G protein-coupled receptor rhodopsin (Rh) in retinal disk membrane is thought to be organized in rows of dimers, thereby providing a signaling platform for G protein transducin (Gt). However, the dynamic nature of the molecular distribution and clustering remains elusive. In singulo and multiplo studies, we found that Rh forms meso-sized raftophilic transient clusters, which are in dynamic equilibrium with non-clustered Rh. These Rh molecules pre-associate with Gt, and are loosely confined in the central area of disks in the dark. Upon activation, Gt dissociates and disperses to disk periphery. These results demonstrate micro- and meso-scale dynamic heterogeneities in retinal disk membranes, and their profound importance in phototransduction system.

1SGP-01 **マイクロチャンバーと融合した大腸菌の生存**

***E. coli* survival in inorganic chamber**

Kazuhito Tabata[1,2,3], Yoshiki Moriizumi[1], Rikiya Watanabe[1], Hiroki Ashikawa[1], Hiroyuki Noji[1,3] ([1]*Grad. sch. eng., Univ. of Tokyo*, [2]*PREST JST*, [3]*ImPACT Cabinet Office*)

E. coli has an inner membrane and outer membrane. The protoplasts can survive the loss of the outer membrane, but perish if the inner membrane is compromised. From this observation, one could propose that maintaining inner membranes is key factor. We prepared a microchamber that had the same volume as an E. coli and also a lipid membrane. By fusing the chamber with an E. coli, we could introduce into the chamber E. coli cytoplasm and into the lipid membrane of the chamber membrane proteins. Prior to the fusion, we had inserted a plasmid into the chamber. Following the fusion, we observed protein synthesis from plasmid. Additionally, we also observed living E. coli-like organisms from the fused cells. These results suggest inanimate conditions from which life can form.

1SGP-02 **1 細胞レベルでの薬剤耐性獲得プロセス**

Acquisition of drug resistance at the single-cell level

Yuichi Wakamoto (*Univ. of Tokyo*)

Bacterial cells are highly adaptive to a wide range of antibiotic stress, and the long-term exposure often results in the emergence of the resistant cells. At a high concentration, only a small fraction of the cells at the beginning of exposure give rise to surviving descendants. To understand the unique features of the cellular histories leading to drug resistance, we observed the both short-term and long-term behaviors of Escherichia coli under drug exposures with microfluidic single-cell time-lapse microscopy. The result shows that some of the initial survivors acclimatize to the drug through multiple rounds of divisions. The tolerance of acclimatized cells is stably heritable under drug exposure, but lost after removal.

1SGP-03 **Plasticity of developmental process that determines floral organ number**

Miho Kitazawa[1,2], Koichi Fujimoto[2] ([1]*CELAS, Osaka Univ.*, [2]*Dept. Biol. Sci, Osaka Univ.*)

Floral organ number is one of the fundamental features that characterises floral morphology that is conserved in the major clades of angiosperms: The basic floral organ number in core eudicots is four or five, whereas that in monocots is three. On the other hand, the basal eudicots show both inter- and intra-specific variation in the number. We have examined the statistical quantities of intra-specific variation in wild populations, and found that the SD is proportional to the square root of absolute difference between the mean and a clade-specific number. We further examined the positioning of excessive sixth organ, and found that a considerable fraction of the flowers showed common arrangement to monocots, which may mimic plasticity at the eudicot-monocot bifurcation.

1SGP-04 **生物システムの可塑性の理解に向けて：理論解析と実験進化**

Toward Understanding of Biological Plasticity: Computational and Experimental analysis

Chikara Furusawa[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sci., Univ. Tokyo*)

Biological systems have the ability to change their state to adapt and to evolve in response to environmental changes. However, despite their importance, studies on biological plasticity have thus far remained at a qualitative level. Here, to unveil the nature of biological plasticity, we performed computer simulations of adaptive evolution using simple cell models and found that cellular state changes in adaptation and evolution are generally restricted to a low-dimensional dynamics in phenotype space. Furthermore, analysis of environmental response and evolutionary dynamics of E. coli also supported that the phenotypic changes were restricted to a low-dimensional dynamics. Based on these results, macroscopic description of adaptation and evolution will be discussed.

1SGP-05 表現型適応と進化のマクロ現象論：揺動応答関係、遺伝的同化、スローマニフォルド仮説

Macroscopic Theory of Phenotypic Adaptation and Evolution: Fluctuation-response, Genetic Assimilation, and Slow-Manifold Hypothesis

Kunihiko Kaneko (*University of Tokyo*)

Characterization of plasticity, robustness, and evolvability is one of the most important issues in biology. Assuming that cells undergo steady growth, protein expression of thousands of genes is shown to change along a one-dimensional manifold in the state space in response to the environmental stress. This leads to a macroscopic law for a cellular-state, as confirmed by adaptation experiments of bacteria. Next, proportionality between phenotypic changes by genetic evolution and by environmental adaptation, uncovered both in experiments and simulations, is formulated by sow-manifold hypothesis to constrain the paths of phenotypic changes. Possible extension of the theory to non-growing cellular states and to multi-level evolution will be briefly discussed.

2SAA-01 光生物分野における新区分の立ち上げ

Launching a new category in photobiology

Yuji Furutani[1], Yuki Sudo[2] ([1]*Inst. Mol. Sci, Nat. Inst. Nat. Sci.*, [2]*Grad. Sch. Med. Dent. Pharm. Sci., Okayama Univ.*)

The Biophysical Society of Japan (BSJ) provides two categories for researchers in photochemistry and photobiology; 18A-Vision & Photoreception, and 18B-Photosynthesis. By the advances of their categories, technologies for visualizing and controlling the cellular activity by light has rapidly expanded over the past decade, paving the way for experiments that would have once seemed impossible. For instance, optogenetics allowed the manipulation of neural activity *in vivo* with millisecond precision. On the basis of the background, the council of the BSJ approved of creating the third category; 18C-Optogenetics & Optical Control. We hope that these three categories fuse among them and cultivate new insights into studies on biophysics and physicobiology.

2SAA-02 オプトジェネティクス革命

Optogenetic revolution

Hiromu Yawo[1,2] ([1]*Tohoku University Graduate School of Life Sciences*, [2]*Center for Neuroscience, Tohoku University Graduate School of Medicine*)

The brain consists of many types of neurons with distinct molecular organizations. Optical stimulation methods have received much attention recently with the technological development of modern optics. They have advantages over conventional electrical stimulation methods: finer spatiotemporal resolution and parallel stimulations at multiple sites. These methods are also less harmful and more convenient than electrical stimulation methods. Another breakthrough combined optical stimulation with genetic engineering technologies, which is otherwise known as optogenetics. Optogenetics is borderless and interacting with a variety of fields.

2SAA-03 新規オプトジェネティクスツール探索：天然および人工の微生物型ロドプシン

Exploration of new optogenetic tools: natural and artificial microbial rhodopsins

Keiichi Inoue[1,2] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*JST PRESTO*)

Microbial rhodopsins are photoreceptive ion-transporting membrane proteins of microorganisms. The optical control of neuronal activity by rhodopsins is one of the most established optogenetic technologies. However, strong demands still exist for more ideal absorption wavelength, selective ion transport, higher ion-flux and so on. To achieve them, gene-search for a new type of rhodopsin is a promising approach. In fact, we recently reported a new functional class of sodium pump rhodopsin (NaR), and it was shown to be able to efficiently control neuronal activity. We have improved the performance of NaR on the basis of biophysical insights for its transport mechanism. In presentation, we will discuss the potential of new rhodopsins for the application to optogenetics.

2SAA-04 レチナールタンパク質を基盤とした光遺伝学ツールの開発に向けて

Towards production of retinal protein-based optogenetic tools

Yuki Sudo (*Div. Pharm. Sci., Okayama Univ.*)

Retinal proteins consisting of a seven-transmembrane domain with a retinal chromophore are widely distributed through all three biological kingdoms. They show a variety of biological functions including vision, ion transportation and photosensing. In addition to the biological aspect, they become a focus of interest in part because of application for optogenetics, a technology for controlling biological activities by light.

Towards production of retinal protein-based optogenetic tools, we are working on microbial retinal proteins with three research topics as follows; (i) functional and structural analysis of novel retinal proteins, (ii) rational design and conversion of them, and (iii) development of optogenetic tools. I will talk about recent progress on them.

2SAA-05 Genetic, biochemical and biophysical studies on flavoprotein photoreceptors applicable for optogenetics

Shinji Masuda (*Center for Biological Resources & Informatics, Tokyo Institute of Technology*)

Flavoprotein photoreceptors are conserved in many organisms, which include cryptochromes, light-oxygen-voltage (LOV) domain-containing proteins and blue-light-using-flavin (BLUF) photoreceptors. Studies with the flavoproteins are basically focused on their unique photochemical reaction, since the flavin cannot be isomerized not as like isomerizable chromophores in other photoreceptors such rhodopsins and phytochromes. One advantage of these flavoproteins in optogenetic studies is that no additional gene is required for chromophore synthesis in vivo, since flavin is a fundamental cofactor in cells. Together with our studies with BLUF proteins, recent progresses on characterization of the flavoprotein photoreceptors and their potential for optogenetics will be discussed.

2SAA-06 Optogenetic potentials of bistable animal opsin-based pigments for regulating GPCR signalings

Mitsumasa Koyanagi[1,2,3] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*OCARINA, Osaka City Univ.*, [3]*JST PRESTO*)

Most animal opsins bind to 11-cis retinal as a chromophore to form a photosensitive pigment. Upon light absorption, opsin-based pigments activate cell signaling through a G-protein-mediated phototransduction cascade. We have investigated various kinds of non-conventional animal opsin-based pigments such as non-visual pigments and invertebrate visual pigments. Our findings revealed that most opsin-based pigments excluding vertebrate visual pigments are bistable pigment; the light-activated state is stable and reverts to the original dark state by subsequent light irradiation. Here we discuss contribution of molecular properties of the non-conventional opsins to the optogenetic applications based on our current studies on several non-conventional opsins.

2SAA-07 生体光操作技術の進展

Technological advances for optical control of living organisms

Yoshinori Shichida (*Dept. of Biophys., Grad. School of Sci., Kyoto Univ.*)

Recent progress in biological science has come along with technological innovations. For example, developments in imaging techniques of cellular activity and regulating biological functions brought rapid progress in understanding of biological phenomena. In particular, optical control for biological functions has brought about a revolutionary shift in the elucidation of neural networks through the expression of channel rhodopsin, a retinal protein, in neuron followed by irradiation with light. In addition, new technologies for optical control of biological functions have begun to appear. In the present talk, I will survey the basic science underling current technologies for optical control and explore the new technologies to be a beginning of the future developments.

2SBA-01 Design of Nucleotide Binding Site Toward Controlling and Understanding Molecular Motor

Takahiro Kosugi (*Institute for Molecular Science*)

Nucleotides (ATP, GTP and so on) play a key role to perform vital tasks in living cells. For example, conformational change of ATP-driven molecular motor is generated by binding this molecule. To understand mechanism of nucleotide binding and consequent conformation change, we designed nucleotide binding proteins. Recently several computational methods have been developed to design artificial protein structures from scratch and small molecule binding proteins in naturally occurring structure. By using these technologies, protein structures with a nucleotide binding site were created from scratch to explore optimal structure and necessary pieces. Also, another binding site in native molecular motor was inserted to control the motor and reveal the molecular mechanism.

2SBA-02 タンパク質分子ブロックを用いた分子モーターのエンジニアリング

Engineering approaches to molecular motors based on protein building blocks

Ken'ya Furuta (*NICT*)

Biomolecular motors have the potential to be used as molecular-scale actuators, switches, and robots in nanoscale devices; however, it is currently not possible to design a novel biomolecular motor from amino-acid sequences. Here, we present an alternative strategy where naturally occurring protein building blocks are combined through protein engineering techniques such as domain swapping and circular permutation to create a new series of biomolecular motors. According to the new bottom-up strategy, we successfully created novel actin-based motors and their derivatives with reversed directionality. Our new strategy combined with structural studies can be a powerful tool to investigate the design principles of biomolecular machines.

2SBA-03 Beyond DNA and RNA: synthetic genetic polymers

Alexander I. Taylor, Philipp Holliger (*MRC Laboratory of Molecular Biology*)

Two of the hallmarks of life, heredity and evolution, can be recapitulated in the test tube using a series of synthetic alternatives to DNA composed of non-natural building blocks, 'xeno nucleic acids' (XNA). We demonstrate that such synthetic genetic systems can be applied to the development of fully-artificial XNA ligands (aptamers), enzymes (XNAzymes), and nanostructures composed of a range of alternative chemical scaffolds with structures and physicochemical properties divergent from DNA and RNA. XNA-based tools and technologies technologies offer many exciting avenues for research, industry and medicine, and suggest the possibility of life based on alternative chemistry ('xenobiology').

2SBA-04 Construction of DNA origami base gene transcription nano chip

Hisashi Tadakuma (*Kyoto Univ, iCeMS*)

In synthetic biology, the design of gene expression requires devices that alter the output depending on the situation. Here, we integrated an enzyme, T7 RNA polymerase, and multiple target gene substrates onto a DNA origami-based nano-chip. This gene nano-chip orthogonally transcribes its own genes, and using this system, we succeeded in a rational design of gene expression from a single nano-chip by controlling the inter-molecular distances between the enzyme and the substrate genes. I would discuss the present and the future of our nano-chip.

2SBA-05 **アクトミオシン細胞骨格の in vitro 再構成**
In vitro reconstitution of contractile actomyosin cytoskeleton

Makito Miyazaki[1,2], Shin'ichi Ishiwata[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*WABIOS, Waseda Univ.*)

Animal cells self-organize various kinds of contractile actomyosin cytoskeleton including actin cortex and cytokinetic rings, which are essential for bleb-driven cell motility and cell division, respectively. To understand the regulatory mechanisms of those processes, we adopted simple *in vitro* model systems composed of purified proteins encapsulated in cell-sized water-in-oil droplets or liposomes. By controlling the protein components and concentrations, ordered patterns reminiscent of actin cortex and cytokinetic rings were self-organized. We will present the formation and contraction processes of actomyosin cytoskeleton in cell-sized confined spaces, and discuss how the cell deformation processes are directly regulated by actomyosin cytoskeleton.

2SBA-06 **Biomolecular Motors: From Cellular Function to Nanotechnological Applications**

Stefan Diez (*B CUBE, TU Dresden, Germany*)

We develop and apply novel optical techniques to investigate molecular transport in cell biology and nanotechnology. Building on our experience in single molecule biophysics and in the in vitro reconstruction of subcellular mechano-systems we study cooperative effects in motor transport. Moreover we aim to apply biomolecular motor systems in a synthetic, engineered environment for the generation and manipulation of nanostructure, optical surface imaging, medical diagnostics and parallel biocomputation. Thereby, our main emphasis is on the development of methods to control the nano-transport sytems by external signals in a spatio-temporal manner.

2SCA-01 **ZMW 法による生命現象の可視化の展開**
Expansion of biological applications using Zero-Mode Waveguides

Sotaro Uemura (*Dept. of Biol. Sci., Grad. Sch. of Sci., The Univ. of Tokyo*)

ZMW is an abbreviation for Zero-Mode Waveguide, a new technique which enables us to visualize single molecules at physiologically relevant concentrations of fluorescent dye. This is impossible to achieve with conventional total internal reflection fluorescence (TIRF) methods due to high background. Using ZMW, our group was the first to successfully visualize the real-time transit of fluorescently labeled tRNAs to the ribosome during protein translation at codon resolution and at physiologically relevant concentrations. We are further expanding this technology to target novel biological phenomena, not only translation but including "Genome editing", "RNA silencing", "Membrane proteins" and "Molecular motors".

2SCA-02 **Observation of single membrane proteins under mechanical tension**

Tae-Young Yoon (*Yonsei University*)

I will talk about our recent efforts, where we endeavor to apply single-molecule methods to membrane proteins. By applying pN-scale force to a single SNARE complex, a main force-generating machine for membrane fusion in all eukaryotes, we reveal large hysteresis in a mechanical unizipping and rezipping cycle of the SNARE complex. With combined application of different single-molecule methods, we show how this rigid SNARE complex is efficiently disassembled by NSF and SNAP. We observed that NSF exploits a spring-loaded mechanism to tightly couple its ATP hydrolysis with unfolding of the SNARE complex.

2SCA-03 **High-speed angle-resolved imaging of catalytic subunit of F1-ATPase**

Sawako Enoki[1], Ryota Iino[2], Yoshihiro Minagawa[1], Yamato Niitani[3], Michio Tomishige[3], Hiroyuki Noji[1] ([1]*Dept. Appl. Chem, Grad. Sch. Eng. Univ. of Tokyo*, [2]*Okazaki Inst. Integ. BioSci., NINS*, [3]*Dept. Appl. Phys, Grad. Sch. Eng. Univ. of Tokyo*)

Many biomolecular machines undergo large conformational change when they function. To monitor these conformational changes at the single-molecule level, we recently developed a dark-field microscopy with microsecond temporal and one-degree angular resolutions, applying a single gold nanorod as an orientation probe. With this method, we observed conformational changes of the catalytic β subunits of the F_1-ATPase from the thermophilic *Bacillus* PS3, a molecular motor driven by ATP hydrolysis. We resolved reversible conformational changes in β subunits among open, closed, and half-closed states. We also estimated the work generated by the β subunits. Based on these results, the mechanism how three catalytic β subunits rotate the central rotor subunit γ will be discussed.

2SCA-04 **Stochastic Regulation of DNA Mismatch Repair**

Jong-Bong Lee (*Dept. of Physics, POSTECH*)

DNA mismatch repair (MMR) system corrects DNA base pair errors. However, critical problems in the mechanism of DNA mismatch repair have been unsolved for decades. Using single-molecule approaches, we have revealed that the coordination of the molecular signaling for far distance transmission of the mismatch finding to the downstream site and the process of the strand excision is regulated stochastically. I will present how single-molecule methods have been used to attack the controversial issues in DNA mismatch repair.

2SCA-05 細胞内一分子計測で探るキネシンの制御機構

Dissecting kinesin regulation through single molecule in cellulo measurements

Yasushi Okada[1,2] ([1]*QBiC, RIKEN*, [2]*Dept. Phys., Grad. Sch. Sci., Univ. Tokyo*)

It has been established that conventional kinesin (KIF5) selectively moves along a specific subset of microtubules in living cells. For example, KIF5 is specifically recruited to the microtubules in the axon initial segment in neurons, which we propose to serve as the road sign for the axonally transported cargoes carried by KIF5. However, the mechanism of this selective binding is still controversial. Here we measured the binding rate constant of kinesin to microtubules in living cells and in vitro using single molecule fluorescence microscopy. The results suggested the existence of four populations of microtubules with different affinity to kinesin.

2SCA-06 Propagation of gene expression noise by RNA polymerase in living cells

Nam Ki Lee (*Dept. of Physics, POSTECH*)

Cell-to-cell variation resulting from noise in gene expression is a general phenomenon in cells, which often determines a cell's variability, and fate. Transcription is the key stage of noise generation in cells. However, how the noise in RNA polymerase (RNAP) is propagated to downstream gene expression noise remains unknown. Here, we quantitatively investigate the propagation of RNAP noise In the second part, I will introduce a novel single-molecule fluorescence approach for studying transcription in living bacterial cells, which show how the transcription and translation are coupled in living bacteria.

2SDA-01 細胞生物学のためのオンチップ高感度熱量センサ

On-chip high sensitive thermal sensors for cell biology

Takahito Ono, Naoki Inomata (*Tohoku University*)

High sensitive thermal sensors on a chip have been developed in order to investigate the heat production of single biological cells. On the detection chip, microchannels and high sensitive temperature sensors are integrated. Produced heat from a trapped cell in the channel is thermally conducted into a temperature sensor via a thermal heat guide. As temperature sensors, a resonant thermal Si sensor and a Si PN junction thermometer are integrated. Heat measurements from a single thermogenic cell (brown fat cell) are demonstrated, which shows that the developed device can detect the heat from the single BFC and will be a powerful tool for the measurements of thermal behavior of biological cells.

2SDA-02 蛍光センサーを利用した一細胞温度計測からわかること

What we see in single-cell thermometry by using fluorescent sensors

Madoka Suzuki[1,2] ([1]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*, [2]*JST, PRESTO*)

The single-cell thermometry is an emerging field at the interface between biology, chemistry and physics. The knowledge on the temperature at the single-cell scale should help us to understand the detailed mechanism of thermogenesis or assess the efficacy and the side effects of externally heating treatments such as hyperthermia at the larger scale. We have developed temperature sensors that are either small fluorescent molecules or fluorescent polymer nanoparticles. In this symposium, I will introduce our recent results of the organelle-targeted thermometry and the microthermography of small living organisms.

2SDA-03 様々な生物種の温度測定に利用でき且つ速い温度変化を測定可能な蛍光性温度プローブタンパク質

Genetically encoded ratiometric fluorescent thermometer with wide temperature range and rapid response

Masahiro Nakano[1], Yoshiyuki Arai[1], Ippei Kotera[2], Kohki Okabe[3,4], Yasuhiro Kamei[5], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ.*, [2]*RIES, Hokkaido Univ.*, [3]*Grad. Sch. Pharma., Univ. Tokyo*, [4]*JST, PRESTO*, [5]*NIBB*)

Temperature is a fundamental physical parameter responsible for biological events. Although conventional thermometers have found several important phenomena such as heat generation in mitochondria, development of a thermometer with a sensitivity to wide temperature range and rapid response is still desired to quantify temperature change in not only homeotherms but also poikilotherms from cellular level to *in vivo* level. Here, we report development of a genetically encoded fluorescent thermometer with the above desired property and demonstrate quantitative monitoring fast temperature changes in mitochondria as well as the thermometry in a living fish embryo.

2SDA-04 機能性磁性ナノ粒子を用いたガン温熱療法

Hyperthermia using functional magnetite nanoparticles

Akira Ito (*Dept. of Chem. Eng., Fac. of Eng., Kyushu Univ.*)

Magnetite nanoparticles-mediated hyperthermia has the potential to achieve tumor-targeted heating without any side effects. The technique consists of targeting magnetite nanoparticles to tumor tissue and then applying an external alternating magnetic field to induce heat generation by the magnetite nanoparticles. Recent years have seen the remarkable advances in development of both functional magnetite nanoparticles and alternating magnetic field generators. Currently, some researchers are attempting to begin clinical trials, suggesting that time may have come for clinical applications. I will review advances in magnetite nanoparticles-mediated hyperthermia.

2SDA-05 人工再構成系を用いた温度感受性 TRP チャネルの機能解析
Single channel analysis of the thermosensitive TRP channels in bilayer lipid membrane

Kunitoshi Uchida[1,2,3], Eleonora Zakharian[2], Makoto Tominaga[3], Jun Yamazaki[1] ([1]*Fukuoka Dent. Coll.*, [2]*Univ. of Illinois Coll. of Med.*, [3]*NIPS*)

Bilayer lipid membrane method is one of the unique electrophysiological techniques that is intended to study specific channel properties of the purified complexes in a well-controlled artificial environment. The advantage of this method reveals the possibility to study and characterize ion channel behavior at the single molecule level. Because the temperature affect anything in living cells, we think that simple model is necessary to understand the gating mechanisms of thermosensitive TRP channel by temperature changes. In this presentation, we will show you one example of the functional analysis of thermosensitive TRP channels by bilayer lipid membrane method and discuss the differences of temperature-dependent activation according to the methods.

2SDA-06 外温性および内温性動物の脳の発生と進化
Brain development and evolution of ectothermal and endothermal animals

Tadashi Nomura (*Dept. Biol. Kyoto Pref. Univ. Med.*)

Organisms live in various environments with different thermal conditions. Homeostatic controls of animals with unique thermal adaptations have been highlighted; however, the regulation of developmental processes of animals in different thermal conditions remain to be elucidated. Recently, we have reported that neural stem cell proliferation and differentiation of developing reptilian brains are much slower than those of mammalian brains. Based on these results, we are currently investigating how environmental temperature affects developmental speeds and pathways of ectothermal and endothermal animals. In this talk, I will introduce unique developmental potentials of reptilian embryos and discuss the evolution of embryogenesis in ectothermal and endothermal animals.

2SEA-01 剛体ドッキングによるタンパク質間相互作用表面のプロファイル解析
Profile analysis of protein interaction surface with rigid-body docking decoys

Nobuyuki Uchikoga (*Dept. of Physics, Chuo Univ.*)

To understand protein-protein interaction mechanisms, protein interaction surfaces are investigated using rigid-body docking process, which generates many possible protein complexes, referred as docking decoys. Protein interaction surfaces of whole decoys would contain clue for understanding protein interaction mechanisms. In this presentation, some works of profile methods are introduced. At first, profile of protein complex, based on amino acid residue sequences of both proteins, was used for searching near-native complexes. Then, this profile was applied to exploring docking space for obtaining more accurate near-native complexes, named as Re-docking scheme. Moreover, differences between true and false partners of protein interaction surfaces were investigated.

2SEA-02 マウスはやはりヒト炎症性疾患のモデルになる ―バイオインフォマティクス的手法によるマウスモデルの再評価―
Genomic responses in mouse models greatly mimic human inflammatory diseases

Keizo Takao[1,2,3] ([1]*Life Sci. Res. Ctr., Univ. Toyama*, [2]*Grad. Sch. Med. Pharm., Univ. Toyama*, [3]*NIPS*)

The use of mice models has been considered essential in modern biomedical research, but the role of them was challenged by a report that genomic responses in mouse models poorly mimic human inflammatory diseases. We reevaluated the same gene expression datasets used in the previous study by focusing on genes whose expression levels were significantly changed in both humans and mice. Contrary to the previous findings, the gene expression levels in the mouse models showed extraordinarily significant correlations with those of the human conditions. These findings demonstrate that gene expression patterns in mouse models closely recapitulate those in human inflammatory conditions and strongly argue for the utility of mice as animal models of human disorders.

2SEA-03 Development of an Efficient Amino Acid Substitution Matrix: MIQS

Kentaro Tomii[1], Kazunori Yamada[1, 2] ([1]*AIST*, [2]*Tohoku University*)

Sequence comparisons are still fundamental ways of finding relatedness of proteins. To improve pairwise amino acid sequence comparison methods, we have developed a novel sensitive matrix, which we designate as MIQS, based on benchmark results with the Structural Classification of Proteins (SCOP) database. We demonstrate performance improvement of classical pairwise sequence comparison methods by utilizing MIQS in terms of detection sensitivity and alignment accuracy. In addition, the performances of pairwise sequence comparison methods with MIQS are superior to that of CS-BLAST, which is one of the most sensitive methods, according to the benchmark on an independent dataset. We show typical examples of sequence comparisons with MIQS.

2SEA-04 ドッキングモデル構造群を用いたタンパク質間相互作用予測
Rigid docking based protein-protein interaction prediction by using high scoring docking models

Yuri Matsuzaki (*ACLS, Tokyo Tech.*)

Protein-protein interaction (PPI) plays key roles in living systems. Predicting relevant interacting partners from their tertiary structure is a challenging topic where computer science methods have potential to contribute. Protein-protein rigid docking has been applied for this purpose by several projects. However, the prediction power is limited mainly because of poor correlation between docking score and actual protein-protein binding affinity. To improve state of the art we introduce several approaches we have been applied to improve docking-based PPI predictions by using residue profiles obtained by rigid docking. We present an evaluation of our method by applying it to proteins of Protein Docking Benchmark ver. 5.0 and bacterial chemotaxis systems.

2SEA-05 An index to collect homologous sequences with the same or similar biochemical functions

Wataru Nemoto[1], Shoichiro Kato[1], Hiroyuki Toh[2] ([1]*Div. of Life Sci. & Eng., Sch of Sci & Eng., Tokyo Denki Univ.*, [2]*Dep. of Biomed. Chem., Sch. of Sci. & Tec., Kwansei Gakuin Univ.*)

For the investigation of the functional mechanisms of a protein by homology modeling, the structure of a protein with the same or similar function to a target amino acid sequence would be appropriate as a template. However, the structures retrieved by sequence similarity search or fold recognition programs do not always have the same or similar function to that of the target, because such programs do not directly evaluate the functional similarity. We recently developed an index to select homologous sequences for functional region prediction by integrating structure and sequence information. The selected sequences are expected to share the same or similar function. Therefore, we applied the index to select homologous sequences appropriate for homology modeling.

2SFA-01 Physical & quantitative aspects of immunology

Tetsuya J. Kobayashi[1,2], Taishin Akiyama[3] ([1]*IIS, Univ. Tokyo*, [2]*JST PRESTO*, [3]*Institute of Medical Science, The University of Tokyo*)

Adaptive immunity is a highly evolved adaptive system.
Despite of its complexity with a variety of cells and molecules involved, basic biophysical processes are fundamental to the immunological functionality. For instance, molecular interactions between immune cell receptors and antigens are responsible for triggering the immune response. Chemotaxis of immune cells are also crucial for detecting foreign antigens and for activating the immune responses.
Investigating such biophysical processes quantitatively are indispensable for understanding how a complex adaptive system operate efficiently and collectively.
In this talk, we show the physical aspects of immunology, and discuss potential contributions of biophysics and quantitative biology to the problems in immunology.

2SFA-02 T細胞活性化の一細胞分子イメージング
Single cell molecular imaging for T cell activation

Takashi Saito[1,2] ([1]*RIKEN-IMS*, [2]*IFReC Osaka Univ.*)

Immune response starts antigen (Ag) recognition by a single T cell upon interacting with an Ag-presenting cell (APC). The interaction induces specific organization of surface receptors called Immune synapse. Ag recognition generates T cell receptor (TCR) microcluster (MC), which recruits kinases and adaptors and serve as the activation site. TCR-MC is dynamically regulated depending on the quality and quantity of activation signals. T cell activation is positively and negatively regulated by co-stimulation and cell adhesion. The molecular interaction of these co-stimulation and adhesion molecules and TCR-MC is critical for the regulation. We show these interactions by quantitative imaging analysis of a single T cell for activation regulation.

2SFA-03 Application of stochastic models in quantitative immunology

Shunsuke Teraguchi, Yutaro Kumagai (*IFReC, Osaka Univ.*)

Many of the physical or fundamental processes in biology are intrinsically stochastic. Movement of immune receptor molecules and their intracellular signaling molecules on a cell surface is inevitably affected by randomness. Single cell behavior including gene or protein expression has large heterogeneity. Moreover, macroscopic phenomena like immune response also depend on stochastic cell-to-cell contact interaction among cells with different immune specificity. Thus, stochastic models, which take account of such intrinsic stochasticity of biological system, have wide application in immunology. Here we shall discuss some of application of stochastic models in our interdisciplinary projects in quantitative immunology.

2SFA-04 動的な誘引場に対する免疫細胞の走化性に見られる共通性と特異性
Generality and specificity in chemotaxis response of immune cells in dynamic gradients of chemoattractant

Akihiko Nakajima[1], Motohiko Ishida[2], Satoshi Sawai[1,2] ([1]*Res. Cent. Comp. Sys. Biol., Grad. Sch. Arts Sci., Univ. Tokyo*, [2]*Dept. Basic Sci., Grad. Sch. Arts Sci., Univ. Tokyo*)

How immune cells integrate temporal and spatial information from the surroundings to determine the direction of movement remains elusive. Here, through microfluidics manipulation of spatio-temporal chemoattractant profiles, we show that, chemotaxis of neutrophil-like HL60 cells exhibit time-scale dependence that highly resembles that previously shown for Dictyostelium cells. Rectified motion towards traveling wave stimulus of 4 min passage time was observed. For slower waves, the migration became symmetric between the wavefront and waveback. Based on pharmacological analysis, we propose that there are likely two layers of rectification mechanism; one that is Dictysotelium-like and the other that is unique in immune cells.

2SFA-05 適応免疫応答を調節するリンパ節内の細胞ダイナミクス
Cellular dynamics shaping adaptive immune responses in the lymph node

Takaharu Okada[1,2,3] ([1]*RIKEN Center for Integrative Medical Sciences*, [2]*PRESTO, Japan Science and Technology Agency*, [3]*Graduate School of Medical Life Science, Yokohama City Univ.*)

The lymphoid organs such as lymph nodes are structurally compartmentalized so that relevant cell types for each mode of immune responses are co-localized and able to access each other. During the last decade, live tissue imaging, in particular using two-photon laser microscopy, has advanced our understanding of immune cell trafficking mechanisms. In this talk, I will discuss the data from our live imaging experiments to visualize migration of B cells, T cells, and dendritic cell subtypes during homeostasis and immune responses in the lymph node. Our imaging data show how migration of these cells are regulated to spatiotemporally organize two different adaptive immune responses, the antibody response and cytotoxic T cell response.

2SFA-06 Quantitative analysis of T cell repertoire and homeostasis

Taishin Akiyama[1], Tetsuya J. Kobayashi[2] ([1]*Institute of Medical Science, The University of Tokyo*, [2]*Institute of Industrial Science, The University of Tokyo*)

Our immunity robustly protects our body from the invasion of unknown pathological organisms while avoiding attacking our body itself.
T cells and their diversity are relevant for discriminating self and non-self and also for controlling immunological responses.
Quantitative characterization of T cell population is, therefore, fundament to understanding how the T cell population is homeostatically maintained and effectively optimized.
In this talk, we show how we can use mathematical modeling and bioinformatics together with various single-cell measurement techniques such as NGS sequencing and cell sorting.

2SGA-01 脂質二分子膜を隔てた情報変換をとらえるクライオ電子顕微鏡単粒子解析法
Single particle cryoEM to elucidate signal transduction through lipiid bilayer membrane

Hideki Shigematsu[1,2,3] ([1]*RIKEN CLST*, [2]*Med. Life Sci., Yokohama City University*, [3]*Yale Univ. Sch. Med.*)

Since cryo-EM meets “resolution revolution” by introducing direct electron detectors, there we have seen a lot of near atomic resolution structures, even for membrane proteins in various conditions including in the lipid bilayer of nanodiscs. To elucidate function and structure relationship of voltage-gated ion channels, we employed liposome as a close to physiological condition to elucidate its functional structures with membrane potential. Here we report our recent progress in so-called random spherically constrained single particle reconstruction for ion channels.

2SGA-02 極低温電子顕微鏡構造に基づいた胃プロトンポンプ—胃酸抑制剤結合モデル
Binding model of the acid suppressant to the gastric proton pump based on cryo-EM structure

Kazuhiro Abe[1,2] ([1]*Cellular and Structural Physiology Institute, Nagoya Univ.*, [2]*Grad. Sch. Pharm.*)

The gastric proton pump, H^+,K^+-ATPase is responsible for the gastric acidification, hence, its inhibitors have been utilized as acid suppressants including recently developing K^+-competitive acid blockers (P-CABs). Here we show the binding model of P-CABs to the H^+,K^+-ATPase, based on 6.5Å electron crystallographic structure of H^+,K^+-ATPase with bound BYK99, a potent P-CAB with fixed ring structure. Together with docking simulations, corroborated by systematic mutagenesis and application of synthesized compounds, our proposed model provides useful information to improve currently developing new class of acid suppressants.

2SGA-03 電子顕微鏡を用いた繊毛の三次元構造解析
Three-dimensional electron microscopy of cilia

Toshiyuki Oda (*Grad. Sch. Med., Univ. Yamanashi*)

Cilia and flagella are conserved motile organelles that play essential roles in cellular motility of eukaryotes and development of higher organisms by generating fluid flow. The beating motion of cilia and flagella is driven by dynein motor proteins. Recent advances in the techniques of cryo-electron tomography and structural labeling revealed the molecular mechanism of the regulation of dynein motor activities as well as the structural configuration of the ciliary macromolecular complexes.

2SGA-04 CryoTEM のための CryoCLEM システムの最新アプリケーション
Latest application of CryoCLEM system for cryo-TEM

Ayumi Ishihara[1], Shinji Aramaki[2], Tomoya Higo[2], Takuo Yasunaga[2] ([1]*Leica Microsystems K.K.*, [2]*Grad. Sch. Computer Sci. & Systems Eng., Kyushu Inst. of Tech.*)

Various kinds of correlative light and electron microscopy; CLEM systems have been proposed. With this technique we can reduce the time of biological analysis from in situ to molecular level in recent years. I will talk about our cryoCLEM system that can be applied to cryo-TEM, one of the methods to analyze protein structure. It can observe vitreous frozen sample in cryogenic temperature with light microscope and cryo-TEM. I will report the correlation accuracy on it.

2SGA-05 Correlative Atomic Force and Transmission Electron Microscopy

Katsuya Shimabukuro[1], Yutaro Yamada[1,2] ([1]*NIT, Ube College*, [2]*Dep. of Bio. Kanazawa Univ.*)

In this study, we present a new technique called correlative atomic force and transmission electron microscopy (correlative AFM/TEM) to observe a targeted region of a sample under both microscopy. Owning to recent advances in time resolution in AFM, dynamic of biomolecules has been revealed, but specifying molecules by AFM alone still remains a challenge. Here, we demonstrate correlative AFM/TEM using actin filaments as a test sample and further show that immune electron microscopy can be integrated into this technique. Therefore specifying molecules imaged in AFM by subsequent immune EM has become possible. In conclusion, correlative AFM/TEM could be one of efficient methods to investigate complex biological system at molecular level in the future.

2SGA-06 Cryo-electron microscopy single particle analysis at near atomic resolution

Naoyuki Miyazaki, Kenji Iwasaki (*IPR, Osaka Univ.*)

Cryo-EM single particle reconstruction has been dramatically developed for the last decade and has been used to determine the near-atomic 3D structures of proteins in a physiological condition without crystallization. Two advanced cryo-electron microscopes have been installed at IPR for the structural determination at near-atomic resolution. One is a JEOL's microscope (JEM2200FS), equipped with an omega-type energy filter and a direct electron detector (Gatan; K2 summit). The other is a state-of-art FEI's microscope (Titan-Krios), equipped with an image corrector, a volta-phase plate, a direct electron detector (Falcon II), and automated data acquisition software. In this talk, we will present our recent results by using our cryo-electron microscopes.

2SAP-01 Cruising inside cells

Atsushi Miyawaki[1,2] ([1]*RIKEN BSI*, [2]*RIKEN RAP*)

Over the past two decades, various genetically encoded probes have been generated principally using fluorescent proteins. I will discuss how the probes have advanced our understanding of the spatio-temporal regulation of biological functions, such as cell-cycle progression, autophagy, and metabolism (retinoic acid and bilirubin), inside cells, neurons, embryos, and brains. I will speculate on how these approaches will continue to improve due to the various features of fluorescent proteins.

2SAP-02 NIR II/III (OTN-NIR)におけるバイオイメージング ー透明性を求めてー

Bioimaging in NIR II/III (OTN-NIR) seeking for transparency

Kohei Soga[1,2], Masao Kamimura[1,2] ([1]*Dept. Mater. Sci. & Tech., Tokyo Univ. of Sci.*, [2]*IFC, Tokyo Univ. of Sci.*)

The loss of electromagnetic wave with a wavelength around visible range consists mainly on scattering and the absorption tail of so-called infrared absorption caused by the atomic vibration. The shorter the wavelength is, the stronger the scattering is. The longer the wavelength is, more intense the infrared absorption is. As a results, the loss curve by them will form a “valley” around the near infrared (NIR) wavelength range, which is called “biological window.” The authors have utilized this biologically transparent window for bioimging by developing both fluorescent materials and imaging systems. The presentation will review a decade of the development together with the next generation imaging, NIR nanothermometry.

2SAP-03 半導体レーザー高機能パルス光源による多光子イメージング

Advanced semiconductor-laser optical pulse sources for multiphoton microscopy

Hiroyuki Yokoyama (*New Industry Creation Hatchery Center (NICHe), Univ. Tohoku*)

High-performance ultrashort optical pulse sources are increasing in importance for deep-site bioimaging via nonlinear multiphoton processes. In general, multiphoton microscopy (in typical, two-photon microscopy) uses femtosecond optical pulses. However, the principle of two-photon absorption and subsequent fluorescence processes tells us that picosecond pulses have the same capability as femtosecond pulses when the peak power and average power are the same. In fact, we have successfully demonstrated the world-deepest in vivo imaging of mouse brain tissues with a picosecond optical pulse source based on semiconductor laser diodes (LDs). Notable benefits of LD-base scheme are the potentialities of compact-size and long-period stable-operation.

2SAP-04 ベクトルビームを用いた共焦点顕微鏡法における分解能向上

Resolution enhancement in confocal microscopy with vector beams

Shunichi Sato, Yuichi Kozawa (*IMRAM, Tohoku Univ.*)

Further improvement of spatial resolution is one of the most important and pressing issues in imaging technology. While super-resolution techniques have achieved extremely high resolution far beyond diffraction limit of light, they brought considerable inconveniences such as sophisticated optical technique and specialized fluorescent material. By contrast, scanning laser microscopy has been well established and widely used in bio- and material- sciences. We have demonstrated that vector beams can create much smaller focal spot than conventional laser beams. We will show the basic ideas of resolution enhancement in confocal microscopy with vector beams, which achieved super-resolution imaging beyond diffraction limit.

2SAP-05 白色レーザーによるコヒーレント非線形光学イメージング

Coherent nonlinear optical imaging using a white-ligtht laser source

Hideaki Kano (*Inst. of Applied Physics, Univ. of Tsukuba*)

Coherent nonlinear optical microscopy has recently emerged as a powerful label-free molecular imaging tool which can detect non-fluorescent species in live cells or tissues. Thanks to recent advances in supercontinuum (white-light laser) generation technology, our system has extended to ultra-broadband spectroscopic live cell imaging. In the present study, the principle, instrumentation, and applications of coherent nonlinear optical microscopy will be described.

2SAP-06 光シート顕微鏡の改良と発生生物学への応用

Light-sheet microscopy: technical development and application for developmental biology

Shigenori Nonaka (*National Inst. for Basic Biol.*)

For measuring biological phenomena, light-sheet microscopy has several advantages such as very low photodamage to the samples, fast image acquisition rate, and relatively deeper penetration depth compared to confocal microscopy. On the last point, the effectiveness of combination with two-photon excitation had been already shown, however, this method suffered with the trade-off of narrow field of view just enough to visualize the width of drosophila embryos (~0.15 mm). We archived much wider field of view enough to visualize medaka (>0.7 mm), by using a high-pulse-power fiber laser. Besides, we will present our efforts to visualize high-speed biological phenomena such as cilia-driven fluid flow in mouse embryos that determine future left-right asymmetry.

2SBP-01 ラマン分光等イメージング技術で紐解く生命現象と情報伝達過程

Bio-imaging without staining: Raman imaging and others

Kotaro Oka (*Dep. Biosci. & Infor., Keio Univ.*)

Several bio-imaging techniques have been developed and used for the understanding the biological phenomena. In neuroscience field, neuroanatomy is fundamental; Golgi method for neuronal structure, immunostaining, and several retrograde- and anterograde-tracing for visualization of neural circuits. Recently, spectral imaging technology has been developed remarkably, and it enables to visualize the biological information without staining. For example, Raman microscopy has been widely used not only for identification of specific chemicals but also to distinguish developmental stages of ascidian embryos (Nakamura et al. 2013). In this talk, I will discuss the application of Raman microscopy and other spectral techniques for investigating the neuronal cytoarchiecture.

2SBP-02 細胞分化のバイオ・ラマン研究： 中間状態の検出

Bio-Raman Research on Cellular Differentiation to Detect the Reversible State

Shin-ichi Morita (*Tohoku Univ.*)

Recently it became possible to measure Raman spectra of a single live cell using a standard Raman microscope. Using Raman signals, we are capable of, for instance, analyzing the distribution of bio-molecules. Also, cutting edge applications were opened. Using the bio-Raman microscope, it is possible to monitor the dynamics of cellular differentiation, proliferation, and apoptosis, defining cellular states in a non-destructive and non-labeling manner. Based on the information of cellular conditions, we are expecting to control cellular destinies. Raman monitoring is probably effective for tissue engineering. The updated results are shown in the talk.

2SBP-03 蛍光プローブの精密設計による迅速癌検出

Rapid cancer imaging by rationally designed fluorescence probes

Mako Kamiya[1,2], Yasuteru Urano[1,3,4] ([1]*Grad. Sch. of Med., Univ. of Tokyo*, [2]*JST PRESTO*, [3]*Grad. Sch. of Pharm. Sci, Univ. of Tokyo*, [4]*AMED CREST*)

It has been a long-term goal to develop tumor-imaging techniques that have sufficient specificity and sensitivity, since early detection and complete resection are an important prognosticator for cancer treatment. Since fluorescence-guided diagnosis is one of the most powerful techniques for real-time in situ tumor detection, we have developed a series of fluorescence probes targeted to aminopeptidases that are overexpressed in tumors. By applying one of these probes to mice model of peritoneal metastasis, tiny disseminated tumors were visualized rapidly and sensitively. Also, by topical application of these probes to resected specimens from human cancer patients, it was revealed that our probes are valid for detecting breast cancer and esophageal cancer.

2SBP-04 バイオラマン顕微鏡を用いた卵子のクオリティー評価

Oocyte evaluation using Bio-Raman microscope

Yumi Hoshino (*Hiroshima University*)

In vitro maturation (IVM) is increasingly being used to treat human infertility, particularly as rescue therapy for patients with polycystic ovarian and ovarian hyperstimulation syndromes. Quality control and determining optimal fertilization conditions-vital to increasing IVM oocyte use for embryo production and improving pregnancy success rates-require thorough understanding of oocyte maturation mechanisms. This knowledge would allow high-quality oocyte production and selection based on predictive markers for successful oocyte maturation. Noninvasive, quantitative, and rapid evaluation is necessary for oocyte selection. I will discuss the applicability of the Bio-Raman microscope in oocyte evaluation.

2SBP-05 二本鎖 RNA オーバーハング構造結合選択性を有する合成蛍光プローブの開発と RNA 干渉研究への応用

Synthetic fluorescent probes capable of selective binding to 3'-overhanging structures in double-stranded RNAs for RNA interference study

Yusuke Sato (*Department of Chemistry, Graduate School of Science, Tohoku University*)

With increasing knowledge about the diverse roles of RNAs within cells, much attention has been paid to RNA-binding fluorescent probes as powerful tools for analyzing biological functions of RNAs. Recently, we developed new class of fluorescent probes that can recognize 3'-overhanging structures of RNAs. Our probes are based on peptide nucleic acid conjugated with fluorogenic intercalator and they exhibit the light-up fluorescence response upon selective binding to overhanging structures. I will present the molecular basis of such fluorescent probes and their application to the analysis of small interfering RNAs (siRNAs) involved in RNA interference mechanism in the living cells.

2SBP-06 細胞膜分子動態が語る細胞の個性

What membrane molecule dynamics tell us about the cell

Hiroko Bannai[1,2], Fumihiro Niwa[2,3], Misa Arizono[2,4], Katsuhiko Mikoshiba[2] ([1]*JST PRESTO*, [2]*RIKEN BSI*, [3]*IBENS, INSERM*, [4]*Univ. of Bordeaux*)

According to the fluid mosaic model, plasma membrane molecules such as lipids and transmembrane proteins have the ability to undergo lateral diffusion freely within the cell membrane. Using a single particle tracking technique with quantum dots (QD-SPT), we found that the mobility of some membrane molecules became abnormal in cellular models of epilepsy and Alzheimer's disease. We also found that cells isolated from animals doomed to develop neuronal disorders preserved such abnormality in membrane molecules behavior, even though they were isolated before symptom onset. Based on these findings, here we discuss the possibility that the behavior of membrane molecules reflects the destiny and properties of individual cells.

2SCP-01 線虫胚における細胞質流動のイメージングとモデリング

Imaging and modeling of cytoplasmic streaming in the *C. elegans* embryo

Akatsuki Kimura[1,2] ([1]*Cell Arch. Lab., Nat. Inst. Genet.*, [2]*Dept. Genet., SOKENDAI*)

Cytoplasmic streaming is a cell-wide flow occurring in various plants and animals. In *Caeorhabditis elegans*, the streaming of cytoplasm is observed upon fertilization. As the polarity of the zygote is not established at this stage, the direction of the cell-wide flow is not pre-determined, but determined in a self-organized manner. Interestingly, the direction of the flow reverses during the streaming. Using a quantitative live-cell imaging, we characterized the dynamics of the flow, and proposed a mechanism for the self-organization of the flow. The proposed mechanism was tested using a theoretical modeling. I will discuss the applicability of our model on cytoplasmic streaming in general.

2SCP-02 Profilin-1 membrane dynamics in live cells

Pierre Moens (*Univ. of New England*)

Profilin is known to bind and sequester G-actin but also to interact with phosphatidylinositol polyphosphate at the cell membrane. We used iMSD, pair correlation and N&B analysis of MDA cells expressing profilin-GFP to investigate its membrane diffusion rate and aggregation. When actin polymerisation is disrupted by Cytochalasin D, we see a decrease in profilin diffusion rate due to the sequestration of fast diffusing profilin by actin monomers. Cell connectivity also shows that the slower diffusion of profilin at the membrane may result from increased obstacles for this population. Finally, the disruption of actin filament leads to an increased rate of profilin cluster assembly/disassembly possibly highlighting a cell mechanism to generate actin filaments.

2SCP-03 細胞機能に関わる細胞内 pH の計測

Fluorescence imaging of cytoplasmic pH associated with cellular functions

Yusuke V. Morimoto[1], Masahiro Ueda[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*)

Intracellular pH plays a key role in a wide range of biological processes. Cytoplasmic pH change is also important for signal transduction in the social amoebae *Dictyostelium discoideum*. Chemotactic stimulation by cAMP was reported to elicit an efflux of protons transiently. On the other hand, cell differentiation into stalk cells causes decrease in cytoplasmic pH. However it remains unclear how the intracellular pH change works in the cellular functions. To investigate the role of intracellular pH, we constructed *Dictyostelium* strains expressing a highly pH-sensitive fluorescent protein and observed the cytoplasmic pH change associated with cell dynamics in both unicellular and multicellular stages.

2SCP-04 Pair correlation microscopy reveals nanoparticle shape to control intracellular transport

Elizabeth Hinde (*Univ. of New South Wales*)

Viruses have developed mechanisms of cellular entry over millions of years and shape has evolved as a critical feature to overcoming barriers which restrict access to target destinations. The efficiency at which pathogens overcome cellular barriers raises the question whether drug carriers can be designed to take an 'optimal' route and enhance chemotherapeutic efficacy. Mapping molecular mobility across cellular compartments is difficult since live cell imaging has insufficient spatiotemporal resolution to follow a population of molecules in real time. Here we used pair correlation microscopy to map the mobility of different shaped nanoparticles across subcellular compartments and measure the impact this has on doxorubicin toxicity.

2SCP-05 Fluidic microenvironment in live cells revealed by standard molecules and nanoparticles

Chan-Gi Pack[1], Min-Kyo Jung[1], Sung-Sik Han[2] ([1]*University of Ulsan College of Medicine & AMC*, [2]*Korea University*)

For quantitative analysis of cellular environments, use of protein molecules and nanoparticles as standard nanoprobes has been demonstrated. Using fluorescence correlation spectroscopy, we compared the diffusional properties of the standard probes in the compartments of live cells as well as in aqueous solution. The probes in each cellular compartments present free diffusion according to Stokes-Einstein relation indicating that cellular compartments can be characterized by a local fluidic viscosity in submicrometer region. Interestingly, the diffusion in a specific compartment is significantly changed by physiological conditions. The study will provide insight into strategies to understanding of molecular function through its hydrodynamic process.

2SCP-06 RGB カラーの蛍光タンパク質センサーによる細胞内 ATP の時空間イメージングと定量解析

Spatiotemporal imaging and quantitative analysis of subcellular ATP using RGB-colorful fluorescent protein based indicators

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Fluorescent indicators capable of optically detecting intracellular adenosine triphosphate (ATP) are promising tools to elucidate its spatiotemporal dynamics in a single cell. Using single fluorescent proteins, we developed red, green, and blue (RGB-color) intensiometric indicators which exhibited the change in the fluorescence intensity at a single wavelength upon sensing ATP. The toolset of the indicators enabled us to simultaneously visualize the dynamics of ATP in cytoplasm and mitochondria at single cells. Furthermore, we found that the fluorescence lifetime of indicators as well as intensity altered in the presence of ATP. Taking advantage of fluorescence lifetime, we propose a novel quantitative method for analyzing subcellular ATP concentration.

2SCP-07 X 線レーザーによる生きた細胞のナノイメージング

Imaging live cell at the nanoscale by X-ray laser diffraction

Yasumasa Joti[1,2] ([1]*JASRI*, [2]*RIKEN SPring-8 center*)

X-ray free-electron lasers (XFELs) with femtosecond pulse duration enable single-shot imaging almost free from sample damage by outrunning major radiation damage processes. In bioimaging, it is important to keep the sample close to the natural condition. Here we present a method, named as pulsed coherent X-ray solution scattering (PCXSS), for capturing a snapshot of live cells kept in a micro-liquid enclosure array (MLEA) by XFEL diffraction. We placed living Microbacterium cells in the MLEA and successively exposed each enclosure to a single XFEL pulse from SPring-8 Angstrom Compact Free-Electron Laser (SACLA). The reconstructed image revealed living cell structure at the nanoscale without any staining, which helps advanced understanding of intracellular phenomena.

2SDP-01 マイクロ秒分解一分子蛍光測定でみる変性タンパク質のダイナミクスとタンパク質折り畳み転移

Microsecond tracking of unfolded protein dynamics and protein folding transitions by single-molecule fluorescence spectroscopy

Hiroyuki Oikawa (*IMRAM, Tohoku Univ.*)

Recently, a surprising advance in the biophysics is the molecular dynamics calculations, which becomes possible to follow the folding dynamics of proteins for more than several milliseconds. However, the experimental verification of the calculations is still difficult. One possibility is the single-molecule fluorescence measurements; however, the time resolution of the conventional method is limited to milliseconds. To obtain the microsecond resolution for tracking protein dynamics, we developed the line-confocal microscope combined with the sample flow system. On our data, unfolded proteins exhibited the submillisecond dynamics and the heterogeneous conformations. In addition, we could track the fast folding/unfolding transitions. We will discuss these results.

2SDP-02 タンパク質凝集体の表面から突出したポリペプチド鎖は分子シャペロンによる脱凝集効率に影響を与える

Polypeptides protruded from the surface of protein aggregation influence the efficiency of disaggregation by molecular chaperones

Yo-hei Watanabe[1,2], Takashi Yamasaki[1], Tatsuya Nojima[3], Akiyoshi Oda[1] ([1]*Dept. Biol., Facult. Sci. Eng., Konan Univ.*, [2]*Inst. Integrated Neurobiol., Konan Univ.*, [3]*IIR, Tokyo Tech.*)

Under various stresses, proteins with organized tertiary structures are disorganized and irreversibly aggregated. Unconventional chaperone ClpB/Hsp104 can reorganize the aggregated proteins. The chaperone forms ring shaped oligomer and threads the aggregated proteins through the central pore of the ring by using energy of ATP. Recently, we evaluated accessibilities of a certain part of the aggregated protein, by using a split-GFP system. The results showed that the significant part of aggregated proteins protruded from the aggregation core and that the extent of the protrusion correlated with the disaggregation efficiency by the ClpB. In this symposium, we discuss how we can control the extent of the protrusion and how the protrusion contributes to the disaggregation.

2SDP-03 Integrated in vivo and in vitro nascent chain profiling reveals widespread translational pausing

Yuhei Chadani[1,2], Tatsuya Niwa[1], Shinobu Chiba[2], Hideki Taguchi[1], Koreaki Ito[2] ([1]*Inst. of Innovative Research, Tokyo Inst. of Tech.*, [2]*Fac. of Life Sci., Kyoto Sangyo Univ.*)

The synthesis of a protein takes tens of seconds to a few minutes, in which amino acids are polymerized linearly. Non-uniform progression of this elongation process is thought to be important for the subsequent fates of newly synthesized proteins. Here we attempted to detect systematically the accumulation of tRNA-linked nascent chain intermediates during the translation of *Escherichia coli* proteins in vivo and in vitro. The results revealed the widespread occurrence of translational pausing in a manner correlated with the subcellular localization and solubility properties of proteins. Our in vivo/in vitro integrated nascent chain profiling provides groundwork information for our understanding of genetic message translation into functional proteins.

2SDP-04 天然タンパク質の分子サイズに関する統計解析

Statistical analysis on the molecular size of native proteins

Hidenobu Kawai, Daisuke Takahashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

A radius of gyration, R_g, is one of the most important parameters characterizing structural properties of a protein. It has been theoretically shown that an R_g of a polymer scales with the number of residues, N, as $R_g = R_0 N^\nu$, where ν is a scaling exponent. To examine whether the scaling relationship holds for native proteins, we calculated R_g and N of protein domains in the SCOPe database. We found that the scaling relationship holds for native proteins, especially for the maximally compact native proteins having the lowest R_g at each chain length. The maximally compact proteins are rich in α/β-structures and disulfide bonds and have functions related to metabolism and hydrolase. These results suggest that protein functions can be restricted by structural constraints.

2SDP-05 ユビキチン化に伴う蛋白質の凝集体形成
Ubiquitylation-induced protein aggregation

Daichi Morimoto[1], Erik Walinda[2], Harumi Fukada[3], Kenji Sugase[1], Masaru Hoshino[4], Takashi Fujii[5], Keiichi Namba[6], Masaaki Komatsu[7], Keiji Tanaka[8], Masahiro Shirakawa[1] ([1]*Eng., Kyoto Uni.*, [2]*Med., Kyoto Uni.*, [3]*Life Envi. Sci., Osaka Pref. Uni.*, [4]*Pharm., Kyoto Uni.*, [5]*Frontier Biosci., Osaka Uni.*, [6]*Frontier Biosci., Osaka Uni.*, [7]*Med., Niigata Uni.*, [8]*Lab. Protein Metabolism, Tokyo Metro. Ins. Med. Sci.*)

Ubiquitin is an extremely rigid protein that tolerates high pressure or temperature. However, ubiquitin-positive protein aggregates are known to be a hallmark of various neurodegenerative diseases. To gain insight into the intracellular aggregate formation of ubiquitin, we examined the physicochemical properties of its polymeric chains. We found that the folding stability of ubiquitin chains decreased with increasing chain length, resulting in amyloid-like fibril formation. When expressed in cells, ubiquitin chains also formed aggregates depending on chain length. Notably, these aggregates were selectively degraded by autophagy. We propose that the instability of ubiquitin chains drives fibril formation, which serve as an initiation signal for autophagy.

2SDP-06 分子シャペロンによるプロテアソームタンパク質分解の制御
Regulation of proteasomal degradation by molecular chaperone

Tomonao Inobe (*Grad. Sch. Sci. and Eng., Univ. Toyama*)

Most unneeded proteins in the cell are degraded by the proteasome. Proteins to be degraded are targeted to the proteasome through the attachment of polyubiquitin chain. Effective proteasome-mediated proteolysis also requires the presence of an unstructured region in the substrate proteins. Such an unstructured region would be recognized by a various cellular molecular chaperones. Here, we present evidence that the molecular chaperone inhibits the proteasome-mediated proteolysis by competitively binding to the unstructured degron on the substrate. Based on this mechanism, we further develop “artificial chaperones” which recognize specific unstructured degrons and thus inhibit degradation of specific proteasome substrates.

2SEP-01 高圧力顕微鏡法で生きた細胞内で働く分子機械を操作する
High-pressure microscopy for controlling molecular machines in living cells

Masayoshi Nishiyama (*The HAKUBI Center, Kyoto Univ.*)

Hydrostatic pressure is an isotropic mechanical action to change the intermolecular interactions between protein and water molecules. This means that applied pressure can induce the structure and function of molecular machines. We have developed a high-pressure microscope that enables us to acquire various microscopic images, regardless of applied pressures. The developed system allowed us to control the molecular machines such as F1-ATPase and kinesin motors. Here, we will show that applied pressures can control the molecular machines in living cells, and then activate the cell motility.

2SEP-02 生細胞内における生体分子動態マッピングに向けて
Researchs towards bio-molecular dynamics mapping in cell

Johtaro Yamamoto (*Faculty of Adv. Life Sci., Hokkaido Univ.*)

Bio-molecules move around randomly or directionally in cell, and that is essential to correctly play their roll because bio-molecules should function in appropriate place and compartment. Most bio-molecules work by interacting with other bio-molecules, and as a result of such interactions the speed of diffusion changes. Thus, analyzing such the dynamics is very important to understand functions of bio-molecules. We have been developing several methods to reveal bio-molecular dynamics distribution in cell towards the bio-molecular dynamics mapping. In this symposium, we will introduce multipoint fluorescence correlation spectroscopy (multipoint FCS) and multiple scan speed raster image correlation spectroscopy (RICS).

2SEP-03 絶対零度で蛍光 1 分子を見る
Fluorescence microscopy of single molecules at a few K.

Satoru Fujiyoshi (*Tokyo Tech*)

We have developed a cryogenic fluorescence microscope for observing individual molecules. The microscope consists of reflecting optics and is perfectly achromatic. In the presentation, I show a recent development of the microscope and discuss the possibility of an imaging of cellular structures.

2SEP-04 位相差法による無染色での試料の同定法の開発
Apodized phase contrast imaging for identification of specimens without staining

Tatsuro Otaki[1,2] ([1]*Core Technology, Nikon Corp.*, [2]*Grad. Sch. Biomed. Eng., Tohoku Univ.*)

We developed a new test method for identification of asbestos by apodized phase-contrast microscopy. The method utilizes dispersion of light and is based on a measurement of refractive indices of asbestos. Apodized phase-contrast microscopy was designed for reducing halo artifacts and imaging fine structures. We assumed that a 0.2 μm of diameter objects can be seen in a liquid immersion media. The newly designed phase plate has a phase ring which alters one quarter phase shift throughout the most of visible region with 2% transmittances. The phase plate also has two apodization areas with 8% transmittances. We propose that this identification method applies to phase specimens such as cellular organelles without staining.

2SEP-05 光の波面を制御して散乱体を透視する

Seeing through scattering media by controlling wavefront of light

Tomohiro Shirai, Kaoru Katoh (*AIST*)

To see inside biological samples clearly and noninvasively is essential for modern biological research. The widely-known techniques to achieve this include optical coherence tomography and confocal microscopy. However, these techniques are applicable only to the surface of the sample, up to at most several hundred micrometers in depth, because they utilize very weak single-scattered light from the sample to form images.

In this talk, we outline a recently-proposed alternative technique using multiple-scattered light which is generally dominant in biological samples. Specifically, we show that it is possible to achieve noninvasive, high-resolution, and deeper imaging through scattering media such as biological samples by controlling wavefront of the incident light.

2SEP-06 構成分子の位置および向きの１分子観察から読み解く分子会合のダイナミクス

Dissection of molecular assembly dynamics by tracking orientation and position of single molecules in live cells

Tomomi Tani (*Marine Biological Laboratory*)

In living cells, the 3D architecture of molecular assemblies such as chromosomes, lipid bilayers and the cytoskeleton is regulated through the interaction among their component molecules. Monitoring the position and orientation of constituent molecules is important for understanding the mechanisms that govern the architectural dynamics of these assemblies. We have developed an instantaneous fluorescence polarization microscope to track the position and orientation of fluorescently labeled particles in real time with single molecule sensitivity in living cells while the particles interact with stable assemblies. Our imaging is broadly applicable to the study of dynamic molecular interactions that underpin the function of micron-scale assemblies in living cells.

2SFP-01 スパースモデリングとデータ駆動科学

Sparse modeling and date driven science

Masato Okada (*Univ. of Tokyo*)

Our project “the Initiative for High-dimensional Data-Driven Science through Deepening of Sparse Modeling” is supported by JSPS Grants-in-Aid for Scientific Research on Innovative Areas. The aim of this project is to establish a novel framework to make the tight connection of information science to the original purpose of data analysis derived from various scientific disciplines, namely, data-driven science. In this talk, I introduce the basic concept of sparse modeling and illustrate the Bayesian spectral deconvolution with the exchange Monte Carlo method. Through the project, we have become convinced that the three levels pointed out by David Marr give a novel insight into data-driven science, and proposed three levels of data-driven science.

2SFP-02 Recent development of Monte Carlo sampling techniques

Koji Hukushima (*Univ. of Tokyo*)

Monte Carlo methods, along with molecular dynamics (MD) method, are considered to be a simulation tool for the equilibrium Gibbs distribution in physics. Protein folding simulations might serve as a prime example in biophysics. Other applications of the MC methods extensively used are Bayesian statistics. Recently, an inverse modeling for a given experimental data set, often called data-driven science, has grown increasingly important in science. The MC methods could be a useful tool, in particular, with increasing the data size. In this talk, we discuss recent development of Markov-chain Monte Carlo method based on a lifting technique which breaks a detailed balance condition. We also discuss a population-type Monte Carlo method for approximate Bayesian computation.

2SFP-03 Fourier imaging with sparse modeling: An application to black hole astronomy

Mareki Honma (*NAOJ Mizusawa*)

Fourier analysis is ubiquitously used in various fields of experimental science raging from biophysics such as NMR, MRI and X-ray diffraction imaging to astrophysics such as radio interferometer. In this talk, I would like to introduce an application example of new technique called “sparse modeling” to effectively solve under-determined Fourier equation and demonstrate that such technique has a potential to provide a breakthrough in Fourier and similar analyses. In particular, I focus on its application to radio astronomy to directly resolve the “shadow” of the super-massive black holes at the centers of galaxies, which still remains as a future milestone in the field of astronomy, and discuss the impact of sparse modeling on Fourier imaging and related fields.

2SFP-04 ベイズ解析を用いるＸ線１分子観察

X-ray Single Molecule Observations using Bayesian Analysis

Yuji Sasaki (*Graduate School of Frontier Sciences, The University of Tokyo*)

Diffracted X-Ray Tracking (DXT) using normal synchrotron orbital radiation source has been developed for obtaining the information of the 3D internal motions of single protein molecules with both high time-resolution and high precision. Recently, we succeeded in observing picometer-scale Brownian motions of individual functional membrane protein channels, and structural fluctuations of intrinsically disordered proteins. Basically, DXT monitor random Brownian motions of inter-molecular dynamics in individual single protein. We need the statistical technique which deals with a great deal of data at high speed. Additionally, in new DXT using a laboratory X-ray source(DXT-lab), dynamic information decreases sharply, so data processing becomes more important.

2SFP-05 スパースNMRデータを用いた細胞内蛋白質立体構造決定
Protein NMR structure determination for sparse data set dervied from living cells

Teppei Ikeya[1,2], Shiro Ikeda[3], Takanori Kigawa[4], Yutaka Ito[1,2], Peter Guentert[5] ([1]*Tokyo Metropolitan University, Graduate School of Science and Engineering*, [2]*CREST, JST*, [3]*The Institute of Statistical Mathematics*, [4]*RIKEN, QBiC*, [5]*Goethe University Frankfurt am Main*)

Investigating three-dimensional (3D) structures of proteins in living cells by in-cell nuclear magnetic resonance (NMR) spectroscopy opens an avenue towards understanding the structural basis of their functions and physical properties under physiological conditions inside cells. Here we introduce methods that enable in-cell NMR protein structure determination for a larger number of proteins at concentrations that approach physiological ones. The new methods comprise advances in NMR data processing of non-uniformly sampled NMR data, automatic chemical shift assignment, and structure refinement with Bayesian inference, which makes it possible to calculate accurate 3D protein structures from sparse data sets of conformational restraints.

2SGP-01 クリプトクロム蛋白質の柔軟なループ構造への多重リン酸化は哺乳類概日時計の周期長を相加的に制御する
Multiple phosphorylation at flexible loops of cryptochrome additively modulates the period of mammalian circadian clock

Koji L. Ode[1,2], Hiroki R. Ueda[1,2] ([1]*Dept. of Sys. Pharm., Grad. Sch. of Med., the Univ. of Tokyo*, [2]*QBiC, RIKEN*)

Mammalian cryptochrome 1 (CRY1) is a transcriptional repressor that closes a negative feedback loop to drive autonomous oscillation of circadian clock. Serial mutagenesis of phosphorylation sites, identified by our phospho-proteomics revealed that phosphorylation near the p-loop/c-lid domains of CRY1 significantly alters the circadian period. Structural information indicates that these loops are highly flexible, and conversely, phosphorylation (or charge) states of modification sites additively regulate the oscillation period from 18 hours to 36 hours. In summary, this study suggests that circadian period is continuous and controllable function, and determined by the inherent structural flexibility of CRY1 protein.

2SGP-02 リン酸化で規定されるタンパク質コンフォメーションから細胞の応答性を予測する
Signaling protein conformation regulated by multiple phosphorylations points in the direction of cell fate

Kayo Hibino[1,2,3] ([1]*NIG*, [2]*SOKENDAI*, [3]*RIKEN*)

Cellular individuality in response to extracellular signals is a major limiting factor in the prediction and manipulation of cellular behaviours. However, origins of the individuality are still unclear. Here, we report detection and manipulation of the initial condition of individual cells, which specifies response to growth factors (GF). The initial condition was described by conformation of RAF, which is a hub for cell signaling. RAF conformation was regulated by multiple phosphorylations. By detecting RAF conformation, the strength of GF-response in individual cells was predicted. Based on the findings, we demonstrate regulation of the conformation and suppression of the excess activation of a disease-related RAF mutant.

2SGP-03 CaMKIIαとカルシニューリンによる神経入力情報のデコーディングと表現
Nonlinear Decoding and Asymmetric Representation of Neuronal Input Information by CaMKIIα and Calcineurin

Hajime Fujii, Masatoshi Inoue, Haruhiko Bito (*Department of Neurochemistry, Grad. Sch. of Medicine, The Univ. of Tokyo*)

How information encoded in glutamate release rates is converted into biochemical activation patterns of CaMKIIα and calcineurin remains unexplored in living neurons. We developed dFOMA (dual FRET with optical manipulation) imaging that permitted simultaneous measurement of CaMKIIα and calcineurin activities in spines while varying glutamate uncaging frequencies. dFOMA imaging during spine morphological plasticity revealed that CaMKIIα and calcineurin, have distinct spatiotemporal kinetics during induction of spine morphological plasticity and different information processing propertie. These results provide evidence that CaMKIIα and calcineurin are fine-tuned to unique bandwidths and compute input variables in an asymmetric manner.

2SGP-04 リン酸化アイソタイプの定量解析によるシナプスリン酸化シグナル伝達の新知見
Novel insight of synaptic phosphorylation signal transduction by quantitative analysis of phosphoisotypes

Tomohisa Hosokawa (*RIKEN BSI*)

Nowadays mass spectrometry and immunoblotting with phosphospecific antibody are major techniques to analyze phosphorylation. However, those techniques are not enough to obtain hidden information about protein phosphorylation. One is a combination of phosphorylation at single protein molecule. When one protein has n of possible phosphorylation site, there are 2n kinds of possible forms (phosphoisotypes) including non-phosphorylated form. One is stoichiometry of those phosphoisotypes, especially proportion of phosphorylated group and non-phosphorylated form among total protein amount. In this presentation, we will show recent progress on quantitative analysis of phosphoisotypes about synaptic protein as an example of novel insight of phosphorylation signal transduction.

2SGP-05 動的蛋白質リン酸化による生命現象の時間スケール調節
Dynamic protein phosphorylation as a time “scale” machine

Tetsuhiro S. Hatakeyama (*Department of Basic Science, The University of Tokyo*)

There are some problems of inconsistencies between timescales in micro and macro biosystems. 1) Timescale separation: Although the characteristic timescales of enzymatic turnovers are typically from sub-milliseconds to seconds, timescales of ordinary biological behaviors are longer and have a wide variety. 2) Inconsistency in environmental dependence: Although the speed of typical biochemical reactions depends on the temperature, timescales of some biological systems, e.g., the circadian clock, are less sensitive to the temperature. To approach these problems, we demonstrate that sequential phosphorylation reactions can work as a global regulator of biological time scales via the enzyme-limited competition. We discuss some examples of such timescale regulation.

2SGP-06 **微小管ダイナミクスを制御する異なる特異的リン酸化カスケード**
Site-specific Phosphorylation Cascades that Differentially Regulate Microtubule Dynamics in Neuron

Tadayuki Ogawa, Nobutaka Hirokawa (*Grad. Sch. Med., Univ. Tokyo*)

Microtubules (MTs) provide dramatic cytoskeletal changes in cells. However, the regulatory mechanisms underlying MT dynamics remain unknown. We identified two different sets of phosphorylation profiles of KIF2A that accelerate (A-type) and brake (B-type) the MT depolymerization activity of KIF2A, respectively. Brain-derived neurotrophic factor (BDNF) stimulates PAK1 and CDK5 kinases, which decrease the MT depolymerizing activity of KIF2A through B-type phosphorylation, resulting in enhanced outgrowth of neural processes. In contrast, lysophosphatidic acid (LPA) induced ROCK2 kinase, which suppresses neurite outgrowth from round cells via A-type phosphorylation. These two exclusive forms of KIF2A phosphorylation differentially regulate MT in neuron.

3SAA-01 **Mitochondrial-mediated antiviral immunity and oxidative phosphorylation**

Takumi Koshiba (*Dep of Biol., Fac. of Sci., Kyushu Univ.*)

Mitochondria, well known as the powerhouse of eukaryotic cells, act as a platform in the innate antiviral immunity of mammals. Because mitochondria are believed to have evolved from organisms such as alpha-proteobacterium, their newly discovered role of branching into the host-cell defense was unexpected. In the symposium, our recent insights into the fundamental phenomenon of mitochondrial involvment in cellular innate antiviral immunity will be discussed.

3SAA-02 **線虫の塩忌避学習による行動変化に関与する神経の同定及び神経回路の解析**
Identification of neurons and analysis of the neuronal circuit involved in the learned salt-avoidance behavior in C. elegans

MoonSun Jang[1,3], Yu Toyoshima[1,3], Hirofumi Kunitomo[1,3], Manami Kanamori[1,3], Takayuki Teramoto[2,3], Takeshi Ishihara[2,3], Yuichi Iino[1,3] ([1]*Department of Biological Sciences, Graduate School of Science, The University of Tokyo*, [2]*Department of Biology, Faculty of Science, Kyushu University*, [3]*CREST, Japan Science and Technology Agency*)

Caenorhabditis elegans, with 302 neurons in total, is a suitable model to understand a complex neural network system in animal brain. Previously, we demonstrated that C. elegans learns to avoid the salt concentration at which it was previous cultivated under starvation. Here, we found that only three sensory neurons, ASER, ASGR and ASGL, contribute to learned avoidance behavior after starvation. Moreover, to determine which neuronal circuit is involved in learned behaviors, we monitored neuronal activity of the whole head neurons in well fed or starved worms expressing the calcium probe, Yellow Cameleon (YC) 2.60, under pan-neuronal promoter using a 4D imaging system. A lot of neurons showed several kinds of responses depending on sensory stimuli or movement state.

3SAA-03 **蛍光イメージングで紐解くインフルエンザウイルス感染の分子基盤**
The molecular basis of influenza virus infection unveiled by fluorescence imaging

Yusuke Ohba, Yoichiro Fujioka, Shinya Nishide, Asuka Nanbo (*Department of Cell Physiology, Hokkaido University Graduated School of Medicine*)

Influenza A virus (IAV) infection causes severe respiratory disorders and its seasonal and pandemic epidemics are significant public health concerns. IAVs are known to be internalized into cells via clathrin-mediated endocytosis; however, the precise molecular mechanism how IAV highjacks the cellular machineries has yet to be elucidated. By using fluorescence imaging, we have demonstrated that the signaling network mediated by Ca^{2+} is demonstrated as a key host-oriented mechanism for viral entry. Recently, we succeeded in identifying a cellular interface through which IAVs induced a Ca^{2+} increase. Our data might provide a better understanding of the molecular bases of IAV infection and a potential therapeutic target in IAV infection.

3SAA-04 **ATP イメージングにより明らかになったアポトーシス細胞における細胞内 ATP 濃度変化の仕組み**
ATP imaging revealed a mechanism of intracellular ATP changes during apoptosis

Hiromi Imamura (*Graduate School of Biostudies, Kyoto University*)

Dynamics of ATP levels in single apoptotic cells was imaged by using a fluorescent ATP biosensor. It was observed that intracellular ATP levels started to decline after activation of caspase-3, and were almost depleted within 1 hour. Either pharmaceutical inhibition or siRNA knockdown of pannexin-1 channel significantly abrogated the decrease in intracellular ATP of apoptotic cells, while overexpression of the channel accelerated it. Interestingly, apoptotic cells treated with pannexin-1 inhibitor continued to consume glucose. It is, thus, likely that apoptotic cells suppress their metabolism by decreasing intracellular ATP in order to reduce wasting nutrients.

3SAA-05 **可溶性因子を介した免疫細胞相互作用の１細胞モニタリング**
Monitoring immune-cell communication via soluble factors at single-cell resolution

Yoshitaka Shirasaki[1,2] ([1]*Grad. Sch. Sci., Tokyo Univ.*, [2]*IMS, RIKEN*)

Secretion of soluble factors, which mediate cell-cell communication, is a key process for maintaining homeostasis and regulating functions of the immune system. Recent advantages of single-cell analysis revealed the heterogeneity and fluctuation of gene expressions including protein secretion response. Our question is how the cells overcome their randomness and autonomously control the immune system. To address this, we have developed a platform for imaging the cellular secretion dynamics of individual cells by fluorescence immunoassay on TIRF microscopy. In this meeting, I will talk about our recent works on visualizing onsets of inflammatory and allergic response by innate immune cells.

3SAA-06 Imaging RNA in living neural circuits with hybridization-sensitive fluorescent probes

Dan Ohtan Wang[1,2] ([1]*Institute for Integrated Cell-Material Sciences, Kyoto University*, [2]*K-CONNEX*)

Photoquenching-based fluorescence imaging techniques are becoming increasingly important in live-cell imaging. Technical improvement has allowed, for the first time, effective RNA molecule detection at a single copy or at thousands of copies in a living cell. We aim to detect activity-triggered changes in gene expression in neuronal circuits using photoquenching techniques in order to dissect the molecular and cellular basis of learning. In my presentation, I will introduce two newly developed RNA labeling technologies that may be potentially used to achieve such goals in living animal brains.

3SBA-01 二次元蛍光寿命相関分光法で観るマイクロ秒領域の生体分子の熱ゆらぎ
Thermal fluctuation of biomolecular conformation on microsecond timescale detected by 2D fluorescence lifetime correlation spectroscopy

Kunihiko Ishii[1,2], Tahei Tahara[1,2] ([1]*Molecular Spectroscopy Lab., RIKEN*, [2]*RIKEN Center for Advanced Photonics*)

In order to study the dynamics-function relationship of biomolecules, it is crucially important to see dynamic events in a native-like condition with the best achievable structural and time resolutions. Two-dimensional fluorescence lifetime correlation spectroscopy (2D FLCS) is a quasi-single-molecule method which enables us to investigate thermal fluctuation of biomolecules in equilibrium utilizing FRET-based structure determination with a microsecond time resolution. We will illustrate the principle and practice of 2D FLCS and demonstrate a few applications in which 2D FLCS uncovers folding dynamics of proteins and nucleic acids taking place within heterogeneous conformational ensembles.

3SBA-02 QM/MM RWFE-SCF 法とマイクロ秒 MD 計算によるタンパク質荷電性残基の pKa 予測
pKa prediction of ionizable residues in proteins by QM/MM RWFE-SCF method combined with microsecond-long MD simulations

Taisuke Hasegawa[1], Shigehiko Hayashi[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Sci., Kyoto Univ.*)

The pKa prediction of ionizable residues in proteins is a critical issue to reveal the atomistic mechanism of protein functions. To accurately predict pKa values, we extended the conventional QM/MM RWFE-SCF free energy optimization method with a multipole operator and a rigidbody dynamics of the QM region. The pKa values of two ionizable residues, D52 and E35, in hen egg white lysozyme were predicted by this method. We found that the optimized protonated and deprotonated states have their distinct conformations. As a result, microsecond-long MD simulations are needed to connect them along the free energy perturbation path. We further show that this method can be applicable to the non-standard residues such as the chromophore of yellow fluorescent protein.

3SBA-03 アンキリンリピートドメインと脂質の相互作用による TRPV1 チャネル活性の制御
Regulatory mechanism of TRPV1 channel activity by the interaction of ankyrin repeat domain with phospholipids

Kazuhiro Takemura[1], Shiro Suetsugu[2], Akio Kitao[1] ([1]*IMCB, Univ. of Tokyo*, [2]*Grad. Sch. Biol. Sci., NAIST*)

The transient receptor potential subfamiliy V (TRPV) is a relatively non-selective cation channel activated by various stimuli. We recently showed that the channel activity of TRPV4 was modulated by direct interactions of the ankyrin repeat domain (ARD) with phosphatidylinositol (4,5)-bisphosphate (PIP2) in the membrane (Takahashi, Nature Comm. 2014). In this study we conducted molecular dynamics (MD) simulations of TRPV1 tetramer in membrane to investigate relation of the aforementioned interactions with channel activities. We observed enhancement of the lipid binding of ARD by PIP2 and change in channel radius by potassium ions. The simulation with the ligand, which is expected to make open state more stable, is currently under consideration.

3SBA-04 タンパク質を基盤とした酸素およびヘム濃度プローブ分子の開発
Protein-based molecular probes for the local concentrations of oxygen and heme

Haruto Ishikawa (*Grad. Scl. Sci., Osaka Univ.*)

Intracellular oxygen and iron concentrations are tightly regulated. Interestingly, iron plays an important role in the oxygen sensor protein, and vice versa. We have developed the protein-based FRET sensors for the concentration of oxygen and heme. Sea lamprey hemoglobin (slHb) is monomeric in oxy form, while slHb self-associates to dimers upon deoxygenation. The oxygen dependent structural transition was applied to the FRET approach. There was no FRET when the oxygen pressure was 0.2 atm, while slHb with the fluorescent dyes exhibited FRET signal in a deoxygenated form. For the detection of heme, we adopted HrtR that controls heme efflux in L. lactis. The HrtR mutants were fused with either GFP or KO. We have succeeded in the observation of heme-dependent FRET signal.

3SBA-05 フラビン結合タンパク質は目的の機能を示すことに対してどの程度「柔らかい」か？
How are flavoproteins “soft” for exhibiting intended functions?

Tatsuya Iwata[1,2] ([1]*Life Sci. Appl. Chem., Grad. Sch. Eng. NITech*, [2]*OptBioTech. Res. Ctr., NITech*)

Some enzymes consist of polypeptides with small organic molecules called coenzymes, thereby such holoenzymes can catalyze reactions that cannot be done by only polypeptides. Of coenzymes, flavins act as mostly redox cofactors, and photoreceptive antenna. I have studied blue-light receptor proteins in which flavins act as chromophores, LOV domain, cryptochrome (CRY) and BLUF domain. Interestingly, the photochemical reactions of flavins in these proteins are different. I have also studied DNA photolyases (PHRs), which are DNA repairing enzymes using light energy, because CRY and PHR have same architecture. Here, I am presenting how proteins are “soft” for using flavins to exhibit intended functions.

3SBA-06 DNA 整列固定技術を用いた DNA 結合蛋白質の単分子機能解析

Single-molecule characterization of DNA-binding proteins with stretchable DNA array

Kiyoto Kamagata (*IMRAM, Tohoku Univ.*)

For regulating a cell function, it is fundamental that DNA-binding proteins bind to target sites of DNA corretly. How do DNA-binding proteins search for and recognize the target sites among a huge DNA within a physiological time? In this study, we have developed a new method for producing the aligned DNA array, DNA garden, and have investigated the target search dynamics of a tumor suppressor p53 by using a single-molecule fluorescence microscopy. p53 possessed two search modes along DNA, and the sliding distance was maintained. The modes and sliding distance were regulated by a disordered DNA-binding domain. Target recognition of p53 was regulated by the actiation and inactivation of p53. Furthermore, I will present the current result of other DNA-binding proteins.

3SCA-01 ナノディスクに再構成した AglB タンパク質の単粒子解析

Single particle analysis of the AglB protein embedded in nanodiscs

Yuki Kawasaki[1], Kouta Mayanagi[1], Ashutosh Srivastava[2], Florence Tama[2,3], Daisuke Kohda[1] ([1]*Div. Struct. Biol. of Med. Inst. Bioreg., Kyushu Univ*, [2]*Dept. of Phys., Grad sch. of Sci., Nagoya Univ*, [3]*AICS., RIKEN*)

Oligosaccharyltransferase (OST) is a membrane protein that catalyzes the transfer of an oligosaccharide chain to the asparagine residues in the acceptor proteins. This study aimed to reveal the structure of archaeal OST called AglB in lipid bilayer environments. We incorporated AglB into nanodiscs, and obtained the 3D structure by the single particle analysis of negatively stained electron microscopic images. The atomic structure was estimated by flexible fitting of the crystal structure into the 3D EM map. We found that AglB took a more open conformation than the crystal structure. We think the dynamic conformational changes are important for AglB to bind the N-glycosylation sequons efficiently during the catalytic cycle of the oligosaccharyl transfer reaction.

3SCA-02 染色体分配を支える CENP-A licensing 複合体の構造基盤

Structural basis of the CENP-A licensing protein complex

Mariko Ariyoshi, Mariko Matsuda, Masahiro Shirakwa (*Grad. Sch. Eng., Kyoto Univ.*)

Centromere, which links two sister chromatids, is an essential chromatin structural unit for dynamic chromatin segregation during mitosis. The functional centromere assembly is required for faithful segregation of sister chromatids, and depends on the accurate deposition of the histone H3 variant, CENP-A. The Mis18 protein complex plays an essential role in CENP-A deposition and maintenance of cell cycle. The molecular mechanism of CENP-A deposition licensing by the Mis18 complex remains unclear. We have investigated inter-subunit interactions and molecular assembly mechanism of the Mis18 complex using biochemical and structural techniques. The structural basis of the Mis18 complex would provide insight into maintenance of centromere and timing of cell cycle.

3SCA-03 フェムト秒 X 線自由電子レーザーによって明らかにされた光化学系 II 複合体の中間体構造

Crystal structure of the oxygen evolving photosystem II in the intermediate state revealed by femtosecond X-ray free electron lasers

Michi Suga (*RIIS, Okayama Univ.*)

Recent crystallographic studies using SR and XFEL have revealed that the OEC of PSII is a Mn4CaO5 cluster coordinated by a well-defined protein environment, and possible reaction mechanisms have been proposed. However, since the water-splitting reaction proceeds through five metastable states, the exact mechanism for O-O bond formation is not clear and under extensive debates. Here we show the crystal structure of PSII trapped in the intermediate state. Difference Fourier map calculated between the intermediate state and the dark-stable state clearly revealed structural changes in the OEC and its vicinity, as well as the region around the QB-binding site during the catalytic cycle. These structural changes provide important clues to the mechanism of water oxidation.

3SCA-04 高分子量タンパク質の機能的運動性を解明するための多量子 NMR 解析法の開発と応用

Developments and applications of multiple quantum NMR methods to characterize functional dynamics of high molecular weight proteins

Yuki Toyama[1,2], Hanaho Kano[1], Yoko Mase[1], Mariko Yokogawa[1], Masanori Osawa[1], Ichio Shimada[1] ([1]*Grad. Sch. Pharm. Sci., the Univ. of Tokyo*, [2]*JBIC*)

Protein structures are assumed to be exist an equilibrium between multiple conformations, and lowly populated conformations in the equilibrium often play critical roles in biological functions. Therefore, to reveal the mechanisms of protein functions, it is important to characterize the conformational equilibrium; i.e., the chemical exchange processes of proteins. Here, we developed novel NMR methods for characterizing chemical exchange processes utilizing multiple quantum relaxation rates of side-chain methyl groups, which can be sensitively observed in large proteins. The methods were applied to the biologically important large proteins, which have been difficult to analyze by conventional NMR methods due to molecular size limitations. (Toyama et al, JACS 2016)

3SCA-05 高速 AFM を用いてタンパク質が動作する姿を活写する

Visualization of protein molecules in action by high-speed atomic force microscopy

Mikihiro Shibata[1,2], Noriyuki Kodera[2], Takayuki Uchihashi[1,2], Toshio Ando[2] ([1]*Dept. Phys., Kanazawa Univ.*, [2]*Bio-AFM FRC*)

Structural biology has long been contributing to our understanding of how proteins function by providing their detailed structures. And yet, the revealed structures have been restricted to static snapshots, limiting the level of our understanding. This restriction is now removed by high-speed atomic force microscopy (HS-AFM) that allows direct visualization of individual protein molecules in action at sub-molecular resolution. HS-AFM studies performed in the last few years have provided new mechanistic insight into the functional mechanism of proteins [T. Ando *et al.*, *Chem. Rev.* 2014, 114, 3120]. In this symposium, we will discuss our latest HS-AFM studies on proteins, including membrane proteins embedded in nanodiscs, a DNA endonuclease and a protein kinase.

3SCA-06 タンパク質膜透過を駆動するモータータンパク質のスナップショット

Snapshots of a protein translocation motor

Arata Furukawa[1], Kunihito Yoshikaie[1], Takaharu Mori[2], Hiroyuki Mori[3], Yusuke Morimoto[2], Yasunori Sugano[1], Shigehiro Iwaki[1], Tooru Minamino[4], Yuji Sugita[2], Yoshiki Tanaka[1], Tomoya Tsukazaki[1] ([1]*NAIST*, [2]*RIKEN*, [3]*Kyoto Univ.*, [4]*Osaka Univ.*)

SecDF is a bacterial membrane protein, which enhances protein translocation via Sec translocon using proton motive force. The 3.3 Å-crystal structure of SecDF revealed that SecDF consists of 12 transmembrane helices and three periplasmic domains (P1-base, P1-head and P4). The following analyses proposed that the P1 head repeats dynamic structural transitions at the periplasmic side to complete the protein translocation. However, limited structural information hampered further analyses for SecDF. Here, we determined crystal structures of SecDF at 2.6-2.7 Å resolution. The higher resolution structures elucidated unique features on SecDF. I would like to introduce our recent findings and discuss the dynamism of the protein translocation by the proton driven motor SecDF.

3SCA-07 分子シミュレーションによる SecDF プロトン透過機構の解明

Molecular mechanisms underlying proton transport in SecDF

Takaharu Mori[1,2], Yoshiki Tanaka[3], Kunihito Yoshikaie[3], Tomoya Tsukazaki[3], Yuji Sugita[1,2,4,5] ([1]*RIKEN Theor. Mol. Sci. Lab.*, [2]*RIKEN iTHES*, [3]*NAIST*, [4]*RIKEN AICS*, [5]*RIKEN QBiC*)

About 30% of proteins are secreted across membranes or integrated into membranes. The Sec translocon plays a central role in these protein translocations. It consists of protein complexes: SecA ATPase, protein channel SecYEG, and membrane chaperone SecDF. Recently, it has been revealed that SecDF undergoes large conformational change by the proton-motive force to enhance protein export. However, the relationship between conformational change and proton transport has not been understood well. To elucidate their molecular mechanisms, we carried out molecular dynamics simulations of SecDF. We found that dynamics of the conserved residues in the transmembrane region is important for water channel formation. We discuss detailed mechanisms for the function of SecDF.

3SCA-08 フレキシブルフィッティングによる電子顕微鏡データからの構造モデリング

Structure Modeling from Cryo-EM Data using Flexible Fitting Approach

Osamu Miyashita (*RIKEN AICS*)

Cryo-EM single particle analysis provides valuable structural information on biomolecular complexes. In particular, it can be used to construct a new structural models of functionally important conformational states. Due the resolution limit, such models are usually constructed utilizing existing atomic structures. An approach for building such atomic models is the use of molecular dynamics simulation with biasing forces that guides the existing model to fit into the low-resolution cryo-EM data. In this talk, we will show the importance of taking statistics from multiple fitting trials and employing multiple biasing forces to improve the accuracy of the atomic models derived from Cryo-EM data. We will also present our recent applications to experimental data.

3SDA-01 イオン駆動型回転モーターにおけるエネルギー変換マシナリーの分子解剖：細菌べん毛モーター固定子の機能と構造

Dissection of the energy-conversion machinery in the ion-driven rotary motor: structural and functional studies of the flagellar stator

Seiji Kojima (*Div. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

Bacterial flagellar motor is an ion-driven rotary machinery and its energy conversion process occurs in the membrane-embedded stator complex. Although many intensive efforts, its high-resolution structure has not been reported so far. To break this situation, we took two approaches: (i) using stator proteins of hyperthermophiles, and (ii) focusing on the soluble part of the stator. In addition, we recently succeeded in obtaining highly pure whole complex of Na^+-driven PomA/PomB stator. Its stability turned out to be better than that obtained by our previous procedures. In this symposium, current understanding for the structure and function of stator revealed by our two approach, as well as updates using the newly purified stators, will be presented.

3SDA-02 好アルカリ性 *Bacillus* 属細菌と枯草菌がもつ Na^+駆動型べん毛モーターの中性環境での Na^+透過性の違いの解明

The elucidation of the Na^+-requirement mechanism for flagellar rotation between alkaliphilic and neutrophilic *Bacillus* at neutral pH

Yuka Takahashi[1], Masahiro Ito[1,2] ([1]*Bio-Nano., Toyo Univ.*, [2]*Faculty of Life Sciences, Toyo Univ.*)

The flagellar motor is generally energized by either a H^+ or Na^+ motive force. MotAB-type stators use H^+, while MotPS-type stators use Na^+ as coupling ions. Alkaliphilic *B. pseudofirmus* OF4 and neutralophilic *B. subtilis* have a MotPS-type stator. Previous study suggested that motility of strain OF4 required a high concentration of Na^+ at neutral pH. Hence, *B. subtilis* requires a lower concentration of Na^+ at the same pH. These results may be due to the difference of optimum growth pH of each bacterium. Here I try to elucidate the mechanism of this difference using site-directed mutagenesis, motility assays and measurement of the expression level of MotPS of wild type and each mutant.

3SDA-03 バクテリアべん毛モーターの回転方向切り替えメカニズム

Switching mechanism of the bacterial flagellar motor

Tohru Minamino (*Grad. Sch. Frontier Biosci, Osaka Univ.*)

The bacterial flagellar motor consists of a rotor and a dozen stators and rotates in both counterclockwise (CCW) and clockwise (CW) directions. The rotor is composed of the MS ring formed by a transmembrane protein FliF and the C ring made of FliG, FliM and FliN. The C ring also acts as a switch to change the direction of motor rotation. The binding of a chemotactic signaling protein to FliM and FliN induces cooperative conformational changes in the FliG ring that allows the motor to spin CW. However, it remains unclear how it occurs. In this symposium, I will provide experimental evidences that conformational rearrangements at an interface between FliG and FliM induce remodeling of the FliG ring and discuss the switching mechanism of the flagellar motor.

Symposium

3SDA-04 The actin-like cytoskeletal protein MamK plays a role in positioning of magnetic organelles for bacterial magnetotactic motility

Azuma Taoka, Yoshihiro Fukumori (*Col. Sci. and Eng., Kanazawa Univ.*)

The function of cytoskeletal elements in positioning of bacterial organelles is still an enigma. We assessed the magnetotactic motility of *Magnetospirillum magneticum* AMB-1 using a modified swimming assay and discovered that the actin-like cytoskeletal protein MamK is necessary for optimal magnetotactic behavior. To determine the function of the MamK cytoskeleton, we developed a live-cell fluorescence imaging technique for examining the subcellular dynamics of magnetosome, which is magnetic organelle and functions as a magnetic sensor for the cell. We found that the function of the MamK cytoskeleton is to position and reliably segregate the magnetosomes. Furthermore, our results showed MamK ATPase activity is necessary for the stable linear positioning of magnetosomes.

3SDA-05 F-ATPase から進化したマイコプラズマ滑走運動 *Mycoplasma* gliding developed from F-type ATPase

Makoto Miyata (*Osaka City University*)

Mycoplasmas mobile, a small bacterium glides with a mechanism unrelated to other motility systems. We clarified energy source, binding target, gliding machinery, component proteins, movements, and mechanics, and then suggested a mechanism. A novel motor complex developed from an F-type ATPase generates a force, which transmits across the membrane and a 512 kDa “crank” protein to a 314 kDa “leg” protein. The leg protein, a receptor repeatedly catches, pulls, and releases sialylated oligosaccharides on host cell surfaces. The movement of leg is featured with 70 nm, 1.6 pN step and directed binding property.

3SDA-06 バクテロイデーテス細菌がスムーズに滑走する仕組み Structure and mechanism of gliding motility of *Bacteroidetes*

Satoshi Shibata (*Graduate Sch. of Biomedical Science, Nagasaki Univ.*)

Many diverse groups of bacteria exhibit gliding motility. The mechanism of bacterial gliding motility remains a mystery because each group seems to have its own unique mechanism. The gliding motility of bacteria belonging to phylum *Bacteroidetes* requires the type IX secretion system (T9SS) and its secreted adhesin. Based on our recent findings about movement of the adhesin on the gliding cell surface and a multi-rail structure for the gliding machinery visualized by electron microscopy, the structure and mechanism of gliding motility of *Bacteroidetes* will be discussed.

3SDA-07 アクチンフィラメントの構造多型性：アクチン結合タンパク質の制御および細胞運動への寄与 Structural polymorphism of actin filaments: its implication in regulation of actin binding proteins and cell motility

Taro Uyeda[1,2], Kien Ngo[1], Taro Noguchi[3], Akira Nagasaki[2], Noriyuki Kodera[4], Kiyotaka Tokuraku[5] ([1]*Dept. of Physics, Waseda Univ.*, [2]*Boomed. Res. Inst., AIST*, [3]*Dept. Chem. Sci. Eng., Natl. Inst. Tech., Miyakonojo Coll.*, [4]*Bio AFM Res. Ctr., Kanazawa Univ.*, [5]*Muroran Inst. Tech.*)

Actin filaments are inherently polymorphic, and interactions with certain actin binding proteins (ABPs) are known to stabilize specific structures. Conversely, actin filaments with different structures would have different affinities for each ABP. Our high speed AFM and TIRF observations showed that cofilin and myosin S1 in the presence of ATP change the structure of actin filaments differently, and the structure induced by transient binding of S1+ATP strongly and cooperatively inhibits cofilin binding without competition for binding sites on actin. Intramolecular FRET analyses showed that actin in different parts of a crawling cell have different structures. We suggest that cooperative conformational changes in actin filaments regulate actin-ABP interactions *in vivo*.

3SEA-01 Design and construction of synthetic microbial communities by combining synthetic biological subsystems

Shotaro Ayukawa (*ACLS, Tokyo Tech*)

Recent progresses in synthetic biology have achieved population-level coordination and control of communities of engineered microbes or synthetic microbial communities. Synthetic microbial communities are expected to be important for many areas of biological sciences and applications. In this session, I will discuss the design and construction of synthetic microbial communities by combining several synthetic biological subsystems. A synthetic microbial community that is programmed to diversify into two phenotypic states starting from one state was realized by combining cell-cell communication and mutual inhibitory subsystems. A mutualistic system in which two different types of cells promote each other’s growth was also realized by combining several other subsystems.

3SEA-02 人工細胞パターン形成 Synthetic cell pattern formation

Miki Ebisuya (*RIKEN QBiC*)

Pattern formation plays crucial roles in multicellular development. One way to understand how cells spontaneously create spatial patterns is to recreate or reconstitute the cellular patterns in vitro. In our lab, we build synthetic gene circuits in cell culture that make neighboring cells communicate with each other, leading to spontaneous pattern formation. I will discuss what we have learned from the synthetic cell pattern formation.

3SEA-03 Microfluidic droplet reactor for artificial/living cellular systems

Masahiro Takinoue[1,2] ([1]*Dept. Comput. Sci., Tokyo Tech*, [2]*PRESTO, JST*)

Artificial cell-like (AC) reactors have played important roles in biophysical research such as cell models and molecular measurements. However, it was difficult to use the previous AC reactors for nonequilibrium reaction systems because the flux of chemicals into/out of the reactors cannot be controlled as intended. Using droplet microfluidics, we recently developed a computer-aided open AC reactor, in which the flux of chemicals is precisely controlled by fusion-fission timing between the AC reactor and chemical-carrier droplets. Thus, our AC reactor can be used for nonequilibrium reaction systems including complex biochemical reactions and cellular systems. We believe that our AC reactor can promote system control biology based on model-driven control of living cells.

3SEA-04 Generation of a self-organizing kidney comprising multiple renal cell types

Minoru Takasato (*RIKEN CDB*)

Directing differentiation of human pluripotent stem cells (hPSCs) into kidney is challenging as an adult kidney comprises 25 distinct cell types, derived from 4 progenitors, including ureteric, nephron, vascular and stromal progenitors. Here we identified the developmental mechanism regulating mesoderm regionalization, in which the duration of exposure to canonical WNT signaling determines cell fate of the primitive streak into either the anterior or posterior intermediate mesoderm. By utilizing this knowledge for hPSCs directed differentiation, we performed a preferential induction of collecting duct versus kidney nephron progenitors that formed kidney organoids when aggregated and grown in 3D. Within kidney organoids, all renal components were self-organized.

3SEA-05 合成生物学研究のための哺乳類の in vitro 生命システム
An in vitro Living System in Mammals for Synthetic Biology Research

Yoh-ichi Tagawa (*Tokyo Institute of Technology School of Life Science and Technology*)

It must be still difficult for synthetic biological research in mammals because of the followings. First, understanding of communications in cell-cell and organ-organ is important because mammal consists of a variety of cells and cell types unlike unicellular organisms. Second, synthetic biological experiments may need a great number of animals, which cannot be used from the view of animal protection. Animal experiment alternatives, i.e., in vitro culture systems, are urgently required for synthetic biology as well as the developments of pharmaceuticals. We succeeded to differentiate murine ES/iPS cells to autonomously beating heart muscles, liver tissues, and pancreatic islets, now are trying to establish an in vitro living system in mammals on microfluidic devices.

3SFA-01 タンパク質と脂質を運んでミトコンドリアをつくる仕組み
Mechanisms of mitochondrial biogenesis by protein and lipid transport

Toshiya Endo (*Kyoto Sangyo Univ., Fac. Life Sci.*)

Normal mitochondrial functions rely on biogenesis of mitochondria by growth and division of pre-exsisting mitochondria that require transport of their major constituents, proteins and phospholipids. In this symposium, I will discuss how the TOM complex, the protein translocator machinery in the outer mitochondrial membrane, can make full use of its structural characteristics to facilitate efficient translocation of over 1,000 different mitochondrial proteins across the outer membrane. I will also discuss how different lipid transport machineries mediate transport of hydrophobic phospholipids between the ER and mitochondria and within mitochondria by different mechanisms.

3SFA-02 精密 X 線結晶構造解析によるチトクロム酸化酵素の酸素還元・プロトンポンプ機構
Detailed crystal structural studies of bovine cytochrome oxidase to elucidate the coupling mechanism of dioxygen reduction and proton pump

Tomitake Tsukihara[1,2], Atsuhiro Shimada[1], Naomine Yano[3], Kazumasa Muramoto[1], Kyoko Shinzawa-Itoh[1], Eiki Yamashita[2], Shinya Yoshikawa[1] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Institute for Protein Research, Osaka Univ.*, [3]*Front. Res. Cen. Appli. Atom. Sci., Ibaraki Univ.*)

The X-ray structures of oxidized/reduced bovine CcO at high resolution reveal that a large water cluster, which includes a Mg2+ ion, is linked to the H-pathway through which protons are pumped. The cluster can contain proton acceptable groups to retain four proton equivalents. The redox-coupled X-ray structural changes around E198, which bridges the Mg2+ and CuA (the initial electron acceptor from cytochrome c) sites, suggest that the CuA-E198- Mg2+ system drives redox-coupled transfer of protons pooled in the water cluster to the H-pathway. Thus, these X-ray structures suggest that the Mg2+-containing water cluster is the crucial structural element providing the effective proton pumping in bovine CcO.

3SFA-03 環境適応における寄生虫ミトコンドリア呼吸鎖のリモデリング
Re-modeling of respiratory chain in the parasite mitochondria during their adaptation

Kiyoshi Kita (*Nagasaki University*)

Parasites have developed a variety of physiological functions necessary for their survival within the specialized environment of the host. Using metabolic systems that are very different from those of the host, they can adapt to low oxygen tension present within the host animals. In addition, parasites have their life cycle where parasite mitochondria play diverse roles. In particular, marked changes in the morphology and components of the mitochondria during the life cycle are very interesting elements of biological processes such as developmental control and environmental adaptation. Recent studies on the dynamic re-modeling of the respiratory chain in the parasitic helminth, *Ascaris suum* and african trypanosome, *Trypanosoma brucei* will be presented.

3SFA-04 どのようにして哺乳類 F_1-ATPase は回転し、そして阻害されるのか? 顕微鏡一分子観察と X 線結晶構造解析による哺乳類 F_1 の角度分割解析

How does F_1-ATPase drive rotation? Angle-divided analysis of mammalian F_1-ATPases by single-molecule and X-ray crystallographic studies

Toshiharu Suzuki (*School of Engineering, The University of Tokyo*)

Rotation of bacterial F_1 has been extensively investigated, but further seeking is prevented because of lacking information of crystal structures. To overcome it, we have established analytical systems of recombinant F_1s of human and bovine, which have provided unexpected valuable clues. Single-molecule analyses of human F_1 revealed a unique rotation scheme different from bacterial F_1 (Suzuki et al, Nature Chem Biol 2014). Crystallographic study of bovine F_1 has given several snapshots for release of product phosphate (Pi), which revealed coupling between Pi-triggered conformation change in Arginine finger residue and rotation. Together with results from several inhibitors, rotation and regulation mechanism of mammalian F_1s well be discussed.

3SFA-05 PINK1 と Parkin によるミトコンドリア品質管理機構は PKA を介した MIC60 のリン酸化により制御されている

PKA-dependent phosphorylation of MIC60 controls mitochondrial clearance regulated by PINK1 and Parkin

Shiori Akabane, Midori Uno, Shunta Shimazaki, **Toshihiko Oka** (*Department of Life Sicence, Rikkyo University*)

A mitochondrial kinase PINK1 and ubiquitin ligase Parkin play a critical role in selective elimination of damaged mitochondria. We revealed that PKA activation decreased PINK1 protein levels, leading to failure of mitochondrial recruitment of Parkin. We identified a mitochondrial protein, MIC60, as a new PKA substrate. MIC60 knockdown caused a reduction in PINK1 protein and mislocalization of Parkin. Phosphorylation-mimic MIC60 mutants failed to restore the defect in Parkin recruitment in MIC60-knocked down cells, whereas a phosphorylation-deficient MIC60 mutant facilitated the mitochondrial localization of Parkin. Our findings demonstrate that PKA negatively regulates PINK1- and Parkin-mediated mitochondrial quality control through phosphorylation of MIC60.

3SFA-06 ミトコンドリア品質管理マシナリーからパーキンソン病の発症機構を明らかにする

How mitochondrial quality control machinery resists a predisposition to Parkinson's disease

Noriyuki Matsuda (*Ubiquitin Project, TMIMS*)

Mitochondria with decreased membrane potential show impairments in ATP synthesis and protein import into matrix. Such low quality mitochondria are marked with ubiquitin for selective degradation. A mitochondrial kinase PINK1 and a ubiquitin ligase (E3) Parkin are key factors in this mechanism, and disruption of which has been implicated in Parkinson's disease onset. Recently we revealed that PINK1 phosphorylates ubiquitin and the resulting phospho-ubiquitin activates and recruits Parkin. However, the mechanism how phospho-ubiquitin associates with Parkin remained largely unknown. We analyzed interactions between Parkin and phospho-ubiquitin by site-specific photocrosslinking, and reveal a novel binding mechanism that might lead to a Parkin conformational change.

3SGA-01 DNA ナノ構造上に構築した化学的に制御可能なナノシステム

Chemically controllable nanosystems constructed in the DNA nanostructures

Masayuki Endo (*WPI-iCeMS, Kyoto University*)

Precise control of the movement of the target molecules is an important issue required for the construction of integrated molecular systems. Using photoresponsive DNA strands, we constructed a rotator system on the DNA origami tile, and the rotary movement of the DNA nanostructure by switching UV/Vis irradiation was observed using high-speed AFM. For control of a linear movement, a pyrene-modified DNA walker and the track were assembled on the DNA origami tile. The photoinduced walking motion of the walker was observed on the DNA origami surface. The photoresponsive plasmonic switching device with gold nanorods was constructed on a cross shaped DNA origami, and the reversible plasmonic switching was controlled by UV/Vis irradiation.

3SGA-02 Organizing DNA origami components into crystalline structures at the lipid/aqueous solution interface

Yuki Suzuki (*FRIS, Tohoku Univ.*)

Molecular self-assembly has attracted great attention as a method to design and construct novel supramolecular architectures. Here, we demonstrate that DNA origami components can self-assemble into two-dimensional crystalline structures on a mica-supported lipid bilayer surface. The adsorption of DNA origami components onto the fluidic lipid bilayer allows them to move freely on the surface, facilitating among interaction to assemble and form micrometer-sized lattices in their lateral dimensions. We anticipate that our lattices will serve as a versatile platform for a diverse range of applications, including periodic arrangement of protein molecules or nanoparticles, scaffolds for nanodevices, and mimics of membrane-cytoskeleton networks.

3SGA-03 生態模倣アクチュエータ作製に向けた試み：液晶中での微粒子運動

Bottom-up technologies for biomimetic actuators: motion of microbeads in liquid crystals

Yoshiko Takenaka[1,2] ([1]*RI for Sustainable Chemistry, AIST*, [2]*JST PRESTO*)

Recently, stimulus responsive soft materials have been studied for the development of future soft actuators. Here, we will show the liquid-crystal actuator which transfers microbeads at room temperature by the irradiation of UV light in liquid crystal media. When UV light was irradiated, a microbead ran away quickly from the light source and returned back gradually toward the original position after the extinction of the light. The mechanism of the motion of microbeads was investigated. We concluded that the thermal expansion of liquid crystal induced by photo-thermal effect would cause the present motion. We also confirmed that the substance which performed the photo-thermal conversion in our experiments was polyimide.

3SGA-04 脂質修飾 DNA ナノ構造体の動的な集合体制御
Dynamic assembly control of lipid-modified DNA nanostructures

Yusuke Yonamine[1,2], Keitel Cervantes-Salguero[3], Waka Nakanishi[2], Kosuke Minami[2], Ibuki Kawamata[3], Satoshi Murata[3], Katsuhiko Ariga[2] ([1]*Grad. Sch. of Eng., Kyushu Univ.*, [2]*NIMS*, [3]*Grad. Sch. of Eng., Tohoku Univ.*)

Recently, precisely designed DNA architectures known as DNA origami or DNA tile can be constructed by configuring the dimensions and programming the sequences. However, the built DNA structure itself is static and does not show dynamic property like self-assembly or taxis. On the other hand, amphiphiles form various kinds of structures including vesicle, Langmuir-film and self-assembled monolayer (SAM) using hydrophobic interaction as driving force, and show dynamic properties like fluidity, fusion or division. In this study, we aimed to endow a DNA structure with hydrophobicity modifying with a cationic lipid and generate a novel lipid-DNA complex that shows self-assembly and chemotaxis.

3SGA-05 DNA のプログラマビリティを利用したカプセル型分子ロボットの創製
Microcapsular robot based on programmability of DNA

Daisuke Ishikawa (*Sch. Comput., Tokyo Tech.*)

The novel concept of molecular robot, approaching methodology to artificial cell-like systems, is an integrated system with sensors, computers and actuators in a soft capsule such as a vesicle or a water droplet to react flexibly to external or internal stimuli. To realize the cell-like systems, it is necessary to construct capsular structures capable of possessing programmable functions based on hybridization of DNA strands and responses to ions or molecules passed through a channel. Here I will talk for capsular structures consist of only amphiphilic DNA nanoplates formed on the basis of the programmability of DNA without any lipid molecules for supporting membrane, which is completely different from the conventional lipid-layer-based capsules.

3SGA-06 自然知能システム：粘菌の計算パワーを活用する
Natural Intelligence System: Exploiting Computational Power of Amoeboid Organism

Masashi Aono[1,2] ([1]*Earth-Life Sci. Inst., Tokyo Tech*, [2]*PRESTO, JST*)

We constructed an experimental system called an “amoeba-based computer,” which embodies the amoeboid organism (Physarum polycephalum) to search for a solution to a combinatorial optimization problem, the traveling salesman problem. The organism explores the state space using fluctuations in the oscillatory dynamics of its pseudopod-like branches that are spatially and temporally correlated. In response to external light stimuli, the organism finds its optimal shape for which the area of the body is maximized and the risk of being illuminated is minimized. Our amoeba-inspired schemes can be implemented based on various nanodevices in a compact and low energy-consumption fashion, leading to a new computing paradigm that exploits computational power of natural phenomena.

1Pos001 Crystal structure of the 11-cis isomer of Pharaonis Halorhodopsin

Siu Kit Chan[1], Haruki Kawaguchi[1], Hiroki Kubo[1], Kunio Ihara[3], Kosuke Maki[1], Tsutomu Kouyama[1,2] ([1]*Grad. Sch. of Sci., Nagoya Univ.*, [2]*RIKEN Harima Branch*, [3]*Center for Gene Research, Nagoya Univ.*)

Halorhodopsin from Natronomonas pharaonis is a retinylidene protein which functions as a light driven chloride pump. Dark adapted pHR consists of all-trans/15-anti and 13-cis/15-syn isomers (pHR'). Upon photon absorption, the retinal chromophore (pHR_578) isomerizes into the 13-cis/15-anti configuration, and the pumping cycle is activated. In this study, we found that 11- and 9-cis isomers accumulated when chloride free pHR was illuminated with red light. Meanwhile, diffraction data from chloride free pHR crystals illuminated with red light showed that the 11-cis/15-syn isomer is produced without a large change in the retinal binding pocket. Combining with the kinetics data, we discuss the interconversion among different isomeric states of pHR.

1Pos002 結晶構造から明らかになった、エンドセリン-1 によるエンドセリン受容体 B 型の活性化機構
Crystal structures of the endothelin receptor type B reveal activation mechanism by endothelin-1

Wataru Shihoya[1,2], Tomohiro Nishizawa[3,4], Akiko Okuta[2], Kazutoshi Tani[2], Yoshinori Fujiyoshi[1,2], Osamu Nureki[3], Tomoko Doi[5] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Cellular and Structural Physiology Institute., Nagoya Univ.*, [3]*Grad. Sch. Sci., Univ. Tokyo*, [4]*JST PRESTO*, [5]*Grad. Sch. Sci., Kyoto Univ.*)

Endothelin, a 21 amino-acid peptide, participates in various physiological processes, such as regulation of vascular tone, humoral homeostasis, neural crest cell development and neurotransmission. Endothelin activate two kinds of class A GPCRs (endothelin receptors, ETAR and ETBR). We report crystal structures of human ETBR in the ligand-free form and in complex with the endogenous agonist endothein-1 (ET-1) at 2.5 and 2.8 Å resolutions, respectively. The ET-1 bound structure elucidated the unique ET-1 binding mode, and Comparison of the ET-1 bound the ligand-free structures revealed the 4 Å inward movement of TM6-7 upon ET-1 binding. A comparison with the rhodopsin and M2 muscarinic receptor suggested a shared mechanism for signal transduction in class A GPCRs.

1Pos003* X 線結晶構造解析による軸糸ダイニン軽鎖 1 の構造評価
X-ray crystallographic characterization of the axonemal dynein light chain-1

Akiyuki Toda[1], Hideaki Tanaka[2], Yosuke Nishikawa[2], Toshiki Yagi[3], Genji Kurisu[2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Institute for Protein Research*, [3]*Facult. Life Environ., Pref. Univ. Hiroshsima*)

Dyneins are microtubule-based molecular motors that consist of heavy, intermediate and light chains. Intermediate and light chains play a regulatory role in cargo binding or ATPase activity. Most of the dynein accessary chains interact with the N-termini of heavy chain. However, it was recently uncovered that axonemal light chain-1 (LC1) is bound to the microtubule-binding domain (MTBD) of outer arm dynein gamma (OADγ). Although the NMR structure of LC1 was available, we determined the crystal structure of LC1 at 1.55 Å to discuss in detail. X-ray structure was different from the NMR structure in secondary structure level implying a considerable conformational flexibility. We will discuss the structural comparison of LC1 and the binding surfaces of its partners.

1Pos004 固定子に作用するべん毛内膜蛋白質 FliL の構造解析
Structural analysis of the Stator Associated Inner Membrane Protein FliL from Vibrio alginolyticus

Miyu Isumi[1], Yuuki Nishino[2], Mayuko Sakuma[2,3], Seiji Kojima[2], Michio Homma[2], Katsumi Imada[1] ([1]*Grad. Sch. of Sci., Osaka Univ.*, [2]*Grad. Sch. of Sci., Nagoya Univ.*, [3]*Radioisotpe Res. Cent.*)

The bacterial flagellum is rotated by a motor embedded in the cell membrane. The flagellar motor consists of the rotor and the stator, and the torque is produced by the interaction between the rotor and the stator. Recently, a single transmembrane protein, FliL, is found to be associated with the stator. FliL is a 18kDa membrane protein consisting of a short cytoplasmic N-terminal region, a single transmembrane region and a large periplasmic region. However its role in the flagellar motor rotation has not yet been completely understood. To elucidate the role of FliL, we purified, characterized and crystalized FliL and its periplasmic fragments. We obtained crystals of a periplasmic fragment diffracted up to 2.1 Å resolution. The structure analysis is now in progress.

1Pos005 Oligomeric structure of the ExbB-ExbD complex revealed by X-ray crystallography and cryo-EM

Saori Maki-Yonekura, Yoshiki Yamashita, Rei Matsuoka, Maiko Tanaka, Fumie Iwabuki, Koji Yonekura (*RIKEN SPring-8 center*)

Gram-negative bacteria import essential nutrients such as iron and vitamin B12 through the outer membrane. This process utilizes the proton motive force supplied by the ExbB-ExbD-TonB system in the cytoplasmic membrane. The ExbB-ExbD complex forms a proton channel and plays a key role in energizing this transport. However, the structural information is very limited.

We have been analyzing the structure of the complex by X-ray crystallography and cryo-EM. We obtain the three-dimensional structure by combining crystal diffraction and a single particle reconstruction. It reveals the stoichiometry and organization of the functional unit. The analysis also suggests distinct conformations in crystal and in solution, which may represent two physiological states.

1Pos006 赤痢菌ニードル複合体の極低温電子顕微鏡による構造解析
Structural analysis of needle complex from *shigella flexneri* by cryo electron microscopy

Naoko Kajimura[1,2], Takayuki Kato[1], Ariel J Blocker[3], Kei-ichi Namba[1,4] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*Res., Center for UHVEM, Osaka Univ.*, [3]*Sch. of Cell. & Mol. Med., Univ. of Bristol*, [4]*RIKEN, QBiC*)

The needle complex (NC) is a huge membrane-embedded complex and is a central component of the Type III Secretion System of pathogenic bacteria. Single particle image analysis (SPA) has shown that NC has a syringe-like structure with three domains: an extracellular needle, a transmembranous basal body and core inner membrane export apparatus. While the advancement in the technology of SPA, determining the high-resolution structure of intact NC is challenging due to the structural heterogeneities, which includes the stoichiometry of component proteins, the symmetry of the major ring in basal body, and so on.

We report the result of structural analysis and characterization of T3SS complex purified from shigella using SPA.

1Pos007 単一ミオシン結合状態のアクトミオシンの高分解能化
F-actin structural changes induced by a single myosin head

Takahiro Namise, Kazuaki Yoshida, Takuo Yasunaga (*Kyushu Institue of Technology*)

It is well-known that muscle contraction is generated by sliding motion between an actin filament and a myosin filament. The molecular mechanism of sliding has been proposed as 'lever arm theory', but the cooperative and unidirectional conformational changes of actin by binding myosin are also reported. However, their functions are not yet understood enough. Here, in order to understand the functional roles of the cooperative conformational changes, we observed each actin filament binding a single myosin head by electron cryo-EM. We have detected an asymmetrical and interesting conformational changes induced by single myosin and reported them before. However, their resolutions were insufficient for elucidating the mechanism, so we try to improvement higher resolution.

1Pos008* NMR analysis of C-terminal periplasmic domain of flagellar motor protein MotB and its active mutant L119P

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MotB protein is a component of Salmonella flagellar motor, which makes the stator with MotA protein, and serves as proton conducting channel. Significant structural change of MotB protein has expected to bind to the peptidoglycan layer and to activate the proton channel. In this study, the structural change of the C-terminal periplasmic fragment of MotB (MotBC2) induced by its active mutant L119P, whose N-terminal helix α1 is disordered in the crystal, was investigated by NMR. NMR analysis of 15NH-Lys labeled MotBC2 showed the significant structural disruption on α1 of L119P mutant. This disruption was localized on N-terminal half of α1 helix, in which structure of WT is stabilized by forming salt bridges with amide group of C-terminal helix α5.

1Pos009 CS-Rosetta 法によるヒストン H2A-H2B ヘテロ二量体の溶液構造解析
Determination of the solution structure of isolated histone H2A-H2B heterodimer by using CS-Rosetta

Tsutomu Yamane, Yoshihito Moriwaki, Hideaki Ohtomo, Mitsunori Ikeguchi, Jun-ichi Kurita, Masahiko Sato, Aritaka Nagadoi, Hideaki Shimojo, Yoshifumi Nishimura (*Graduate School of Medical Life Science, Yokohama City University*)

CS-Rosetta is a program for protein structure determination from NMR experimental parameters and is able to determine solution structures of proteins only from the chemical shift values. However, to determine structures only from the chemical shift values, the target protein have to be a monomer, less than about 120 residues, and smaller flexible region. Histone H2A-H2B heterodimer is out of this limitation. In the present study, we applied CS-Rosetta to determine the solution structure of isolated histone H2A-H2B heterodimer only from chemical shift values. As the results, our solution structures are in good agreement with the chemical shifts values, the chemical shift indices, and the dynamical properties obtained from NMR experiments.

1Pos010 時間分解 EPR 法によるヒトインスリンのアミロイド線維化による構造変化の観測
Effects of amyloid fibrillations on geometries of human insulin as studied by time-resolved EPR spectroscopy

Tomoka Abe[1], Takashi Tachikawa[1], Eri Chatani[1], Paul Zierep[2], Stefan Weber[2], Toshifumi Mori[3], Shinji Saito[3], Yasuhiro Kobori[1] ([1]*Grad. Sch. Sci., Kobe Univ.*, [2]*Freiburg Univ.*, [3]*IMS*)

Protein's aggregation generates 'amyloid fibrils' which have been implicated to cause the Alzheimer's diseases. However, the protein structures have been still unclear in the atomic levels on the fibril states. We have investigated influences of the fibrillations on the protein local structures. Time resolved EPR measurements were performed to characterize the structure and the geometries of photo-induced radical pairs generated in protein-ligand complex for the native and fibril states of human insulin. We have also carried out NMR measurements to identify the binding site of the ligand, which is necessary to clarify the docking region observed by the EPR. Based on these magnetic resonance studies, we will discuss the fibrillation effects on the protein structures.

1Pos011 溶液 NMR 法を用いた長距離情報の取得による Nrd1 のドメイン間配向の決定
Structural analysis of a multi-domain protein using long-range distance information derived by solution NMR

Kan Nagai, Ayaho Kobayashi, Yutaka Ito, Masaki Mishima (*Graduate School of Science and Engineering, Tokyo Metropolitan University*)

Negative regulator of differentiation 1 (Nrd1) is known as a negative regulator of sexual differentiation in fission yeast. Nrd1 consists of four RRMs (RNA recognition motif).
In an effort to delineate the relationship between Nrd1 structure and function, we prepared each RRM and tandem region of Nrd1. The structure of the second RRM of Nrd1 has been determined. Structural analyses of RRM1-2 and RRM3-4 are also in progress. To perform accurate measurements, we have succeeded in protein ligation between RRM1-2 and RRM3-4. By the use of PRE (paramagnetic relaxation enhancement), PCS (pseudo-contact-shift), and RDC (residual dipolar coupling), determination of the inter-domain orientation and the full length structure has been expected.

1Pos012 サイズ排除クロマトグラフィー／X 線小角散乱法に基づいたニトリラーゼ会合体プロトマーの構造特性
Structural characterization on nitrilase protomers analyzed by size-exclusion chromatography/small-angle X-ray scattering (SEC-SAXS)

Masatoshi Usui[1,2], Ryo Ishiguro[1,2], Homare Yokota[1,2], Yusuke Takeda[1,2], Takaaki Hikima[2], Tetsuro Fujisawa[1,2] ([1]*Grad. Sch. Eng., Gifu Univ.*, [2]*SPring-8 Center, Harima Inst., RIKEN*)

Microbial nitrilase hydrolyzes nitrile compounds into useful intermediates. It is self-associating system and forms oligomers in activated state. Since nitrilase solution always comprises more than two association states, it is complicated to structurally characterize. We, therefore, employed size-exclution chromatography in line with small-angle X-ray scattering (SEC-SAXS) method [1] at BL45XU, SPrng-8. Under dissociative solvent condition, the SEC-SAXS column profiles displayed a few composite peaks whose smallest one was assigned to protomer. In the present paper, we report the structural characterization on various microbial nitirlase protomers by globally analyzing these SAXS profiles.

[1] Malaby *et al.*, J. Appl. Cryst. (2015).

1Pos013* 様々な炎症物質を認識する NLRP3-LRR ドメインの構造基盤の解明

Investigation of molecular basis underlying the recognition of various inflammatory substances by NLRP3-LRR domain

Ryota Yamamoto[1], Kazuto Yamashita[1], Hiroshi Imamura[2], Motonari Tsubaki[1], Eri Chatani[1] ([1]*Grad. Sch. Sci., Univ. Kobe.*, [2]*AIST*)

NLRP3 is proinflammatory protein which recognizes various damage-associated molecular pattern molecules. To investigate molecular details about sensing the ligands, we have produced a recombinant leucine-rich repeat domain of NLRP3 (NLRP3-LRR), a C-terminal domain proposed to play a role as a receptor. As a result of construction of an *E.coli* expression system in which NLRP3-LRR was fused with glutathione-S-transferase, a purified protein was obtained successfully. Small angle X-ray scattering measurement suggested that NLRP3-LRR obtained had a folded structure, and its conformational stability was also evaluated by guanidium hydrochloride-induced unfolding. On the basis of the obtained results, structural features of NLRP3-LRR will be discussed.

1Pos014 PRP の匂い分子結合における構造変化

The structural changes of the peri-receptor protein (PRP) on the odorant-binding process

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Odorant receptors are usually covered with mucous or lymph solution. Thus, the hydrophobic small molecules would be difficult to approach the receptors. The two types of soluble proteins, odorant-binding protein and chemosensory protein (peri-receptor protein; PRP) would dissolve the hydrophobic small molecules in the hydrophilic environment and carry them up to the receptors. The molecular mechanisms of the process, however, are not well elucidated. We measured the structural change of PRPs under different pH and temperatures by synchrotron radiation small- and wide-angle X-ray scattering (SR-SWAXS) and succeeded mostly to measure the WAXS curve covering from the tertiary structure to the secondary one. The detailed analysis and discussion will be presented.

1Pos015 放射光小角散乱データに基づく対称性を考慮したニトリラーゼオリゴマーのモデリング

Modeling of Nitrilase oligomer with flexible symmetry based on synchrotron small-angle scattering data

Tetsuro Fujisawa[1,2,3], Keiichi Kameyama[1], Ryo Ishiguro[1,2] ([1]*Dep. Chem. & Biomol. Sci., Fac. Eng., Gifu Univ.*, [2]*Spring-8 Center, RIKEN Hrima Inst.*, [3]*Synchro. Center, Nagoya Univ.*)

Nitrilase oligomer is very unique enzyme because the enzymatic activity is tightly linked with its association states. We have extensively studied the temperature-pressure behavior of its association by x-ray small-angle scattering (SAXS) and found several intermediates. *Ab initio* modeling methods are generally used for SAXS data, which cannot be applicable for our case because of lack of necessary symmetric options. One of the alternative modeling methods, Integrating Modeling Platform (IMP) is highly flexible modeling platform developed by Prof. A. Sali, UCSF, with which one can model protein complex imposing arbitrary symmetry[1]. In the present paper, we report the application of IMP on SAXS data from nitiralse oligomers.

[1] D. Russel et al., PLoS Biology, (2012).

1Pos016 β-シート中におけるアミノ酸トリプレットパターンの解析

Analysis of amino acid triplet patterns in β-sheets

Hiromi Suzuki (*School of Agri., Meiji Univ.*)

We selected 21,312 proteins showing less than 30% sequence identity from PDB and analyzed patterns of amino acid triplets (combination of two amino acid pairs) in β-sheets taking hydrogen bonding pattern into account. For the parallel triplets, ratios of Arg, Glu, Gln, and Lys located at the center strands were less than half of expected values. For the anti-parallel triplets, charged amino acids such as Arg, His, Lys, Asp and Glu located at the center strands, preferred alternatively charged amino acids and avoid hydrophobic or aromatic amino acids for both HB and nHB partners. Among amino acid triplets, 7 patterns, Cys-Cys-Cys, Trp-Cys-Cys, Lys-Cys-Trp, Glu-Lys-Glu, Lys-Glu-Lys, Glu-Arg-Glu, Ala-Cys-Trp, were observed more than 10 times of expected values.

1Pos017 タンパク質の構造コンプライアンス特性とドメイン間運動の関係性解析

Analysis of the Relationship Between Structural Compliance Properties and Inter-domain Motion of Proteins

Keisuke Arikawa (*Fcl. Eng., Kanagawa Inst. of Tech.*)

The application of force-moments in static equilibrium to the focusing domains of a protein model results in the decomposition of inter-domain motion into motion modes according to the magnitude of structural compliance. The elastic-network model (ENM) is a protein model where domains are treated as deformable bodies. The inter-domain motion of real proteins can be approximated using a combination of the softer motion modes. Moreover, differences in the compliance properties are evaluated based on the canonical angles between the subspaces spanned by the force vectors corresponding to the motion modes. If a compliance property greatly changes when a part in the ENM is constrained, that part plays an important role for the motion.

1Pos018 自由エネルギー変分原理に基づく Pim-1 キナーゼ阻害剤系の相対的結合自由エネルギーの予測

Prediction of the relative binding free energies for Pim-1 kinase - inhibitor systems based on the free energy variational principle

Anna Hirai (*Dept. of Bioinfo., Col. Life., Ritsumeikan Univ.*)

It is important for drug discovery to calculate values of the binding free energy between a protein and a ligand. In this study, we calculated the relative binding free energy for the Pim-1 kinase system using a method based on the free energy variational principle. It does not need to make sample of the intermediate states and to invoke empirical parameterizations. Pim-1 kinase is the protein found in patients suffering with human hematopoietic malignancies (leukemia and prostatic cancer), and thus its inhibitor can be medicine for the cancers. The details of the results of the calculated relative binding free energy in several ligands of Pim-1 kinase will be presented in the conference.

1Pos019 Flexible docking between cyclin-dependent kinase 2 and its inhibitor using multicanonical MD

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We have executed an accurate flexible docking experiment using multicanonical molecular dynamics (McMD) simulations. McMD dynamically biases the system based on the potential energy, enabling a random walk through the energy space. Here, we have performed a long timescale McMD simulation of the docking of CDK2 and one of its inhibitors, CS3. CDK2, also known as cyclin-dependent kinase 2, is involved in cell cycle regulation. We were successfully able to predict the correct binding pose with respect to the X-ray structure using the PMF along two reaction coordinates based on the position of the COM of CS3 and the RMSD of CS3. Furthermore, we have potentially found a secondary binding pose which is similar to how ATP, CDK2's natural ligand, binds.

1Pos020* 高濃度リガンド条件による蛋白質-リガンド結合部位および経路の効率的探索

Accurate and efficient protein-ligand docking method using all-atom molecular dynamics at high concentration of ligands

Chika Sato[1], Akio Kitao[1,2] ([1]*Grad. Sch. Front. Sci., Univ. Tokyo*, [2]*IMCB, Univ. Tokyo*)

We propose a more accurate and efficient method for protein-ligand docking using all-atom molecular dynamics simulation (MD), compared to conventional docking methods. All-atom models can give more accurate results than implicit solvent models, however, it typically requires simulation time of micro second order. In this method, ligands are placed around a protein at high concentration (~120 mM), which drastically increases the probability of protein-ligand binding. We will show that this method predicted correct binding positions in at least three protein-ligand complexes within 100 ns MD. We also identified typical ligand binding pathways and transient binding sites, indicating that this method can predict protein-ligand complex structure as well as binding pathways.

1Pos021 自由エネルギー変分原理を用いたタンパク - リガンド間相対的結合自由エネルギー計算の DHFR-TMP 系への応用

Calculation of relative binding free energy between DHFR-TMP system on the basis of free energy variational principle

Naoto Nishimura (*Grad. Sch. bioinfo., Univ. Ritsumeikan*)

In the drug discovery stage, it is important to predict the binding free energy of a drug and a protein. The purpose of this study is to establish a new in silico method which is low cost and no parameter tuning. Our target protein is dihydrofolate reductase with 18 inhibitors which is the derivative of Trimethoprim. In this study, an approximated method is used to obtain relative binding free energy on the basis of the free energy variational principle with molecular dynamics simulation. The correlation coefficient between experimental and calculated values is improved by classification of 18 ligands, based on physical similarity.

1Pos022 巨大ヘモグロビン酸素解離中間体の X 線結晶構造と分光学的解析

Crystallographic and spectroscopic analysis of the oxygen-dissociation intermediate of the giant hemoglobin

Nobutaka Numoto[1], Taro Nakagawa[2], Nobutoshi Ito[1], Yoshihiro Fukumori[3], Kunio Miki[4] ([1]*Med. Res. Inst., Tokyo Med. & Dent. Univ.*, [2]*Nagahama Inst. of Bio-Sci. & Tech.*, [3]*Coll. of Sci. & Eng., Kanazawa Univ.*, [4]*Grad. Sch. of Sci., Kyoto Univ.*)

Allosteric oxygen-binding mechanism of hemoglobin (Hb) has been widely discussed whereas the structure of intermediate form between the oxy and deoxy states without any artificial modification of the Hb molecule is still unclear. We demonstrate that the oxy crystals of the extracellular giant Hb (400 kDa) of a tubeworm, Oligobrachia mashikoi can transform to the oxy/deoxy intermediate form in keeping a crystalline structure by the soaking methods. X-ray crystallographic and microspectrophotometric analyses revealed that ternary and quaternary structural changes of some intermediate forms of the giant Hb.

1Pos023 Structural analysis of Calredoxin from *Chlamydomonas reinhardtii*

Ratana Charoenwattanasatien[1,2], Risa Mutoh[2], Hideaki Tanaka[2], Takashi Matsumoto[3], Takashi Oda[4], Mamoru Sato[4], Michael Hippler[5], Genji Kurisu[1,2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Inst. Protein Res, Osaka Univ.*, [3]*Rigaku*, [4]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*, [5]*Inst. Plant Biol. and Biotech., Univ. of Munster*)

Calredoxin (CRX) is a novel chloroplast-localized protein from *Chlamydomonas reinhardtii* consisting of the two calmodulin (CaM) domains and the thioredoxin (TRX) domain. We try to clarify the structural mechanism of Ca^{2+} and redox signaling mediated by CRX. The crystal structure of CRX with four bound Ca^{2+} ions was solved at 1.6 Å resolution showing tandemly linked CaM and TRX domains. At the interface of two domains, we found the hydrogen bond network and confirmed its importance by functional analysis with a chloroplast-type 2-Cys peroxiredoxin. We also studied the conformational change of CRX upon Ca^{2+}-binding using the SAXS and NMR. The dummy atom models with and without Ca^{2+} and NMR chemical shifts change upon Ca^{2+}-removal showed the significant structural change.

1Pos024 分子動力学による溶液中 *Agaricus brasiliensis* 由来 β-グルカンの構造解析

Conformational analysis of β-glucans from *Agaricus brasiliensis* revealed by molecular dynamics in solution

Yoshitaka Matsumura[1], Kodai Inoue[1], Makoto Suminokura[1], Mikako Kubo[1], Mariko Demura[1], Takayuki Ichioka[1], Yasumasa Morimoto[1], Mitsuru Tashiro[2], Ken-ichi Ishibashi[3], Naohito Ohno[3], Masaki Kojima[1] ([1]*Sch. of Life Sci., Tokyo Univ. of Pharm. and Life Sci.*, [2]*Dept. of Chem., Coll. of Sci. and Tech., Meisei Univ.*, [3]*Sch. of Pharm., Tokyo Univ. of Pharm. and Life Sci.*)

β-glucan derived from fungus has multiple bioactivities such as immunostimulation and tumor suppression. We previously analyzed β-glucan from mushroom *Agaricus brasiliensis* by NMR and mass spectroscopy, and demonstrated that it was mainly composed of β-1,6 glycosidic linkage with a small amount (less than 10%) of β-1,3 glycosidic branches. Based on these results, we performed molecular dynamics (MD) simulation, of β-glucans with various primary structures so as to obtain conformation ensemble equilibrated in solution. The results indicated that the β-glucan molecules dominantly form helical structures, but that the detailed features can be further classified based on their primary structures.

1Pos025 **Aβ conformation on a hydrophilic/hydrophobic interface by molecular dynamics simulations**

Satoru Itoh[1,2], Hisashi Okumura[1,2] ([1]*IMS*, [2]*Sokendai*)

The amyloid-β peptides (Aβ) form amyloid fibrils which are associated with Alzheimer's disease. It was reported that amyloidogenesis is accelerated on a hydrophilic/hydrophobic interface such as an air/water interface or an interface between sugar-head groups and hydrocarbon chains of glycolipids. In order to investigate amyloidogenesis on the hydrophilic/hydrophobic interface, we performed molecular dynamics simulations for a full-length Aβ molecule in the presence of the interface. We will show Aβ conformations on the hydrophilic/hydrophobic interface and discuss effects of the interface on the amyloidogenesis in our presentation.

1Pos026 **分子動力学シミュレーションを用いた四量体型サルコシン酸化酵素における生成物の選択的移動の解明**
Selective transport of product in heterotetrameric sarcosine oxidase by molecular dynamics simulation

Go Watanabe[1], Takami Saito[2], Daisuke Nakajima[1], Akinori Hiroshima[1], Haruo Suzuki[1], Shigetaka Yoneda[1] ([1]*Sch. Sci., Kitasato Univ.*, [2]*Grad. Sch. Sci., Kitasato Univ.*)

Heterotetrameric sarcosine oxidase (HSO) contains a large cavity near the active site and several tunnels from the cavity to the outer region. In order to investigate how substrates and products migrate to or from the active site through the tunnels, we performed molecular dynamics (MD) simulation. From the trajectories of water molecules in the simulation, we determined the channels for the water transport. Moreover, potential of mean force (PMF) for the transport of the product (imino intermediate) was calculated for each channel using Steered MD simulation and umbrella sampling. The comparison of the PMF values for the channels clearly shows that one specified channel is selected for the transport of the imino intermediate.

1Pos027 **レオニン合成酵素における生成物支援機構の理論的解明**
Theoretical elucidation on the product assisted catalysis of threonine synthase

Yuzuru Ujiie[1], **Mitsuo Shoji**[1], Ryuhei Harada[1], Takeshi Murakawa[2], Yasuteru Shigeta[1], Hideyuki Hayashi[2] ([1]*Univ. of Tsukuba*, [2]*Osaka Medical College*)

Threonine Synthase (ThrS) catalyzes a L-threonine formation from O-phospho-L-homoserine. A series of reactions catalyzed by ThrS are very complicated; full of regiospecific and stereospecific steps. Therefore, the molecular mechanisms of the reaction control (product-assisted catalysis) are not yet clarified. In this study, molecular dynamics (MD) simulation with the thermodynamics integration approach were performed to elucidate the accurate free energy differences among the key intermediate states of ThrS. By performing 100ns MDs, we found that the major conformations are changed by the reaction-controlling ions. Calculated results suggest that the controlling of the substrate conformation is one of the important molecular mechanisms in the product-assisted catalysis.

1Pos028 **Hybrid ab initio molecular dynamical simulation of cytochrome c oxidase: Mechanisms of structural changes by dynamical ligand recognition**

Ryuichiro Terada, Kang Jiyoung, Masaru Tateno (*Grad. Sch. Sci., Univ. Hyogo*)

Cytochrome c oxidase (CcO) is the terminal enzyme of the respiratory chain, catalyzing the oxygen reduction in the binuclear center (BNC), which is composed of the CuB and heme a3 sites. The crystal structures of CcO showed the translocation of heme a3 upon the ligand binding, which resulted in the structural changes of a nearby helix. In this study, to elucidate the mechanisms of these structural transitions, we conducted hybrid ab initio quantum mechanics (QM) molecular dynamics (MD) simulations. The analysis revealed that repulsive interactions between the ligand and His240 were induced, translocating heme a3. In the subsequent changes of the helix, the intermediate state was identified, hindering the high energy states, which thus acted as a switch in CcO.

1Pos029 **スレオニル tRNA 合成酵素におけるアミノ酸選択機構の理論的研究**
Theoretical study on the molecular mechanism of amino-acid selection in threonyl-tRNA synthetase

Yoshiharu Mori[1], Hisashi Okumura[1,2] ([1]*IMS*, [2]*SOKENDAI*)

We studied the selection mechanism of a threonine molecule in threonyl-tRNA synthetase (ThrRS) using several theoretical methods. First, we modeled the catalytic site of ThrRS. ThrRS has a zinc ion in the catalytic site. The coordination structure of the catalytic site was modeled and confirmed by molecular dynamics simulations and quantum chemical calculations. We found that molecular dynamics simulations can give accurate results, which were confirmed by the QM/MM calculations.
We performed further MD simulations to calculate the free energy profile of amino-acid binding to the catalytic site of ThrRS. To enhance sampling efficiency, we used the umbrella sampling method. The free energy profile for several amino acids was obtained.

1Pos030 **量子化学計算ソフトウェアへのレプリカ交換法の導入**
The implementation the Replica-Exchange Umbrella Sampling in the quantum mechanical simulation packages

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We implemented the Replica-Exchange Method (REM), the Umbrella Sampling (US), and the Replica-Exchange Umbrella Sampling (REUS) in the DFTB+ simulation package. Furthermore, we try to implement these methods in the other quantum mechanical simulation packages. These useful methods have not been used well in the quantum chemistry fields, especially these method is not available in the quantum mechanical simulation packages. Therefore, at the beginning we implemented these methods in one of the famous quantum mechanical simulation package, DFTB+. We carried out the two different molecules simulation, the one is the malonaldehyde, the other is the phthalocyanine by DFTB+ with these methods and we will show the results of these simulations in the poster.

1Pos031 Hydrogenase : ab initio quantum mechanics study of oxygen-tolerance mechanism

Jae Hyun Kim, Jiyoung Kang, Masaru Tateno (*Graduate school of Life Science,University of Hyogo*)

Hydrogenase catalyzes the reversible redox reactions of hydrogen in the microorganism. In membrane bound hydrogenase (MBH), the [4Fe-3S] cluster, which is located in the proximity of the catalytic center, has been suggested to play a critical role to preserve the catalytic activity even in the presence of oxygen (i.e., the oxygen tolerance). Very recently, the hydroxide bound forms of the proximal cluster were proposed experimentally. In the present study, we conducted ab initio electronic structure calculations in the presence and absence of the hydroxide ion, and thereby identified definite differences in the features of the reactive orbitals, which may act as a switch of the electron transfer between the proximal and catalytic clusters in the catalytic cycle.

1Pos032* アルカン合成酵素 AD の NMR と分子動力学シミュレーションによるダイナミクス解析
Structural dynamics of an alkae synthase, AD, studied by NMR and molecular dynamics simulations

Yuma Suematsu[1], Yuji O. Kamatari[2], Yuuki Hayashi[1], Munehito Arai[1] ([1]*Dept. of Life Sci., Univ. of Tokyo*, [2]*Life Sci. Res. Center, Gifu Univ.*)

Cyanobacteria synthesize alkanes using an aldehyde deformylating oxygenase (AD). To study the structural dynamics of AD, here we measured NMR TROSY-HSQC spectra and the R_1 , R_2 , and heteronuclear *NOE* of AD. Whereas the free form of AD had both fast and slow dynamics, the substrate analog-bound form of AD showed uniform dynamics throughout the molecule. We also performed molecular dynamics simulations of AD. Although the structure of the N-terminal 20 residues was not determined in the crystal structures of AD, we found by simulations that the N-terminal region is flexible but forms helical structures and interacts with the substrate-entry site. We propose that low activity of AD is attributed to the dynamic nature of the regions around the substrate-entry site.

1Pos033* エンド-1,3-β-グルカナーゼ触媒ドメインの構造ダイナミクス解析
Structural dynamics analysis of catalytic domain of endo-1,3-β-glucanase

Ayako Miki[1], Satomi Inaba[1], Kazumasa Sakurai[2], Masayuki Oda[1] ([1]*Grad. Sch. of Life and Environ. Sci., Kyoto Pref. Univ.*, [2]*High Pressure Protein Res. Center, Kindai Univ.*)

We have analyzed structural and functional properties of endo-1,3-β-glucanase from *Cellulosimicrobium cellulans* DK-1, which has a catalytic domain (26.5 k) and a carbohydrate-binding module (13.1 k), connected by a Gly/Ser-rich linker (1.2 k). In this study, we analyzed structural dynamics of inactive mutant of catalytic domain, E119A, using NMR; 1) signal assignment, 2) signal change upon the substrate binding, 3) signal change under high pressure conditions. Despite of its high molecular weight, the NMR signals were well dispersed, and about 50% of them could be assigned. Upon the binding of laminarioligosaccharides, some signals of residues around catalytic motif were shifted. Together with the results of high-pressure NMR, we discuss the structural dynamics.

1Pos034 カルモジュリン結合ペプチドはミッドカインに親和性を示し、その一アミノ酸変異による立体構造変化が結合親和性の向上に寄与する
Secondary structure change by single alanine substitution in Calmodulin-binding peptide improved the binding affinity with Midkine

Hidenao Arai, Koji Matsuoka, Naoto Nemoto (*Grad. Sch. Sci. Eng., Saitama Univ.*)

We discovered that the Calmodulin (CaM)-binding peptide containing alanine substitution, named mtCBP, has a specific affinity with Midkine despite in the presence or absence of Ca^{2+} from SPR analysis using BIAcore. Dissociation constant of mtCBP with Midkine was lower by twenty-fold than that of wild-type CBP (wtCBP). It have been reported that mtCBP enhanced the binding affinity with CaM in the presence of Ca^{2+} as compared to wtCBP [*JMB*, 258, 6-13 (1996)]. The secondary structure predictions of these peptides revealed that the ratio of helix-coil-helix structures of CBP changed from 71% in wtCBP to 95% in mtCBP. We suggested that CBP was disordered by a single substitution, and the binding manner of mtCBP with Midkine might be different from that with CaM.

1Pos035 ラン藻由来アルカン合成酵素のアラニンスキャン変異解析
Alanine scanning mutagenesis of a cyanobacterial alkane synthase

Keigo Shimba, Fumitaka Yasugi, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

Cyanobacterial alkane biosynthesis is an attractive way of producing renewable substitutes for fossil fuels. Alkanes are produced from aldehydes by aldehyde deformylating oxygenase (AD) in cyanobacteria. Crystal structures of AD have suggested a reaction center and a substrate binding channel. However, which residues are catalytically more important and whether other residues have a role in the catalysis are unknown. To clarify these issues, we performed alanine scanning mutagenesis of AD. We found that the residues in iron binding sites are essential and that five residues at the substrate binding site are more important for the catalysis. We will discuss the contribution of the residues in AD to the activity, structural stability, and substrate specificity.

1Pos036* 複数のエピトープを認識する G2 の 1 本鎖抗体の構造解析
Structural analysis of a single-chain Fv antibody of G2 that recognizes multiple epitopes

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A monoclonal antibody (mAb) G2 can interact with two completely different peptides with high affinity but different binding kinetics and thermodynamics. In this study, to reveal the unique antigen recognition mechanism of G2, we generated its single-chain Fv (scFv) and analyzed the structural and functional properties. The antigen binding experiments using Biacore showed that binding kinetics of G2 scFv to the two antigen peptides are similar to those of G2 mAb. In NMR experiments for ^{15}N-labeled G2 scFv, $^{15}N/^{1}H$ HSQC signals sharpened by the addition of each antigen peptide, indicating that the free form of G2 scFv has a flexible conformation and can change into rigid conformations in the antigen bound states. The signal change was different between the two peptides.

1Pos037 複数の抗原を特異的に認識する抗体 G2 の 3 つめのエピトープの同定

Identification of the third epitope recognized by multispecific antibody G2

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A monoclonal antibody (Ab) G2 possesses an unusual characteristic; it can react with at least three proteins (ATP6V1C1, SEPT3, and C6H10orf76) other than the original antigen, chicken prion protein ($ChPrP^C$) (Kamatari et al. 2014). The epitopes in $ChPrP^C$ and ATP6V1C1 have been identified, but those in the others not yet. In this study, we identified the third epitope in SEPT3. We inserted the DNA segments with different sizes into pRSET-B vector, expressed in E. coli BL21, analyzed the binding of the expressed proteins with G2 by Western blotting, and identified the epitope in SEPT3. Intriguingly, there is no amino acid sequence similarity between the epitopes in $ChPrP^C$, ATP6V1C1, and SEPT3. This is the first report on the Three-in-One type Ab.

1Pos038* ラン藻でのアルカン合成に必要な 2 つの酵素間の結合部位の探索

Search for the binding sites between two enzymes essential for cyanobacterial alkane biosynthesis

Mari Chang[1], Keigo Shimba[2], Yuuki Hayashi[2], Munehito Arai[1,2] ([1]*Department of Physics, University of Tokyo*, [2]*Department of Life Sciences, University of Tokyo*)

Cyanobacterial alkane biosynthesis is catalyzed by a two-step reaction involving acyl-(acyl carrier protein) reductase (AAR) and aldehyde deformylating oxygenase (ADO). Recently, both enzymes are reported to interact with each other, but their binding sites are unknown. To elucidate the AAR binding site on ADO, we carried out an alanine scanning mutagenesis around the substrate-entry site of ADO and found that six mutants greatly reduced the alkane production in E. coli coexpressing AAR and ADO. Size exclusion chromatography of AAR-ADO mixtures shows that two of the six mutants, in which mutation sites are located on the protein surface, have reduced affinity with AAR, suggesting that AAR binds the substrate-entry site of ADO to efficiently deliver the substrate.

1Pos039* ケモカイン受容体制御因子 FROUNT－制御化合物間の立体構造情報に基づく相互作用解析

Structure-based analyses of the interaction between the chemokine receptor-regulator FROUNT and anti-inflammatory compounds

Soichiro Ezaki[1], Sosuke Yoshinaga[1], Norihito Ishida[1], Mitsuhiro Takeda[1], Kaori Yunoki[1], Yuya Terashima[2], Etsuko Toda[2], Kouji Matsushima[2], Hiroaki Terasawa[1] ([1]*Faculty of Life Sciences, Kumamoto University*, [2]*Graduate School of Medicine, The University of Tokyo*)

Leukocyte chemotaxis is induced when chemokines bind to their receptors during the inflammation response. We previously identified the cytosolic regulator, FROUNT, and obtained two compounds that inhibit the FROUNT-receptors interaction and exert anti-inflammatory effects. The aim of this study is to optimize the compounds, based on the structural information of FROUNT. We performed NMR titration analyses using 15N-labeled FROUNT and the two compounds. Although their chemical structures are different, a similar chemical shift perturbation pattern of the FROUNT peaks was observed, in a slow-exchange manner on the NMR time scale. These data suggested that the two compounds function via a common mechanism to inhibit FROUNT.

1Pos040 CD28 と SH2 ドメインとの相互作用における構造熱力学的解析

Structural and thermodynamic analysis of interactions between CD28 and SH2 domains

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Co-stimulatory signals from CD28 receptor and ligand interactions are required for full activation of T-cells. CD28 has no enzymatic activity and its cytoplasmic region consists of 41 amino acids that contain the sequence YMNM, whose phosphorylation of the tyrosine triggers the recognition by SH2 adaptor proteins. In this study, we determined the crystal structures of Gads SH2, p85 N-terminus SH2, and C-terminus SH2 in complex with the CD28-derived phosphopeptide at 0.90 - 1.20 Å resolution. The bent conformation of peptide bound to Gads SH2, different from those in the others, is mainly due to the bulky side-chain of Gads Trp118. The recognition modes of the three proteins can explain the binding thermodynamics analyzed using isothermal titration calorimetry.

1Pos041 T 細胞受容体による特異的および交差反応的な抗原認識機構の解明

Analyses of the structural mechanisms of specific and crossreactive recognitions of peptide-MHC by TCRs

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T cell receptor (TCR) recognizes and binds to a complex of foreign-peptide and MHC molecules, peptide-specifically or broadly (crossreactively), which leads to T cell activation. TCR also has an ability to recognize self-peptide-MHC complexes, which may cause autoimmune diseases. In this study, we analyze the difference between the mechanisms of specific and crossreactive recognitions of a peptide-MHC complex by TCRs, based on the observations from MD simulations and FMO method. Because the double mutations of the specifically recognizing TCR is known to cause autoimmune diseases, we also analyze the mutant TCR to elucidate the mechanism of autoimmune-related recognition and the relationship to the corssreactive one.

1Pos042 レプリカ交換分子動力学シミュレーションによって明らかになった HP1αCD/histone H3 tail 複合体形成の仕組み

Mechanism of the complex formation of HP1αCD/histone H3 tail revealed by the replica-exchange molecular dynamics simulations

Satoshi Omori[1], Nobuto Hashiguchi[2], Kei Moritsugu[2], Yoshifumi Nishimura[2], Akinori Kidera[2] ([1]*GSIS, Tohoku Univ.*, [2]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*)

Recognition of methylated lysine 9 of histone H3 by chromodomain of heterochromatin protein 1α (HP1αCD) leads to heterochromatin formation, which represses gene expressions. Recent studies have revealed the increase in binding affinity by phosphorylation of four consecutive serine residues in the N-terminal disordered loop of HP1αCD. To clarify the associated molecular mechanism, configurational samplings of HP1αCD in the H3 tail unbound/bound forms with/without serine phosphorylation were performed by replica-exchange molecular dynamics simulation. The phosphorylation was found to strongly affect the structural ensemble of the N-terminal loop, which stabilized the bound form, and destabilized the unbound form to promote the complex formation of HP1αCD/H3 tail.

1Pos043 Go-like モデルを用いたプラストシアニンとシトクロム f 複合体の構造安定性に関する理論的研究

Theoretical study on the structural stability of plastocyanin and cytochrome f complex by using Go-like model

Satoshi Nakagawa, Shogo Kinoshita, Makoto Wada, Kazutomo Kawaguchi, Hidemi Nagao (*Grad. Sch. Nat. Sci. Tech., Kanazawa Univ.*)

In photosynthesis, plastocyanin(PC) transfers one electron from cytochrome f(Cytf) in the cytochrome b6/f complex to P700+ in Photosystem I. Although the structure of PC-Cytf complex has already been analyzed by NMR measurements, the structure of PC-P700+ complex is not analyzed yet. In this study, we propose a theoretical and computational approach to explore the stable structure of complex by using langevin dynamics simulation consisting of Go-like model. In our approach, intermolecular interaction is considered as only hydrophobic interaction. From our results of PC-Cytf complex simulation, the present complex structure becomes similar to the experimental results. We discuss the effect of hydrophobic interaction in the complex system by using free energy profile.

1Pos044 粗視化分子動力学シミュレーションを用いた MVM の生物物理学的特性の解明

Elucidating biophysical properties of the Minute Virus of Mice capsid: Coarse-Grained Molecular simulation

Koji Ono, Shoji Takada (*Dept. Biophys., Grad. Sch. Sci., Kyoto Univ.*)

Viruses are deeply related to human diseases. For example, Zika virus recently has been reported in many countries and causes a serious disease in newborns. Currently, we can use much information about viral biophysical properties. Viral capsid shows high symmetry structure which is key to performing viral self-assembly. Viral self-assembly is one of the most important part in viral life cycle. In this study, We focused on Minute Virus of Vice (MVM) which is one of the simplest non-enveloped icosahedral single-strand DNA virus. To deeply understand viral biophysical properties for investigating viral self-assembly process at molecular level, we performed coarse-grained molecular dynamics simulations of atomic force microscopy(AFM) nanoindentations of MVM capsid.

1Pos045 粗視化力場を用いたタンパク質-リガンド結合シミュレーション：結合経路上の変異がリガンド結合に及ぼす影響の解析

Coarse-grained simulations of protein-ligand binding: effect of mutations near the ligand-binding pathways

Tatsuki Negami, Tohru Terada, Kentaro Shimizu (*Grad. Sch. of Agri. and Life. Sci., The Univ. of Tokyo*)

Based on the results of coarse-grained molecular dynamics (CGMD) simulations performed on various protein-ligand systems, we have hypothesized that the ligands move along specific pathways to reach the ligand-binding pockets. This hypothesis implies that substitutions of the residues near the pathways should alter the binding kinetics. In this study, we therefore designed several mutants of a protein that had amino-acid substitutions near its ligand-binding pathway. For each mutant, 5-μs CGMD simulations were performed 100 times with different initial velocities and initial ligand placement. Based on the analysis of the trajectories, we discussed the effect of the mutations on the binding/unbinding kinetics and the ligand-binding pathway.

1Pos046 タンパク質-リガンドドッキング計算における最適パラメータの同定による分子設計の拡張

Identification of optimal parameter values in ligand-receptor docking calculation to extend applicability

Takuya Sumi[1], Hiroshi Yamaguchi[2], Ryuichiro Terada[1], Jiyoung Kang[1], Masaru Tateno[1] ([1]*Grad. Sch. Sci., Univ. of Hyogo*, [2]*Grad. Sch. Med., Nagoya Univ.*)

For molecular design, such as discovering of new pharmacological drugs, docking simulations of ligand and receptor protein have played a crucial role. However, some available programs are not satisfiable in the precision: In fact, for some ligand-receptor complexes that we are working on, ASEDock, which was implemented in MOE, did not generate the correct structures, while for other complexes, it successfully did. In order to extend the applicability, we examined some parameter values, most of which were relevant to the conformational/configurational spaces to be explored. The calculations with the refined optimal parameter values provided better structures than those obtained by other programs such as AutoDock4.2, preserving the balance with the computational costs.

1Pos047 Analysis of protein complexes structures towards rational design of inhibitors of Protein-protein interactions (PPIs)

Daisuke Kobayashi, George Chikenji (*Nagoya Univ.*)

Recently the increasing number of inhibitors for PPIs have been found, and for some proteins, several protein-inhibitors complexes have become available. It is believed that only a small part of interface residues, the so-callled “hot spots”, contribute a lot to the binding energy in PPIs. However, it is still unclear if hot spots actually play key roles in binding with the partner proteins or the inhibitors against the proteins in the structurally characterized complexes. Here, we analyze the protein-inhibitors structures, and investigate the key structures of the interacting sites of PPIs, which could be templates to mimic for designing novel inhibitors for PPIs. Moreover, we aim to clarify that to what extent those crucial structures overlap with hot spots.

1Pos048* 天然タンパク質の立体構造物性に関する統計解析

Statistical analysis on the structural properties of native proteins

Hidenobu Kawai, Daisuke Takahashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

Both a protein density, ρ, and a radius of gyration, R_g, are important parameters characterizing structural properties of a protein. However, how R_g and ρ are related to protein structure and function is poorly understood. To clarify this issue, here we calculated R_g and ρ of protein domains in the SCOPe database. We found that the maximally compact native proteins are rich in α/β-structures and disulfide bonds and have functions related to metabolism and hydrolase. On the other hand, small proteins with disulfide bonds such as toxins are most frequently observed in high density proteins, while α/β-structures and enzymes are frequent in low density proteins. These results suggest that protein functions can be restricted by structural constraints.

1Pos049 膜貫通βバレルにおけるβストランドのねじれと曲りに関する解析
Twisting and bending of β-strand in the transmembrane β-barrel

Nobuaki Kikuchi, Shinichi Ebisawa, Yuka Watanabe, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. Bioinfo., Grad. Sch. Eng., Soka Univ.*)

It is well known that the majority of β-sheets consist of the twisted and the bended β-strands. In our previous study for soluble proteins, Ser, Thr and Asn were found to suppress twisting β-strand. In this study, we calculated twist and bend angles of β-strands in transmembrane β-barrel. The average twist and bend angles of β-strands were analyzed on the basis of the number of β-strands present in the barrel, and it was found that they decrease with increasing the number of β-strands in the barrel. We also analyzed amino acid composition in membrane barrel proteins and found that the fractional contents of Ser and Thr residues increase with the number of β-strands. These results indicate that Ser and Thr residues suppress the twist also in the transmembrane β-barrel.

1Pos050 球状蛋白質の構造的性質とフォールディング速度との相関
Relationship between the Folding Rate and Structure-based Properties of Globular Proteins

Balachandran Manavalan[2,3], **Kunihiro Kuwajima**[1,2], Jooyoung Lee[2,3] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Comput. Sci., KIAS*, [3]*Center In-Silico Protein Sci., KIAS*)

We constructed a non-redundant database, which contains the data for folding kinetics of 77 two-state and 46 non-two-state proteins. The temperature correction was introduced on the basis of the Eyring equation of protein folding, so that all the rate constant values were standardized at 25°C. We studied the correlation between the folding rate and structure-based properties of proteins. We found that long-range contacts between residues at least 16 residues apart from each other along the primary sequence control the folding of the non-two-state proteins, while more short-range contacts and simultaneous organization of the whole structure predominate the folding of the two-state proteins.

1Pos051* 金属イオンがα-ラクトアルブミンのフォールディング中間体の熱力学的安定性に与える影響
Effects of metal ions on thermodynamic stability of folding intermediates of α-lactalbumin

Reina Shinozaki, Michio Iwaoka (*Dep. Chem., Sch. Sci., Tokai Univ.*)

α-Lactalbumin (αLA), which has four native disulfide bonds (SS), is stabilized by a calcium ion (Ca^{2+}) bound in the β-domain. It was reported that the oxidative refolding from the fully reduced form (R) to the native state (N) proceeds at pH 8.4 through characteristic intermediates, 2S* and 3S*, which are metastable intermediates among the ensembles of the SS intermediates. In this study, to reinvestigate the oxidative folding pathways, we carried out the experiments by application of a selenium oxidant, DL-*trans*-3,4-dihydroxy-1-selenolane oxide (DHS^{ox}), which has high oxidizing ability in a wide applicable pH range (pH 3-10). The results will be discussed focusing on the effects of a metal ion on the thermodynamic stability of the folding intermediates.

1Pos052 ウマアポミオグロビンの pH によるフォールディング機構
Mechanism of pH-induced folding of horse apomyoglobin studied by a statistical mechanical model

Takuya Mizukami, Yosuke Sakuma, **Kosuke Maki** (*Grad. Sch. Sci., Nagoya Univ.*)

We exploited a statistical mechanical model that incorporates the protonation mechanism of conventional models, the Monod-Wyman-Changeux and the Linderstrom-Lang smeared charge models, to investigate the folding of horse apomyoglobin over a wide pH range with a time window of ~40 µs to ~100 s, using continuous-/stopped-flow fluorescence. Quantitative analysis assuming a five-state sequential scheme indicated that 1) pH-induced folding/unfolding is represented by both specific binding and Coulombic interactions; 2) kinetic folding/unfolding intermediates share kinetic mechanisms with the equilibrium intermediate, indicating their equivalence; and 3) native-like properties are acquired successively during folding by intermediates and in transition states.

1Pos053 SWAXS 解析によるトレハロースがミオグロビン構造へ与える効果の解明
SWAXS analysis on effect of trehalose on myoglobin structure

Satoshi Ajito, Mitsuhiro Hirai (*Grad. Sch. Sci., Univ. Gunma*)

Trehalose is one of the disaccharides consisting of glucoses. Microorganisms become to produce trehalose under extreme environments such as high and/or low temperature, oxidation, desiccation, and so forth. Thus, trehalose is considered to have distinct bio-protective effects compared with other sugars. However, the molecular mechanism underling the bio-protective function of trehalose in living cells is still under intensive issues. We have studied the structure of myoglobin dissolved in trehalose solutions using X-ray scattering method. The pH, trehalose concentration, and temperature of the solutions were varied. The effect of the denaturant (GndHCl) under the presence of trehalose has also been studied. The details under various conditions will be presented.

1Pos054* Conformational Diversity in the Intrinsically Disordered HIV-1 Tat Protein induced by Zinc and pH

Tomoko Kunihara, Yuuki Hayashi, Hisashi Kudo, Hidenobu Kawai, Yoshiki Oka, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

HIV-1 transactivator of transcription (Tat) is an intrinsically disordered protein, and it involves in various functions. Previous studies show that Tat binds to zinc ions both in isolation and in the transcriptional complex. However, effects of zinc binding on Tat conformations remain unclear. Here, we studied structures of the zinc-bound Tat isolate by circular dichroism, NMR and X-ray scattering. We revealed that the zinc-bound Tat at neutral pH has slightly compact structures with residual helices, whereas Tat is fully unfolded at acidic pH or in the zinc-free state. Moreover, we found a pH-induced equilibrium folding intermediate of Tat. Such conformational diversity induced by zinc and pH may cause multiple functions of Tat.

1Pos055 ＮＭＲ測定と同期したアゾベンゼン架橋剤の光異性化反応によるＧＢ１タンパク質のフォールディング操作

Manipulating Protein GB1 Folding Using Photoisomerization of an Azobenzene Cross-Linker Synchronously with NMR Observation

Toshio Nagashima, Keisuke Ueda, Toshio Yamazaki (*RIKEN CLST*)

Azobenzene (AB) is used as a molecular device, which owns a property of large geometry change by *cis-trans* photoisomerization. The photoisomerization induces a conformational change of an AB-cross-linked protein through two cysteine residues separated by an appropriated distance. We published a method to detect this dynamic process by NMR spectroscopy in last year. In this presentation, we show several mutants of AB-cross-linked protein GB1 indicate distinguishing unfolded structures coupled with photoisomerization of the AB. These results contain significant information of the site-specific stability of the folding. This technique may provide a novel method of the artificial site-specific manipulation of protein structure and a new perspective on the protein folding.

1Pos056 レプリカ交換分子動力学シミュレーションによる pH に依存したポリグルタミン酸の構造変化の研究

Replica-exchange molecular dynamics study of pH dependent structural changes of polyglutamic acids

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The solution pH plays important roles in the protein structure formation. During the past several decades, the relationship between structures of polyglutamic acids (PGA) and the solvent pH has been extensively studied. However, the nature of the dominant structure under acidic condition is still controversial. In this study, we evaluated the helix stability of the PGA taking the full- and non-protonated states, by using replica exchange molecular dynamics simulations. As a result, the helix content of PGA was qualitatively reproduced, while there is some inconsistency with another part of experimental data. The charge of partially deprotonated states may have a critical role in the reproduction of experimental data completely.

1Pos057* Secondary structural change of glucagon during fibril formation process with DMPC lipid bilayers as revealed by ^{13}C solid-state NMR

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Glucagon is a 29-amino acid peptide hormone secreted in the pancreas to control the level of blood sugar and easily forms amyloid fibrils at acidic condition. ^{31}P solid-state NMR experiments of the glucagon-membrane system at neutral pH indicated that glucagon forms some types of aggregations and the intermediates of glucagon fibrils disrupt the membrane. Here, we measured structural change of Gly^4 and Ala^{19} during glucagon fibrillation in the glucagon-membrane system at neutral pH by ^{13}C solid-state NMR. We found that the secondary structure around both residues changed from α-helix to β-sheet. These results provide structural insight into glucagon fibril formation in the lipid bilayers. We will discuss the detailed interactions between glucagon fibrils and membrane.

1Pos058 天然変性タンパク質 c-Jun と転写コアクチベータ CBP の KIX ドメインの相互作用

Interaction of the intrinsically disordered c-Jun with the KIX domain of the transcriptional coactivator CBP

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Intrinsically disordered proteins play an important role in cellular functions by binding their targets and folding into ordered structures. However, the mechanism by which they recognize their targets is poorly understood. To understand the target recognition mechanisms of intrinsically disordered proteins, we studied the interaction of c-Jun with the KIX domain of the transcriptional coactivator CBP. Here, we overexpressed the N-terminal activation domain of c-Jun in *E. coli* and purified the protein. Circular dichroism spectra show that c-Jun is intrinsically disordered. Using NMR, we will study the KIX-binding site of c-Jun. We will also study the structure and dynamics of c-Jun in the free and the KIX-bound forms.

1Pos059 ヒト α-シヌクレインのダイナミクスとアミロイド線維形成のしやすさの関係

Relationship between the dynamics of human α-synuclein and its propensity to form amyloid fibrils

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Amyloid fibrils of α-synuclein (α-Syn) (and/or its intermediate structures toward the mature fibrils) is involved with pathogenesis of Parkinson's disease. Elucidation of the mechanism of amyloid fibril formation of α-Syn is thus important. Here we compared the dynamic behavior of α-Syn under various conditions which alter the propensity of the protein to form amyloid fibrils by quasielastic neutron scattering experiments using the backscattering spectrometer BL02 (DNA) at MLF/J-PARC. It was shown that the internal dynamics of α-Syn is enhanced when the proteins are under the conditions where they show an increased propensity to form amyloid fibrils. This implies that the fibril formation of α-Syn is entropically driven.

1Pos060* 中間状態で阻害するフィブリノーゲンのアミロイド線維化抑制効果

Fibrinogen inhibits amyloid fibrillation by stopping at the stage of intermediates

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Amyloid fibrils, abnormal protein aggregates, are associated with a number of pathological diseases. The formation of amyloid fibrils is usually suppressed by certain mechanisms in vivo, which might be owing to its multi-protein coexisting environment. In this study, focusing on fibrinogen (Fg), originally known to acting as a vital part in blood coagulation, we investigated effects of Fg on amyloid formation of an insulin-derived peptide fragment. As a result, we found that Fg strongly inhibited the fibrillation by lengthening the lifetime of intermediate species. Together with the result of NMR measurement, it was indicated that the amyloid intermediates attach to surface of Fg, which thereby retarded the subsequent formation of fibrils markedly.

1Pos061 ポリグルタミン酸の構造特性に関する陽溶媒における効率的な分子動力学による研究

Structural feature of polyglutamic acids studied by enhanced molecular dynamics with explicit solvent

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As the polyglutamic acids exhibit the helix-coil transition with respect to the pH, this peptide has been extensively studied. Experiments such as CD spectra demonstrated that it assumes the coil and helix structures in the neutral and acid solutions, respectively. Interestingly, the back bending behavior of pKa at decreasing pH can be seen as an example of allosteric effect over the proton binding affinity. Nevertheless, even the dominant structure in acid solution is still controversial. In this study, we performed replica-exchange with solute tempering simulations of the fully protonated polyglutamic acids in the explicit solvent and shed the light on the structural feature of the peptide.

1Pos062 Control and biophysical characterization of soluble protein oligomers using short peptide tags

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Here we report the effects of short peptide tags, consisting of 5 consecutive single type amino acids, attached to a model BPTI variant, on sub-visible soluble oligomer formation. Dynamic Light Scattering (DLS) indicated that highly solubilizing K, D and E tags favored 3-4mers, whereas the S, N, T and Q tags, which moderately affect protein solubility, produced dimers and hydrophobic I and L produced 15-7 mers along with very large soluble oligomers. The DLS observations were almost fully corroborated by size-exclusion chromatography. Furthermore, the biochemical and biophysical stability of the tagged BPTIs were influenced by oligomer sizes. These observations clearly suggest that peptide tags can be used for controlling the formation of sub-visible soluble oligomers.

1Pos063* 複数のアミロイド性ペプチドを含む複雑な系におけるアミロイド線維形成

Amyloid Fibrillation in Promiscuous Systems Containing Various Amyloidogenic Peptides

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While there are many studies addressing amyloid fibrillation of a single peptide, amyloid fibrillation in promiscuous systems, i.e., mixture of various peptides, has not been studied. We studied the amyloid fibrillation of K3, which is one of the proteolytic fragments of β_2-microglobulin (β2m) digested by Achromobacter protease I. Two systems were examined in our research; amyloid fibrillation of a mixture of the proteolytic fragments and that of K3 in the presence of β2m. Solution NMR combined with kinetic measurements revealed that the protein-protein interactions retarded the amyloid fibrillation of K3 in both systems. Our results suggest a novel mechanism of amyloid fibrillation in promiscuous systems, especially those under physiological conditions.

1Pos064 光合成生物及び非光合成生物由来 ferredoxin-NADPH 酸化還元酵素触媒反応の可逆性

Reversibility of the redox reactions catalyzed by ferredoxin-NADPH oxidoreductases from phototroph and heterotroph

Daisuke Seo (*Nat. Sci. Tec., Kanazawa Univ.*)

Ferredoxin-NADPH oxidoreductase (FNR) is a member of the flavin-dependent NADPH oxidoreductase family catalyzing reversible redox reaction between ferredoxin and NADPH. In this presentation, reversibility of FNR catalyzing reactions is discussed in the view of structure function relations based on the pre-steady state reaction data. The reaction of FNR from heterotroph *Bacillus subtilis* was optimized to NADPH oxidation direction whereas that from photoautotroph *Chlorobaculum tepidum* was rather reversible. The results of the truncation mutational works on the C terminal residues suggested that the residue stacked on the re-face of the isoalloxazine ring is involved in the control of the redox properties of FNRs.

1Pos065 高活性型 CaMKIδ(1-299)のキナーゼ研究への活用

Application of high active form CaMKIδ(1-299) for the study of protein kinase

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We describe here the expression and characterization of a constitutively active fragment of CaMKIδ(1-299). We used a simple one-step purification method to isolate the recombinant enzyme at high yield from Escherichia coli. CaMKIδ(1-299) exhibited broad substrate specificity highly similar to that of wild-type CaMKIδ, and complementary to that of the PKAc. The protein kinase activity of CaMKIδ(1-299) was higher compared with that of PKAc as well as CX-30K-CaMKII that comprises a constitutively active fragment of CaMKII. Furthermore, kinase activity was highly stable against thermal inactivation and repeated freeze-thawing. Thus, CaMKIδ(1-299) represents a readily available alternative that can be used as a “phosphorylating reagent” alone or in combination.

1Pos066 システイン残基修飾によるピルビン酸デヒドロゲナーゼキナーゼ 2 の動的構造変化

Dynamical structural changes of pyruvate dehydrogenase kinase 2 by modification of cysteine

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Human pyruvate dehydrogenase kinase 2 (PDK2) regulates the pyruvate dehydrogenase complex (PDC). PDK2 exists as a dimer, and C-terminal tail structure of PDK2 binds to other subunit (cross arm structure). Forming cross arm structure has been suggested to closely relate with the regulation of PDK2 function. In this study, we revealed the molecular mechanism of the PDK2 regulation by modification of Cys384 in the cross arm structure. Structural changes of the cross arm structure was characterized by measuring the time-resolved fluorescence spectroscopy of Trp383. Modification of Cys384 induced the motional flexibility of cross arm structure. This finding suggests that modification of Cys384 disrupted the cross arm structure, which caused the dissociation from PDC.

1Pos067 光照射を利用した硫酸還元菌由来[NiFe]ヒドロゲナーゼの活性化機構の FT-IR 研究

FT-IR studies on the activation mechanism of [NiFe] hydrogenase from *Desulfovibrio vulgaris* Miyazaki F using light irradiation

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[NiFe] hydrogenase is a metalloenzyme which catalyzes the reversible H_2 oxidation reaction. The acid-base equilibrium between the ready Ni-SI_r and active Ni-SI_a states is a common feature among [NiFe] hydrogenases, but the mechanism of the acid-base equilibrium remains unrevealed. We have shown for the first time that the Ni-SI_r state of [NiFe] hydrogenase is photo-activated to its Ni-SI_a state by laser light irradiation at 514.5 nm. From the pH-dependent light-reactivity of the Ni-SI_r state, we propose that the bridging OH^- ligand between Ni and Fe ions dissociates as a H_2O molecule from the Ni-Fe active site by light irradiation at low pH. These findings provide new insights into the activation mechanism of [NiFe] hydrogenase.

1Pos068 アフリカツメガエル由来(6–4)光回復酵素の 4 番目の電子移動トリプトファンの解析

Analysis of the fourth electron-transferring tryptophan in Xenopus laevis (6-4) photolyase

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(6-4) photolyases ((6-4)PL) are flavoproteins using blue light to repair the UV-induced pyrimidine(6-4)pyrimidone DNA lesions. Their flavin cofactor (FAD) can be reduced to repair-active FADH- by a photoinduced electron transfer. In (6-4)PLs, the FADH- is stabilized by the subsequent electron transfer via a chain of four Trp residues.
Here, we investigated the effect of mutation of the 4th Trp on the DNA repair activity of Xenopus laevis (6-4)PL (Xl64) in vivo. Photoreduction properties of this mutant was independently characterized in vitro.
Our results indicate that the 4th Trp in Xl64 is essential for photoreduction of FAD. However, Arabidopsis thaliana (6-4)PL (At64) doesn't have the 4th Trp. We discuss the role of residues around the 3rd Trp in Xl64 and At64.

1Pos069 分子シミュレーションによるエピジェネティックな酵素に対する基質の結合選択性の研究

Study for the Ligand Binding Selectivity of Epigenetic Enzymes by using Molecular Simulations

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Epigenetic Enzymes have important roles in regulation of gene expression. These enzymes control chromatin states by DNA metylation or histone modifications like metylation and acetylation. These proteins relate to several diseases including cancer and neurological diseases. These are thus potential targets of drugs for these diseases. Many inhibitors are proposed and some of them are used as drugs practically. In this study, we focused on selectivity of the inhibitors. Small change in interaction between inhibitor and enzyme causes difference of efficiency as inhibitor. We performed generalized-ensemble molecular dynamics simulations. And we reproduced the selectivity which observed in experiments.

1Pos070 分子動力学シミュレーションによる MutS の homoduplex DNA と mismatch DNA の認識メカニズム解析

Analysis of recognition of homoduplex and mismatched DNA by MutS by MD simulations

Hisashi Ishida, Atsushi Matsumoto (*National Institutes for Quantum and Radiological Science and Technology, Molecular Modeling and Simulation Group*)

In order to understand how MutS recognizes mismatched DNA and induces the reaction of DNA repair using ATP, the dynamics of the complexes of MutS (bound to the ADP and ATP nucleotides, or not) and DNA (with mismatched and matched base-pairs) were investigated using molecular dynamics simulations. As for DNA, the structure of the base-pairs of the homoduplex DNA which interacted with the DNA recognition site of MutS was intermittently disturbed, indicating that the homoduplex DNA was unstable. MM-PBSA/GBSA showed that the MutS-homoduplex DNA complex bound to two nucleotides was unstable because of the unfavorable interactions between MutS and DNA. This would trigger the ATP hydrolysis or separation of MutS and DNA to continue searching for mismatch base-pairs.

1Pos071 ONIOM 法を用いたアデニル酸キナーゼ反応機構に関する計算化学的研究

Computational Study on the Reaction Mechanism of Adenylate Kinase with ONIOM method

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Adenylate kinase catalyzes the reaction: ATP + AMP + Mg2+ -> ADP + ADP + Mg2+. We have been studying the theoretical model of the reaction using MM/QM(ONIOM) method. We constructed the model of complex structure of enzyme and substrates, ATP and AMP with Mg ion with some water molecules surronding the active center using MM or MD calculation with AMBER99 force field. The truncated models were used for the calculations with ONIOM method, and the reactant, product, transition structures were optimized. The highest level of the theory(B3LYP/6-31+G(d):Amber(embed)) with 853 atoms(89 atoms in QM) gives the reaction barrier of about 19 kcal/mol. The details about the model size, the conformational differences, or the free energy profiles, will be discussed.

1Pos072 Evolutionary optimisation of elastic network structures: Models of allosteric proteins

Holger Flechsig (*Hiroshima University*)

The functional activity of proteins often relies on allosteric coupling between functional regions within its structure. To elucidate general aspects of allosteric systems we have designed through evolutionary optimization elastic-network structures which, as the principal element of allostery, encode coupling among two remote binding sites. In the designed structures allosteric communication is analyzed in terms of strain propagation, used to identify pathways and subnetworks through which remote interactions are established. The effect of mutations is demonstrated and robustness of allosteric performance in the designed structures examined. Finally, we discuss the relevance of our model system in the light of actual allosteric proteins.

1Pos073 バクテリオロドプシンの構造・機能特性に対する物理架橋PVAハイドロゲル中への固定の影響
Effects of Immobilization of Bacteriorhodopsin with Poly(Vinyl Alcohol) Hydrogels on Its Structural and Functional Properties

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We previously reported the correlation between light-induced bleaching of a photochromic protein bacteriorhodopsin (bR) and change in its 2D-crystalline state. Immobilization of bR with keeping its structure and function is necessary to apply this relation to a novel optical device. In this work, structural and functional properties of bR in purple membrane (PM) in poly(vinyl alcohol) (PVA) hydrogel were studied since it can possess abundant water in matrix. Visible circular dichroism and X-ray diffraction analysis showed native-like 2D-crystal as well as PM stacking during gelation. Transient absorption exhibited little influence on bR photocycle in immobilized PM/PVA samples. A model for PM stacking during gelation bR will be presented from these experimental results.

1Pos074 **Thermo-induced phase separation dynamics of a biopolymer model on water-in-oil droplets**

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It is difficult to understand the living cell properties since they contain a variety of biopolymers. Therefore, simple model systems (e.g. water-in-oil lipid droplet) have been used in previous studies as a micro-cell like container. As a part of such studies, we investigated thermo-induced phase separation behaviors of Hydroxypropyl Cellulose (HPC), which was contained in the water-in-oil droplets with or without lipids as a biopolymer model. We found the start time of phase separation in a heating process had a dependence on the HPC concentration on the lipid droplets. Moreover, we examined the phase separation patterns on the droplets, which were coated with and without lipids respectively, so as to reveal the interaction between lipids and the biopolymer model.

1Pos075 タンパク質中に生成した金ナノクラスターの発光特性
Emission property of Au Nanoclusters Formed in Protein

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Protein-encapsulated Au nanoclusters (NC) have attracted much interest for their interesting optical properties. Depending on the reaction condition, different sizes of Au NC, Au25 and Au8, are formed within a packed of Bovine Serum Albumin (BSA). The emission lifetime of Au25-BSA was reported to be around several μs and become longer upon lowering temperature. We measured the excitation power dependence of Au25-BSA emission up to much higher range than reported so far. The emission intensity depended linearly on the excitation power up to 1.4×105 W/cm2 at 80 K, whereas it saturated already around 2.0×103 W/cm2 at room temperature. This behavior seems inconsistent with the previews report of the fluorescence lifetime. We will discuss the mechanism of our finding.

1Pos076 青色光によるバクテリオロドプシン色素再生に対する脂質膜相転移の影響
Effects of Lipid Phase Transition on Chromophore Regeneration of Bleached Bacteriorhodopsin in Bilayer Vesicles by Blue Light Irradiation

Shunsuke Yano[1], Kentarou Motegi[1], Hikaru Tanaka[1], Yasunori Yokoyama[1], Masashi Sonoyama[2], Koshi Takenaka[1] ([1]*Grad. Sch. Eng., Nagoya Univ.*, [2]*Grad. Sch. Sci. & Tech., Gunma Univ.*)

Bacteriorhodopsin (bR), a photochromic membrane protein, is one of candidate for industrial applications. We found out that reconstituted bR undergoes light-induced bleaching above lipid phase transition temperature where bR crystalline array is melt. Though complete regeneration of the bleached chromophore is preferred for the application, only ~70% regeneration by blue light irradiation was reported for bR in purple membrane. We elucidate the effects of lipid phase transition on the blue light regeneration of bR in proteoliposomes to improve the regeneration rate. The regeneration rate reached above 90% in the liquid crystalline phase, whereas it bottomed out at ~70% at gel phase at which bR forms native-like crystal. The cause of high regeneration rate is discussed.

1Pos077 ATP結合タンパクのゼロからのデザイン
Design of ATP-binding protein from scratch

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ATP hydrolysis plays important roles in various proteins such as molecular motor and kinase. To explore a minimal set for ATP hydrolysis in protein structures, we started to computationally design ATP-binding proteins from scratch. For a phosphates binding site of ATP, we used the P-loop, which is a conserved phosphate binding motif in native. Conducting folding simulations, we found optimal secondary structure lengths and loop types to build structures with the P-loop. Based on these, we generated various structures with different topologies and shapes, and tested how much the pocket for nucleoside of ATP is created. We report and discuss the designed simple ATP-binding proteins.

1Pos078* 合理的設計による抗体精製用リガンドFPAの開発
Rational design of FPA, a ligand for antibody purification

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Protein A (PA) is frequently used as an affinity ligand for purification of antibodies (Abs). However, acidic pH (typically pH 3-4) is required to dissociate Abs from PA, which may cause immunogenic aggregation of Abs. To dissociate Abs at more neutral pH, we designed a novel affinity ligand, FPA, which is a fusion of the B domain of PA and the coiled-coil region of c-Fos. Here, we confirmed by circular dichroism, small-angle X-ray scattering, and HPLC measurements that FPA associates Abs at pH 7 and dissociates them at pH 5 by forming a homodimeric coiled-coil. Moreover, to dissociate Abs at pH above 6, we have rationally introduced multiple amino acid substitutions into the c-Fos region of FPA. Thus, FPA is potentially useful for purification of antibody medicines.

1Pos079 新規酸化還元応答蛍光タンパク質の作成

New design of redox sensitive fluorescence proteins

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Redox homeostasis is critical for cell viability. For example, activities of various thiol-modulated enzymes are regulated by change in redox states. In addition, redox state of an organelle in the cell is regulated in conjunction with other organelles. Simultaneous observation of redox states in multiple organelles is therefore important to study the maintenance of redox homeostasis. For the purpose, we have invented new redox sensor proteins. Our sensor proteins designated as Oba-Q (oxidation balance sensed quenching proteins) and Re-Q (reduction sensed quenching proteins) show rapid quench of their fluorescence responding to the redox states. We successfully applied these proteins simultaneously to monitor redox changes in different organelles of HEK-239 cells.

1Pos080 自然界のタンパク質を大きく改造して創るヘム結合タンパク質

Computational design of heme-binding proteins by largely remodeling naturally occurring proteins

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One of the most important challenges for creating functional proteins is to design binding pockets for any given small-molecule ligands. We aim to develop a computational approach for engineering the binding pockets by remodeling both side-chains and main-chain of naturally occurring protein structures. Here, we try to design heme-binding proteins based on a protein (pdb: 1z1s) that does not bind heme. The scaffold protein has relatively large pocket inside the structure, but the size is not large enough to bind heme. Removing the C-terminal α-helix and a long loop of the protein, we computationally generated a large pocket for heme, which enable to build sidechains interacting heme in the pocket. The produced protein can be a base for targeting other small-molecules.

1Pos081 Selection of Ru(bpy)32+ motifs from a randomized peptide library

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Ru(bpy)32+ complex has become an important molecule in research. This attention results from unique attributes of the complex. Introduction of these attributes into proteins provides effective means for potential applications. Thus motivated, we aim to discover a ruthenium- binding motif by incorporating 3 bipyridylalanines (BpyAlas) into a library DNA contains 3 amber codons and using a mutant of an archaeal TyrRS which charges BpyAla to amber suppressor tRNA. After encapsulation of the library and translational system within liposome, the fluorescent positive liposome was isolated by FACS and recovered DNA was used for the next round of the selection. Finally, we read the sequence and synthesized a few peptides. The investigation of the ruthenium-ion binding is ongoing.

1Pos082 複数の遺伝子群の共進化を可能とする完全試験管内選択系の開発

Development of a totally in vitro selection system for co-evolution of plural genes

Asuka Ueki, Kei Fujiwara, Nobuhide Doi (*Grad. Sch. Sci. Tech., Keio Univ.*)

Directed evolution is a powerful tool to improve a single gene. With the emergence of synthetic biology, a technique for co-evolution of plural genes in a complicated system has been desired. Here we report our recent trial to construct a cell-free method for in vitro selection of a gene circuit by combining cell-free protein synthesis in liposomes and emulsion PCR. We found that several indispensable molecules for cell-free protein expression inhibit PCR reaction. This problem was solved by constructing a mutated thermophilic DNA polymerase. We applied this system for in vitro selection of a model circuit consisting of a sigma factor gene and its regulatory sequence located at the upstream of the reporter gene.

1Pos083* ファージディスプレイ法を用いたタンパク質デザインへの応用を目指した蛍光一分子ソーターの開発

Development of a single-molecular sorting system based on fluorescence detection for protein design using phage display method

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We constructed a single-molecule sorting system based on the confocal fluorescence optics and microfluidics, and aim to apply the system for the selection process in the phage display. We first introduced phages expressing five GFPs to the system; however, the number of phages counted by fluorescence signals was smaller than that estimated by the colony formation assay. We next introduced a quartz objective having low background and used a new phage expressing the larger number of GFPs, and suppressed adsorption to the flow path by using silica tubes. After these improvements, we could obtain the coincidence in the numbers of phages counted by fluorescence and colonies. We are conducting the separation of two phages, one expressing GFP and the other expressing YFP.

1Pos084 進化分子工学に向けたスクリーニングシステムの開発

Development of an integrated femtoliter chamber array system for directed evolution of protein molecules

Yi Zhang, Hiroto Kizoe, Yoshihiro Minagawa, Kazuhito Tabata, Hiroyuki Noji (*Grad. Sch. Eng., Univ. Tokyo*)

There is a great deal of interest in the development of screening system that combines protein synthesis, phenotype characterization, and genotype identification for directed evolution of proteins. We enabled massively parallel protein synthesis on femtoliter droplet array, where single DNA molecules can be encapsulated, transcribed, assayed, and recovered. The well-isolated droplets enabled the linkage of genotype and phenotype, and the stationary property of the chamber facilitated the characterization for protein functions. The quantity and activity of proteins synthesized in each droplet can be evaluated with a coupled fluorogenic assay, and the mutations responsible for the improved function can be determined with droplet recovery followed by sequencing.

1Pos085 **チトクロム *c* とチトクロム酸化酵素の複合体構造が示す新しいタンパク質間相互作用様式**

Complex structure of cytochrome *c* and cytochrome *c* oxidase shows a novel inter-protein interaction mode

Satoru Shimada[1], Kyoko Shinzawa-Itoh[1], Junpei Baba[1], Shimpei Aoe[1], Atsuhiro Shimada[1], Eiki Yamashita[2], Jiyoung Kang[1], Masaru Tateno[1], Shinya Yoshikawa[1], Tomitake Tsukihara[1,2] ([1]*Picobiology Inst., Grad. Sch. Life Sci., Univ. Hyogo*, [2]*Inst. Protein Res., Osaka Univ.*)

Cytochrome *c* oxidase (C*c*O) transfers electrons from cytochrome *c* (Cyt.*c*) to O_2 to make H_2O. To elucidate the electron transfer mechanism, we determined the structure of the mammalian Cyt.*c*-C*c*O complex at 2.0 Å resolution and identified an electron transfer pathway from Cyt.*c* to C*c*O. The novel inter-molecular interaction between Cyt.*c* and C*c*O is characterized by mutual recognition mediated by the long arms of a few hydrophilic amino acids; small contact surface; a long span between the proteins; the presence of three water layers between the proteins; and a large fluctuation of Cyt.*c*. The "soft and specific" interaction is predicted to contribute to the rapid association/dissociation of the Cyt.*c*-C*c*O complex, which facilitates sequential supply of electrons.

1Pos086 **蛍光偏光解消度を利用したシトクロム c-シトクロム c 酸化酵素間電子伝達複合体形成における相互作用解析**

Interaction analysis of electron transfer complex formation between cytochrome c-cytochrome c oxidase using fluorescence anisotropy

Hiroshi Kagaya[1], Wataru Sato[1], Takeshi Uchida[1,2], Kyoko Itoh－Shinzawa[3], Shinya Yoshikawa[3], Koichiro Ishimori[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*, [3]*Grad. Sch. of Life Sci., Univ. of Hyogo*)

The electron transfer (ET) reaction from cytochrome c (Cyt c) to cytochrome c oxidase (CcO) is an essential process for the energy generation in cells. Although the specific interprotein interactions are supposed to regulate the ET reaction, the molecular mechanism is still unclear. To identify key interactions for the ET complex formation between Cyt c and CcO, we prepared some Cyt c mutants and estimated the dissociation constant for the complexes in both of the reduced and oxidized states using fluorescence anisotropy measurements. The mutation at Ile81 of Cyt c increased the binding affinity to CcO, revealing that the hydrophobic interaction near the exposed heme periphery regulates the complex stability to facilitate the ET reaction from Cyt c to CcO.

1Pos087 **チトクロム c 酸化酵素の水素結合状態変化の酸素還元反応への影響**

The effect of the hydrogen bond network on the oxygen reduction of cytochrome c oxidase

Yudai Aoyagi, Tatsuhito Nishiguchi, Kyoko Shinzawa-Itoh, Shinya Yoshikawa, Satoru Nakashima, Takashi Ogura (*Grad. Sch. Lif. Sci., Univ. Hyogo*)

Cytochrome c oxidase (CcO) is the terminal enzyme of the respiratory chain and catalyzes the reduction of molecular oxygen to water. The oxygen reduction reaction is coupled with proton pumping across the mitochondrial membrane.

We have proposed a mechanism in which CcO senses the enough amounts of protons before starting oxygen reduction. To clarify the mechanism, we performed time-resolved visible absorption measurement at pH 9.0 where supplied protons are deficient. In this study, we used oxygen lung flow system that supplied oxygen, which was necessary for the reaction. As a result, it was found that the reaction rate became slower at pH 9.0 than that at pH 6.8. And in initial stage of O2 binding, time constants are more complicated than that observed at pH 6.8.

1Pos088 **時間分解共鳴ラマン分光法によるチトクローム酸化酵素の共役機構**

Coupling mechanism of Cytochrome c oxidase studied by time-resolved resonance Raman spectroscopy

Satoru Nakashima, Yoshiyuki Nakagawa, Kyoko Itoh-Shinzawa, Shinya Yoshikawa, Takashi Ogura (*Grad. Sch. Sci., Univ. Hyogo*)

Cytochrome c oxidase (CcO) catalyses the oxygen reduction reaction coupled with proton pumping. The coupling between oxygen reduction site (heme a3 with CuB) and the proton pump pathway is the most crucial point that should be clarified in detail. To elucidate these proton transfer dynamics, time-resolved resonance Raman spectroscopy was applied to the CO photolysis in different pH. Whole dynamics of the protein accompanied with ligand dissociation was observed and step-by-step mechanism was revealed. Among the dynamics of the protein, heme a3 shift was affected by altering pH. The sensing of water pool environment by heme a3 was suggested and these finding suggested the coupling between the reaction site and proton pumping pathway.

1Pos089* **シトクロム c–シトクロム c 酸化酵素間の電子伝達複合体形成における脱水和の機能的意義**

Functional significance of dehydration for formation of electron transfer complex between cytochrome c and cytochrome c oxidase

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In the respiratory chain, cytochrome c oxidase (CcO) interacts with cytochrome c (Cyt c) to accept electrons. Our calorimetric experiments showed that the Cyt c-CcO complexation is entropically driven, suggesting dehydration associated with formation of hydrophobic interactions. Here, we examined the dehydration by osmotic pressure analysis, revealing that about 20 water molecules are dehydrated, which corresponds to 70% of the total entropy change for the binding. We also found that the primary dehydration site is around Ile81 located near the exposed hydrophobic heme periphery on Cyt c. Such dehydration would expel water from the interaction site to form the hydrophobic electron transfer pathway between two proteins, promoting the effective electron transfer reaction.

1Pos090 **呼吸鎖ヘム・銅酸素還元酵素スーパーファミリーのプロトン輸送経路の構造解析**

Structural analysis of the proton transfer pathway in respiratory heme-copper oxygen reductase superfamily

Kazumasa Muramoto (*Grad. Sch. of Life Sci., Univ. of Hyogo*)

Respiratory chain generates proton motive force coupled to electron transfer at high energy efficiency. Respiratory O_2 reductases and NO reductases belong to evolutionally related heme-copper O_2 reductase (HCOR) superfamily.

In most of HCOR structures, interface between the transmembrane and the extracellular domains containing water cluster is suggested to be a part of proton transfer pathway. In this study, to compare the interface structures of nine HCOR members, I performed molecular surface analysis. The results suggest that the water cluster is isolated from external solvents and local conformational change or protonation/deprotonation of specific amino acid is necessary for proton access between protein interior and exterior.

1Pos091 ナノリットルフロー時間分解可視・赤外分光法を用いた一酸化窒素還元酵素の短寿命反応過渡種の計測

Detection of Short-Lived Reaction Species of Nitric Oxide Reductase Using Nanoliter-Flow Time-Resolved Visible/IR Spectroscopy

Hanae Takeda[1], Tetsunari Kimura[2], Shoko Ishii[1], Takehiko Tosha[3], Yoshitsugu Shiro[1,3], Minoru Kubo[3,4] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Grad. Sch. Sci., Kobe Univ.*, [3]*SPring-8 Center, RIKEN*, [4]*JST PRESTO*)

Nitric oxide reductase (NOR) is a membrane protein that catalyzes the reduction of 2NO to N_2O in the active site, composed of heme and non-heme irons. Recently, we succeeded in detecting the NO-bound state upon caged-NO photolysis by time-resolved (TR) IR spectroscopy, using a novel nanoliter flow system. In the next step, to detect the transient hyponitrite ($N_2O_2^{2-}$) species, we are trying to control the reaction kinetics by the mutation. By TR UV-vis measurements, we confirmed that recombinant WT NOR exhibits the same reaction kinetics as the native enzyme. We are currently preparing variants on the proton pathway, which are expected to slow down the hyponitrite decomposition and thus allow for direct observation of the N-N bond formation.

1Pos092 金電極上に固定化した一酸化窒素還元酵素の電気化学的還元活性

Electrochemical reduction activity of nitric oxide reductase immobilized on Au electrodes

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Nitric oxide reductase (NOR) is an enzyme that catalyzes the reduction of nitric oxide (NO) to nitrous oxide (N_2O). Since the NO reduction mechanism on NOR is still unclear, it should be elucidated. To elucidate the NO reduction mechanism, the electrochemical method combined with surface spectroscopic analyses is attractive because the change of protein structures can be observed *in situ*.

We will report NO reduction activity of the NOR immobilized Au electrodes by using cyclic voltammetry. We will also report surface spectroscopic analyses to monitor the interfacial molecular structures and adsorbed species at NOR immobilized Au electrodes.

1Pos093 ナノディスクに再構成した Vibrio cholerae 由来シトクロム cbb3 の構造，機能的評価

Structural and functional characterization of nanodisc-reconstituted cytochrome cbb3 oxidase from Vibrio cholerae

Masanao Inoue[1], Akihiro Shibata[1], Mizue Imai[1], Takeshi Uchida[2], Kazumasa Muramoto[3], Noritsugu Shiro[3], Shinya Furukawa[3], Koichiro Ishimori[2] ([1]*Graduate School of Science, Hokkaido University*, [2]*Graduate School of Science, Hokkaido University*, [3]*Graduate School of Life Science, Hyogo University*)

Cytochrome cbb3 (cyt cbb3) is one of the respiratory chain enzymes, which promotes the key events of the aerobic respiration, O2 reduction and transmembrane proton pumping, by accepting electrons from cytochrome c4 (cyt c4). To characterize the structure and function of cyt cbb3 under physiological conditions, we reconstituted cyt cbb3 into nanodisc and examined the heme environmental structure and the ligand binding kinetics by resonance Raman spectroscopy and flash photolysis, respectively. While the heme environmental structure was quite similar to that observed for detergent-solubilized cyt cbb3, the higher oxygen affinity was suggested for nanodisc-reconstituted cyt cbb3, providing clues to explain the high activity of cyt cbb3 at low oxygen concentrations.

1Pos094* シトクロム *c* とカルジオリピン含有バイセルの相互作用の溶液 NMR 解析

Solution NMR characterization of the interaction between cyt *c* and cardiolipin-incorporated bicelles

Hisashi Kobayashi, Satoshi Nagao, Shun Hirota (*Grad. Sch. Mat. Sci., Nara Inst. Sci. Tech.*)

Cytochrome *c* (cyt *c*) is a heme protein located in mitochondria, where it transfers electrons in the respiratory chain. Cyt *c* associates with the inner mitochondrial membrane by interaction with cardiolipin (CL), which may increase the peroxidase activity of cyt *c*. Although the cyt *c*-CL interaction plays an important role to alter the cyt *c* function, the detailed mechanism of the function conversion is unclear. Solution NMR is a powerful method for analyzing molecular interactions in proteins. However, it is difficult to study membrane-associated proteins by solution NMR due to their high molecular weight. In this study, we investigated the CL-interaction site of cyt *c* by solution NMR using CL-incorporated bicelles.

1Pos095* **Molecular mechanism of the ATP-dependent modulation of the Mg^{2+} channel MgtE for Mg^{2+} homeositasis**

Atsuhiro Tomita[1], Mingfeng Zhang[2], Hironori Takeda[3], Fei Jin[2], Tatsuro Maruyama[4], Masanori Osawa[4], Ryuichiro Ishitani[1], Ichio Shimada[4], Zhiqiang Yan[2], Motoyuki Hattori[2], Osamu Nureki[1] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Sch. Life Sci., Fudan Univ.*, [3]*Fac. Life Sci., Kyoto Sangyo Univ.*, [4]*Grad. Sch. Pharm., Univ. Tokyo*)

Mg^{2+} is essential for numerous physiological processes, and Mg^{2+} homeostasis is crucial for life. MgtE is a Mg^{2+} channel widely distributed in all domain of life and is involved in the maintenance of intracellular Mg^{2+} homeostasis. Previous reports suggested the intracellular Mg^{2+}-dependent gating mechanism of MgtE. Here, we report the functional and structural analyses of MgtE, revealing ATP as another regulatory factor of MgtE to switch the mode of the channel gating. The ATP binding to MgtE enhances the affinity of MgtE for Mg^{2+} within a physiological range, enabling MgtE to act as a Mg^{2+} sensor in vivo, whereas the ATP dissociation from MgtE upregulates the Mg^{2+} influx even at the high intracellular Mg^{2+} concentration, possibly facilitating the ATP synthesis.

1Pos096 チトクロム酸化酵素の高分解能結晶構造から明らかとなった高効率プロトンポンプ機構

High-resolution crystal structure of cytochrome c oxidase reveals the mechanism of highly efficient proton pumping

Atsuhiro Shimada[1], Naomine Yano[1], Kazumasa Muramoto[1], Eiki Yamashita[2,3], Kyoko Shinzawa-Itoh[1], Tomitake Tsukihara[1,2], Shinya Yoshikawa[1] ([1]*Picobiol. Inst., Univ. Hyogo*, [2]*Inst. Protein Res., Osaka Univ.*, [3]*SPring-8, RIKEN*)

Cytochrome c oxidase (CcO) pumps 4 protons sequentially from N-side to P-side through the proton-pump pathway, called H-pathway, composed of water cavities and hydrogen bond networks. Although CcO stores 4 protons before proton pumping, there is no enough capacity storing the protons in H-pathway. Our improvement in the resolution of X-ray structures of fully oxidized/reduced CcOs from 1.8/1.9 Å to 1.5/1.6 Å identifies the presence of an isolated water cluster which preserves 4 pumping protons and donates them actively into H-pathway. Furthermore, multiple conformers of D51 at H-pathway exit suggesting a significant proton affinity upon reduction are consistent to the reported proton-pump timing. These findings show a well-ordered proton storage and release mechanism.

1Pos097 立体構造に基づいた原核生物由来ナトリウムチャネルにおける選択性フィルターの変異体解析

Structural and mutational analysis of the selectivity filter of prokaryotic sodium channel

Katsumasa Irie[1,2], Yukari Haga[2], Shun Nakamura[2], Yoshinori Fujiyoshi[1,2] ([1]*CeSPI, Nagoya Univ.*, [2]*Grad. Sch. Pharm. Nagoya Univ.*)

The selective permeation of sodium ion by voltage-gated sodium channel (Nav) plays the main role in the transition of the action potential in the neural cells.
We evaluated the electron density in the selectivity filter of the crystal structure of NavAb, which is homologue of prokaryotic Nav (NavBac), under various cationic conditions. Depends on the ionic radius or hydrogen exchange ratio of each cation, the different election density was observed. Especially, in the high concentration of sodium ion, asymmetrical density was found in the four-fold symmetric selectivity filter.
By the mutational analysis based on the structural information, the residues were determined that discriminates the ionic radius and dehydrates the hydrated cation in the selectivity filter.

1Pos098 X 線 1 分子動態計測法への試料温度ジャンプシステムの導入

The introduction of temperature-jump system to the Diffracted X-ray Tracking (DXT)

Hirofumi Shimizu, Masayuki Iwamoto (*Univ. Fukui. Fac. Med. Sci.*)

The DXT is a powerful method to measure the conformational changes of proteins in the single molecule level. We have previously achieved the recordings in a sub-millisecond time resolutions by adopting a high-speed camera system, which enabled us to evaluate the single-molecular fluctuations of KcsA potassium channels. Here we introduced IR laser and thermal camera system to change their temperature during the recordings. The temperature can be changed in the range of Δ60 degree by the laser flush and was monitored by the thermal camera system. We observed enhancement of the fluctuations responding to the upper-shift of the temperature. This preliminary data suggested that the temperature-jump system will be an efficient tool to study molecular fluctuations of proteins.

1Pos099 原子分解能のシミュレーションによって明らかになった ADP/ATP 膜輸送体の交互アクセス機構

Deciphering Alternating Access Mechanism of a Mitochondrial ADP/ATP Membrane Transporter with Atomistic Simulations

Koichi Tamura[1], Shigehiko Hayashi[2] ([1]*RIKEN AICS*, [2]*Grad. Sch. Sci., Kyoto Univ.*)

ADP/ATP carrier (AAC) is a membrane transport protein embedded in the mitochondrial inner membrane. Its physiological role is to exchange cytosolic ADP for matrix ATP. Although X-ray structures representing an outward-facing (OF) state have been available since 2003, structure corresponding to an inward-facing (IF) one is yet to be found. Utilizing a recently developed enhanced sampling molecular dynamics (MD) method, linear response path following (LRPF), we have revealed an atomic structure of the IF conformation of AAC. The IF structure we found was shown to bind a well-known inhibitor, bongkrekic acid, revealing its mechanism of inhibition in atomic details. Details of the structure will be discussed.

1Pos100 サーモフィリックロドプシンの極めて高い熱安定性に対する統計熱力学

Statistical Thermodynamics for Remarkably High Thermal Stability of Thermophilic Rhodopsin

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Thermophilic rhodopsin (TR) and Xanthorhodopsin (XR) are photoreceptor proteins which possess seven transmembrane domains. Although TR and XR have high amino-acid sequence similarity and almost the same three-dimensional structure, the thermal stability of TR is much higher than that of XR. Using our theory based on statistical thermodynamics, we show that a large solvent-entropy gain upon protein folding is responsible for the high stability of TR. The solvent is formed by hydrocarbon groups constituting nonpolar chains of the lipid bilayer within a membrane as well as water molecules. TR has more aromatic side chains with large sizes than XR. Moreover, these side chains efficiently participate in close packing of the protein, leading to the large solvent-entropy gain.

1Pos101* 脂質二分子膜中におけるポア形成ペプチドの分子メカニズム解明に向けたモデルペプチドのチャネル電流測定

Systematically designed model pore-forming peptides study on molecular mechanism in lipid bilayers using channel current recording

Yusuke Sekiya[1], Hirokazu Watanabe[1], Kenji Usui[2], Ryuji Kawano[1] ([1]*Tokyo Univ. Agr. Tech.*, [2]*Konan Univ.*)

Pore-forming antimicrobial peptides (AMPs) are expected to be novel antimicrobial agents because their action mechanism is applicable to broad spectrum microorganism. However, the detail of pore-forming mechanism associated with their structures has been still unclear. Here, to understand the relationship between the structure and the pore-forming activity, we measured channel current of 4 systematically designed model AMPs named LK peptides, which have 1, 2, 3 or 4 repeats of “LKKLLKL”, consist of leucine (L) and lysine (K) in peptide sequences. From analysis of channel current, it seems that we determined these peptides form pores through similar mechanism, however, longer peptides form more rapidly than shorter ones and it should reflect the pore-forming activity.

1Pos102* 補酵素フラビンの置換による微生物外膜シトクロムのプロトン移動の発見

Proton transfer reaction in outer-membrane flavocytochromes revealed by replacement of flavin cofactor

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Iron reducing bacteria (e.g. *Shewanella oneidensis* MR-1) have an ability to transport respiratory electrons from cell inside to extracellular solid substrates via flavocytochromes located outer-membrane (OMFs). Recently we have identified various small molecules that could replace flavin cofactor in OMFs. We, herein, report that our kinetic analysis with the alternative cofactors shows that proton transfer reaction is associated with the electron transfer in OMFs and possibly protons are exported via OMFs, suggesting that OMFs might act as a novel proton channel. In the poster, we will discuss about the detailed mechanism with the data about pH dependency and kinetic isotope effect on the electron transfer in flavin-replaced OMFs.

1Pos103 **Toward the elucidation of structure/function relationship of transport proteins**

Naoki Soga[1], Rikiya Watanabe[1,2], Hiroyuki Noji[1] ([1]*Dept. of App. Chem., The University of Tokyo*, [2]*PRESTO, JST*)

Transporter proteins play physiologic roles by transporting ions across bio-membranes. The ion transport is mediated via conformational change of transporters, and therefore, the elucidation of structure/function relationship, which would be provided by the simultaneous detection of conformational change and ion transport, is crucial to understand the operating principle of transporters. Here, we attempted to develop a novel platform for the simultaneous detection using lipid bilayer chamber system (ALBiC). The results showed that ALBiC allowed measuring conformational change and ion transport of transporter, e.g., F_oF_1-ATPase, at single molecule level. Thus, ALBiC would be applied as a basic tool for the elucidation of a structure/function relationship of transporters.

1Pos104 **生細胞における G タンパク質共役型受容体の拡散・機能連関の比較解析**
Comparative analysis of diffusion-function relationship of G protein-coupled receptors on the living cell surface

Masataka Yanagawa[1], Michio Hiroshima[1,2], Yuichi Togashi[3], Takahiro Yamashita[4], Yoshinori Shichida[4], Masayuki Murata[5], Masahiro Ueda[2,6], Yasushi Sako[1] ([1]*Cellular Informatics Lab., RIKEN*, [2]*QBiC, RIKEN*, [3]*RcMcD, Hiroshima Univ.*, [4]*Dept. Biophys., Grad. Sci., Kyoto Univ.*, [5]*Dept. Life Sci., Grad. Arts and Sci., Univ. Tokyo*, [6]*Grad. Frontier Biosci., Osaka Univ.*)

G protein-coupled receptors (GPCRs) are major drug targets. The development of a method for measuring the activities of GPCRs is essential for pharmacology and drug discovery. However, current drug screening methods require monitoring of the downstream intracellular reactions specific to each GPCR. Here, we show that single-molecule imaging provides an alternative method for assessing GPCR activity. First, we demonstrate that the diffusion coefficient of a class C GPCR, metabotropic glutamate receptor 3, is tightly coupled with its functional states such as G protein binding and clathrin-dependent endocytosis. Then, we compare the diffusion changes of various GPCRs upon ligand stimulation to verify the generality of the diffusion-function relationship of GPCRs.

1Pos105 **Trafficking of endocytic PAR-1 carrier vesicles in cancer cell**

Seohyun Lee[1], Kohsuke Gonda[2], Hideo Higuchi[1] ([1]*Graduate school of science, University of Tokyo*, [2]*Graduate school of medicine, Tohoku university*)

Endocytic machinery of membrane protein plays essential roles in understanding not only the feature of internalization of a specific target protein but also the signal transfer that is mediated by the protein. Here, we imaged the movement of Protease-activated receptor-1 (PAR-1) at the moment of endocytosis in three dimensions using dual-focus optic system. Since PAR-1 is activated by thrombin, we compared the difference of PAR-1 movements during endocytosis between activated state and inactivated state of PAR-1. To investigate the contribution of motor protein in endocytosis, the adaptor protein BicD1, which recruits dynein in vesicle transport is silenced in order to study how dyneins are related to the machinery of PAR-1 endocytosis as well as its signal transfer.

1Pos106 **ミトコンドリア内膜タンパク質のマイクロ流路デバイスによる実時間解析**
The real-time analysis of respiratory chain complex I on mitochondrial inner membrane by using microfluidic device

Yuji Kimura, Sayaka Kazami, Yu Hashimoto, Hiroyasu Itoh (*Tsukuba Research Center, Hamamatsu Photonics KK*)

Mitochondria play critical role in energy generation in cells, and their dysfunction is closely related with diseases and aging process. To establish evaluation system for mitochondria, we assessed activity of mitochondrial complex I (CI) by using microfluidic device: two flow channels connected with an aperture (2 μm) where a large-sized mitochondrial inner-membrane (giant mitoplast, GM) of porcine heart (10 μm) is captured. Because the binding site of NADH, a substrate for CI, is inside of the GM, an electrical pulse was applied to perforate the GM at the aperture, to get access to the binding site. As NADH diffused into the GM via the aperture, pH fluorescent indicator became bright at another channel. It suggests proton was transferred across the membrane by CI.

1Pos107 **細胞シグナリングに関与する上皮成長因子受容体クラスターのコレステロールを介した形成メカニズム**
Cholesterol Mediated Mechanism for Signaling Cluster Formation of Epidmal Growth Factor Receptorer

Michio Hiroshima[1,2], Masahiro Ueda[1], Yasushi Sako[2] ([1]*RIKEN QBiC*, [2]*RIKEN Cellular Informatics Laboratory*)

Activation of epidermal growth factor (EGF) signaling has been suggested to be inextricably concerned with EGF receptor clusters. We previously revealed the coupling between the receptor behavior and clustering by using single-molecule imaging, however, the relationship between clustering and membrane condition remains unknown. When membrane cholesterol which is one of the primary components of plasma membrane was depleted, the receptor mobility changed from confined to free diffusion and the cluster size became smaller. As the result, the downstream signaling to a cytoplasmic protein, Grb2, significantly changed. The correlation between the observed phenomena will be discussed to understand the clustering mechanism, especially from the aspect of signaling regulation.

1Pos108 **ヨクトリットルスケール空間において粘性が DNA の運動に与える影響の評価**
Evaluation of viscosity effect on DNA movement in yocto (10^{-24}) liter space

Masaki Matsushita, Hirokazu Watanabe, Masayuki Ohara, Ryuji Kawano (*Life Sci. Biotech., Tokyo Univ. Agri. Tech.*)

Alpha-hemolysin (αHL) has a cylindrical nanospace (vestibule) that is 2.6 nm in diameter and 5.0 nm height. A hairpin DNA (hpDNA) with 9 bp fits in the vestibule space and moves in the nanospace under applying voltages. At that time, we can observe three different types of current levels that are associated with three different types of the hpDNA states. In this study, we considered that the evaluating current fluctuations will make physicochemical properties clear in nanospace because the fluctuation can reflect the physical condition in the nano-environment. We attempted to study the viscosity effect of the aqueous solution in the nanospace. The αHL with hpDNA can be applied for evaluating the environment effects in nanospace as the “biological nanopore probe”.

1Pos109 ヌクレオソームスライディングの分子機構に関する分子シミュレーション研究

Molecular Mechanisms of Nucleosome Sliding Revealed by Coarse-Grained Molecular Dynamics Simulation

Toru Niina, Shoji Takada (*Graduate School of Science, Kyoto Univ.*)

Nucleosome, a complex of DNA and histone proteins, is a fundamental unit of chromatin. The positions of nucleosomes on genomic DNA play an important role for chromatin structure and gene regulation. Some recent studies experimentally revealed the nucleosome positions on DNA and other recent studies enabled to predict the positions theoretically. However, the dynamics of nucleosome movement is not well known. In this research, we used coarse-grained molecular dynamics simulation to investigate the dynamics and mechanism of nucleosome sliding. In simulation, we observed sliding that depends on DNA sequence. Then we analyzed the molecular mechanism of the sliding and found there are several movement modes of sliding.

1Pos110* 多分子及び一分子測定により解明されたがん抑制タンパク質p53の超高速セグメント間移動

Ultrafast intersegmental transfer of a tumor suppressor p53 investigated by ensemble and single-molecule measurements

Yuji Itoh[1,2], Agato Murata[1,2], Satoshi Takahashi[1,2], Kiyoto Kamagata[1,2] ([1]*IMRAM, Univ. Tohoku*, [2]*Grad. Sch. Sci., Univ. Tohoku*)

The intersegmental transfer (IT) is a process where DNA-binding proteins transfer from one site of DNA to other without dissociation, and is assumed to be important in the target search of a tumor suppressor p53. Based on stopped-flow measurements, we demonstrated that the kinetic rate of IT for p53 was more than 10 times faster than that of other DNA-binding proteins. The rate of IT was diminished in the mutant without the disordered C-terminal domain but not in the mutant without the core domain. Thus, the ultrafast IT is facilitated by the unique structural property of p53 and may account for its efficient target search. We succeeded in immobilizing DNAs crisscross and are currently trying to observe IT directly by using single-molecule fluorescence microscopy.

1Pos111 大腸菌非六量体型 DNA ヘリカーゼ UvrD 多量体の 1 分子 FRET イメージング

Single-molecule FRET imaging of the oligomeric form of the non-hexameric *Escherichia coli* helicase UvrD

Hiroaki Yokota (*Biophotonics lab, GPI*)

Escherichia coli UvrD protein is a non-hexameric superfamily I DNA helicase which plays a crucial role in nucleotide excision repair and methyl-directed mismatch repair. We performed direct single-molecule fluorescence visualization of the helicase and reported that the helicase unwinds DNA in the form of an oligomer (dimer or trimer) (*Biophys. J.* 2013). Although the oligomeric form is responsible for the unwinding of DNA, only monomeric structures of the helicase are available (*Cell* 2006). Here, to address the dynamics of the oligomer, single-molecule FRET imaging was performed. FRET was observed between UvrD monomers that were labeled with either a donor or an acceptor fluorophore.

1Pos112 Identification of initial ES complex of topoisomerase IIβ and target DNA employing molecular dynamics docking simulation

Kakeru Sakabe, Hiroshi Nisihigami, Jiyoung Kan, Masaru Tateno (*Grad. Sch. Sci., Univ. Hyogo*)

The double-stranded DNA (dsDNA) prefers to form highly-ordered complicated topological structures, such as the supercoil in genome DNA, despite of the linear DNA chains. DNA topoisomerase IIB (topoIIB) is an elaborate machine that acts as a scissors-and-paste of the phosphate backbones in dsDNA, resulting in passing another dsDNA chain through the breakage of the phosphate backbone. In this study, we built the initial DNA-topoIIB complex (an ES complex) by our sophisticated structural modeling and molecular docking techniques, since its 3D structure is impossible to be determined by the current crystallographic analysis. In the session, we discuss the dynamical concerted mechanisms that were elucidated to be achieved by the DNA recognition and catalysis by topoIIB.

1Pos113* Elongation of Intrinsically Disordered Linker in p53 and the Effects on DNA Binding and Sliding Ability

Dwiky Rendra Graha Subekti[1,2], Agato Murata[1,2], Yuji Ito[1,2], Satoshi Takahashi[1], Kiyoto Kamagata[1] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Sci., Tohoku Univ.*)

In tumor suppressor p53, the intrinsically disordered region (IDR) is considered only as a flexible linker connecting two DNA binding domains during the search of its target DNA sequence. However, a possible involvement of IDR in the activity of p53 is suggested due to the conservation of IDR length across different species. To address this issue, we prepared p53 mutants with doubled (DL-p53) and tripled (TL-p53) IDR length and investigated their DNA binding ability and sliding dynamics along DNA. WT-p53 possessed different dissociation constants (Kd) to target and non-target DNA, while DL and TL-p53 possessed similar Kd. The elongation of IDR didn't affect the sliding dynamics. These results suggest that IDR length of p53 is important for differentiating DNA sequence.

1Pos114 Nucleoprotein Filament Assembly Dynamics of Dmc1 and Rad51 Recombinases

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Eukaryotic cells contain two recombinases (Rad51 and Dmc1) involved in homologous recombination. In mitotic cells, only Rad51 is present, but in meiotic cells, both Rad51 and Dmc1 are required. We compared the kinetics of nucleoprotein filament assembly of these two recombinases in Saccharomyces cerevisiae using single-molecule tethered particle motion experiments. ScRad51 forms more stable filaments than ScDmc1 due to a larger association rate constants (kon), and both ScRad51 and ScDmc1 nucleate on DNA in dimers. We also found that ScDmc1 prefers to nucleate on ds/ssDNA junctions, while ScRad51 prefers to nucleate on ssDNA. These kinetic parameters allow to elucidate the interaction of Rad51 and Dmc1 recombinases in cellular conditions.

1Pos115 リバースジャイレースによるバブル DNA 超らせん導入の物理機構

Physical mechanism of introducing positive supercoils into bubble DNA by reverse gyrase

Ryota Moritake, Takato Sato, Yuta Suzuki, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

Reverse gyrase (RG) is a hyperthermophille-specific DNA topoisomerase that binds to partially heat-denatured DNA (bubble DNA) and overwinds (introduces positive supercoils into) DNA using ATP hydrolysis energy. Although it has been suggested that RG introduces positive supercoiling through large conformational changes (Lulchev & Klostermeier (2014) Nucl. Acids. Res.), the RG-bubble DNA complex structure and the mechanism of how ATP energy is used remain elusive. In this study, by conducting molecular dynamics simulation, we found RG stably binds to bubble DNA and the binding of negatively charged ATP to RG induces dielectric response. Our results suggest the vital role of electrostatics in the function of RG.

1Pos116 Single molecule FRET measurements of Cas9 conformation change

Kazushi Isomura[1], Shohei Kajimoto[2], Saki Osuka[3], Hiroshi Nishimasu[2], Tomohiro Shima[1], Tomotaka Komori[1], Osamu Nureki[2], Sotaro Uemura[1] ([1]*Uemura lab., Grad. Sch. Sci., Univ. Tokyo*, [2]*Nureki lab., Grad. Sch. Sci., Univ. Tokyo*, [3]*Dep. Bio., Sci., Univ. Tokyo*)

CRISPR-Cas9 is a widely used genome editing tool. Structural studies have shown that Cas9 undergoes sequential conformational changes that couple with single-guide RNA binding and subsequent DNA binding. However, the dynamic transition process during these conformational changes remains unobserved. To tackle this issue, we performed single molecule FRET measurements on Cas9 using site-directed fluorescent pairs at several locations. Our results suggest that after nucleotide binding, Cas9 retains a transition state for several seconds, but then fluctuates between structural polymorphisms. At this meeting, we will discuss how Cas9 dynamically changes its conformation to process guide RNA-directed DNA cleavage.

1Pos117* Investigate how mSWI5-SFR1 complex facilitates mRAD51 presynaptic filament formation using single-molecule approaches

Chih-Hao Lu[1], Guan-Chin Su[2], Peter Chi[2], Hung-Wen Li[1] ([1]*Dept. of Chemistry, Nat'l Taiwan Univ.*, [2]*Institute of Biochemical Science, Nat'l Taiwan Univ.*)

RAD51 is essential for DNA homologous recombinational (HR) repair. Single-stranded overhang in damaged DNA allows RAD51 to nucleate and extend into presynaptic filament. Nucleation is kinetically slower and several accessory proteins have been identified to regulate RAD51 nucleation. SWI5-SFR1 (S5S1) is a heterodimeric protein, and biochemical work showed that S5S1 stimulates RAD51-mediated HR preliminarily by stabilizing RAD51 filament. Our single-molecule expt. show that mS5S1 interacts with mRAD51 to form complex and efficiently triggers nucleation. Moreover, mS5S1 stimulates nucleation by reducing mRAD51 seeding size and increasing mRAD51-ssDNA affinity. Our research provides mechanistic details of how accessory proteins stimulate recombinase activity.

1Pos118 粗視化シミュレーションによる障害物存在下での DNA 結合タンパク質挙動研究

The movement of DNA binding protein including obstacles along DNA

Mami Saito[1], Shoji Takada[1], Tsuyoshi Terakawa[2] ([1]*Grad. Sch. Sci., Uni. Kyoto*, [2]*Medical Center, Uni. Columbia*)

There are many obstacles such as transcription factors and histones along DNA. For many processes in cells, such as transcription, DNA repair, and DNA replication, DNA binding proteins have to overcome many obstacles; almost all are roadblock on DNA and a few might occupy target sites. Some single molecular experiments reveal that DNA binding protein can bypass or push obstacles. However the detail of DNA binding protein movement is unclear. We address the movement of DNA binding protein under obstacles along DNA by performing coarse-grained molecular dynamics simulation software CafeMol.

1Pos119 結晶構造中で観察される DNA 構造ゆらぎの網羅解析

DNA conformational transition inferred from re-evaluation of m|Fo|-D|Fc| electron density maps

Tomoko Sunami[1], Toshiyuki Chatake[2], Hidetoshi Kono[1] ([1]*National Institutes for Quantum and Radiological Science and Technology*, [2]*Kyoto University Research Reactor Institute*)

Flexibility of DNA plays important roles on biological processes. Recent analyses have indicated that traditional refinement does not always give us accurate crystallographic heterogeneity and some information of DNA flexibility is possibly overlooked in previous crystallographic studies. We re-calculated m|Fo|-D|Fc| electron density maps of double helical DNA crystal structures in PDB to utilize the overlooked information. We observed potential conformational transitions in 27% of DNA phosphates analyzed. The present results indicate that peaks around phosphates in the m|Fo|-D|Fc| maps of Z-DNA and B-DNA sometimes corresponds to ZI ZII and A/B -> BI transitions, respectively. We also discuss effect of metal-coordination on conformational transitions in Z-DNA.

1Pos120 ＤＮＡ高次構造の捩じれ速度依存性

Dependence of twisting velocity on higher order structure of DNA

Kotaro Yoshida, Yoshihiro Murayama (*Tokyo Univ. of Agri. and Tech.*)

Higher order structure of DNA such as supercoils is dynamically controlled in a living cell, which affects the regulation of gene expression. We have investigated the process of the formation and deformation of DNA supercoils, and its twisting velocity dependence. One end of a single DNA molecule was attached to glass surface and another end was attached to a 3 μm diameter magnetic bead. We used a novel tweezer consists of a focused laser and two pairs of electric coils, which can apply force and torque to the bead independently. We have found that large structural changes often appear at same number of rotation due to the formation and deformation of supercoils. We will discuss the dependence of the twisting velocity on the structural change.

1Pos121 マイクロ液滴界面を利用した RNA 転写配列を有する DNA マイクロ構造体の構築

Construction of DNA micro-structures with RNA transcription sequences using the interface of microdroplets

Risa Watanabe[1], Masamune Morita[1], Miho Yanagisawa[2], Masahiro Takinoue[1] ([1]*Dept. Comput. Sci., Tokyo Tech.*, [2]*Dept. Appl. Phys., Tokyo Univ. Agri. Tech.*)

DNA nanotechnology has achieved the construction of DNA origami and DNA hydrogels based on DNA self-assembly. This technology has been developing rapidly and has received attention in various fields such as molecular robotics and biomedical applications. However, control technologies of functional DNA structures responsive to external stimuli have been underdeveloped. In this study, we succeeded in constructing hydrogel-like DNA microstructures with RNA transcription sequences by DNA self-assembly on interface of water-in-oil microdroplets covered with cationic lipids. Because hydrogels generally have the stimulus-responsive property, we believe that this structure can be used as intelligent materials such as stimuli-responsive drug gene expression systems.

1Pos122 Phase transition of genomic DNA molecules in solutions with different concentration of propanol

Yue Ma[1], Yuko Yoshikawa[2], Koichiro Sadakane[1], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Doshisha University*, [2]*Ritsumeikan University*)

In order to evaluate propanol's effects on DNA duplex stability and structure, a long chain DNA, λDNA (48 kbp), was observed through single-molecule by fluoresce microscopy. The average length of DNA molecules has been used to evaluate the compaction of DNA molecules. For DNA molecules in different concentrations of 1-propanol solutions, two minimum of average length showed at 60% (v/v) and 80% (v/v). In 2-propanol solutions, the average length of DNA decreased and transited from elongated coil state to folded globule as the concentration of 2-propanol increased until 75% (v/v). From the concentration of 75% (v/v), DNA maintained as folded globule and the average length of DNA molecules did not show any obvious change.

1Pos123 Nucleic Acid Folding Revealed From Replica Exchange Molecular Dynamics

Jacob Swadling (*University of Tokyo*)

Hairpin loops are one of the most common secondary structural motifs found in RNA. DNA hairpins, on the other hand, rarely occur in nature and as a consequence are far less studied. This work represents an in-depth computational comparison of the thermodynamics, structure formation and kinetics of these important secondary structures.

We have used Replica Exchange Molecular Dynamics (REMD) to elucidate the free energy landscapes of single-stranded RNA and DNA, of sequence 5'-UUUAACC(U)18GGUU-3' and 5'-TTTAACC(T)18GGTT-3' respectively. Each model consisted of 108 replicas of 32,522 atoms, simulated at temperatures ranging from 270 - 500 K for 1μs with an MD timestep of 2fs.

1Pos124 荷電脂質膜表面上での自己組織化 DNA マイクロ構造の形成

Formation of self-assembled DNA microarchitectures on a cationic lipid membrane surface

Masamune Morita[1], M. Shin-ichiro Nomura[2], Satoshi Murata[2], Miho Yanagisawa[3], Masahiro Takinoue[1] ([1]*Dept. Comput. Sci., Tokyo Tech.*, [2]*Dept. Robotics, Tohoku University*, [3]*Dept. Appl. Phys., Tokyo Univ. Agri. Tech.*)

Based on DNA nanotechnology, various-sized DNA architectures such as nanometer-sized DNA origami and macroscopic DNA hydrogels have been constructed. Self-assembled DNA microarchitectures has potential application in various fields such as molecular robotics and nucleic acid therapeutics. However, it is difficult to control the self-assembled DNA microarchitectures under conventional methods. Here, we present the generation method of self-assembled DNA microarchitectures with fractal-like patterns on a cationic lipid membrane surface of micrometer-sized water-in-oil droplets. This fractal-like pattern was formed by kinetic process of the DNA self-assembly. We hope that this technology can lead to novel generation method for construction of DNA microarchitectures.

1Pos125 DNA 光修復活性を有する DNA 酵素の赤外分光解析

FTIR spectroscopic analysis of a DNAzyme possessing DNA photorepair activity

Yuhi Kurahashi, Wijaya I M. Mahaputra, Tatsuya Iwata, Hideki Kandori (*Nagoya Institute of Technology*)

DNAzyme is a DNA that has enzymatic activity. In 2004, DNAzyme that repairs UV-induced damaged DNA, named UV1C, was reported. UV1C has guanine rich sequence and shows enzymatic activity in the presence of Na^+. Therefore, UV1C is considered to form a higher order structure, G-quadruplex (G4). To investigate repair mechanisms of UV1C, DNA repair activity was compared in the presence of monovalent cations (Li^+, Na^+, K^+). DNA repair was observed in the presence of not only Na^+ or K^+, but also Li^+. CD spectra showed different structure of UV1C with Li^+. We will present light-induced difference Fourier transform infrared (FTIR) spectra for the DNA repair by UV1C with such cations.

1Pos126 Sub-millisecond folding dynamics of $preQ_1$ riboswitch studied by two-dimensional fluorescence lifetime correlation spectroscopy (2D FLCS)

Bidyut Sarkar[1], Kunihiko Ishii[1,2], Tahei Tahara[1,2] ([1]*Molecular Spectroscopy Laboratory, RIKEN*, [2]*RIKEN Center for Advanced Photonics*)

The $preQ_1$ (7-aminomethyl-7-deazaguanine) riboswitches regulate the expression of genes involved in the biosynthesis of the nucleoside queuosine from GTP in bacteria. The $preQ_1$ riboswitches comprise some of the smallest metabolite sensing RNA sequences or 'aptamer' domains. Here we investigate the folding dynamics of the aptamer domain of $preQ_1$ riboswitch from B. *subtilis* using two-dimensional fluorescence lifetime correlation spectroscopy (2D FLCS)[1]. The ligand free 'apo'-state of $preQ_1$ riboswitch, strategically labeled with a FRET pair, shows sub-millisecond dynamics among multiple conformational states. We also study the dynamics of folding induced by ligand-binding of the riboswitch in the presence of $preQ_1$.

[1]Ishii, K.; Tahara, T. *J. Phys. Chem. B* 2013, 117, 11414.

1Pos127 F_1-ATPase の回転における加水分解待ち状態から ATP 結合待ち状態への構造遷移

Conformational transition from catalytic dwell to ATP-binding dwell in F_1-ATPase rotation

Kei-ichi Okazaki[1,2], Mitsuhiro Sugawa[3], Gerhard Hummer[2] ([1]*IMS*, [2]*MPI Biophysics*, [3]*Univ. of Tokyo*)

F_1-ATPase (F_1) is the catalytic part of ATP synthase that synthesizes most of ATP in living organisms. Extensive crystallographic and single-molecule studies have clarified atomic structures and mechanochemical coupling of this unique rotary motor, respectively. However, connection between these two aspects are not well established: structural basis of the functional cycle has been controversial. Here, we use molecular dynamics simulations to clarify conformational cycle of the molecular motor in atomic detail, integrating information from recent crystallographic and single-molecule studies. Especially, we resolve conformational dynamics from the catalytic dwell to the ATP-binding dwell that involves a 40 deg. rotation of the rotor.

1Pos128 a-subunit ヘリックスが傾いた新構造における FO 回転分子モーターのイオン伝導経路解析

Analysis of the ion pathway of FO molecular motor using the revised structure with tilted a-subunit helices

Kota Tezuka, Ryoichi Kiyama, Daiki Yamakoshi, Dan Parkin, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

FO motor is the membrane-embedded part of ATP synthase, which is composed of the stator a and b subunits and the rotor c-ring. FO converts the ion translocation into rotation of the c-ring. This mechanism is explained by the half-channel model that assumes two different ion channels that are formed by c-ring and a-subunit, and our previous MD study indicated the existence of the half-channels. In this study we extend our previous study by employing the recently-proposed structure in which a-subunit helices are considerably tilted. We found that water channels are formed depending on the rotary state of c-ring, as was observed in the vertical a-subunit helices. We examine the role of charged residues in b-subunit, as well as charged groups of lipids.

1Pos129 好熱菌由来の回転モーター F_1 の燐酸解離のタイミング

Timing of Pi release in the rotary motor thermophilic F_1 (TF_1)

Kengo Adachi[1], Kazuhiro Oiwa[2], Masasuke Yoshida[3], Taro Uyeda[1], Kazuhiko Kinosita, Jr.[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*Adv. ICT Res. Inst., NICT*, [3]*Dept. Mol. Biosci., Kyoto Sangyo Univ.*)

TF_1 is a rotary molecular motor driven by sequential hydrolysis of ATP in three catalytic sites. In the standard coupling scheme, ATP binding starts rotation at 0°, and at ~200° the ATP is cleaved, and then the ADP is released at ~240° after a third ATP is bound. However, whether Pi is released at 200° or 320° has not been settled. Here, ATPγS is a slowly hydrolyzed ATP analog and moreover thio-Pi release is ~two-fold slower than Pi release in TF_1, irrespective of temperature. During rotation of TF_1 attached with a 40-nm gold bead in ATPγS + ATP, slow thio-Pi release could not be discerned at 320° after long dwells at 200°, which correspond to ATPγS cleavage, at 18, 23, and 30°C. This suggests that both ATP cleavage and Pi release occur at 200° after ATP is bound at 0°.

1Pos130* F1-ATPase の制御因子としての ε サブユニットの ATP 解離反応における役割

Role of ε subunit on ATP dissociation as a regulator for F1-ATPase

Makoto Genda[1], Rikiya Watanabe[1], Yasuyuki Yamada[2], Hiroyuki Noji[1] ([1]*Graduate School of Engineering, University of Tokyo*, [2]*Department of Life Science, Rikkyo University*)

F1-ATPase is a rotary motor protein that catalyzes ATP synthesis via mechanical rotations. This chemo-mechanical coupling is regulated by ε subunit of F1, i.e. it enhances ATP synthesis efficiency by 4.6 folds; however it remains elusive how ε modulates at elementary step resolution. In this study, we conducted a stall-release experiment on the same F1 molecule before and after reconstitution of ε, which was confirmed using the fluorescent-labeled ε, and investigated the impact of ε on the rate-limiting step of ATP synthesis, i.e., ATP dissociation. After reconstitution of ε, the equilibrium of ATP binding / dissociation was inclined to ATP dissociation, suggesting that this equilibrium shift due to ε highly contributes to the enhancement of ATP synthesis efficiency.

1Pos131 F1-ATPase の P-loop 変異体におけるリン酸解離の機構

The kinetics of Pi release in F1-ATPase investigated with P-loop mutations

Hiroka Narita[1], Hitoshi Hoshina[1], Hikaru Yoshida[1], Yohei Nakayama[1], Shoichi Toyabe[2], Hiroshi Ueno[3], Eiro Muneyuki[1] ([1]*Dept. of Phys., Chuo Univ.*, [2]*Dept. Appl. Phys., Tohoku Univ.*, [3]*Dept. Appl. Chem., Tokyo Univ.*)

For a rotary molecular motor, F1-ATPase, two models are considered for the timing of ADP release and Pi release. In 2010 Watanabe et al. reported that Pi dissociates after ADP [Nat Chem Biol.6 814-820(2010)]. On the other hand, Shimo-Kon et al. has reported that ADP dissociates after Pi [Biophysical.J.98 1-10(2010)]. Thus, more experimental studies are required. In this study, we focused on P-loop mutant TF1 (βG158A) where dissociation of Pi is slow. By single molecule rotation experiment using gold colloid, long dwell time was observed at an angle of ATP hydrolysis and Pi dissociation pauses. We are trying to examine the rotation of hybrid F1 created from wild-type and P-loop mutant to see if the Pi dissociation and ATP hydrolysis on the mutant β are separated or not.

1Pos132* 腸球菌 V-ATPase の Na^+濃度依存 ATPase 活性を促進する化合物の同定

Identification of accelerators on Na^+-depending ATPase activity of Enterococcus hirae V-ATPase

Senka Gi[1], Lica Fabiana Yakushiji[1], Hiroshi Ueno[2], Hiroyuki Noji[2], Takayoshi Arai[1], Katsuhiko Moriyama[1], Hideo Togo[1], Takeshi Murata[1,3] ([1]*Grad. Sch. Sci., Univ. Chiba*, [2]*Grad. Sch. Eng., Univ. Tokyo*, [3]*PRESTO, JST*)

V-ATPase is an ion transport molecular motor found in various organelle membranes within the cell. V-ATPase consists of two parts, a hydrophilic part V1 hydrolysing ATP and a hydrophobic part Vo transporting the ion. Na^+-transporting V-ATPase enables E. hirae to grow in high salt and high pH condition. We performed the screening and searched for the compounds that affected the ATPase activity of E. hirae V-ATPase from Chiba Chemical Library. Several compounds accelerating ATPase activity were identified and the accelerators raised the Na^+-depending ATPase activity to 1.5 times. We also examined single molecular observation of V-ATPase with the accelerator. In our poster, we would like to discuss about the mechanism of the acceleration.

1Pos133 キネシンによる微小管の構造変化

A novel function of kinesin-1: changing microtubule conformation that accelerates successive kinesin binding

Tomohiro Shima[1,2], Manatsu Morikawa[3], Junichi Kaneshiro[1], Taketoshi Kambara[1], Shinji Kamimura[4], Toshiki Yagi[5], Hiroyuki Iwamoto[6], Taro Ichimura[1], Tomonobu Watanabe[1], Sotaro Uemura[2], Ryo Nitta[7], Yasushi Okada[1,2], Nobutaka Hirokawa[3] ([1]*RIKEN QBiC*, [2]*Grad. Sch. Sci., Univ. Tokyo*, [3]*Grad. Sch. Med., Univ. Tokyo*, [4]*Dept. Biol. Sci., Chuo Univ.*, [5]*Dept. Life Sci., Pref. Univ. Hiroshima*, [6]*SPring-8, JASRI*, [7]*RIKEN CLST*)

KIF5C, a member of kinesin-1, transports various cargoes along microtubules (MT). Despite abundance of MT in cytoplasm, KIF5C has often shown selective binding to a subset of MT. This selectivity has been explained by existence of different types of MT but the cause of such difference remains obscure. Here, we show that binding of KIF5C accelerates successive binding of KIF5C, suggesting that KIF5C changes MT into a high-affinity form. Consistent with the idea, our structural analysis demonstrate that binding of KIF5C elongates the axial tubulin-pitch of MT strongly resembling a high affinity form of MT. These results suggest positive feedback relation between KIF5C binding and MT conformation which leads the cooperative and selective KIF5C binding to a subset of MT.

1Pos134 二量体分子モーターの歩行に関する統一モデル

A unified walking model for dimeric motor proteins

Kazuo Sasaki[1], Motoshi Kaya[2], Hideo Higuchi[2] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Sci., Univ. Tokyo*)

Dimeric motor proteins, such as kinesin-1, cytoplasmic dynein-1, and myosin-V, move along protein filaments unidirectionally in a stepwise manner. Experiments have shown that these proteins take backward steps when large load forces are applied. The ratio of the number of backward steps to that of forward steps (“step ratio”) and the “dwell time” between steps were observed to increase as the load force increases. In contrast to kinesin and myosin-V, the experimental data for dynein available in the literature are not enough for quantitative analysis. We collected additional data for dynein, and propose a unified theoretical model to explain the dependences of the step ratio and of the dwell time on the load force measured for kinesin, dynein, and myosin-V.

1Pos135 タンデムに２つの頭部をつないだキネシンを用いた選択的な前方へのステップの研究

Preferential forward stepping mechanism of kinesin-1 studied using tandemly joined two-headed monomer

Kohei Matsuzaki[1], Hiroshi Isojima[1], Sawako Enoki[2], Hiroyuki Noji[2], Michio Tomishige[1] ([1]*Dept. Appl. Phys., Grad. Sch. Eng., Univ. Tokyo*, [2]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*)

Kinesin-1 moves along microtubule by alternately moving two motor domains (“heads”); the unbound trailing head is prohibited from rebinding to the rear-binding site and is allowed to binds to the forward-binding site 16-nm ahead. However, the mechanism of preferential stepping is still unclear. Previously we engineered asymmetrically joined two-headed monomer, in which the neck linker (C-terminus) of the first head is connected to N-terminus of the second head, to distinguish between biased binding and biased diffusion-based stepping. In this study we observed the motion of each heads using the high-speed dark field microscopy and found that the biased diffusion based stepping is less efficient because the unbound head frequently rebinds to the rear-binding site.

1Pos136 光応答性蛋白質 Dronpa を利用したキネシン運動活性の光可逆的阻害

Photo-reversible inhibition of kinesin motor activity utilizing photochromic protein Dronpa

Kohei Uchida, Shinsaku Maruta, Kazunori Kondo (*Grad. Sch. Bioinfo., Univ. Soka*)

Structure and energy transducing mechanism of ATP driven motor kinesin have been well studied. Previously we have demonstrated that incorporation of photochromic molecules into the microtubules binding site of kinesin enabled photo-reversible regulation of kinesin ATPase activity. In this study, we prepared the photochromic protein “Dronpa derivative 145K-145N” fused with kinesin microtubule binding site, “Loop11” and “Loop12” in order to inhibit kinesin motor activity photoreversibly. The Dronpa 145K-L11-145N showed the reversible absorption spectral changes upon 400nm and 500nm light irradiation in a same manner with original Dronpa tandem dimer 145K-145N. 145K-L11-145N inhibited microtubles gliding on the conventional kinesin in the invitro motility assay.

1Pos137* ゆらぎの定理を用いた非侵襲な力測定によるメラニン色素顆粒輸送のメカニズムの解明

Application of the fluctuation theorem for the non-invasive measurement of force to pigment transport in melanophores

Shin Hasegawa[1], Kazuho Ikeda[2], Takashi Sagawa[3], Yasushi Okada[2,4], Kumiko Hayashi[1,5] ([1]*Sch. Eng., Tohoku Univ.*, [2]*QBiC, RIKEN*, [3]*NICT*, [4]*Sch. Sci., Univ. Tokyo*, [5]*AMED, PRIME*)

Zebrafish rapidly alter their pigmentation in response to environmental changes. This change is mediated by aggregation and dispersion of melanosomes filled with melanin pigments in pigment cells of scales. Melanosomes are transported by kinesin and dynein. In particular, we studied the aggregation of melanosomes transported by dynein motors. In our work, we examined the motion of melanosomes. Analyzing the motion based on the fluctuation theorem of non-equilibrium statistical mechanics, the motor force was measured. The distribution of force thus measured has multiple peaks. It indicates that melanosomes were transported by multiple force-producing units (FPUs). Then we discuss the effect of inhibitor of dynein(ciliobrevin) on the number of FPUs.

1Pos138 DNA オリガミを用いた速度の異なるキネシンによる協調運動の観察

Cooperative transport by two kinesin motors with different velocities studied using programmable DNA origami

Ryosuke Masuda[1], Tsuyoshi Omi[1], Yamato Niitani[1], Mitsuhiro Iwaki[2], Michio Tomishige[1] ([1]*Department of Applied Physics, Univ. of Tokyo*, [2]*QBiC, RIKEN*)

Kinesin moves along microtubules to transport cargoes inside the cell. During the transport, multiple motors are bound to a cargo, however the mechanism of cooperativity between the motors is still unclear. Recently, DNA origami has been used to study cooperative transport by kinesin and dynein molecules. Here, we applied DNA origami to study cooperative transport by kinesin molecules with different velocities. When fast (kinesin-3) and slow (kinesin-5) kinesins were attached to both ends of rigid DNA origami scaffold, most of the scaffold moved with slow velocity, presumably by slowing down the fast motor. By using spring-like DNA origami as a scaffold, we could identify detachment and reattachment of the leading fast motor during the cooperative transport.

1Pos139 Inhibitory mechanism for photochromic kinesin Eg5 inhibitor composed of spiropyran derivative

Ryoma Yamamoto, Kei Sadakane, Shinsaku Maruta (*Grad. Sch. Bioinfo., Univ. Soka*)

Eg5 is one of the mitotic kinesin that maintain spindle formation and separate spindle. Inhibition of Eg5 results in and inducing cytostasis and apoptosis. It is well known that Eg5 has some specific potent inhibitors, e.g. STLC, Monastrol and Ispinesib. Previously we designed and synthesized a novel photochromic inhibitors of Eg5, Spiropyran-Diaminopropionic Acid (SP-APA) and Spiropyran-Lysine (SP-Lys). The inhibitors showed photo reversible inhibition of Eg5 ATPase activity. In this study, we analyzed the step influenced by the novel photochromic inhibitors in the ATPase kinetic pathway using stopped flow apparatus with fluorescent labeled ATP derivative, Mant-ATP. SP-Lys inhibited the initial binding step in the absence of microtubules.

1Pos140 1分子 FRET 観察による細胞質ダイニンの構造変化の計測 Single molecule FRET observation of cytoplasmic dynein's conformational change

Mikiya Sakata[1], Takuya Kobayashi[1], Mitsuhiro Sugawa[1], Tomohiro Shima[2], Junichiro Yajima[1], Yoko Y. Toyoshima[1] ([1]*Grad. Sch. Arts and Sci., Univ. of Tokyo*, [2]*Grad. Sch. Sci., Univ. of Tokyo*)

Single molecules of cytoplasmic dynein exhibit the diffusive motion on microtubules. This behavior is different from the unidirectional movement shown by collective molecules and considered to be an autoinhibited state. This state is related to the intramolecular head-head stacking, however, it is unclear the transition and the equilibrium between the stacking and separating states. Here, we attempted to observe the dynamics between the two states by single molecule FRET. Dynein motor head was engineered to be labeled with fluorescent dyes. The optical system was optimized to detect the FRET. By the FRET observation, the conformational change of dynein will be revealed, and we hope to see the state of dynein molecules in autoinhibited state on microtubules.

1Pos141 微小管系モーターダイニンを基に新規アクチン系モーターをエンジニアリングする Engineering Novel Actin-Based Molecular Motors from the Microtubule-based Motor Dynein

Akane Furuta, Kazuhiro Oiwa, Hiroaki Kojima, Ken'ya Furuta (*Frontier Research Lab, NICT*)

Our questions are: "what factors are critical for a linear molecular motor to simply move forward?" and "what determines the direction of movement?" To address these questions, we took a bottom-up approach where protein building blocks from different cytoskeletal systems are combined to create a new series of molecular motors. We show that the hybrid motors–combinations of a motor core derived from dynein and non-motor actin-binding proteins–robustly drive the sliding movement of actin filament. Moreover, the direction of movement is reversible by simply changing the geometric arrangement of these building blocks. Our synthetic strategy will provide the design principle of molecular motors that work, for example, along artificial tracks at nanometer dimensions.

1Pos142* 細胞質ダイニンのマルチスケールシミュレーション:全原子から連続体へ Multiscale Simulations of Cytoplasmic Dynein: From All-atom to Continuum Mechanics

Shinji Iida[1,3], Benjamin Hanson[4], Narutoshi Kamiya[2], Genji Kurisu[1], Takahide Kon[3], Haruki Nakamura[1], Sarah Harris[4] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. SS, Univ. Hyogo*, [3]*Grad. Sch. Sci., Osaka Univ.*, [4]*Sch. Phys. Astro., Univ. Leeds*)

As well as providing detailed information on the dynamics of macromolecules, all-atom molecular dynamics (MD) can also be used to parametrize coarse-grained (CG) models. One such model is Fluctuating Finite Element Analysis (FFEA), a simulation technique which represents large, globular proteins as continuum mechanical objects.
Our aim is to illustrate that this kind of multiscale modelling allows us to accurately track the dynamics of macromolecules. All-atom MD simulations of a dictyostelium discoideum dynein were performed, from which the effective Young's modulus was calculated and used to initialize a low resolution continuum representation. FFEA simulations were then performed using this CG model and were shown to successfully reproduce the relevant dynamics.

1Pos143 Diffusive Component in Directed Movements of Cytoplasmic Dynein

Takayuki Torisawa, Ken'ya Furuta, Kazuhiro Oiwa, Hiroaki Kojima (*NICT, Advanced ICT Research Institute*)

Motor proteins are enzymes that generate forces and directed movements under thermally fluctuating environments. Cytoplasmic dynein is a microtubule-based motor involving in almost all of the minus-end directed transports in the cell. Previous studies have demonstrated that dynein changes its behavior from diffusion to directed movement upon clustering or binding to the regulatory proteins. We observed the directed movement of dynein both in vitro and in vivo, and found that all the directed movement contained a diffusive property characteristic to the movement of single dynein molecules. Together with force-response measurements and numerical simulations, we propose that dynein rectifies thermal diffusion to achieve efficient directed transport.

1Pos144 細胞質ダイニンの構造変化を伴う運動メカニズムに関する分子シミュレーション研究 Molecular simulation study on the working mechanism with structural changes of cytoplasmic dynein

Shintaroh Kubo, Shoji Takada (*Grad. Sci., Univ. Kyoto*)

Dynein is one of the molecular motors which transport cargo on the microtubules(MTs) to the minus end by using ATP hydrolysis energy. Roughly, dynein consists of AAA+ ring (ATP hydrolysis sites) and microtubule binding domain(MTBD). Each domains have two different conformations. AAA+ ring takes pre or post state and MTBD shows different affinity to the MTs; low or high. It has already been reported that one motor domain takes several conformations. However, the molecular mechanism of conformational changes are not enough understood.
In this study, we focus on the conformational changes of one motor-domain between pre-low state and post-high state by using CafeMol which is coarse-grained MD simulation system developed in our laboratory.

1Pos145* ヒト細胞質ダイニン１分子のパワーストローク運動距離の測定

Measurement of the power stroke distance of cytoplasmic dynein motor

Yoshimi Kinoshita[1], Taketoshi Kambara[1,2], Kaori Nishikawa[1], Motoshi Kaya[1], Hideo Higuchi[1] ([1]*The University of Tokyo*, [2]*QBiC, Riken*)

Cytoplasmic dynein is a motor protein moving along microtubules, and plays important roles in vesicle transport and mitosis. To understand the conformational changes of dynein, we measured the efficiency of FRET from ring-BFP to linker-GFP, and the distance driven by power stroke of single-headed dynein interacting with microtubules by optical tweezers. The efficiency and the apparent stroke distance depend on ATP concentration. High efficiency and distance at ~1mM ATP indicate that the 9-nm power stroke is generated by swing of linker. This is supported by the result that the dynein mutated at its linker did not generate the power stroke. The power stroke driven by structural change of linker will be fundamental mechanism of dynein motility.

1Pos146 ミオシン S1 によって誘起されるアクチンフィラメントの協同的構造変化の高速 AFM による観察と、その生理的意義

High-speed AFM demonstration of cooperative structural changes in actin filaments induced by myosin S1 and physiological implication

Kien Xuan Ngo[1,2], Noriyuki Kodera[3], Toshio Ando[3], Taro Ueda[1,2] ([1]*Biomed. Res. Inst., AIST*, [2]*Dept. Phys., Waseda Univ.*, [3]*Dept. Phys., & Bio-AFM FRC, Kanazawa Univ.*)

Using high speed AFM, we imaged real-time binding of myosin subfragment-1 (S1) to actin filaments in solutions containing different nucleotides, and analyzed structural changes in actin. Helical pitch of actin filaments became longer by ~8% when they were experienced repetitive transient binding of S1 in the presence of ATP. Thus, cofilin, which shortens the helix by ~25%, and myosin II clearly induce different cooperative conformational changes in actin. We suggest that these structural differences in actin caused by binding of S1+ATP is the basis for cooperative inhibition of cofilin binding to actin filaments, which we established recently, implicating the structural polymorphism of actin filaments in their functional regulation in cells.

1Pos147 単一フィラメントにおけるアクチン重合・脱重合ダイナミクスの力学的制御

Mechanical manipulation of polymerization dynamics of individual actin filaments

Hiroaki Kubota[1], Makito Miyazaki[1,2], Taisaku Ogawa[3], Togo Shimozawa[4], Kazuhiko Kinosita Jr.[1], Shin'ichi Ishiwata[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*Waseda Bioscience Research Institute in Singapore, Waseda Univ.*, [3]*QBiC, Riken*, [4]*Dept. Life Sci. Med. Biosci., Waseda Univ.*)

The roles of mechanical force on the dynamics of the actin cytoskeleton are closely related to diverse cellular functions. Information regarding the effects of force applied to polymerizing/depolymerizing actin filaments is important to uncover the mechanical properties supporting cell dynamics such as deformation, motility, and division. One class of prospective key players involved in actin dynamics are formin family proteins (formins), which are the barbed-end actin-binding proteins, and regulate actin polymerization dynamics. Here, using a single-molecule strategy with optical tweezers, we demonstrate that formin mDia1-mediated actin polymerization has the potential to be accelerated in an "all-or-none fashion" by stretching tension.

1Pos148 Actomyosin contraction with a contractile ring related cross-linker in an *in vitro* active gel model system

Kyohei Matsuda, Takuya Kobayashi, Mitsuhiro Sugawa, Yoko Y. Toyoshima, Junichiro Yajima (*Department of Life Sciences, Graduate School of Arts & Sciences, The University of Tokyo*)

A single cell is divided into two daughter cells by shrinking a contractile ring in cytokinesis. This ring structure is constructed by actin cytoskeleton, myosin II motor protein and contractile ring related cross-linkers (CRC). However functional correlation of these protein elements is not yet understood. Here we use an *in vitro* active gel model system to show that myosin II and CRC actively reorganize actin filaments into a variety of mesoscopic patterns. One solution containing G-actin and myosin II show that actomyosin form the rigid network as ATP decrease. The solution in addition of CRC show that the three components make different networks and start contracting. These results suggest that the CRC plays important roles of structural formation and contraction.

1Pos149 混み合い環境でのアクチン線維の集団運動による秩序構造の出現

Appearance of ordered structure by collective motion of actin filaments in crowded environments

Takahiro Iwase, Yasuhiko Sasaki, Kuniyuki Hatori (*Dept. Bio-Systems Eng., Yamagata Univ.*)

Recently collective motion of actin filaments interacting with myosin has been reported. Here we show ordered structures of actin filaments under viscous medium; methylcellulose. Actin filament at 5 μM was added to a heavy meromyosin-fixed glass slide. Subsequently, fluorescently-labeled actin filaments and ATP solution containing 1% methylcellulose were added. We observed various ordered structures formed with actin filaments, which was dependent on KCl concentration. At 25 mM KCl, actin filaments repeated the bundle formation and their collapse. Above 150 mM KCl, a stripe pattern of the bundle array appeared. As KCl concentration was increased, the gap intervals of the array narrowed. Crowded environments may induce certain ordered patterns in active matters.

1Pos150 Self-organizations of actin filament networks in confined spaces: A simulation study

Takahiro Nitta (*Gifu Univ.*)

Actin filaments take a variety of forms of networks, such as cortex, filopodium and contraction ring. The networks play essential roles in cell morphology and motility. The networks self-organized under the influence of associated proteins and surrounded boundaries, such as cell membranes. However, complication in cellular environments has hampered quantitative and systematic investigations on what are essential ingredients for the self-organizations of interests. Here, by using computer simulations, we investigated the self-organizations of actin filament networks in confined spaces. We found that various actin networks formed depending on associate proteins and the size of the confinements.

1Pos151 蛍光顕微鏡および高速 AFM によるミオシンと F-アクチン間の協同的結合の経時的観察

Real-time observation of cooperative binding between myosin and F-actin by fluorescence microscopy and high-speed atomic force microscopy

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Cooperative binding of myosin to F-actin is attributed to cooperative conformational changes of actin subunits induced by binding of myosin head to neighboring actin subunits. The purpose of this study is to analyze the propagation of the cooperative conformation change evoked by HMM binding. Here, we continuously observed the binding of HMM to F-actin loosely immobilized on lipid membrane using fluorescence microscopy (FM) and high-speed AFM (HS-AFM). In FM observation, small HMM clusters were observed immediately after the addition of HMM-GFP, and the clusters gradually spreaded along the filaments. In HS-AFM observation, we found that a HMM molecule attached to F-actin and moved along the filament toward a neighboring cluster, and the cluster consequently grew.

1Pos152* 枯渇力が誘起する微小管集団運動に関する研究

Study of the collective motion of microtubules induced by depletion force

Ai Saito[1], Ryuhei Suzuki[1], Tamanna Ishrat Fahana[1], Arif Md. Rashedul Kabir[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*)

Collective motion is a fascinating display of coordinated behavior of self-propelled objects such as animals, birds, fishes, etc. Being inspired by the collective motion of the living organisms swarm robotics has emerged as an active research field. The biomolecular motor system microtubule-kinesin is an example of self-propelled object using which recently we have demonstrated collective motion in vitro. In this work, we perform detail investigation on the effect of various parameters such as microtubule density, depletion force, etc. on the collective motion of the microtubules. This work would help understand the coordinated behavior of living organisms observed in nature and at the same time might be beneficial for the swarm robotics.

1Pos153 神経細胞における細胞骨格アクチンの修復の分子メカニズム

Molecular mechanism of cytoskeletal Actin repairing in nerve cells

Tomboy Higo[1], Ayumi Ishihara[2], Shinji Aramaki[1], Yoshiko Itou[2], Takuo Yasunaga[1] ([1]*Kyushu Institute of Technology*, [2]*Leica Microsystems*)

It has been thought that when a nerve cell is damaged one, it can never be repaired. However, the recent neuroscience research reported that broken nerve system also has the potential of recovery. Also, recent electron microscopy (EM) can elucidate the molecular architecture of the cell under the near physiological conditions. Thus, we observed cytoskeleton recovery process in filopodia and lamellipodia. After inhibition of polymerization of actin filaments by cytochalasin D, which is corresponding to damage states, we observed the nerve cells in the recovery process with a confocal laser and a cryo-light microscope. Then, We observed them by cryo-electron microscopy and correlated both images. Thus, we try to unveil molecular mechanism of cytoskeleton rescuing.

1Pos154 超解像光学顕微鏡で観察した収縮環の計測と解析

Measurement and analysis of contractile ring observed with STED and Sim

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Super-resolution microscope opened a new window to observe biological events of 50-100 nm level. We, therefore, observed cytokinesis of cultured cell with SIM and STED. Many authors reported the presence of filamentous actin and myosin II in the contractile ring but it still remains unknown how the actin filament and myosin II are arranged to generate force for division.

Detail of the contractile ring was observed with SIM and STED. We mesured angle and length of each bipolar myosin molecules and count the number of the bipolar myosins. We also observed relationship between bipolar myosin and filamentous actin. Dynamics of fine structure of contractile ring will be discussed in relation to the actin asociate proteins.

1Pos155 アクチンフィラメントの協同的構造変化を阻害するアクチン変異の遺伝子内サプレッサー解析

Intragenic suppressor analysis of actin mutation that impairs cooperative conformational change of actin filament

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We recently showed that G146V mutant actin, which dominantly impairs cooperative conformational changes of actin filaments, is non-functional in budding yeast, supporting the idea that cooperative conformational changes are essential for actin functions. To further gain insight into this issue, we performed intragenic suppressor screening against G146V actin, and found yeast cells expressing G146V/V152A double mutant actin are viable. Yeast cells expressing V152A actin grew slowly, were rounder and larger, with fewer, misoriented actin cables. These phenotypes were rescued by the double mutation. We suggest that both mutations each impairs interaction with actin binding proteins that form actin cables (e.g., fimbrin and tropomyosin) in a mutually suppressible manner.

1Pos156 クライオ電子線トモグラフィ法で明らかにした，フィロポディア内におけるファシンによるアクチンフィラメント束化メカニズム

F-actin bundling mechanisms by fascin in filopodia was revealed by cryo-ET

Shinji Aramaki[1], Kouta Mayanagi[2], Kazuhiro Aoyama[3,4], Takuo Yasunaga[1] ([1]*Department of Bioscience and Bioinformatics*, [2]*Medical Institute of Bioregulation, Kyushu University*, [3]*FEI Japan*, [4]*Research Centre for Ultra-High Voltage Electron Microscopy, Osaka University*)

Filopodia are membrane protrusion structure which are involved in many crucial cell functions. Despite of importance of them, molecular mechanisms have remained elusive. In this study, we revealed intracellular structure of filopodia with cryo-ET and an averaging technique. In filopodia, f-actin were bundled by fascin and they had highly ordered architecture. In addition, structural variation of f-actin was smaller than in vitro. Finally, our 3D map, which crosslinking structure of f-actin and fascin revealed that fascin bind to f-actin with different types of binding manner. The bundling mechanisms must be implicated in function of filopodia. We are also trying to be introduced a CLEM technique to expand the ability of whole cell structural analysis with cryo-ET.

1Pos157　MEA システムを用いたニワトリ胚由来心臓組織片の薬剤応答

Drug response of embryonic chick heart tissue pieces using multi electrode array system

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Cardiotoxicity testing is the most important in drug development because there is a lethal cause such as arrhythmia. Therefore, multi electrode array (MEA) system is currently expected as a new tool to measure extracellular potential with non-invasively and long term, additionally indicating the index of causing arrhythmia. We put the heart tissue pieces isolated from chick embryos on the MEA system, followed by recording the response before and after drug administration using MEA system. We analyzed the inter-spike interval and field potential duration from the field potential recording. By using cardiac tissues, it would be expected to the development of accurate cardiotoxicity measurement system.

1Pos158　心筋細胞の集合体に対する機械的刺激の影響

Influence of mechanical stimulus on embryonic chick heart cell aggregates

Shin Arai, Ayaha Tsuyuki, Takahiro Uehara, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

Numerous studies have indicated cardiac mechano-electric coupling in order to regulate the heartbeat and revealed an important role of stretch activated channels. To investigate the coupling between cardiomyocytes, we have made PZT driven probes to apply mechanical stimulus on the cardiac cell aggregates. Our approach is providing mechanical tapping to cell aggregates by tungsten probes and investigating the response, especially how the local mechanical stimulus affects the intrinsic spontaneous beat activity, especially inter-beat intervals (IBI) by observing the beat motion and the fluorescence images with calcium-sensitive dyes. We will present the results of the IBI behavior under the probe tapping and discuss the IBI vs. the period of the external stimulus.

1Pos159　フィードバックマイクロレオロジーによる細胞骨格の非線形力学挙動計測

Nonlinear mechanical properties of Cytoskeletons measured with Dual-Feedback Microrheology

Natsuki Honda, Kenji Nishizawa, Takayuki Ariga, Daisuke Mizuno (*Kyushu University, Department of Physics*)

Mechanics of cells are largely governed by cytoskeletons. Since cytoskeletons supply scaffolds for motor proteins, motor-generated forces directly act to localized portion of cytoskeletal filaments. Here we investigate non-linear response of cytoskeletons to forces locally applied in a scale similar to occur in living cells, by developing a novel optical-trap-based microrheology implemented with dual-feedback control. We observed that thermal fluctuations of probes embedded in various cytoskeletons were reduced significantly when subjected to localized forces. Although such micro-scale response of cytoskeletons must be non-affine at low frequencies, our high-bandwidth experiments enabled to investigate the stiffening behavior in combination with numerical simulations.

1Pos160　ケラトサイトの運動方向を決定する２つのメカノセンシング機構

Keratocytes have hybrid mechanosensing system to decide their migration direction

Chika Okimura, Yoshiaki Iwadate (*Fac. Sci., Yamaguchi Univ.*)

Crawling cells can generate polarity for migration in response to forces applied from the substratum. Such reaction varies according to cell type, fast- and slow-crawling cells. Fish epidermal keratocytes are a type of fast-crawling cell. However, they have stress fibers in the cell body, a typical slow-crawling cell structure. Under periodic stretching of the elastic substratum, intact keratocytes rearrange their stress fibers perpendicular to the direction of stretching in the same way as slow-crawling cells and migrate parallel to the stretching direction, while stress fiber-less keratocytes migrate perpendicular to it, in the same way as fast-crawling cells. Keratocytes have a hybrid mechanosensing system to decide their migration direction.

1Pos161*　非熱的な力に駆動された細胞内部の混み合い状態

Intracellular crowding mechanics driven by athermal force

Kenji Nishizawa, Daisuke Mizuno (*Grad. Sch. Sci., Kyushu Univ.*)

Mechanics of cells plays crucial roles in many biological processes. The effect of “crowding” in cells are not fully investigated. We prepared several models of cytoplasm which lacks cytoskeletons. Viscosity of cell extracts rapidly increased as the protein concentration becomes higher. The critical concentration where viscosity diverges was found to be ~0.3 g/ml which is physiological concentration in living cells. Furthermore, viscoelastic properties in the cultured cells were measured. The volume fraction of intracellular macromolecules was changed (0.3 ~ 0.6 g/ml) by adding sucrose to the culture media and glass like behavior in living cells were measured. We speculate the effect of the athermal drive on the out-of-equilibrium mechanics in crowding materials.

1Pos162　心筋細胞の集合体群に与えるマルチプローブ機械的刺激の影響

Effect of Multi-probe stimuli on cardiac cell aggregates with spontaneous beat

Ayaha Tsuyuki, Shin Arai, Takahiro Uehara, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

In the heart, the beat motion and its propagation are anisotropic in order to optimize the pump activity. In contrast, cardiac cell aggregates on a dish are presumably isotropic and gradually generate spontaneous beats followed by their synchronization in 24h of incubation. Previously, we have applied a mechanical stimulus by a probe on a dish and analyzed the effects on the intrinsic beats and their synchronization. The stimulus enhanced or varied the beat activities. Furthermore, we have fabricated a multi-probe stimulus system for a wide variety of stimuli, such as push/stretch and directional anisotropy. We will investigate the effects of these stimuli on cardiac aggregates and explore the origin of the anisotropic beat propagations and their synchronization.

1Pos163 魚類表皮細胞ケラトサイトのかたち・サイズと牽引力
Relationship between traction forces, and shape and size of keratocytes

Ayane Sonoda, Chika Okimura, **Yoshiaki Iwadate** (*Fac. Sci., Yamaguchi Univ.*)

Fish epidermal keratocytes maintain an overall fan shape during their crawling migration. Migrating keratocytes can be divided into fragments by treatment with the protein kinase inhibitor staurosporine. Fragments containing a nucleus and cytoplasm behave as mini-keratocytes and maintain the same fan shape as the original cells. The relationship between shape and traction forces has not been clarified. The distributions of traction forces exerted by mini-keratocytes and keratocytes were similar. The magnitude of the traction forces was proportional to the area of the keratocytes and mini-keratocytes, suggesting that keratocyte shape depends on the distribution of the traction forces, and that the magnitude of the traction forces depends on the area of the cells.

1Pos164 アメーバ運動する線虫精子の牽引力測定
Measurement of Traction Force generated by Amoeboid Sperm of C. elegans

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C. elegams sperm, which use a unique cytoskeleton, called MSP (Major Sperm Protein) instead of actin, crawl over the substrate in a similar fashion to other amoeboid cells. To analyze its mechanical properties, traction forces of C. elegans sperm was measured by using a thin layer of elastic sheet as the substrate. Deformation of elastic substrate by sperm allowed detail analysis of traction forces, showing that traction forces distributed in two distinctive zones; one at the front and the other at the back part of the cell. Both traction forces pointed toward the center of the cell, which well agrees with our model that C. elegans sperm is driven by two types of forces, one at the front to pull the leading part forward and the other to shrink the back part of the cell.

1Pos165 細胞周期進行に伴う細胞内部環境のダイナミクス
The dynamics of Intracellular Environments during Cell-cycle progression

Katsuhiro Umeda, Kenji Nishizawa, Daisuke Mizuno (*Grad. Sci., Univ. Kyushu*)

Cell physiology is cell-cycle dependent in general, and so may be the mechanics. So far, however, measuring the intracellular mechanics by itself has been challenging. Here, we succeeded in measuring the cell-cycle dependent mechanics of HeLa cells (HeLa.S-Fucci) utilizing the dual-feedback microrheology. Cytoplasm in a confluent epithelial-like sheet shows glassy behavior; its viscoelasticity is drastically affected by a slight change of macromolecule concentration and/or metabolic activity. We found that elastic plateau is greater and high-frequency power-law is smaller in S-phase compared to G1. This result seems reasonable since both protein synthesis and metabolism in cell body may be less active in S-phase since cells focus to duplicate DNA in nucleus.

1Pos166* Actin Cytoskeleton Remodeling Dynamics of Adherent Cells Under Mechanical Strain of Gelatin Substrate

Kwokhoi Ng[1], Kentaro Iketaki[1], Ryuzo Kawamura[1], Seiichiro Nakabayashi[1], Yosuke Yoneyama[3], Fumihiko Hakuno[3], Shin-Ichiro Takahashi[3], Fumiki Yanagawa[2], Toshiyuki Takagi[2], Shinji Sugiura[2], Toshiyuki Kanamori[2], Hiroshi Yoshikawa[1] ([1]*Dept. Chem.,Saitama Univ.*, [2]*BRD.,AIST.*, [3]*GASLS., The Univ. of Tokyo*)

Recent studies clearly revealed that network structure of cytoskeletons sensitively response to external mechanical stimuli. So far, such dynamic remodeling of cytoskeletons has mainly been studied by uni- or bi-axial deformation of rubber-based culture substrates with external actuators. Alternatively, we recently established a new class of gelatin substrate which is capable of applying anisotropic and multidirectional mechanical strain to cells without the necessity of external actuators. In this work, we carried out time-lapse confocal imaging of LifeAct-mRFP visualized actin cytoskeleton in myoblasts cultured on the gelatin substrate to clarify actin cytoskeleton remodeling dynamics with respect to various mechanical strains.

1Pos167 血管壁内力学環境を考慮したコラーゲン微細溝基質による血管平滑筋細胞の分化制御
Control of vascular smooth muscle cell differentiation using a novel micro-grooved collagen substrate

Kazuaki Nagayama, Keiichi Uchida, Saki Takeuchi (*Micro-Nano Biomechanics Laboratory, Department of Intelligent Systems Engineering, Ibaraki University*)

Vascular smooth muscle cells (SMCs) change their phenotype from contractile to synthetic under pathological conditions. To understand the smooth muscle pathophysiology, such as arteriosclerosis, it is important to understand the mechanism of smooth muscle cell differentiation. The mechanical environments around SMCs in vivo are quite different from those of the cultured cells: they show elongated shape and form a tissue aligned in the circumferential direction of the arterial walls. This micro-scaled mechanical environment might affect cell differentiation processes. Thus in this study, we developed a novel micro-grooved collagen substrate to induce cell elongation and alignment like in vivo, and investigated their effects on smooth muscle differentiation processes.

1Pos168 力学的強度の制御を可能とする光架橋性コラーゲンゲルの開発
Development of photo-cross-linked collagen gels with tunable mechanical property

Takahiro Fujisawa[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*IMCE, Kyushu Univ.*)

In general, cells sense mechanical property of extracellular milieu and alter their functions depending on the level of the matrix elasticity. To investigate the mechanobiologic behaviors of cells, use of the elasticity-tunable hydrogels for cell culture is essential. In this research field, One of the most widely-used gels for this purpose is collagen-coated poly(acrylamide) gels. However, the gels do not necessarily provide rigorous tuning of elasticity of collagen matrix itself. In this study, we tried to construct the elasticity-tunable collagen gels keeping the native triple helix. We introduced photo-crosslinking styrene group to collagen (StC) while keeping triple helix, demonstrated photo-induced gelation of the StC, and measured elasticity of the StC gel.

1Pos169 組織切片の伸展性応答: ひび割れパターンと病態

Response of Tissue Slice to Mechanical Stretching: Characteristic Cracking Pattern Reflecting Disease State

Keisuke Danno[1], Takuto Nakamura[1], Naohiko Nakamura[2], Kota Iguchi[2], Masaya Ikegawa[3], Kenichi Yoshikawa[3] ([1]*Doshisha Univ.*, [2]*Kyoto Univ.*, [3]*Doshisha Univ.*)

Pathological diagnosis is usually performed based on morphological observation on tissue section by use of optical microscopy. Here, we will report a new methodology to evaluate the disease state through the application of mechanical stress on tissue slice. It will be shown that characteristic cracking pattern is generated in accordance with the development of the disease. Through the quantitative analysis of the cracking pattern, it becomes possible to make the diagnosis of the disease state in a quantitative manner. Possible further extension of such novel method as the diagnosis tool will be discussed.

1Pos170 タリンとビンキュリンによる力と硬さの感知

Force- and rigidity-sensing by talin and vinculin

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Talin and vinculin are major adaptor proteins at focal adhesions (FAs), supra-molecular complexes that mediate cell adhesion to the extracellular matrix (ECM). In vitro studies have shown that talin works as a potential mechanosensor; mechanical extension of talin exposes cryptic binding sites for vinculin. Here, we show that vinculin binding to talin is indeed force-dependent at FAs in living cells. In addition, we reveal that the talin-vinculin binding is required for anchoring the actin cytoskeleton to FAs, which ensures advancement of leading edges of cells. Using mechanical modeling, we further show that the rigidity of ECM also influences the vinculin binding to talin, suggesting that talin and vinculin play a key role in sensing the ECM rigidity.

1Pos171 マイクロメートルスケールの足場構造に依存した細胞性粘菌の細胞遊走

Migration of Dictyostelium cells on micro-scale ridge structures

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Recent studies are uncovering migratory behaviors of various cell types induced by fabricated structures on a cell-substrate whose mechanisms and functions remain largely unexplored. Here, we show that in Dictyostelium, there is a high tendency for the cells to migrate along micrometer-size ridges. At the protruding ends of the cells was enrichment of F-actin that propagated on top of the ridges. Upon application of an attractant gradient, in more than a 1/3 of the cells, the characteristic F-actin patterns disappeared and the cells moved towards the direction of higher attractant concentration. These observations suggest that chemotaxis can override topography-induced migration, and because they sometimes coexist, the two processes can operate independently.

1Pos172* 膜タンパク質による細胞間相互作用の定量的解析

The quantitative analysis of the intercellular interaction by membrane proteins

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The cell-cell interaction by membrane proteins are concerned with vital phenomena like growth and immunology. However, in general, conventional biochemical methods measure protein interactions alone, without considering the effect of native membrane structure on the binding reactions. Here, we have tested the effectiveness of flow cytometry as a measurement of the intercellular binding by membrane proteins. Flow cytometry was able to measure the specific binding of membrane proteins in living cell, and successfully defined the apparent binding constants. This method detects the cell-cell interaction closer to in vivo than previous one, leading to contribute to the elucidation of biological mechanisms and the development of the immune cell therapy.

1Pos173 重力下での形態形成・維持に対するアクトミオ シンネットワークの寄与

Teoretical study of contribution of YAP-dependent actomysin network to morphogenesis under gravity

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Multicellular organisms form their specific body shapes against the gravity. Recently, YAP was reported as a key transcriptional regulator to keep their body shape under gravity, where the knockout mutant of YAP of medaka was observed to form the flattened body or misalignment of the organ, and YAP knockout spheroid formed cell group cultured in human also exhibited the flattened shape. Then, we construct a model of dynamics of YAP-dependent actomyosin network and interactions among cells to consider the mechanism of the abovementioned phenomena. We particularly focus on the roles of the interplays among intra- and inter-cellular molecular dynamics and the translation and deformation dynamics of cells for their body shape regulations.

1Pos174 Divergence of structural strategies for E-cadherin homophilic binding among bilaterians

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Homophilic binding of E-cadherins through their ectodomains is fundamental to epithelial cell-cell adhesion. Of the five rod-like, tandemly aligned extracellular cadherin domains (ECs) of vertebrate E-cadherin, the tip EC plays a pivotal role in binding interactions. Comparatively, the N-terminal six consecutive ECs of Drosophila E-cadherin, DE-cadherin, can mediate adhesion; however, the underlying mechanism is unknown. Here we report atomic force microscopy imaging of DE-cadherin ECs. We identified a tightly folded globular structure formed by the four N-terminal-most ECs. Analysis of hybrid cadherins of different hexapods indicated association of the E-cadherin globular portion with the determinants of homophilic binding specificity.

1Pos175* **DNA hybridization を介した細胞-細胞間接着ダイナミクスの解明**

Dynamics of cell-cell adhesion via DNA hybridization

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In recent years, the use of specific molecular recognition via synthetic DNA or biotin/streptavidin has shown promising for the spatial regulation of cell-cell attachment between same and/or different cell lines, which can potentially be applied to fabrication of complex and heterogeneous 3-dimensional tissue. However, how such functional molecules showing specific molecular recognition influence native cellular interactions (e.g., via cadherin) remains still unclear. In this study, to clarify the impact of DNA hybridization on cell-cell adhesion, we have developed cell-cell adhesion models by using a cell and a planar lipid bilayer of which surfaces were functionalized with DNA-PEG-lipids and/or E-cadherin.

1Pos176 **膵島 α 細胞の分泌顆粒動態に及ぼす接着分子 CADM1 の影響**

Effect of cell adhesion molecule 1 expression on intracellular granule movement in pancreatic α cells

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CADM1, identified as an adhesion molecule in pancreatic α cells, has been reported not only to communicate among α cells and between nerve fibers, but also to prevent excessive glucagon secretion from α cells. Here, we investigated the effect of CADM1 expression on the movement of intracellular secretory granules in α cells. Spinning disk microscopic analysis showed that a mean velocity of secretory granules was significantly decreased in CADM1-knockdown αTC6 cells compared to wild type cells. The velocity of granule movement decreased greatly in αTC6 cells treated with nocodazole but not cytochalasin D. These results suggest that intracellular granules in α cells move along the microtubule network, and that CADM1 influences their velocity.

1Pos177 **分散培養 iPS 細胞の増殖応答性に対するハイドロゲル表面へのラミニン修飾状態の本質的効果**

Essential role of mannter of laminin-modification for hydrogel surface on the prolifelation activity of dissociated iPS cells

Kenta Mizumoto[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Univ. Kyushu*, [2]*IMCE., Univ. Kyushu*)

Feeder-free dissociating culture of iPS cells has been required for stable and easier supply of them for further clinical applications. One of the problems with the dissociating culture is that iPS cells undergo apoptosis not only due to the loss of cell-cell adhesion but also depending on the quality and strength of cell-substrate adhesion. To clarify the optimized physicochemical conditions for the dissociating culture of iPS cells, we have scrutinized the effect of manner of laminin-modification for hydrogel surface on proliferation activity of iPS cells: chemical immobilization with or without spacers and physical adsorption. We discuss how the mobility and density of laminins on hydrogel contribute to survival and proliferation of dissociated iPS cells.

1Pos178 **細胞内ナノ粒子導入・細胞間相互作用制御のための材料工学**

Materials engineering approaches to modulate cell membrane structures for nanoparticles delivery and regulation of cell-cell interactions

Yoshihisa Kaizuka, Tomoto Ura, Hidenobu Nakao (*NIMS*)

Modulations of cellular functions with metal, inorganic, or polymer materials provide means of biophysical characterizations. First, we developed techniques for internalizing metal and inorganic nanoparticles in cytoplasm of live cells through pressurized plasma membranes. These technologies may be useful for molecular delivery and bio-optical applications. Second, we characterized macromolecules that interact with plasma membranes. We found that the incorporation of those molecules could alter macroscopic mechanical properties of plasma membranes. Such mechanochemical modulation of cell membranes may be used for various biophysical analyses, including characterizations of cell-cell interactions.

1Pos179* **脂質膜に覆われた細胞サイズ生体高分子ゲルの弾性率**

Elasticity of biopolymer gel in cell-sized droplet covered with a lipid membrane

Atsushi Sakai, Yoshihiro Murayama, Miho Yanagisawa (*Tokyo university of Agriculture and Technology*)

Elasticity of cytoplasmic gel is essentially important for shape regulation, cell motility, etc. The gel phase localizes underneath biomembrane, and strongly interacts with the biomembrane. We investigate the interaction between the biomembrane and elasticity of the cytoplasmic gel by using biopolymer microdroplets covered with a lipid layer as a cell model. We found the membrane contact of the biopolymer gel increases the elasticity. In addition, the gel elasticity becomes higher with a decrease in gel size. These results suggest that cytoplasmic gel and biomembrane jointly regulate elastic property of the gel.

1Pos180 **原始真核生物の細胞内小器官の 3D 構造モデルから得られる生物の新たな情報**

New obvious information obtained from cell organelle 3D-structural models of primitive eukaryote

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EM offers exceptional resolution of extremely small biological specimens, providing images for fundamental cellular phenomena. Especially, sequential 2D-EM images and 3D reconstruction will provide us several information not only to the 3D-structural shape and connection. We selected *C. merolae*, a primitive eukaryote, as a model organism for mitosis. Although the size of the cell is 2-5 micron, one cell is surely to divide into two daughter cells. In this meeting, in addition to analyze the interaction between individual several organelles during mitosis cycle, we reveal individual morphology details and compute the volume occupancy of each organelle. We will discuss you a new view about the volume correlative relationship between cell and its organelles.

1Pos181 Accumulation of cargo proteins can physically trigger vesiculation in membrane trafficking system

Masashi Tachikawa (*RIKEN*)

Membrane trafficking is a process to transport cargo proteins from endoplasmic reticulum to various destinations via deformations of endo-membranes. Three protein complexes are known to catalyze this membrane deformation: COP I, COP II and clathrin. How do these protein complexes evolve independently? Here we propose a novel interpretation for this problem. Cargo accumulation itself catalyze deformation of surrounding membrane as physical process. To test this hypothesis, we performed coarse-grained physical simulations of membrane and cargo particles. We found that cargo particles inside membrane tend to assemble and the increase of them form tubular structures at the surrounding membrane.

1Pos182 多突起型チューブリン封入リポソームの形態形成メカニズム
Morphogenetic mechanism of tubulin-encapsulating giant liposomes with a hundred of membrane spines

Masahito Hayashi, Kingo Takiguchi (*Grad. Sch. Sci., Nagoya Univ.*)

We have demonstrated that tubulin-encapsulating giant liposomes (TEGLs) show reversible elongation of their bipolar membrane spines as a result of polymerization and depolymerization of inside microtubules caused by change of pressure or temperature. Here, we found poly-spine TEGLs, which had a hundred of membrane spines, when microtubules elongated after hypertonic treatment. The spines had nearly the same length and separated each other by nearly the uniform distance. Microtubules at the center of a TEGL formed mesh-like structure. Several microtubules growing from the mesh converged on and supported each membrane spine from inside. These results suggest that microtubules on separated spines interact dynamically through the membrane tension on a liposome.

1Pos183 アクトミオシンネットワークの収縮による細胞サイズ液滴の運動
Directed motion of cell-sized droplets driven by actomyosin network contraction

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Actin cortex is a two-dimensional actomyosin network assembled at the inner surface of the cell. Rupture of actin cortex by myosin induces bleb formation, and cells migrate by blebbing. Although several models of cell migration have been proposed, living cells are too complex to quantify the contributions of each protein such as actin, myosin and actin cross-linkers to cell migration. Here, we developed a simple system mimicking the cell by encapsulating purified proteins in water-in-oil droplets. We observed spontaneous formation of actin cortex beneath the droplet surface. Actomyosin contraction ruptured the cortex, which induced directed motion of the droplets. We will present how the contractility of actomyosin networks regulates the droplet motility.

1Pos184 高濃度アクチン繊維が引き起こす立体的集団運動
The collective motion and band pattern formations of sliding actin filaments driven by HMM

Yuuji Setoguchi, Hirotaka Taomori, Masayuki Hoshida, Yu Ichinose, Hajime Honda (*Dept. Bioeng., Nagaoka Univ. Tech.*)

High concentration of F-actin was known to indicate collective motion when driven by HMM on the glass surface. We have found that the motion includes bi-directional flow of filaments even apart from the HMM surface and that the filaments gradually assembled to form band patterns of about 80 micrometers in width and several millimeters in length. 1) Methyl cellulose was found to facilitated the collective motion but not band pattern formation. 2) Decreasing the width of assay-chamber less than 300 micrometers loss collective motion. 3) Movements of filaments were affected by indented pattern of the surface in a concentration dependent manner. These results indicate that this phenomenon might be caused not only by hydrodynamic but also inter-filamentous interactions.

1Pos185 細胞に優しい三次元組織体の構築：レーザートラップと高分子の混雑効果の活用
Constructing stable cellular assembly in the absence of artificial scaffold by use of laser tweezers

Shoto Tsuji, Aoi Yoshida, Taeko Ohta, Hiroaki Taniguchi, Kenichi Yoshikawa (*Doshisha University*)

Construction of stable cellular assembly of desired arrangement is one of the most important targets in modern cellular biology and also in regenerative medicine. Here, we will report a noninvasive method to create stable 3D cellular assembly without using any artificial scaffold. The essence of our method is to utilize laser tweezers to assemble desired targeted cells in an aqueous cell culture in the presence of solvable polymer, such as dextran and PEG. It is shown that stable cell-cell contact is generated spontaneously and such contact persisting even when the assembly is transported to the medium free from the polymer.

1Pos186 ヒト疾患診断マーカーとして有用なエキソソームの生物物理解析に適した新規分離調製法の開発
A novel isolation and preparation method for the biophysical analyses of useful exosomes as diagnostic markers for human diseases

Noriyuki Ishii, Mitsushi J. Ikemoto, Takayuki Odahara (*Biomedical Research Institute, National Institute of Advanced Industrial Science and Technology (AIST)*)

Exosomes are extracellular vesicles secreted from biological cells. Those from diseased cells contain disease-specific nucleic acids and proteins. Therefore, they have attracted great interest as high sensitive diagnostic biomarkers at early stages of human diseases. However, their physiological and biophysical features remain largely unknown. In order to clarify the molecular mechanisms underlying exosome functions, we have developed a novel isolation method suitable for biophysical analyses that can resolve physicobiochemical features of exosomes by electron microscopy and particle counting techniques. We demonstrate the usefulness of our isolation method in conjunction with electron microscopic analyses of exosomes from human embryonic kidney cells.

1Pos188* アクチンの示すミクロ顆粒系での局在性転移：細胞の混雑環境モデリング

Selective Localization of Actin in Micro-Domains under Molecular Crowding: Difference among Monomeric, Linear-Polymeric and Bundling State

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Living cells maintain their lives by utilizing micro-compartments entrapping various macromolecules under crowding condition. Currently, such micro-compartmentalization has often been interpreted in relation to phospholipid bilayer membrane. On the contrary, a few studies have argued that the origin of compartmented living cells is attributable to the spontaneous micro-segregation in a crowding environment with macromolecules.
Here, we examined the crowding binary polymer solution as the possible origin on the micro compartmentalization of primitive life.
We found biomolecules, such as actin, are specifically entrapped within the inner portion of cell-sized spheres rich in bulky polymers under the condition of micro-segregation with binary polymer solution.

1Pos189 ジャイアントベシクルにおけるアミロイド繊維の形成

Amyloid fibril formation in giant vesicle

Tong Zhu[1], Kensuke Kurihara[1,2,3] ([1]*Okazaki Institute for Integrative Bioscience*, [2]*Institute for Molecular Science*, [3]*Research Center for Complex Systems Biology, The Univ. of Tokyo*)

To explore the boundary between living and non-living matters, the constructive biology approach is fascinating. By means of extracting the minimum factors of a certain cellular phenomenon into the cell-mimicking vesicles so that the universal principle of life could be inspected. In this study we focus on the actin filaments, which is one of the three cell skeleton components and allows the cells to adopt different shapes and perform different functions. Since the amyloid beta peptides tend to form fibril that is similar with actin filaments in terms of structures and sizes, here we attempted to use amyloid beta peptide as a model for studying the influence of the fibril formation on giant vesicles.

1Pos190* 人工細胞モデルを用いた生体膜融合機構の解明

Biophysical principle of membrane fusion revealed by artificial lipid vesicles

Yui Suzuki[1], Ken Nagai[1], Anatoly Zinchenko[2], Tsutomu Hamada[1] ([1]*JAIST*, [2]*Grad. Sch. of Environmental Studies, Nagoya Univ.*)

We have developed a novel system for the photocontrol of fusion of lipid vesicles through the use of a photosensitive surfactant containing an azobenzene moiety (AzoTAB). Real-time microscopic observations clarified a change in surface area and internal volume of vesicles during a fusion. We also determined optimal cholesterol concentrations and temperature to induce fusion. The mechanism of the fusion event is attributed to a change in membrane tension, which is caused by solubilization of lipids through isomerization of AzoTAB. We utilized a micropipette technique to measure membrane tension, and discussed the fusion mechanism in terms of membrane elastic energy with the experimentally obtained membrane tension.

1Pos191 *in vitro* 1分子イメージング解析により明らかになったPI(4,5)P2依存的なPTENの膜結合の促進・安定化

Phosphatidylinositol lipid PI(4,5)P2 enhances membrane binding of PTEN revealed by *in vitro* single-molecule imaging analysis

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PTEN, a 3-phosphatase of PI(3,4,5)P3, is a peripheral membrane protein undergoing the shuttling between cytoplasm and cell membrane. Posterior localization of PTEN on plasma membrane is an essential factor for cellular polarization. To elucidate the mechanisms of the interaction between PTEN and lipid molecules, we have developed *in vitro* assay system for single-molecule analysis on artificial lipid bilayers. By comparing PI(4,5)P2 and phosphatidylserine, as representatives of negative charge lipids, it was shown that increase of PI(4,5)P2 is more effective to recruit and stabilize PTEN on the membrane. These findings illustrate a positive feedback mechanism for PTEN membrane binding, in which its enzymatic product PI(4,5)P2 enhances the membrane localization of PTEN.

1Pos192 Measurements of mitochondrial motility in cell body

Hyunjin Choi, Yuki Sugimoto, Yoshihiro Ohta (*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*)

Mitochondrial motility is necessary for cellular activities. However, since the mitochondrial motility in the cell body has not been quantitatively analyzed, the regulation mechanism of mitochondrial motility has not been clarified in the cell body. The aim of this study is to quantitatively analyze the mitochondrial motility and to examine the effect of phosphatases on mitochondrial motility. Although mitochondrial was not stable, it significantly depended on the temperature and ATP concentration in cells. Further, the inhibitors of phosphatases affected mitochondrial motility. These results mitochondrial motility was regulated by phosphorylation and dephosphorylation. The details will be discussed.

1Pos193 人工脂質膜小胞内におけるPIP3/PTEN traveling waveの再構成

Reconstitution of traveling wave of PIP3/PTEN on membrane in GUVs

Hitomi Matsubara (*Lab. Single Molecule Biology, Grad. FBS., Osaka Univ.*)

Phosphatidylinositol (PtdIns) lipids signaling system generates an anterior-posterior polarity in migrating *Dictyostelium discoideum* cells. It forms two domains on the membrane where PtdIns 3,4,5-trisphosphate (PIP3) is enriched at the front and where phosphatase and tensin homolog (PTEN), a PtdIns 3-phosphatase, is enriched at the tail. These domains are mutually exclusive, and when cell migration is inhibited they oscillate spontaneously on the membrane forming traveling wave pattern. To understand how this pattern forms, I chose a method of reconstitution *in vitro* by using giant unilamellar vesicles (GUVs). I will discuss the production and degradeation of PIP3 by purified PtdIns 3-kinase and PTEN, respectively, successfully monitored with PHD-GFP in GUVs.

1Pos194 細胞透過ペプチド・オリゴアルギニンの単一ベシクルへの侵入に対する脂質組成の効果

Effects of lipid compositions on the entry of cell-penetrating peptide oligoarginine into single vesicles

Sabrina Sharmin[1], Md Zahidul Islam[1], Mohammad Abu Sayem Karal[1], Sayed Ul Alam Shibly[1], Hideo Dohra[2], Masahito Yamazaki[1,3,4] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Green Sci. Tech., Shizuoka University*, [3]*Res. Inst. Ele., Shizuoka Univ.*, [4]*Grad. Sch. Sci., Shizuoka Univ.*)

To elucidate the mechanism of the entry of R_9 into cells, we investigated the interactions of CF-R_9 with GUVs of various lipid compositions containing AF647 using the single GUV method for CPPs.[1] For DOPG/DOPC- and DLPG/DTPC (2/8)-GUVs, CF-R_9 entered the GUV lumen from the outside by translocating across the lipid bilayer without pore formation through which AF647 leaked. In contrast, high concentration of CF-R_9 induced pores in DLPG/DTPC (4/6)-GUVs, through which CF-R_9 entered the GUV lumen. In contrast, CF-R_9 could not enter DOPG/DOPC/cholesterol (2/6/4)-GUVs as a result of no translocation of CF-R_9 across their bilayers. We discuss the elementary processes of the entry of CF-R_9 into GUVs.[2]

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1Pos195 Functional significance of trimerization in Cl$^-$ pumping properties of halorhodopsin examined by nanodisc reconstitution

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Halorhodopsin (HR), a light-driven chloride pump, forms a homotrimer in the cytoplasmic membranes, which is essential for its chloride transport. Although the functional significance of trimerization in HR has been examined, previous studies used the detergent-solubilized systems, where non-native interactions might be formed between the protein and detergent. To clarify the functional significance of the trimarization under physiological conditions, we reconstituted trimeric and monomeric HR into the "nanodisc", and applied flash photolysis to analyze the Cl transport process. The life time of the intermediate of the photocycle in monomeric HR was found to be changed, suggesting that the trimerization regulates the stability of the intermediates in the Cl transport.

1Pos196 脂質膜の伸展の分光学的研究

Spectroscopic Investigation of Osmotic Pressure-Induced Membrane Stretching

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Osmotic pressure Π induces lateral tension σ in lipid bilayers of vesicles. Here, we investigated the effects of Π on physical properties of bilayers such as membrane fluidity. For this purpose, first we measured the GP value of Laurdan in DOPC-GUVs and -LUVs when they were transferred into a hypotonic solution with initial concentration difference ΔC^0. For both the GUVs and the LUVs, the GP value decreased monotonously with ΔC^0, indicating an increase in fluidity of their bilayers. Inducing the same decrease in GP values required 10 times higher ΔC^0 for the LUVs compared with the GUVs. After conversion of ΔC^0 to σ using a theory, we found that the decrease in GP values over σ was almost the same for both vesicles, supporting the theory on the radius dependence of σ.

1Pos197 細菌-膜小胞間相互作用の動態と情報伝達機構の解析

Dynamics and signal transduction of the interaction between bacteria and membrane vesicles

Yosuke Tashiro, Yusuke Hasegawa, Kotaro Takaki, Hiroyuki Futamata (*Dept. of Eng., Shizuoka Univ.*)

Membrane vesicles (MVs) are secreted from a range of microbial species and transfer their content to other cells. Although MVs play important roles in bacterial communication, little is understood whether MVs selectively interact with bacterial cells. Here we investigated the specificity of MV-cell interaction. MVs derived from an enterobacterium *Buttiauxella agrestis* specifically interact with cells of the same species. The specific interaction of *B. agrestis* MVs with bacterial cells was explained in terms of interaction energy based on DLVO theory. *B. agrestis* MVs enabled plasmid DNA in MVs to be transferred to the same species at high efficiency. These results suggest a novel characteristic that MVs selectively associated with bacterial cells.

1Pos198 マガイニン２が誘起する脂質膜中のポア形成に対する抗菌ペプチド・PGLa の効果

Effect of Antimicrobial Peptide PGLa on Magainin 2-Induced Pore Formation in Lipid Membranes

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Synergistic effects of antimicrobial action of PGLa and magainin 2 have been reported. To elucidate these effects, we investigated the interactions of PGLa or PGLa/magainin 2 mixture with single DOPG/DOPC-GUVs. Coexistence of low concentrations of PGLa increased the rate constant of magainin 2-induced pore formation in single GUVs. Interaction of the same concentrations of PGLa with the GUVs increased the fractional area change of the GUV membrane greatly. In the interaction of carboxyfluorescein-labeled PGLa (CF-PGLa) with single GUVs, CF-PGLa translocated from the outer to the inner monolayer before pore formation. On the basis of these results, we discuss the mechanism of the effect of PGLa on the magainin 2-induced pore formation.

1Pos199 大腸菌異物排出系トランスポーター MdtB, MdtC 会合の可視化

Assembly of the xenobiotic efflux transporters MdtB and MdtC of *Escherichia coli*

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Among five RND-type xenobiotic efflux systems of *Escherichia coli*, only the AcrAB-TolC complex is constitutively expressed. The MdtABC-TolC complex is unique in that it has two inner membrane transporters MdtBC whose genes are induced in response to indole. Here we visualized how the MdtB and MdtC transporters are assembled *in vivo* using GFP and TagRFP. When expressed alone, MdtB-GFP formed a homotrimer, whereas MdtC-TagRFP did not. When co-expressed, a heterotrimer consisting of two MdtB-GFP molecules and one MdtC-TagRFP molecule was formed predominantly. These results suggest that MdtB and MdtC favor to form the 2:1 heterotrimer, which has been proposed, by the study on tandem-linked trimers, to have the highest drug efflux activity among all possible trimers.

1Pos200 外部環境を感知する磁場駆動型リポソームの創製
Magnetically-driven moving liposomes that sense environmental information

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Cell-type robot based on liposome must have three important functions; sensing, thinking and moving. In this study, we firstly aimed to construct temperature-sensitive moving liposome driven by external magnetic field, by encapsulation of magnetic nanoparticles modified with thermosensitive polymer gel (Therma-Max) into them. The velocity of the liposome under a magnetic field gradient was increased in accordance with external temperature. Moreover, we developed liposome that sense external salt concentration by adding a pore-forming peptide (Magainin 2) into the external solution under a similar magnetic field gradient. These moving liposomes that moved by the combination of the external stimuli could be a novel prototype of the cell-type robots for molecular robotics.

1Pos201 ミトコンドリア密集による ATP 産生の効率化
Effective ATP generation by closely located mitochondria

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Mitochondria are the main organelles that produce ATP and reactive oxygen species in cells and are not uniformly distributed in cells. In this study, we aimed to examine the effect of close location of mitochondria on their activities. For this purpose, we isolate mitochondria from porcine heart and adsorbed them on dishes. ATP production rate significantly increased with the density of mitochondria adsorbed on a dish. Microscopic analysis of the adsorbed mitochondria showed that mitochondria located closely were more polarized than mitochondria located sparsely. The computer simulation of the generation of mitochondrial membrane potential supported that closely located mitochondria have a higher proton motive force. The details will be discussed.

1Pos202 高いトポロジー種数を持つベシクルの形態：核膜形状の形成
Morphology of high-genus vesicles under pore-size constraint: Construction of nuclear envelope shape

Hiroshi Noguchi (*ISSP, Univ. Tokyo*)

Nuclear pores have an approximately uniform distribution in the nuclear envelope of most living cells. Hence, the morphology of the nuclear envelope is a spherical stomatocyte with a high genus. We will present the morphology of high-genus vesicles under pore-size constraint by dynamically triangulated membrane simulations. Bending-energy minimization without volume or other constraints produces a circular-cage stomatocyte, where the pores are aligned in a circular line on an oblate bud. We have clarified the conditions for the formation of a spherical stomatocyte: a small perinuclear volume, osmotic pressure within nucleoplasm, and repulsion between the pores.

1Pos203 両親媒性 DNA オリガミによる W/O マイクロエマルションの光応答性観察
Photo-responsive water-in-oil microemulsion made of amphiphilic DNA origami

Misato Tsuchiya[1], Daisuke Ishikawa[1], Yuki Suzuki[2], Masayuki Endo[3], Masahiro Takinoue[1] ([1]*Dept. Comput. Sci., Tokyo Tech.*, [2]*Fronti. Res. Inst. Interdiscip. Sci., Tohoku Univ.*, [3]*WPI-iCeMS., Kyoto Univ.*)

Water-in-oil (W/O) microemulsion has been used in a variety of fields including cell-size chemical reactors and biomedical applications, and is expected as a highly-functional material that is responsive to stimuli such as light, temperature and chemicals. However, it is not easy to programmably change the functions of W/O microemulsions. In this study, we propose W/O microemulsion made of an amphiphilic DNA origami whose functions can be changed programably by DNA sequence design. Here, we aim the photoregulation of the stability of the microemulsion by introducing modified DNA that is responsive to ultraviolet irradiation. In this presentation, we report analysis results of the time variation of the microemulsion collapse.

1Pos204 合成高分子による膜曲率の認識
Recognition of membrane curvature by synthetic amphiphilic polymers

Naho Sunagawa[1], Manami Tsukamoto[1], Kenichi Kuroda[2], Jun-ichi Kikuchi[1], Kazuma Yasuhara[1] ([1]*Grad. Sch. Mat. Sci., Nara Inst. Sci. Tech.*, [2]*Sch. Dentistry Univ. Michigan*)

Generation of membrane curvature is essential for various cellular dynamics such as cell division, vesicle trafficking, and protein sorting. Some proteins are known to have a curvature-sensing motif, which recognizes packing defects in a lipid membrane. In this study, we have designed synthetic curvature-sensing polymers inspired by the amphiphilic structure of the curvature-sensing motif in the proteins. The curvature-sensing feature of the polymers was evaluated by microscopic observation using several model membrane systems. Through the structural tuning of the polymers, we have obtained a curvature-sensing polymer, which selectively bind to highly curved membrane with a smaller diameter than several micrometers over low-curvature membrane.

1Pos205 膜貫通タンパク質の細胞膜上二次元拡散における二段階緩和の理論解析
Theoretical analysis of a two-step relaxation on protein diffusion in the plasma membranes

Tomonari Sumi[1], Atsushi Okumoto[2], Hitoshi Goto[2], Hideo Sekino[2] ([1]*Res. Inst. Interdisciplinary Sci., Okayama Univ.*, [2]*Toyohashi Univ. Tech.*)

A two-step relaxation on translational diffusion of a transmembrane protein in the plasma membrane has been observed by single-molecule experiment. To explain this anomalous diffusion, a nested double compartment model consisted of small and large compartments has been proposed. It has also been pointed out that each compartment is delimited by the actin filament “fence” and transmembrane protein “pickets” associated with the fences. In this study, we apply a diffusion equation approach to investigate the diffusion dynamics of a protein in a model membrane and demonstrate that the two-step relaxation can be reproduced without the nested double compartments using only the fence and pickets model combined with the lowering in a local diffusion around the pickets.

1Pos206 **肺サーファクタントタンパク質 B の N 末端による脂質単分子膜の構造変化**
Morphology changes in lipid monolayers induced by the N-terminal segments of surfactant protein B

Hideyuki Nagatsuka, Masahiro Hibino (*Div. Sust. Enviro. Eng., Muroran Inst. Tech.*)

The lung surfactant is a complex lipid-protein film and prevents the alveoli from collapsing during exhalation. The surfactant protein B (SP-B) helps spread the surfactant across the surface of the alveoli. However, the function of SP-B in the monolayer or replacement surfactants is not well understood. Here, we describe that interactions between lipids and SP-B peptides in the model lung surfactant create localized monolayer-to-multilayer transitions that provide low surface tensions on compression and rapid and repeatable respreading on expansion. The SP-B peptides used for the studies are N-terminal segments of full-length SP-$B_{1\text{-}78}$, namely, SP-$B_{1\text{-}25}$, SP-$B_{6\text{-}25}$, SP-$B_{10\text{-}20}$ and fluorescenin-labeled SP-$B_{1\text{-}25}$. The details of the results will be discussed.

1Pos207 **シトクロム P450 基質薬剤クロルゾキサゾンとホスファチジルエタノールアミン・モデル膜との相互作用**
Interaction between cytochrome P450 substrate drug chlorzoxazone and phosphatidylethanolamine model membranes

Hiroshi Takahashi (*Grad. Sch. Sci. Tech., Gunma Univ.*)

To clarify the roles of membrane lipids in cytochrome P450 (CYP)-drug metabolism process, we have been studying the interaction between chlorzoxazone (CZX) and model biomembranes. In the meetings of recent two years, we reported the inhibition effect of cholesterol on the binding of CZX to phosphatidylcholine (PC) bilayers. In this study, we have investigated on the interaction between CZX and 1-palmitoyl-2-oleoyl-phosphatidylethanolamine (POPE) model membranes by using X-ray diffraction and DSC measurements. PE is the second major phospholipid in biomembranes. Addition of CZX reduced the bilayer-to-inverted hexagonal phase transition temperature and induced the coexistence of the gel and liquid-crystalline phases.

1Pos208 **脂質キュービック相間の相転移における方位関係**
Orientational Relationships In Transformations Between Three Inverse Bicontinuous Cubic Phases of a Lipid

Toshihiko Oka[1,2] ([1]*Faculty of Science, Shizuoka University*, [2]*Research Institute of Electronics, Shizuoka University*)

The transformation between three inverse bicontinuous cubic (Q_{II}) phases of a lipid in the single crystal region of monoolein was studied. X-ray diffraction data reveal that the crystallographic orientation of Q_{II}^P rotates 55 degree around the [0-11] axis from Q_{II}^D. It showed one direction of the four-branched water channels in the Q_{II}^D phase is preserved in the six-branched water channels of the Q_{II}^P phase. I built a transformation model that would keep the direction of the water channels preserved in both phases and cause the water channels along other direction in Q_{II}^D to shrink and disappear. I also studied the Q_{II}^D-Q_{II}^G transformation and built a transformation model which has the same relation in the Q_{II}^D-Q_{II}^P transformation.

1Pos209 **ジミリストイルホスファチジルコリン−コレステロール二成分二分子膜中における構成脂質の部分モル体積**
Partial molar volumes of constituent lipids in the binary bilayer of dimyristoylphosphatidylcholine and cholesterol

Nobutake Tamai[1], Naohiro Takeshita[2], Masaki Goto[1], Hitoshi Matsuki[1] ([1]*Grad. Sch. Biosci. Bioind., Tokushima Univ.*, [2]*Grad. Sch. Adv. Tech. Sci., Tokushima Univ.*)

In this volume study on the dimyristoylphosphatidylcholine (DMPC)-cholesterol (Chol) binary bilayer using densitometry, we estimated the mean apparent molar volume of the constituent lipids at 30 °C for the binary bilayers with different Chol concentrations. It decreased monotonically with increasing Chol concentration. This monotonic decrease is due to the fact that the volume that a Chol molecule occupies within the binary bilayer is smaller than that of DMPC. We found that the concentration dependence of the mean apparent molar volume can be expressed by a convex curve in the Chol concentration region below ca. 8 mol %. This suggests that attractive interaction works between DMPC and Chol molecules within the binary membrane in this region.

1Pos210 **部分フッ素化リン脂質と DMPC の二成分系混合膜物性に関する系統的研究**
Mixing behaviors in the binary membrane of DMPC and its partially fluorinated analogues with different perfluoroalkyl chain lengths

Miki Horikoshi[1], Kohei Morita[1], Toshinori Motegi[1], Hiroshi Takahashi[1], Hideki Amii[1], Toshiyuki Takagi[2], Toshiyuki Kanamori[2], Masashi Sonoyama[1] ([1]*Div. Mol. Sci., Gunma Univ.*, [2]*AIST*)

We have recently reported that F4-DMPC, a partially fluorinated dimyristoylphosphatidylcholine (DMPC) with a perfluorobutyl group at the end of the acyl chains, shows lower phase transition temperature and non-ideal mixing in the binary membrane with DMPC, indicating the membrane properties of F4-DMPC are significantly different from those of DMPC. In this study, we focus on partially fluorinated DMPCs with longer perfluoroalkyl (Rf) chains (F6-DMPC and F8-DMPC) and effects of the Rf chain lengths on non-ideal mixing behaviors in the binary phospholipids with DMPC. The composition phase diagrams of the binary F6-DMPC/DMPC and F8-DMPC/DMPC membrane obtained by DSC measurements will be discussed.

1Pos211 **多電極アレイ上でのエレクトロフォーメーション法によるリポソームの作製**
Preparation of liposomes by electro formation method on multi electrode array

Hayato Akizuki, Tomoyuki Kaneko (*LaRC, Grad. Sci. Eng., Hosei Univ.*)

Liposomes are artificial vesicles made by lipid bilayer, and are used in study as a model for biological membranes *in vitro*. The major methods of liposome preparation are gentle hydration, spontaneous transfer, electro formation, and so on. Electro formation method is known to prepare giant liposomes easily. In this study, we applied lipids (phosphatidylcholine and phosphatidylglycerol) on multi electrode array (MEA) and tried to prepare the liposomes by electro formation method. Electric pulse was loaded with square wave, voltage and frequency were changed. As a result, we succeeded in preparing many giant liposomes rapidly. This method could be applied to the study of membranes model for cell scale.

1Pos212 PEG 脂質を導入した支持脂質二重膜の拡散特性

Effect of PEG-lipid on diffusion properties of supported lipid bilayer

Moeko Saruta, Takuhiro Otosu, Shoichi Yamaguchi (*Grad. Sch. Sci. Eng., Saitama Univ.*)

Model lipid membranes are often utilized to elucidate the complex structural as well as functional properties of cell membranes. Among them, supported lipid bilayer (SLB) is a widely-used planar membrane on a solid substrate. However, utilization of SLB is limited because a narrow space between SLB and the solid substrate makes it difficult to incorporate transmembrane proteins. In this regard, introduction of polymer cushions in between the lipid bilayer and the solid substrate is attracting much attention. To fully utilize such polymer-cushioned lipid bilayers, the effect of polymer cushions on physical properties of lipid bilayers must be evaluated. In this study, we analyze the diffusion properties of SLB containing PEG-lipid as a cushion.

1Pos213 プログラマブルな性質を有する DNA ナノプレートからなるマイクロカプセルの形成

Microcapsular compartments composed of programmable DNA nanoplates

Daisuke Ishikawa[1], Yuki Suzuki[2], Chikako Kurokawa[3], Masayuki Ohara[4], Masamune Morita[1], Miho Yanagisawa[3], Ryuji Kawano[4], Masayuki Endo[5], Masahiro Takinoue[1] ([1]*Sch. Comput., Tokyo Tech.*, [2]*FRIS, Tohoku Univ.*, [3]*Dept. Appl. Phys., Tokyo Univ. of Agri. and Tech.*, [4]*Dept. Life Sci. and Biotech., Tokyo Univ. of Agri. and Tech.*, [5]*WPI-iCeMS, Kyoto Univ.*)

Living systems consist of an enormous number of compartmentalized spaces where incompatible or opposing reagents are spatially isolated to avoid mutual deactivation. This is also a key concept to realize artificial cell-like systems furnished with sensing, actuating and computing functions. Water droplets stabilized with surfactants or particles in organic phase can be a simple system for creating the artificial cell-like systems because they have confined spaces separated by immiscible phase, which allows such individual reactions based on DNA and other biomolecules as occur within living cells. Herein, to construct artificial cell-like systems, we propose water microdroplets stabilized by amphiphilic DNA nanoplates equipped with pores that work as a molecular channel.

1Pos215 コリネ細菌の機械受容チャネルによる細胞力覚とグルタミン酸放出機構

Bacterial mechanosensation and glutamate export by mechanosensitive channels in Corynebacterium glutamicum

Yoshitaka Nakayama[1], Kosuke Komazawa[2], Navid Bavi[1,3], Ken-ichi Hashimoto[2], Hisashi Kawasaki[2], Boris Martinac[1,3] ([1]*Victor Chang Cardiac Research Institute*, [2]*Tokyo Denki University*, [3]*University of New South Wales*)

Corynebacterium glutamicum secretes a massive amount of glutamate through the mechanosensitive (MS) channel MscCG, and the gating of MscCG is affected by the membrane lipid components. Here we show a successful preparation of C. glutamicum giant spheroplasts that has enabled us to record MS channel activities and characterize mechanical properties of the membrane. Using the patch-clamp technique we have identified two types of MS channels, the MscCG and an MscL-like channel. Our findings demonstrate that MscCG has evolved to transport ions and other osmolytes across the C. glutamicum membrane. Furthermore, our study indicates that interactions between the MscCG and the lipid bilayer present the key factor towards improving the glutamate export in C. glutamicum.

1Pos216 全反射赤外分光で見る電位依存性プロトンチャネル VSOP への金属結合

Metal binding to the voltage-gated proton channel VSOP studied by ATR-FTIR

Masayo Iwaki[1], Kohei Takeshita[2,3,4], Yasushi Okamura[5], Atushi Nakagawa[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*Inst. Protein Res., Osaka Univ.*, [3]*Inst. Acad. Initiat., Osaka Univ.*, [4]*JST-PRESTO*, [5]*Grad. Sch. Med., Osaka Univ.*)

The voltage-gated proton channel (VSOP) has a Zn^{2+}-binding site in the extracellular region of voltage-sensing transmembrane helices. The present study aimed to elucidate the molecular mechanism of channel inactivation controlled by Zn^{2+}, by using ATR-FTIR. Zn^{2+}-induced difference ATR-FTIR spectra of VSOP showed IR features that can be assigned to the His-CN and carboxylate-OCO^- stretches as well as Amide I changes likely in α-helical peptide bonds. The effects of mutation of ligand candidates of E115 or D119 on the structural changes induced by Zn^{2+}-binding will be discussed in relation to the atomic structure and electrophysiological studies [1]. (1, Takeshita et al. (2014) Nat. Struc. Mol. Biol., 21, 352.)

1Pos217 ナノキャビティでの K^+の占有が Kv1.2 チャネルを通る K^+の滑走を引き起こす

Occupancy of a K^+ in the nanocavity induces K^+ ions' run through the Kv1.2 channel

Takashi Sumikama, Shigetoshi Oiki (*Univ. of Fukui*)

The molecular dynamics (MD) simulations have shown that ions permeate through K^+ channels via the knock-on mechanism happening near the selectivity filter (SF), but this oversimplified view is insufficient to account for experimentally observed single-channel currents. Here, we analyzed MD-simulated ion trajectories through the Kv1.2 channel using an event-oriented analysis and found that the nanocavity (NC) connecting the intracellular bulk to the SF governs ion permeation in a digital manner. The ions in the SF are mostly immobilized when the NC is unoccupied by a K^+, while they run outwardly when occupied. Thus, the one ion in the NC serves as a catalytic intermediate for permeation, which quantitatively explains the conductance-concentration relations.

1Pos218 KcsA チャネルの細胞内領域の荷電状態がチャネル開閉に与える影響

Effects of the electrostatic state of the cytoplasmic domain in the KcsA channel on its gating

Minako Hirano[1], Toru Ide[2] ([1]*GPI*, [2]*Okayama Univ.*)

We show effects of the electrostatic state of the cytoplasmic domain (CPD) in a proton-activated K^+ channel (KcsA) on its activation gate and inactivation gate. Previously we revealed that the CPD of the KcsA regulates its gating by sensing pH. Here, we show that E146 is the most effective amino acid on the gating and suggest that electrostatic repulsions between R142s from different strands make the activation gate open when E146 is protonated. In addition, the inactivation gate was K^+-conductive in CPD-protonated mimicking mutants and the electrostatic state of D149 affected fast inactivation and high K^+ selectivity, two properties characteristic of wild-type KcsA.

1Pos219 Discrimination between mitochondrial rounding and permeability transition

Takahiro Shibata, Yoshihiro Ohta (*Grad. Sch. Life Sci. & Bio Tech., TUAT*)

Mitochondria show the elongated structure in cells. Once mitochondria get damaged, mitochondria change their morphology and show the round structure. Although mitochondrial permeability transition (MPT) is often observed as mitochondrial damage, the relationship between MPT and the rounding has not been clarified. The aim of this study is to examine whether MPT is necessary for mitochondrial rounding. Elongated mitochondria were isolated from C6 glioma cells. MPT and rounding was induced by t-butyl hydroperoxide (t-BuOOH). When Ca^{2+} concentration is high, t-BuOOH induced both MPT and rounding. At physiological concentration of Ca^{2+}, t-BuOOH induced the rounding but not MPT. This suggest that rounding does not require MPT.

1Pos223 Artificial bilayers formed on a solid substrate for ion-channel recordings

Toru Ide[1,2], Saki Nomura[1], Minako Hirano[2], Junnya Ichinose[1], Hiroaki Yokota[2] ([1]*Grad. Schl Sci. Tech., Okayama Univ.*, [2]*GPI*)

We have developed a simple technique in which a bilayer is made by contacting a solid substrate to a lipid-solution interface and ion-channels immobilized on the surface of the solid substrate is inserted into the bilayer mechanically. Using this technique, we measured single channel currents of several types of ion-channel protein including KcsA, MthK, BK, and P2X4 along with those of two channel-forming peptides and a channel-forming protein. The technique requires only one action that simultaneously forms the bilayers and inserts the channels into the bilayers. The simplicity of this technique allows it to potentially be combined with high-throughput screening devices. This is also applicable to develop a single channel imaging apparatus.

1Pos224 電位依存性ホスファターゼ VSP の酵素ドメインにおける膜相互作用部位の役割
The role of membrane interacting region of phosphatase domain in voltage-sensing phosphatase (VSP)

Akira Kawanabe[1], Masaki Hashimoto[1], Tomoko Yonezawa[1], Yuka Jinno[2], Souhei Sakata[2], Yasushi Okamura[1] ([1]*Osaka Univ.*, [2]*Osaka Med. Col.*)

Voltage-sensing phosphatase (VSP) consists of a transmembrane voltage sensor domain and a PTEN-like cytoplasmic region, which dephospholyrates PI(4,5)P2 regulated by membrane potential (Murata et al. 2005). The mechanism of the coupling between the two modules in VSP has been enigmatic so far.
In this study, we examined the phosphatase activity of VSP with mutation in the putative membrane interacting region of the phosphatase domain (PD) (L284 and F285). The drastic changes of the voltage-dependent phosphatase activities were shown with mutation of these sites. These findings suggest that the membrane interacting region in the PD plays important roles in the voltage dependent regulation of phosphatase activity in VSP.

1Pos225 イオンチャネル機能に対する膜脂質効果の解析に向けた脂質二重膜組成の迅速変更法
Rapid replacement of the lipid bilayer composition for the analysis of the lipid-effect on the ion channel function

Masayuki Iwamoto, Shigetoshi Oiki (*Dept. Mol. Physiol. & Biophys., Univ. Fukui Facult. Med. Sci.*)

In this study we established an instantaneous lipid change method for the contact bubble bilayer (CBB) method. Two strategies were examined. First, desired lipids were served as liposomes, and suspensions of liposomes were perfused toward the vicinity of the CBB to promote spontaneous membrane fusion with the CBB. Second, desired lipids were dissolved in the oil, which were injected close to the annular oil phase of the CBB. The lipids were spontaneously transferred to the CBB. Changes in the lipid composition of the CBB were detected as the lipid-dependent changes in the gating of the KcsA potassium channel and in the pore-forming activities of some antibiotics. Applicability of the presented method to the ion channel study will be discussed.

1Pos226 高速原子間力顕微鏡による K^+チャネル KcsA とポア結合性サソリ毒アジトキシン-2 の結合ダイナミクスの一分子解析
Single-molecule blocking dynamics of a scorpion toxin on the KcsA potassium channel revealed by HS-AFM

Ayumi Sumino[1,2], Takayuki Uchihashi[3], Takashi Sumikama[2], Shigetoshi Oiki[2] ([1]*JST/PRESTO*, [2]*Facult. Med. Sci., Univ. Fukui*, [3]*Depart. Phys., Kanazawa Univ.*)

Agitoxin-2 (AgTx2) is a small protein (38 a.a.) extracted from scorpion venom, and blocks potassium channels through docking to the extracellular surface of the channel. In this study, we directly observed the binding dynamics of AgTx2 to the KcsA channel by HS-AFM. The KcsA channel is a homo-tetramer, and the AFM image observed from the extracellular side showed a square shape. When an AgTx2 bound on the channel surface, the channel changed the shape from square to round with increased height. We analyzed time course of the binding transitions, and found that there are two bound states with high and low affinity. The mechanism underlying the heterogenous binding will be discussed.

1Pos227* 線虫（C. elegans）嗅覚感覚神経細胞内の領域特異的なにおいに対する cGMP 応答
Compartmentalized cGMP responses to odor in Caenorhabditis elegans' olfactory sensory neurons

Hisashi Shidara, Keita Ashida, Kohji Hotta, Kotaro Oka (*Grad. Sch. Sci. and Tech., Keio Univ.*)

Cyclic guanosine monophosphate (cGMP) is one of the essential second messengers for sensory signal pathways in many animals. According to molecular genetic studies, cGMP could have region-specific roles for C. elegans' olfaction: olfactory sensation in cilia (at the top of the head) and adaptation in soma of AWC olfactory neurons. However, it unrevealed how cGMP actually behaved region-spatially in AWC. Here, we visualized intracellular cGMP by using a genetically encoded cGMP indicator, cGi500, and examined spatial and temporal responses of cGMP in AWC. The cGMP imaging elucidated that odor stimulus induced a transient cGMP fall in cilia, but gradual increase in soma. These results indicated compartmentalized cGMP responses in olfactory neurons.

1Pos228* *C. elegans* の低温適応における温度情報伝達の分子ロジック
Molecular logic for temperature signaling in cold tolerance of *C. elegans*

Tomoyo Ujisawa[1], Misato Uda[1], Akane Ohta[1], Katsushi Arisaka[2], Atsushi Kuhara[1] ([1]*Inst. for Integrative Neurobiology, Konan University.*, [2]*Dept. of Physics and Astronomy, UCLA, U.S.A.*)

Temperature is essential for life and proliferation of animal. We previously reported that ASJ photo-sensoryneuron senses temperature and releases insulin to regulate cold tolerance. We are analyzing molecular physiological neural coding of temperature sensation in ASJ sensoryneuron. Previously, we revealed that TAX-4 cGMP-gated channel is required for temperature signaling in ASJ. Also, we found that cold tolerance is regulated by redundant functions of three Gα proteins in ASJ. Optical recording of calcium concentration in ASJ upon temperature-changes described that three Gαs, two GCs and four PDEs acts in the respective diverse aspects in temperature signaling.

1Pos229 蛍光温度計シートを用いた神経細胞の熱発生計測
Detection of Neural Thermogenesis with Fluorescent Thermometer Sheet

Mizuho Gotoh[1,2,3], Kotaro Oyama[1,4], Yuki Kawamura[1], Hideki Itoh[1,5], Shin'ichi Ishiwata[1,6] ([1]*Sch. Adv. Sci. Eng., Waseda Univ.*, [2]*Grad. Sch. Comp. Human Sci., Tsukuba Univ.*, [3]*HIRI, AIST*, [4]*Cell Physiol., Jikei Univ.*, [5]*Inst. Med. Biol., A*STAR, Singapore*, [6]*WABIOS, Waseda Univ., Singapore*)

The goal of this research is to detect thermogenesis involved in firing of neurons. Here we cultured rat hippocampal neurons on a "thermometer sheet" consisting of thin polymer (PMMA) sheet, in which temperature-sensitive fluorescent dye (Eu-TTA) (cf., Itoh, H. et al., BIOPHYSICS, 2014) was embedded to image two-dimensional temperature distribution. Ca^{2+} bursts in neurons were repetitively evoked by electrical stimulations to examine heat production due to Ca^{2+} uptake by SERCA (cf., Suzuki, M. et al., Biophys J, 2007, Itoh, H. et al., Chem Commun, 2016). We found that the thermometer sheet could detect changes in local temperature increase in neurons with high accuracy (less than 0.01℃), and succeeded in observing the correlation with cytosolic Ca^{2+} elevation.

1Pos230* 線虫の早期嗅覚順応における感覚・介在神経細胞の部分特異的可塑性
Compartmentalized modulations of sensory and interneurons for early adaptation in C. elegans

Keita Ashida, Hisashi Shidara, Kohji Hotta, Kotaro Oka (*Keio University*)

Single neurons consist of compartmentalized units, which are characterized by physical-chemical features and showed compartmentalized modulations by environmental stimuli. Caenorhabditis elegans adapt to odors by 5-min odor pre-exposure; however, modulations between sensory and interneurons have not been examined. Simultaneous Ca2+ imaging of AWC sensory neuron and AIY interneuron revealed that neural activities did not change in AWC by adaptation; AIY showed region-specific modulations. Neurotransmitter from AWC to AIY is glutamate, and glutamatergic inputs to interneurons also showed region-specific modulations. This study showed compartmentalized modulations of glutamate (input) and neuronal activity (output) in C. elegans, for the first time.

1Pos231* イベルメクチンによる α7 ニコチン性アセチルコリン受容体の分子内動態増大の発見
Discovery of the internal motion enhancement of α7 nAChR with Ivermectin

Tomoyuki Baba[1], Keigo Ikezaki[1], Hiroshi Sekiguchi[2], Tai Kubo[3], Yuji C. Sasaki[1,2] ([1]*Grad. Sch. Front. Sci., Univ. Tokyo*, [2]*JASRI/SPring-8*, [3]*MolprofRC/AIST*)

α7 nAChR has an important role as the cholinergic neurotransmission in the central nerve system. In the case of Alzheimer disease, this nerve system is thought to be disordered. Thus, α7 nAChR is one of the most important drag targets for Alzheimer disease and required further investigations of its mechanism. Recently, structural information of α7 nAChR in open, close and desensitization state was revealed. However, the dynamic of each state is still unclear. Here, we used Diffracted X-ray Tracking (DXT) method to measure the dynamics of α7 nAChR. Using DXT measurement, we investigated the effect of Ivermectin which is one of the Positive Allosteric Modulator for α7 nAChR. We found that Ivermectin enhanced the tilting motion (3.98 mrad/2.0 ms) of α7 nAChR dramatically.

1Pos232 ミミズ体壁刺激に関する慣れとその回復
Establishment and recovery of habituation by repeated tactile stimulus in earthworm

Yoshiichiro Kitamura[1], Hitoshi Aonuma[2], Hiroto Ogawa[3], Kotaro Oka[4] ([1]*Dept. Math Sci. Phys, Kanto Gakuin Univ.*, [2]*Res Inst Elect Sci., Hokkaido Univ.*, [3]*Dept. Biol. Sci., Hokkaido Univ.*, [4]*Dept. Biosci Info, Keio Univ.*)

We previously reported that habituation by repeated tactile stimulus to the body wall in the earthworm is occurred assumedly due to via nitric oxide (NO) signaling, because relatively high concentration of NO or cGMP accelerated decrease of number of action potentials (APs) by tactile stimulus. In this study, we investigated recovery period of habituation by repeated tactile stimulus. After establishment of habituation, AP generation by tactile stimulus fully recovered after 120 min. From these results, non-associative learning such as habituation in the earthworm continue for several hours.

1Pos233 シャルコマリートゥース病の原因遺伝子の一つであるダイナミン２の変異は細胞の異常なアクチン動態とラメリポディア形成の減少をもたらす
Expression of a dynamin 2 mutant associated with Charcot-Marie-Tooth disease leads to aberrant actin dynamics and lamellipodia formation

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The effects of dynamin 2 mutations linked to Charcot-Marie-Tooth disease (CMT), an inherited peripheral neuropathy, on the actin cytoskeleton remain unclear. Immunofluorescence and live-imaging showed that the K562E mutation resulted in formation of aberrant F-actin clusters with a significant loss of serum stimulation-dependent lamellipodia formation and disappearance of radially aligned actin bundles. The K562E mutant colocalized with the F-actin clusters, but not with clathrin-coated pit marker proteins. The present study is the first to show the association of dynamin CMT mutation with aberrant actin dynamics, which may contribute to defective endocytosis and myelination in Schwann cells in CMT.

1Pos234 ヒト苦味受容体の基質認識の分子機構研究
Ligand-induced structural changes of human bitter taste receptor

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The bitter taste receptors (TAS2Rs) belong to a family of the G protein-coupled receptors. The role of TAS2Rs is to transfer the bitter information to the inner cell without intake of bitter substances. Our goal is to elucidate the molecular mechanisms of ligand-induced structural changes of human TAS2R by using ATR- FTIR spectroscopy. The challenge ahead is to obtain enough amount of the purified proteins, which are suitable to the ATR-FTIR measurements.
In this study, we report our current status of protein sample preparations, which shall be suitable to IR spectroscopy. His-tagged hTAS2R16 was expressed in yeast *Pichia pastoris*, and then purified after a range of screening of genetic modifications, cell expressions and detergent solubilization.

1Pos235 性ホルモンによる海馬神経シナプスの制御：オスとメスの性差
Effect of estrogen and androgen on hippocampal synapses : gender difference of male and female

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Sex steroids (androgen and estrogen) are locally synthesized in hippocampal neurons and behave like neurotrophic factor. Recently, evidence is accumulated on the rapid effect of sex steroids on synaptic plasticity including spinogenesis and long-term potentiation (LTP) in the hippocampus. We demonstrated these rapid effect of androgen (testosterone: T and dihydrotestosterone: DHT) and entrogen (17β-estradiol: E2). Kinase networks (Erk MAPK, PKA, PKC and LIMK) are activated by sex steroids, leading to actin polymerization resulting in spine increase within 2 hours. In LTP induction, under weak-theta burst stimulation > E2 binds to ER > activation of kinase network (MAPK, PKA, PKC) > phosphorylation of NMDA receptor leading to activation of NMDA receptor and induced LTP.

1Pos236 ナトリウムポンプ型ロドプシンの光反応中間体の発色団構造
Structure of retinal chromophore of the photointermediates in sodium ion pump rhodopsin

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Krokinobacter eikastus rhodopsin 2 (KR2) is a light-driven sodium ion pump. Similarly to other microbial rhodopsins, a series of intermediates appear in its photocycle. To elucidate the ion pumping mechanism of KR2, we explored the chromophore structures of the intermediates using time-resolved resonance Raman spectroscopy.
We obtained the spectra of K, L, M, and O intermediates. For O intermediate, we revealed that the retinal chromophore is reprotonated in the Schiff base and is in 13-*cis* form. This means that retinal configuration changes from 13-*cis* form to all-*trans* form in the recovery to the ground state. This feature is contrast to that of bacteriorhodopsin, a well-known microbial rhodopsin, and may be characteristic of KR2.

1Pos237 固体 NMR を用いたミドルロドプシンのレチナール結合ポケットの構造解析
Solid-state NMR structural study of retinal-binding pocket in middle rhodopsin

Izuru Kawamura[1], Hayato Seki[1], Arisu Shigeta[1], Yoshiteru Makino[1], Takashi Okitsu[2], Akimori Wada[2], Yuki Sudo[3] ([1]*Grad. Sch. Eng., Yokohama Natl. Univ.*, [2]*Kobe Pharm. Univ.*, [3]*Okayama Univ.*)

Microbial rhodopsins contain an all-*trans* retinal chromophore within seven transmembrane helices and the retinal photoisomerization triggers structural change of protein which leads to expression of the function. Middle rhodopsin (MR) is known for having 11-*cis* retinal. Solid-state NMR spectroscopy is a powerful technique to investigate membrane protein structure and dynamics. In this study, based on ^{13}C spin diffusion NMR spectra, we observed cross peaks of ^{13}Cζ Tyr185 with ^{13}C20 retinal in (MR) which has bR-like photocycle but SRII-like color. We will show the comparison of the Tyr and retinal structure among microbial rhodopsins and would like to discuss the contribution of the Tyr interaction with retinal to the color and retinal configuration.

1Pos238* 酸性及び中性におけるナトリウムイオンポンプ KR2 のレチナール結合ポケットの固体 NMR 構造解析
Solid-state NMR analysis of retinal binding pocket structure of sodium ion pump, KR2, at acidic and neutral pH

Arisu Shigeta[1], Shota Ito[2], Takashi Okitsu[3], Akimori Wada[3], Keiichi Inoue[2,4], Hideki Kandori[2], Izuru Kawamura[1] ([1]*Graduate School of Engineering, Yokohama National University*, [2]*Nagoya Institute of Technology*, [3]*Kobe Pharmaceutical University*, [4]*JST PRESTO*)

Krokinobacter rhodopsin 2 (KR2) is the first rhodopsin which transports non-proton cation. We aimed to reveal the structure of retinal and residues around retinal by solid-state NMR. Retinal-, Tyr- and Lys-labeled WT-KR2 in membrane was applied to Dipolar Assisted Rotational Resonance (DARR) at pH 8, 6 and 4. Comparison of these data allowed to give the following insights. Chemical shift value of retinal Schiff base indicated the torsion around Schiff base which is caused by the unique location of Asp116, a counterion of Schiff base. We also determined the chemical shift of Tyr218 which forms rather weak hydrogen bond with Asp251. These features may give us new insights about unique mechanism of sodium ion pump.

1Pos239 Na+/H+ハイブリッドポンプロドプシン KR2 における His30 の役割
Role of His30 in Na+/H+ Hybrid Pumping Rhodopsin KR2

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KR2 functions as a light-driven outward Na+/H+ hybrid pump. His30 is located in the extracellular side and ion transport activity measurement showed that KR2 H30A lost H+ pumping activity, while it kept Na+ pumping activity. It indicates that His30 is only important for H+ pump of KR2, whose mechanism is unclear.
In this study, we applied low-temperature light-induced different FTIR spectroscopy at 200-277 K to study structural changes from the resting state to the O-intermediate. We analyzed protonation state of side-chains including His30 and structural change of peptide backbone. We also measured ion transport activity of KR2 H30X mutants to obtain further insight on H+ pump function. Role of His30 in KR2 will be discussed.

1Pos240* FTIR 分光法によって明らかになった光駆動内向きプロトンポンプの輸送機構

Transport mechanism of light-driven inward proton pump revealed by FTIR spectroscopy

Shota Ito[1], Sahoko Tomida[1], Yoshitaka Kato[1], Yurika Nomura[1], Satoshi Tsunoda[1], Keiichi Inoue[1,2], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*PRESTO, JST*)

Mechanism of active transport of ions is one of the central questions in biophysics. It is best studied for a light-driven proton pump bacteriorhodopsin (BR), where pKa of key residues are well controlled spatially and temporally. Although people believed such elaborate mechanism only possible for outward proton pump, we have recently discovered the presence of oppositely directed proton pump, named *Po*XeR. Detailed FTIR analysis of photoreaction intermediates revealed that protein controls thermal isomerization of retinal and pKa of different residues from those of BR, leading to inward proton pump in *Po*XeR. Highly elaborate molecular design of *Po*XeR for reverse-directed proton transport based on similar protein architecture of BR will be discussed.

1Pos241 H⁺ポンプ型ロドプシンの比較研究：H⁺ donor 残基の相互置換による検討

Replacements of "donor" residues in the light-driven H^+-pump rhodopsins

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Many H^+-pump rhodopsins conserve a "H^+ donor" residue in the cytoplasmic channel. For conventional H^+ pumps, this residue is conserved as Asp or Glu, but is replaced by Lys in *Exiguobacterium sibiricum* rhodopsin (ESR). We examined the replacement effects of the donor residues. Judging from the M decay, the embedded Lys in Asp-type deltarhodopsin (DR) did not function, but Lys in Glu-type proteorhodopsin (PR) and Asp/Glu in Lys-type ESR showed proper donor functions. Next, we determined the activation volumes of M decays. The value for DR was about two-fold larger than those of PR, ESR and their donor replacement mutants. These probably reflect that Glu and Lys-type pumps share common H^+-transfer machineries, but which are different from that of Asp-type pump.

1Pos242 アセタブラリアロドプシン II のプロトン移動における D92 および C218 残基間の相互作用の役割

Role of the interaction between D92 and C218 in the proton transfer reaction in *Acetabularia* rhodopsin II

Jun Tamogami[1], Takashi Kikukawa[2], Keisuke Okawa[1], Noboru Ohsawa[3,4], Kohei Date[1], Toshifumi Nara[1], Makoto Demura[2], Tomomi Kimura-Someya[3,4], Mikako Shirouzu[3,4], Shigeyuki Yokoyama[3,5], Seiji Miyauchi[6], Kazumi Shimono[6], Naoki Kamo[2] ([1]*College Pharm. Sci., Matsuyama Univ.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*, [3]*RIKEN SSBC*, [4]*RIKEN CLST*, [5]*RIKEN Structural Biology Laboratory*, [6]*Fac. Pharm. Sci., Toho Univ.*)

Acetabularia rhodopsin II (ARII) is a eukaryotic light-driven proton pump from *Acetabularia acetabulum*. The previous determined X-ray crystal structure of ARII implied the formation of an interhelical hydrogen bond between D92, a proton donor to the retinal Schiff base, and C218 located in the cytoplasmic region. Here we investigated the effect of the replacement of C218 on the photochemical reaction. The C-to-A or -S mutation of this residue caused the delay of the proton uptake during N-decay, which may imply the importance of the weak interaction between these two residues in the reprotonation of D92 from the intracellular surface. Based on the observations, a putative role of the hydrogen bond between D92 and C218 in the proton transfer is discussed.

1Pos243* 固体 NMR によるバクテリオロドプシンの暗順応状態における Tyr185 の構造解析

Structure of Tyr185 in dark-adapted bacteriorhodopsin as studied by solid-state NMR

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Dark-adapted state bacteriorhodopsin (bR) has two retinal configurations of 13-*cis*, 15-*syn* and all-*trans*, 15-*anti* as a ratio 1:1. Tyr185 is highly conserved among all microbial rhodopsins and forms a hydrogen bond with Asp212. Here, we observed the solid-state NMR spectra of ^{13}C14,20-retinal, ^{13}Cζ-Tyr labeled bR. The cross peak of Tyr185 with two retinal states appeared at a lower magnetic field at 157.6 ppm. We reported the difference of two backbone structures of Tyr185 corresponding to two retinal configurations in the dark [1]. It is indicated that there is non-alternation of hydrogen bond of Tyr185 with Asp212 even though the change of backbone structure. We will show the comparison of Tyr185 NMR signal between wt-bR and its mutants.

[1] I. Kawamura (2007) JACS.

1Pos244 光駆動ナトリウムポンプ KR2 の多量体形成に重要なアミノ酸残基

Oligomerization of light-driven sodium pump KR2 is important for ion transport activity

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Nature created two kinds of light-driven cation pumps. A light-driven proton pump bacteriorhodopsin (BR) forms a trimer in 2-D hexagonal lattice of purple membrane, which is important for thermal stability and function. A light-driven sodium pump KR2 forms a pentamer in 3-D crystal, whose role has been unclear. In this study, we found that oligomerization of KR2 is dependent of pH in a detergent DDM. To identify important amino acids for oligomerization, several mutants were analyzed by size-exclusion chromatography. Among them, H30A and Y154A showed difference elution profile, together with lowered pump activity. Role of oligomerization for the function of KR2 will be discussed based on various methods including high-speed atomic force microscope.

1Pos245* Theoretical study on molecular mechanism of a light-driven ion transport of Halorhodopsin

Ryo Oyama, Shigehiko Hayashi (*Grad. Sch. Sci., Kyoto Univ.*)

Halorhodopsin from Natronomas pharaonis functions as an inward light-driven chloride pump and is utilized to silence neurons in optogenetics technique in neuroscience. The chromophore retinal isomerizes from all-tran conformation to 13-cis one upon photoabsorption, and triggers a photocycle during which one chloride ion is transported across the membrane. In this study, we performed QM/MM RWFE-SCF calculations to examine the functional coupling of the structural change of the chromophore isomerizing from all-trans conformation to 13-cis one described at the quantum chemistry level of theory with protein large conformational changes of alternating access for the active transport of the ion described with MD simulations with a MM force field.

1Pos246 **Rhodobacter capsulatus PYP の複合体形成に伴う構造変化**
Complex induced structural changes of Rhodobacter capsulatus Photoactive Yellow Protein

Yoichi Yamazaki, Yohei Shibata, Hironari Kamikubo (*Grad. Sch. Mat. Sci. NAIST*)

Photoactive Yellow Protein in Rhodobacter capsulatus (Rc-PYP) is a light receptor which binds to interaction protein named PBP. Interaction mechanism of Rc-PYP and PBP has not been clarified yet. Different FT-IR spectrum at the formation of the complex state revealed that Glu45 which makes hydrogen bonding with chromophore was deprotonated in the complex state, even though UV-Vis spectrum is alike to that of protonation. FT-IR measurement also showed that cysteine vibrational change at the complex formation. Rc-PYP and PBP have one and two cysteine residues respectively. Cysteine vibrational changes are induced only at the complex formation. Then to identify this structural changes, we investigated that complex formation of cysteine substituted Rc-PYP and PBP.

1Pos247 **Rc-PYP の光依存的に形成する複数種の複合体解析**
Analysis of light dependent multiple complex formation of Rc-PYP

Yohei Shibata, Yoichi Yamazaki, Keito Yoshida, Shoki Nakata, Hironari Kamikubo (*Analysis of light dependent multiple complex formation of Rc-PYP*)

Rhodobacter capsulatus PYP (Rc-PYP) is a light receptor protein which binds with PYP binding protein (PBP) in a light-dependent manner. Titration small angle X-ray scattering (tiSAXS) experiments have suggested that Rc-PYP and PBP can form several complexes with different stoichiometry. To investigate the complex formation mechanism, we identified UV-vis spectroscopic properties of the complexes. We found at least two complexes accompanied by different resistance against TCEP. PBP titration measurement against Rc-PYP also revealed that the ratio of the complexes depends on [Rc-PYP]/[PBP]. These results partly support the previous equilibrium model. Combining the results from tSAXS and spectroscopic experiments, we will show the multiple complexes formation mechanism.

1Pos248* **同位体標識試料を用いた BLUF ドメインの水素結合環境の解明**
Analysis of a hydrogen bonding network of the BLUF domain using isotope-labeled samples

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BLUF (blue-light sensor using FAD) domain is a photoreceptor domain that binds flavin (FAD) as a chromophore. A conserved Gln residue near the FAD is prerequisite for the photoreaction and keto-enol tautomerization of Gln has been proposed. To clarify hydrogen bonding network around FAD and Gln by FTIR spectroscopy, we tried to identify the signals from C=O, C=N stretches of Gln and C4=O stretch of FAD. We prepared a combination of ^{15}N-Gln and/or ^{13}C-FAD labeled proteins. Using Gln auxotrophic *E. coli* strain in the medium containing 0.5 g/L amino acids, labeling efficiency was estimated as almost 100% for Gln and <10% for other amino acids by MALDI-MS. The model of the hydrogen bonding network around FAD will be discussed from the FTIR results.

1Pos249 **ホタルオキシルシフェリン吸収・蛍光スペクトルにおける水和効果**
Hydration effects on absorption and fluorescence spectra of firefly oxyluciferin

Miyabi Hiyama[1], Yoshifumi Noguchi[1], Hidefumi Akiyama[1], Kenta Yamada[2], Nobuaki Koga[2] ([1]*ISSP, Univ. Tokyo*, [2]*Grad. Sch. Info. Sci., Nagoya Univ.*)

Recently, the absorption and fluorescence spectra of firefly oxyluciferin and its conjugate acids and bases were resolved. These spectral shapes of each chemical species must have reflected the vibronic and/or hydration effect depending on their structures and charge distributions. However, experimental studies alone do not show whether the shape of these spectra originate from the molecular vibrations. Therefore, the aim of this study is to elucidate the vibronic effects on the absorption and fluorescence spectra of these chemical species. The Franck-Condon factors between the ground and excited states were obtained using Barone's method. Theoretical spectra are in good agreement with the experimental ones.

1Pos250 **光化学系 II におけるクロロフィル励起三重項状態の赤外分光解析**
FTIR analysis on the localization of the excited triplet state of chlorophyll in photosystem II

Tatsuya Mitomi, Ryo Nagao, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

The excited triplet state of chlorophyll in photosystem II is a precursor of harmful singlet oxygen, and its quenching mechanism is important for photoprotection. However, the chlorophyll molecule where the triplet state is localized has not been definitely identified. To investigate the triplet localization, we introduced a site-directed mutation at D2-His197, which interacts with the keto C=O of Chl_{D1} via a water molecule, and studied its effect on the triplet state using FTIR spectroscopy. Triplet/singlet FTIR difference spectra showed that the keto C=O bands of Chl_{D1} were upshifted by D2-H197A mutation. The upshift of the keto C=O frequency of Chl_{D1} was also reproduced by DFT calculation. It is thus concluded that the triplet state is mainly accumulated on Chl_{D1}.

1Pos251 **光合成水分解反応におけるメタノール阻害機構の赤外分光解析**
FTIR study on the mechanism of methanol inhibition in the S-state cycle of photosynthetic water oxidation

Haruna Yata, Tatsuki Shimizu, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

The mechanism of methanol inhibition of photosynthetic water oxidation in photosystem II is still unresolved. We investigated the effects of methanol on water-oxidizing reactions using FTIR spectroscopy. It was shown that methanol addition decreased mainly the S_3-to-S_0 efficiency, and altered protein conformations near the Mn cluster. In addition, the CO band of methanol was identified in the S_1/S_2 spectrum using ^{18}O-methanol. However, methanol did not affect the OH bands of a water cluster adjacent to the Mn cluster. These results suggest that methanol is located near the Mn cluster, but not affecting the adjacent water cluster. It may perturb H-bond networks leading to the lumen, causing inhibition of proton release and/or water insertion in the S_3-to-S_0 transition.

1Pos252* **Structure and Function of Novel Carbonyl-Carotenoid bound to Light-Harvesting Complex II from Transplastomic Lettuce**

Nami Yamano[1], Kentaro Ifuku[2], Hideki Hashimoto[1,3,5], Norihiko Misawa[4], Ritsuko Fujii[1,5] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*Grad. Sch. Biostudy, Kyoto Univ.*, [3]*Sch. Sci. Tech., Kwansei Gakuin Univ.*, [4]*Res. Inst. Biores. Biotech., Ishikawa prefectural Univ.*, [5]*OCARINA, Osaka City Univ.*)

Light harvesting complex II (LHCII) binds four carotenoids at the specific binding sites in protein for regulating solar energy. CGM lettuce is produced by chloroplast genome modification and specifically accumulates a variety of non-native carbonyl carotenoids, but is deficient in most of the natural carotenoids instead. We determined the chemical structure of carotenoids bound to LHCII in CGM, and evaluated their dissociation behaviors upon rising temperature. As results, we successfully concluded that a novel carbonyl carotenoid was bound in both 9'-cis and all-trans forms in specific binding sites, respectively. Fluorescence-excitation spectra confirmed the LHCII with carbonyl carotenoids can utilize 80% of the solar energy utilized by control LHCII.

1Pos253 **偏光全反射赤外分光法による光合成水分解 Mn クラスター周辺のプロトン化構造の解析**

Protonation structure around the water-oxidizing Mn cluster in photosystem II revealed by polarized ATR-FTIR spectroscopy

Shin Nakamura, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

Photosynthetic water oxidation is performed at the Mn cluster in photosystem II. In this study, we investigated protonation structures around the Mn cluster, which play an important role in water oxidation mechanism, using polarized ATR-FTIR spectroscopy. An ATR-FTIR difference spectrum upon the S_1-to-S_2 transition exhibited a broad negative band around 2600 cm^{-1} with a large dichroic ratio. QM/MM calculations showed that the frequency and dichroic property of this band are consistent with the NH stretching vibration of a protonated cation of D1-H337, which is H-bonded to an oxo bridge of the Mn cluster having four H_2O ligands. It was suggested that the cationic form of this His is an important factor to determine the redox potential of the Mn cluster.

1Pos254 **酸素発生系マンガンクラスターの配位水分子の化学的性質**

Chemical properties of terminal water ligands of the Mn cluster

Hiroki Nagashima, Hiroyuki Mino (*Grad. Sch. Sci. Nagoya Univ.*)

Mn cluster acts as a catalyst of water oxidation in photosynthesis. Recent X-ray crystallography revealed the structure of the Mn cluster. X-ray studies identified water sites (W1-W4 and O1-O5), which are the candidates for substrates. In the previous study, we measured ENDOR on the Mn cluster and identified the water protons surrounding Mn cluster. In this study, we applied ENDOR to NH3 bound and deuteron exchanged Mn cluster to identify substrate water sites. The ENDOR results indicated that W1 is exchangeable with ammonia and W2 has remarkable slow exchange rate in both S1 and S2 states. Therefore, it is suggested that terminal water ligands W1 and W2 were excluded from candidates for substrate water molecules.

1Pos255* **光化学系 II と阻害剤アジ化物イオンとの共結晶化と X 線結晶構造解析**

Co-crystallization of photosystem II with an inhibitor NaN_3, and its structural analysis

Shoya Tamaru[1], Yasufumi Umena[2], Jian-Ren Shen[2] ([1]*Graduate School of Natural Science and Technology, Okayama University.*, [2]*Reseach Institute for Interdisciplinary Science, Okayama University.*)

In photosystem II (PSII), chloride (Cl-) is required to achieve the maximum rate of oxygen evolution and binds at 2 sites near the Mn4CaO5 cluster. Azide (N3-) is a competitive inhibitor that binds to the Cl- binding sites (Cl-1 and/or Cl-2 site) and inhibits the water oxidation reaction. In order to elucidate the molecular mechanism for the N3-induced inhibition, we crystallized cyanobacterial PSII in the presence of NaN_3 but in the absence of Cl-, and collected X-ray diffraction data at 2.2 Å resolution. Structural changes around the Cl-1 binding site were observed, which showed that the distance of D2-K317 and D1-D61 was shortened. This may be one of the major reasons for the inactivation of oxygen evolution upon binding of N3-.

1Pos256 **時間分解 EPR 法を用いた PSII 反応中心に生成する初期電荷分離構造・電子的相互作用の解析**

Time resolved EPR study on orientations and electronic couplings of the primary charge-separated state in the PSII reaction center

Reina Minobe[1], Masashi Hasegawa[1], Shusuke Katagiri[3], Takahiro Sakai[2], Hiroki Nagashima[2], Takashi Tachikawa[1], Hiroyuki Mino[2], Yasuhiro Kobori[1] ([1]*Graduate School of Science, Kobe Univ.*, [2]*Graduate School of Science, Nagoya Univ.*, [3]*Graduate School of Science, Shizuoka Univ.*)

The photosynthesis has attracted a lot of attentions for a long time in terms of the utilization of the sunlight for the highly efficient solar energy conversion. PSII reaction center plays an important role on the efficient electron transfer process in the process of photosynthesis. Although kinetics of the primary charge separation and the recombination processes have been observed, no experimental studies have been performed to unveil the geometries and the electronic couplings of the primary charge-separated states. We have herein employed the time resolved EPR (TREPR) method to clarify the geometry and the electronic coupling of the primary charge-separated state in which the secondary electron acceptors of quinones are doubly-reduced in the membrane.

1Pos257 **FTIR 分光電気化学法を用いた光化学系 II における第一キノン電子受容体 Q_A の酸化還元電位計測**

Measurement of the redox potential of the primary quinone electron accepter Q_A in photosystem II by FTIR spectroelectrochemistry

Ayaka Ohira, Ryo Nagao, Takumi Noguchi, Yuki Kato (*Grad. Sch. Sci., Nagoya Univ.*)

Electron transfer energetics in PSII has been discussed mainly on the basis of the redox potential E_m of Q_A. Although, in almost all the recent works on $E_m(Q_A)$, redox states of Q_A were monitored indirectly with a fluorescence method, the fluorescence yield does not necessarily reflect only the redox states of Q_A. Thus, a fluorescence method has a problem for measuring $E_m(Q_A)$. In this work, we established a new method using spectroelectrochemistry and light-induced FTIR difference spectroscopy, which can detect directly the Q_A reaction. The $E_m(Q_A)$ value was determined to be ca. -100 mV. In comparison with a previous work (Shibamoto et al., 2010), we conclude that a $E_m(Q_A)$ value obtained using fluorescence is more negative by 30 mV than the true value of $E_m(Q_A)$.

1Pos258 光化学系 II におけるキノン電子受容体の電子移動制御機構

Regulation mechanism of electron transfer between quinone electron acceptors in Photosystem II

Yosuke Nozawa, Takumi Noguchi (*Division of Material Science, Graduate School of Science, Nagoya University*)

The primary quinone electron acceptor Q_A in PSII accepts an electron from pheophytin and transfers it to the secondary quinone electron acceptor Q_B. Little is known about the regulation mechanism of electron transfer between Q_A and Q_B. In this study, we investigated the pH dependence of Q_B^- relaxation using FTIR spectroscopy to reveal the effect of pH on the equilibrium between Q_A and Q_B. The results showed that Q_B^- decayed faster at a higher pH. It is thus suggested that the electron-transfer equilibrium of quinone electron acceptors is shifted to the Q_A side as the pH is increased, facilitating charge recombination with the water-oxidizing center. We propose that this is one of photoprotection mechanisms of PSII to prevent excessive electron-transfer reactions.

1Pos259 Energy gap dependence for the exciton relaxation rate using Time-dependent renormalized Redfield theory

Akihiro Kimura (*Graduate School of Science, Nagoya Univeristy*)

Study about the photosynthetic antenna is focused on the long-lived exitonic coherence at room temperature. To analyze it, we constructed the rate formula for exciton relaxation by time-dependent renormalized Redfield theory (TRRT) [1,2].

Since TRRT is only expressed for diagonal transition in a reduced density matrix, we extend it to analyze the off-diagonal transition.

In this presentation, we first introduce the detail of the formalism [3], and next discuss the applicability of TRRT with the energy gap dependencies of the relaxation rate by using TRRT and modified Redfield theory.

References

[1] A. Kimura, The 53rd Annual Meeting of the Biophysical Society of Japan, 2Pos182.

[2] A. Kimura, Chem. Phys. Lett. 645 (2016) 123

[3] A.Kimura, submitted to Chem. Phys. Lett.

1Pos260 Photozipper-DNA 複合体平衡の定量的モデル

Quantitative modeling of the equilibria among Photozipper-DNA complexes

Yoichi Nakatani, Osamu Hisatomi (*Grad. Sch. Sci., Osaka Univ.*)

Photozipper (PZ) is a blue light-regulated basic leucine zipper (bZIP) module consisting of a bZIP and a light-oxygen-voltage-sensing (LOV) domains. In this study, we quantitatively analyzed the DNA-binding of PZ and its mutants by electrophoretic mobility shift assay in the dark and light states. The half-maximal effective concentration (EC50) of PZ for the target sequence was estimated to be ~ 40 nM in the light state, which was more than 10-fold smaller than that in the dark state. Then, we estimated the dissociation constant (Kd) of each PZ complex, and constructed a model representing the equilibria among PZ and PZ-DNA complexes. Our model suggested the key role of the light-induced dimerization on the affinity change of PZ for the target sequence.

1Pos261 光制御型 bZIP モジュール Photozipper の構造変化の変異体解析

Mutational analyses of the conformational switching of a light-regulated bZIP module, Photozipper

Osamu Hisatomi (*Graduate School of Science, Osaka University*)

A synthetic gene encoding a bZIP domain and a LOV domain of aureochrome-1 was inserted into an expression vector, and the recombinant protein named Photozipper (PZ) was isolated from *E. coli* cells. Upon blue light (BL) illumination, LOV domain undergoes a reversible conformational change, which induces dimerization and subsequent binding of PZ to the target sequence. To elucidate the molecular mechanism of PZ, we substituted amino acids in the LOV domain and investigated the dimerization and DNA-binding of PZ mutants. Our results suggested that the conformational rearrangement of hydrophobic residues on the β sheet surface of LOV core induced the dimerization of PZ.

1Pos262 水晶微量天秤による光制御型 bZip モジュール photozipper の DNA 結合の解析

The DNA-binding of a light-regulated bZIP module, photozipper, analyzed by quartz crystal microbalance

Samu Tateyama, Osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

Photozipper (PZ) is a light-regulated basic leucine zipper (bZIP) module composed of a bZIP domain and a light-oxygen-voltage-sensing domain of aureochrome-1. Blue light induces the dimerization and the subsequent binding of PZ to the target DNA sequence. In this study, we attempted to quantify the DNA binding of PZ and its site-directed mutants in which Asn131 in the basic region were substituted to Ala, Ser and Gln. From the analyses of quartz crystal microbalance, the half maximal effective concentration (EC_{50}) of PZ to the target sequence was estimated to be ~ 250 nM in the presence of 200 mM KCl. The EC_{50} was consistent with that obtained from electrophoretic mobility shift assay, and Asn131 was suggested to play a crucial role on its binding to DNA.

1Pos263* 真正細菌のポンプ型ロドプシンの機能転換およびその分子メカニズムについての研究

Functional conversion of eubacterial pump rhodopsins and the investigation of the molecular mechanism

Yurika Nomura[1], Keiichi Inoue[1,2], Shota Ito[1], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*JST PRESTO*)

Light-driven ion pump rhodopsins are classified into proton, chloride and sodium pumps, whose characteristic motifs are DTD(E), NTQ and NDQ, respectively. These motifs must be important for their functions, but, there could be additional residue(s) determining functional differences. Here, we attempted the functional conversions of pumps by mutations of the motifs and additional residues to identify critical amino acids for each function. As the result, the function of some mutants were successfully converted, whereas the others were not. We also measured flash photolysis of these mutants. Only functionally converted mutants show similar photoreactions to natural pumps. We'll discuss molecular mechanism of successful and unsuccessful functional conversions.

1Pos264 光駆動内向きプロトンポンプの発見
Natural light-driven inward proton pump

Keiichi Inoue[1,2], Shota Ito[1], Yoshitaka Kato[1], Yurika Nomura[1], Mikihiro Shibata[3,4], Takayuki Uchihashi[3,4], Satoshi Tsunoda[1], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*JST PRESTO*, [3]*Faculty Sci., Kanazawa Univ.*, [4]*Bio-AFM Frontier Research Center, Kanazawa Univ.*)

Light-driven outward proton pumps are widely distributed in nature, converting sunlight energy into proton motive force. In contrast, the presence of inward proton pumps is highly unlikely, as it is energetically unfavourable. Here we report that nature also created inward proton pump, named *Poi*XeR, in a deep-ocean marine bacterium. An inward proton transport by light was detected when *Po*XeR was heterologously expressed in *E. coli* cells. In addition, electrophysiological measurements using mouse neuronal cells showed inward photo-current over a wide-range of membrane potential independent from outer pH. Various spectroscopic and high-speed AFM measurements were applied to this new class of rhodopsin function, which will be presented in the poster.

1Pos265 電気生理実験により解析した光駆動型ナトリウムポンプの輸送機構
Transport mechanism of NaRs studied by electrophysiology measurement

Yuko Kozaki[1], Satoshi Tsunoda[1], Keiichi Inoue[1,2], Rei Abe-Yoshizumi[1], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*JST PRESTO*)

NaRs are a new class of microbial rhodopsin family which actively transport sodium and proton depending on ionic condition. KR2 is the first NaR isolated from a flavobacteria, *Krokinobactar eikastus*, whose crystal structure was determined recently.
Many NaRs possess a sodium binding pocket at the extracellular side, whereas the role of the sodium binding is unknown. To address this, we studied the ion transport activity of NaRs by electrophysiology, which can control different ionic concentrations between cytoplasmic and extracellular sides. We found that the activity is dependent on the extracellular [Na^+], suggesting that two different pumping modes are switched by the sodium binding in the pocket.

1Pos266 An index to select homologous sequences with the same functional region

Shoichiro Kato[1], Hiroyuki Toh[2], Wataru Nemoto[1] ([1]*Life Sci. & Eng., Grad. Sch. of Sci. & Eng*, [2]*Dept. Biomed. Chem., Scl. of Sci. & Tech., Kwansei Gakuin Univ.*)

One of the strategies to predict functional regions requires a structure and a set of homologous sequences. A certain degree of sequence divergence in the sequence set is effective to calculate residue conservation. However, the development of a method to select homologous sequences appropriate to calculate residue conservation has not been sufficiently addressed. An objective method to select appropriate homologous sequences is desired. We developed an index [1] to select the sequences with the same functional region from the set of homologous sequences. We will introduce a server to predict functional regions by using the index, and the extension of the index to another bioinformatics problem, homology modeling.
[1] BMC Structural Biology. 2012;12,11

1Pos267 GWAS データによる疾患関連複合体モデルの予測
Prediction of disease related supramolecule models using GWAS data

Toshiyuki Tsuji[1,2], Atsushi Hijikata[1], Takao Yoda[1], Tsuyoshi Shirai[1] ([1]*Nagahama Institute of Bio-Science and Technology*, [2]*MITA International School*)

We have developed the Sugorok method, which was reported in last annual meeting BSJ 2015, to predict and model the structures of supramolecules in a biological network by combining structural data of the Protein Data Bank and interaction data in IntAct databases.
In this presentation, we have attempted to construct the disease related supramolecule models using genome-wide association study (GWAS) data. GWAS data was mapped on the constructed models, and when the mutants from a GWAS were mapped at least two subunits in a model, it was judged as a disease related model. As a result, over five hundreds models were predicted to be disease related. The most frequent GWAS subjects assigned to the models were Urinary metabolites, Type 2 diabetes, and Coronary heart disease.

1Pos268 Predictions of cancer causing mutations that potentially affect GPCR-GPCR interaction

Shunsuke Fujishiro[1], Vachiranee Limviphuvadh[2], Sebastian Maurer-Stroh[2], Yoshihiro Yamanishi[3], Hiroyuki Toh[4], Wataru Nemoto[1] ([1]*Life Sci. & Eng., Grad. Sch. of Sci. & Eng., Tokyo Denki Univ.*, [2]*BII, A*STAR.*, [3]*Med. Ins. of Bioreg., Kyushu Univ.*, [4]*Dept. Biomed. Chem., Sci. & Tech., Kwansei Gakuin Univ.*)

Somatic hotspot mutations that are associated with cancers are frequently found in GPCRs. Their cancer causing molecular mechanisms has not been clarified, yet. A number of these mutations may cause cancers through promoting or inhibiting interactions between GPCRs. Previously, we predicted GPCR-GPCR interaction pairs among 828 wild type GPCRs by GGIP [1]. In order to examine the effect of the mutations on GPCR-GPCR interaction, we investigated if the prediction results would change when the mutant sequence is used for the prediction. As a result, we found 620 cancer causing mutations whose prediction results are different from those of their wild types. We discuss several mutations that cause cancers by affecting GPCR-GPCR interaction.
[1]. Proteins. 2016 (in press).

1Pos269 ベクトル表現化したアミノ酸残基のマッチングによるタンパク質ーリガンド結合予測
A new approach for protein-ligand binding predictions based on matching of vector-represented amino acid residues

Atsushi Hijikata, Masafumi Shionyu, Tsuyoshi Shirai (*Nagahama Inst. Bio-Sci. Tech.*)

Predicting potential ligands and binding sites of uncharacterized proteins is one of the most challenging tasks in bioinformatics. We present a new approach for fast predictions of ligand type and binding sites of proteins based on a docking simulation using a matching of vector-represented amino acid residues to those empirically defined. We tested the presented method to discriminate various ligands on “uncharacterized” protein targets. For each of the target proteins, 33 different types of ligands were examined. When the ligand(s) of top-ranked and within top 3 were picked up, they were correct (actual ligand type) for 71 (14.1%), and 147 (29.2%) out of 503 targets, respectively. The results demonstrated a high performance of the method in predicting ligand types.

1Pos270 β-Trefoil タンパクのフォールディングに重要な残基に関する残基間平均距離統計に基づく解析

Analysis of residues significant for folding of beta-trefoil proteins based on the inter-residue average distance statistics

Takuya Kirioka, Takeshi Kikuchi (*Dept. of Bioinfo., Col. Life Sci., Ritsumeikan Univ.*)

It is an important problem how information about protein folding is encoded in amino acid sequence. A beta-trefoil fold protein consists of 3-repeating subdomains with a pseudo 3-fold symmetry structure and has very similar structures, but their sequences have no significant homology. In this study, we try to identify significant regions and residues for folding of beta-trefoil proteins from the prediction of compact region by contact map based on inter-residue average distance statistics (Average Distance Map), the prediction of high interacting residues (F value analysis) and conservation of hydrophobic residues. We also speculate the folding pathway of a beta-trefoil protein based on the present results.

1Pos271* 多剤認識転写因子 LmrR における薬剤分子認識機構の計算化学的解析

Computational study on the mechanism of multidrug recognition by a transcriptional repressor LmrR

Kazuho Cryershinozuka, Tadaomi Furuta, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech.*)

We focused upon a transcriptional repressor, LmrR, which is a cause of multidrug resistance in Lactococcus lactis. According to an experimental study, LmrR recognizes various drugs by an entropically driven binding process. Here, to elucidate the underlying mechanism of such an interesting drug-binding, we performed MD simulations for three kinds of LmrR-drug complexes and analyzed their binding free energies by using a MM/3D-RISM method. As a result, it was found that the hydration entropy is a main contributor of binding, and interestingly protein's fluctuation is increased by the binding of a drug molecule, especially in a hypothetical DNA-binding site. It was inferred that the dissociation of LmrR from DNA is triggered by the increase of such a protein fluctuation.

1Pos272 Lysozyme superfamily のアミノ酸配列解析によるフォールディング領域予測

Folding region predictions by amino acid analysis of lysozyme superfamily proteins

Takuto Nakashima, Michiro Kabata, Takeshi Kikuchi (*Dept. of Bioinfo., Col. Life Sci., Ritsumeikan Univ.*)

The aims of this research are to infer folding mechanism of lysozyme superfamily and discuss whether common folding core exists. In this research, we treated C-type lysozyme proteins and superfamily proteins of C-type (I-type, G-type, L-type). In order to achieve our aims, we used the methods based on the inter-residue average distance statistics in addition to the standard bioinformatics tools. As a result of our analyses, the existence of structurally common folding core is detected, and the core includes conserved hydrophobic residues which indicate high contact frequency. In other words, our research showed that the conservation of the folding mechanism of common structural elements among evolutionally distant lysozyme superfamily.

1Pos273 二次構造順序の変化によって起こる蛋白質フォールドの多様化

Loop connectivity change drives protein fold divergence

Shintaro Minami[1], George Chikenji[2], Motonori Ota[1] ([1]*Grad. Sch. of Comput. Sci., Nagoya Univ.*, [2]*Grad. Sch. of Eng., Nagoya Univ.*)

Protein evolution gives extend to the protein fold space. Several mechanisms of evolutionary fold change (indel or circular permutation (CP)) have been studied comprehensively. However, fold change is not limited to them, and other types such as β-swap or β-flip, in which the loop connectivity is changed, still remain to be unexplored. We investigated connectivity change shown in the homologous proteins. The connectivity changes were found in 8% of homologous groups and it can be generally classified into 4 types; CP, swap (SW), flip (FL), and complicated shuffling (CM). Most of the CM can be factorized into 2-4 simple changes (CP, SW, FL) implying evolutionary accumulation of the changes. These results shed new light for understanding of the fold evolution.

1Pos274 タンパク質の立体構造とアミノ酸配列間の疎水性の関係

The relationship between hydrophobicity in amino acid sequence and three-dimensional structure of a protein

Kohei Ohnishi, Takeshi Kikuchi (*Dept. of Bioinf., Col. of Life Sci., Ritsumeikan Univ.*)

Bioinformatics techniques have been developed to compute a protein structures from amino acid sequences. We have empirically believed that globular protein forms a hydrophobic core that has important roles for the structure formation.

In this study, we performed more advanced sequence analysis by combining their empirical knowledge and Average Distance Map (ADM), and examined the relationships between amino acid sequences and three dimensional structures. We treated a several family from each superfold in CATH and focused on sites with high hydrophobic appearance frequency in family. We infer the residues significant for folding of the family. The results will be presented at the society.

1Pos275 Role of interdomain communication in pacemaking circadian rhythm studied by a single molecule model of KaiC

Shota Hashimoto, Sumita Das, Masaki Sasai, Tomoki P. Terada (*Dept. Comput. Sci. Eng., Grad. Sch. Eng., Nagoya Univ.*)

By mixing three cyanobacterial proteins, KaiA, KaiB, and KaiC with ATP in vitro, phosphorylation level of KaiC can be reconstituted in vitro. In addition, it has been suggested that the period length of KaiC phosphorylation cycle is determined by the ATPase activity of KaiC without KaiA and KaiB, but the mechanism underlying this correlation has not been revealed yet. By assuming the interdomain structural communication between the N-terminal C1 domain and the C-terminal C2 domain of KaiC, we constructed a structure-based model of KaiC single hexamer and carried out a stochastic simulation using Gillespie algorithm. We show the requirements for the correlation and discuss the mechanism of temperature compensation of circadian oscillation.

1Pos276* A stochastic simulation study on the correlation between circadian oscillation and ATPase activity of KaiC hexamer

Sumita Das, Shota Hashimoto, Tomoki P. Terada, Masaki Sasai (*Department of Computational Science and Engineering, Nagoya University,Nagoya*)

KaiC is a hexameric protein, and each of its six subunits comprises two ATPase domains: CI and CII. It has been reported that the ATPase activity of KaiC is correlated with the oscillation frequency of the KaiABC system. Here, we developed a model of KaiC single hexamer and examined the coupled oscillations of ATP hydrolysis and phosphorylation/ dephosphorylation. We found that the correlation between the ATPase activity in CI and the circadian frequency in CII is mediated by the structure change in each CI-CII subunit in the model. Based on this result, we discuss how the temperature insensitivity of the ATPase rate establishes the temperature compensation phenomenon of the circadian oscillation.

1Pos277 数理モデルによる心筋細胞の集団効果の解析 Community effect of cardiomyocytes in beating rhythms is ruled by stable cells

Tatsuya Hayashi[1], Tetsuji Tokihiro[1,2], Hiroki Kurihara[2,3], Fumimasa Nomura[4], Kenji Yasuda[2,5] ([1]*Grad. Sch. Math. Sci., The Univ. of Tokyo*, [2]*JST, CREST*, [3]*Grad. Sch. Med., The Univ. of Tokyo*, [4]*Inst. Biomat. Bioeng., Tokyo Medical and Dental Univ.*, [5]*Fac. Sci. Eng., Waseda Univ.*)

Community effect of cardiomyocytes is investigated in silico by changing number and features of cells as well as configurations of networks. The theoretical model is based on experimental data and accurately reproduces the recent experimental results about coupled two cultured cardiomyocytes that proved the fact that the synchronized beating of two coupled cells is tuned not to the cell with faster beating rate but to the one with more stable rhythm. In a network of cardiomyocytes, beating fluctuation rapidly decreases with increase of the number of cells, almost irrespective of the configuration of the network. The universality of this community effect lies in the fluctuation dissipation theorem in statistical mechanics.

1Pos278 Discreteness-induced transition in multi-body reaction systems

Yohei Saito, Yuki Sughiyama, Tetsuya Kobayashi (*IIS, Univ. Tokyo*)

Qualitative change in dynamical behaviors owing to finite system size attracts much attention in stochastic chemical reactions. Noise-induced transition is a well-known mechanism of this change, where increase in multiplicative noise intensity due to smaller system size induces change in stationary distribution. Although finite system size not only increases noise but also emphasizes the state space discreteness, the latter effect has been overlooked. In this work, we identify another mechanism, “discreteness-induced transition”, in a simple 1- and 3-body reaction system, and derive a sufficient condition under which it arises. Furthermore, we show that the critical size of discreteness-induced transition can become quadratically large in multi-body reaction systems.

1Pos279 Probability Eddy currents in stochastic gene expression dynamics in eukaryotes

Bhaswati Bhattacharyya[1], Masaki Sasai[1,2] ([1]*Department of Computational science and engineering, Nagoya University*, [2]*Department of Applied Physics, Nagoya University*)

Eukaryotic genes are regulated not only by the transcription-factor binding/ unbinding (TF-b/u) but also by epigenetic mechanisms including histone modification (Hm). We develop a model to describe a coupled dynamics of TF-b/u and Hm as stochastic transitions among discrete states. Coupled dynamics of the faster TF-b/u and the slower Hm gives rise to two basins of attraction for the gene activity of a self-activating gene for a broad range of parameters. By using a mean-field representation of TF-b/u, Hm, and gene-activity, we investigate the flow of probability in the three-dimensional space. We analyze the features of eddy current of the probability flow around these basins to analyze the effects of timescale separation in processes of eukaryotic gene switching.

1Pos280 集団増殖系における定常状態熱力学 Steady State Thermodynamics in Population Dynamics

Yuki Sughiyama, Tetsuya J. Kobayashi (*IIS, Univ. Tokyo*)

Methods of thermodynamics can contribute to solving problems in evolution of microorganisms, because there exists a mathematical similarity between population dynamics and statistical physics. In this study, we evaluate population growth (expansion rate of population size) by using a framework of steady state thermodynamics (SST).
In this talk, according to the SST framework, we decompose the population growth into two parts, housekeeping and excess growths, and we construct a Clausius inequality in population dynamics, which gives the upper bound of the excess growth. The equality is achieved in quasistatic environmental changes. We also clarify that this bound can be evaluated by “lineage fitness”, which is an experimentally observable quantity.

1Pos281 上皮陥入過程における三次元多細胞動態の力学制御機構 Mechanical regulatory mechanism of 3D multicellular dynamics during epithelial invagination

Satoru Okuda, Mototsugu Eiraku (*RIKEN Center for Developmental Biology*)

During morphogenesis, various cellular activities are spatiotemporally regulated on the protein regulatory background to construct the complicated, three-dimensional (3D) structures of organs. To approach the mechanisms underlying 3D multicellular constructions, we have developed the general-purpose method using 3D vertex model. The model enables the quantitative simulation of morphogenesis on the basis of single-cell mechanics, with complete control of various cellular activities such as cell contraction, growth, rearrangement, division, and death. By combining computational simulations with experimental observations using embryonic stem cell-derived tissues, we have analyzed the regulatory mechanism of the epithelial invagination during the 3D optic-cup formation.

1Pos282 Modeling folding of epithelial cell sheets

FuLai Wen[1], YuChiun Wang[2], Tatsuo Shibata[1] ([1]*RIKEN Quantitative Biology Center*, [2]*RIKEN Center for Developmental Biology*)

Epithelial sheets comprising interconnected polarized cells serve as barriers to protect underlying tissues and undergo folding to form organ structures. While numerous molecular machineries are identified for epithelial folding, the mechanical mechanisms underlying cell deformation and tissue bending still remain unclear. Based on a vertex model, we systematically analyze the change of cell shape and sheet morphology due to the modulation of each cell surfaces. In particular, we find that the differential basal-lateral mechanics within sheets can initiate folding, producing morphology distinct from those induced by apical constriction, thus revealing the importance of mechanical modulation at the basal-lateral surface.

1Pos283 等方的なアクチンミオシン細胞骨格におけるモーター誘起応力に関する理論
Theory on motor-induced stress in an isotropic actomyosin cytoskeleton

Tetsuya Hiraiwa (*Dept. Sci., Univ. Tokyo*)

We focus on active mechanics of a cortical cytoskeleton. Mechanical properties of a cortical cytoskeleton govern motor-induced contractility of a cell, which plays crucial roles in dynamic cellular behaviors like cytokinesis and cell migration.
In the presentation, we will explain our theoretical work on motor-induced contractility in a cortical cytoskeletal network [1]. We will propose a mechanical model of an isotropic stiff F-actin network with crosslinkers and study how and when the contractility occurs. In particular, since a cortical cytoskeleton in a living cell should be flowable, we consider the network in which there are few amount of crosslinkers and/or crosslinkers and F-actins can turn over.

[1] T. Hiraiwa and G. Salbreux, PRL, 116, 188101 (2016).

1Pos284 Phase-field simulations of the basic cell-cell effects of adhesion and chemoattractant on multi-cellular interaction

Daisuke Imoto[1,5], Satoshi Sawai[1,2,3], Shuji Ishihara[4] ([1]*Dept. Basic Sci., Grad. School of Arts and Sci., Univ. of Tokyo*, [2]*Research Ctr for Complex Systems Biology, Univ. of Tokyo*, [3]*JST PRESTO*, [4]*School of Sci. Eng., Meiji Univ.*, [5]*National Research Institute of Police Science*)

Phase-field has an advantage over other formalisms of being able to simulate cell morphology, motility, and collective cell movements with continuously deforming boundaries. However this very feature makes it difficult to describe close-contacts between the cells often observed in tissues and group of migratory cells. Here, using a generalized 2-cell system, we numerically examined combinations of two types of volume exclusion and three types of adhesion formulations, and clarified conditions that allow co-occurrence of long-time scale cell-cell attachment and coordinated movements. The formalisms were further coupled with a directional sensing model to study how adhesion and chemotaxis must be coordinated to give rise to behaviors observed in Dictyostelium.

1Pos285 様々な回転する自走粒子の集団運動
Collective motion of various kinds of rotating self-propelled particle

Ken Nagai[1], Yutaka Sumino[2], Chate Hugues[3,4], Kazuhiro Oiwa[5,6], Takuma Sugi[7], Hideo Iwasaki[8] ([1]*Sch. Mater. Sci., JAIST*, [2]*Dep. Appl. Phys., Tokyo Univ. Sci.*, [3]*CEA-Saclay*, [4]*Beijing Comp. Sci. Res. Ctr.*, [5]*Adv. ICT Res. Inst., NICT*, [6]*Grad. Sch. Sci., Univ. Hyogo*, [7]*Mol. Neurosci. Res. Ctr., Shiga Univ. of Med. Sci.*, [8]*Sch. Adv. Sci. Eng., Waseda Univ.*)

There are various kinds of self-propelled particle that keeps its rotation rate for a long time such as an E. coli close to wall, and a microtubule running on glass in an in vitro motility assay. Using an agent-based model, we elucidated the role of the memory of rotation rate in collective motion of self-propelled particles. We found that a hexagonal lattice of vortices, vortices located randomly, global nematic phase, and density soliton waves were formed only when the rotation rate was kept for a while. The same phases as in the model formed in the collective motion of microtubules running on a glass, C. elegans and Geitlerinema on agar in the real world, which indicates that our model catch the universal properties of collective motion.

1Pos286 蛍光分光法による時計タンパク質 KaiC の動的構造変化の解析
Spectroscopic characterization of the conformational change of the cyanobacterial clock protein KaiC

Atsushi Mukaiyama[1,2], Jun Abe[1], Yoshihiko Furuike[1,2], Eiki Yamashita[3], Takao Kondo[4], Shuji Akiyama[1,2] ([1]*IMS, CIMoS*, [2]*SOKENDAI*, [3]*IPR*, [4]*Naogya Univ.*)

KaiC is composed of an N-terminal C1 and a C-terminal C2 domain and forms double-ring hexamer. A cyclic phosphorylation/desphosphorylation at the C2 ring proceeds with a period of ~24h in the presence of other two kinds of clock proteins, KaiA and KaiB, whereas recent studies show that the C1 ring is also functionally important. To reveal a dynamic aspect of the C1 ring, we examined structural transitions of KaiC and its mutant carrying a fluorescent probe in its C1 ring using fluorescence spectroscopy. The C1 ring undergoes a tiny conformational change through fluorescently-distinct conformations that are likely essential for a cyclic conformational change of KaiC.

1Pos287* 高速原子間力顕微鏡によって明らかにする Kai タンパク質間の動的相互作用のリン酸化状態依存性
HS-AFM images reveal dynamic interaction between Kai proteins dependent on phosphorylation states of KaiC

Shogo Sugiyama[1], Tetsuya Mori[2], Takayuki Uchihashi[1,3], Johnson Carl H.[2], Toshio Ando[1,3] ([1]*Dept. of phys., Kanazawa Univ.*, [2]*Dept. of Biol. Sci., Univ. Vanderbilt*, [3]*Bio-AFM FRC., Kanazawa Univ.*)

The circadian rhythm in cyanobacteria is essentially generated by an oscillator composed of three Kai proteins (KaiA, KaiB and KaiC). The Kai system is very unique because the self-sustainable oscillation of KaiC phosphorylation can be reconstructed in vitro only by incubating KaiC with KaiA, KaiB and ATP. However, detailed molecular mechanism is still unclear. Here, we observe the interaction between KaiC and KaiA/B by using high-speed AFM to investigate the interaction dynamics. HS-AFM image clearly shows that KaiA and KaiB bind to KaiC and dynamic interaction between Kai proteins dependent on phosphorylation states of KaiC. We discuss our recent findings about dynamic interactions among the Kai proteins that may significantly impact on the oscillatory stability.

1Pos288 生命システムの振動現象における頑健性と可塑性の互恵的関係

Reciprocity between robustness and plasticity in biological oscillators

Tetsuhiro S. Hatakeyama, Kunihiko Kaneko (*Department of Basic Science, The University of Tokyo*)

Circadian clocks exhibit the robustness of period and plasticity of phase against environmental changes such as temperature and nutrient conditions. However, it is unclear how both are simultaneously achieved. By investigating distinct models of circadian clocks, we demonstrate reciprocity between robustness and plasticity: higher robustness in the period implies higher plasticity in the phase, where changes in period and in phase follow a linear relationship with a negative coefficient. Generality of reciprocity in clocks with the adaptation mechanism is confirmed with theoretical analysis of simple models. We also discuss that reciprocity between robustness and plasticity can be obtained in a wide variety of biological oscillators not limited to the circadian clock.

1Pos289* 自律的な振動運動を示す微小管リング状集合体

Mechanical Oscillation of Dynamic Microtubule Rings

Masaki Ito[1], Kabir Arif Md. Rashedul[2], Md. Sirajul Islam[1], Daisuke Inoue[2], Shoki Wada[1], Kazuki Sada[1,2], Akihiko Konagaya[3], Akira Kakugo[1,2] ([1]*Grad. of CSE, Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*, [3]*DIS, TITECH*)

Mechanical oscillation is a unique phenomenon observed in living systems, which emerges from a wide range of self-assembled structures, and plays important roles in many biological processes. Although many efforts have been devoted to demonstrate the mechanical oscillation of organized structures produced through self-assembly in vitro, it has rarely been documented. We report the mechanical oscillation of ring-shaped structures, composed of multiple microtubule (MT) filaments, obtained through energy dissipative self-assembly of MT filaments beneath an air-buffer interface. The MT rings exhibit autonomous oscillation manifested through periodic changes in the size and shape. In the presentation, proposed mechanism of the oscillation of the MT rings is discussed.

1Pos290* キネシン駆動微小管のパターン形成と局所相互作用

Configuring Dynamic Patterns of Microtubules Driven by Kinesins

Sakurako Tanida[1], Ken'ya Furuta[2], Kaori Nishikawa[1], Hiroaki Kojima[2], Masaki Sano[1] ([1]*Graduate School of Science, The University of Tokyo*, [2]*National Institute of Information and Communications Technology*)

It is known that a variety of individual gather and exhibit characteristic dynamical patterns. In the context of non-equilibrium physics, these are regarded as an example of collective motions of active particles with local interactions. To investigate the relation between patterns and interactions, we observed collective patterns of microtubules driven by kinesins on flat glass surface. As the kinesin-density increases, microtubules adhere to the surface more closely, and this strengthens excluded volume effect in a collision interaction. Using this method, we found two types of patterns which have different sizes and orders and this suggests that those can be explained from a single mechanism with excluded volume effect.

1Pos291* 離散的な相互作用を行う振動子ネットワークの解析

Analysis of nonlinear oscillator network with discrete interactions

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Networks of the nonlinear systems have been studied as a biological population model. Recent studies enable the analysis of the directed and undirected networks of the nonlinear systems. However such studies assume that interactions are continuous.
Our objective is to develop the networks of nonlinear oscillator systems with discrete interactions and to analyze them. We use the fusion and fission of water-in-oil droplets to create the discrete interactions. We numerically analyzed the nonlinear oscillator system with discrete interactions. We believe that our study will be applied to a wide range of studies in biophysical models of multicellular systems such as neural systems.

1Pos292* 混み合い状況下におけるマイクロ粒子のラチェット輸送

Ratchet transport of microparticles in crowded conditions

Masayuki Hayakawa[1], Yusuke Kishino[2], Masahiro Takinoue[1,2,3] ([1]*Dept. of Comput. Intell. and Syst. Sci., Tokyo Tech.*, [2]*Dept. of Engineering, Tokyo Tech.*, [3]*Dept. of Computer Science, Tokyo Tech.*)

Studies of transports in a crowded condition have been attracting attention for the understanding of the cellular transports in a molecule-crowded cells. Previous studies have focused on observations of high-density kinesins on single microtubules. However, because a whole picture of molecular motors is not clear yet, it is hard to extract essences of directed transports. Thus, a study of simple experimental model with the crowdedness is desired. Here we report the experiments in which microparticles are moved by steady electric fields. Only in the crowding area, we found that particles were collectively transported due to the pushing by other particles. We believe that our study will contribute to develop the study of the effective transport in the crowded condition.

1Pos293 「ゆらぎ」が創り出す「秩序構造」：細胞の混雑環境のモデリング

Fluctuation Creates Exotic Spatial-Order: Verification with a Simple Crowding Cellular-Model

Soutaro Oda[1], Chwen-Yang Shew[2], Kenichi Yoshikawa[1] ([1]*Faculty of Life and Medical Sciences, Doshisha University*, [2]*Department of Chemistry, CSI, City University of New York*)

Fluctuations are ubiquitous in any size-scale systems, and an investigation of confined particles under fluctuations is relevant to how living cells on the earth maintain their lives. Inspired by biological cells, we conducted the model experiment through a very simple fluctuating system containing one or several large spherical granular particles and multiple smaller ones confined on a cylindrical dish under vertical vibration. We found a universal behavior that large particles preferentially locate in cavity interior due to the fact that large particles are depleted from the cavity wall by small spheres under vertical vibration through the real-world simple experiment. We would like to propose a novel hypothesis in terms of entropy.

1Pos294 置かれた環境を感応する自発運動系：化学的非平衡性により駆動する生物らしさを示す実空間モデル

Smart response of chemically driven self-motile object: Real-world modeling

Shiho Sato[1], Hiroki Sakuta[2], Kenichi Yoshikawa[1,2] ([1]*Facul. Life Med. Sci., Univ. Doshisha*, [2]*Grad. Sch. Life Med. Sci., Univ. Doshisha*)

We report a simple artificial model of self-propelled active matter. We found the occurrence of spontaneous regular motion for a nitrobenzene droplet floating on an aqueous phase. Depending on the initial position of the droplet in a vessel, model of self-propelled motion exhibits either reciprocal or rotational motion. When the droplet put at the center, back and forth repetitive motion is caused. Whereas, when the initial position is taken near the edge of the vessel, circular orbital motion is generated. We report our experimental results on the dynamic behavior of nitrobenzene droplet by changing the chemical composition of the aqueous phase. The mechanism of the self-propelled motion will be discussed in terms of interfacial instability, i.e., the Marangoni effect.

1Pos295 Shot noise free number and brightness 解析法による生細胞内グルココルチコイド受容体二量体化過程の時空間分布解析

Spatio-temporal distribution analysis of glucocorticoid receptor dimerization in cells by shot noise free number and brightness analysis

Ryosuke Fukushima[1], Jotaro Yamamoto[2], Masataka Kinjo[2] ([1]*Grad. Sch. Life Sci., Hokkaido Univ.*, [2]*Faculty of Adv. Life Sci., Hokkaido Univ.*)

Glucocorticoid receptor (GR) associates the nuclear receptors superfamily. GR translocates from cytoplasm to nucleus by stimulated of ligands from outside of the cell. This is well-known scheme, however, it is still unclear whether GRs are monomers or dimers when these translocates to nucleus. In this study, we aimed to reveal when and where GR dimerize in cell.
To visualize the movement and the dimerization of GR, we developed a new method which gives the number of molecules and molecular brightness in each pixel of confocal fluorescence microscopy images by using two detector system. New model on the GR nuclear translocation will be proposed according to experimental results. This would be the first step towards understanding of GR regulatory mechanism in living cell.

1Pos296 蛍光異方性を用いた免疫センサの試作

Development of Fluorescence Anisotropy Immunosensor

Seiichi Suzuki, Sena Hasegawa, Maki Takagi, Takuya Ito, Toshinori Kojima (*Faculty Sci. Tech., Seikei Univ.*)

Immunosensor is highly sensitive and specific detector of antigen. But it is difficult to apply for mixture of biological materials, because of nonspecific adsorption on sensor surface. In this study, we detected the binding of antibody to specific antigen directly with fluorescence anisotropy method. Fluorescence anisotropy is anisotropy of fluorescence polarization in different directions. Fluorescence anisotropy reflects the rotational motion of fluorophore or molecule on which fluorophore is attached. Antigen binding on antibody molecule reduces fluorescence anisotropy without disturbance of nonspecific surface adsorption. With this method, antigen binding on surface immobilized labeled antibody molecule was detected with high sensitivity.

1Pos297* 実時間選択的回収による免疫細胞の１細胞遺伝子発現解析

Single cell gene expression analysis of stimulated immune cells with real-time selection

Yumiko Tanaka[1], Yoshitaka Shirasaki[1,2], Mai Yamagishi[1,2], Kaede Miyata[1], Nobutake Suzuki[1], Osamu Ohara[2], Kazuyo Moro[2], Sotaro Uemura[1] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*IMS., Riken*)

Stimulated immune cells secrete cytokines to regulate the immune systems. Our previous data showed cytokine mRNA levels driven by the stimulation, leading to the cell-to-cell heterogeneity. To clarify what factors generate the heterogeneity, we combined our real-time secretion imaging system with a single-cell selectively picking technique to collect cells in a specific secretion state, and we analyzed cytokine mRNA levels of these picked cells whose secretion state were in synchrony. In contrast to cells picked regardless of secretion timing, cells picked 30 min after the observation of the beginning of secretion showed homogeneous cytokine mRNA levels. Our results suggest that the cell-to-cell heterogeneity mainly originates from fluctuations in response timing.

1Pos298 カップ形状 AFM チップを用いた簡便な細胞間相互作用計測法の開発

Easy Measurement of Cell-Cell Interactions Using Cup-Shaped AFM Chip

Hyonchol Kim[1], Ayana Yamagishi[1], Miku Imaizumi[2], Chikashi Nakamura[1,2] ([1]*Biomed. Res. Inst., AIST*, [2]*Grad. Sch. Eng., Tokyo Univ. Agric. Technol.*)

A new method to measure cell-cell interactions using an AFM chip on which cup-shaped hemisphere was immobilized was proposed. For the fabrication of the cup-shaped particle, polystyrene microsphere templates were coated with nickel and the templates were removed by burning. The cup was picked up using micromanipulator and put it on the end of AFM cantilever through epoxy resin. The fabricated AFM chip (referred as "cup-chip") was approached on a cell which was not attached strongly on a substrate, then, the cell was captured to inner cavity of the cup-chip. The cup-chip was approached on another cell and cell-cell adhesive forces were measured. These results indicate cup-chip is convenient and universal tool to pick up a cell and to measure cell-cell interactions.

1Pos299 アンルーフ法を用いた水溶液環境下における細胞内骨格のＡＦＭイメージング

An Unroofing Method to Observe the Cytoskeleton Directly at Molecular Resolution Using Atomic Force Microscopy

Eiji Usukura[1], Akihiro Narita[1], Akira Yagi[2], Shuichi Ito[2], Jiro Usukura[1] ([1]*Grad. Sch. Sci., Univ. Nagoya*, [2]*Olympus Co., Ltd.*)

An improved unroofing method enabled the cantilever of an atomic force microscope to reach into inside of cell and thereby to visualize the intracellular cytoskeleton directly at high resolution comparable to conventional electron microscopy. Actin filaments clearly exhibited a short periodicity of as well as a long helical periodicity. The polarity of the actin filaments appeared to be determined by the shape of the periodic striations. Microtubules were identified based on their thickness. Clathrin coats and caveolae were observed on the cytoplasmic surface of cell membranes. The area containing clathrin molecules and their terminal domains was directly visualized. Characteristic ridge structures located at the surface of the caveolae were observed at high resolution.

1Pos300 原子間力顕微鏡による細胞機能と力学特性の単一細胞相関解析法

Atomic force microscopy for single-cell correlation analysis between cellular function and cell mechanical property

Ryosuke Tanaka[1], Yoshikatsu Akiyama[2], Jun Kobayashi[2], Masayuki Yamato[2], Okajima Takaharu[1] ([1]*Grad. Sch. Info. Tech. Univ. Hokkaido*, [2]*Inst. Adv. BioMed. Eng. Sci. Univ. Tokyo Women's Med.*)

It has been recognized that the ensemble-averaged mechanical property of cells is intimately related to their various biological functions [1]. However, such a relationship has not been fully understood at single-cell level due to the lack of nano-mechanical measurement method for a large number of cells. We report an atomic force microscopy (AFM) system that allows measuring cells scattered in an extremely wide range. Using the AFM system with an immunofluorescence technique, we can estimate the correlation between cell mechanical property and cell function at the single cell level. We will show the detailed setup of AFM system and experimental results of primary hepatocyte with the system. [1] Gonzalez-Cruz RD et al., Proc Natl Acad Sci U S A, 2012, 12;109(24)

1Pos301 デジタルマイクロ流体技術によるデジタルバイオアッセイ

Digital bioassay in digital microfluidic platform

Ryohei Kobayashi[1], Sadao Ota[1,2], Hiroyuki Noji[1,3] ([1]*Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*JST, PRESTO*, [3]*ImPACT, JST*)

Digital ELISA has enabled highly sensitive detection of target molecules, but not widely spread yet. In this work, we combined digital bioassay platform with digital microfluidics (DMF). DMF is a technology capable of electrical manipulation of individual microliter droplets. First, we demonstrated the DMF's basic operation of moving and splitting droplets. Next, we confirmed that a solution of fluorescent beads was transported by the DMF system and sealed in microchambers by oil. Finally, we conducted a digital counting experiment. The result showed that this integrated system is able to detect molecules at very low concentration (< several pM). Our device has great potential of enabling sensitive, portable immunoassay and will be useful in the field of medical care.

1Pos302 左右両耳内部における脈波計測

Measurements of Pulse Waves in the Both Ears

Yoshitomi Morikawa (*AIST*)

We have developed a wearable measuring instrument for pulse waves in the ears. The instrument can measure pulse waves in the both ears for a moving person. This remarkable feature has a great worth in various real life scenes. We report the measuring results of pulse waves in the both ears for a person in a sitting state and also slowly moving states.

1Pos303 ラボＸ線光源を用いたＸ線１分子動態観察

X-ray Single Molecule Observations using Laboratory X-ray Generator

Keigo Ikezaki[1], Ken Matsubara[1], Yuhuku Matsushita[1], Jae-won Chang[1], Hiroshi Sekiguchi[2], Yuji Sasaki[1] ([1]*University of Tokyo*, [2]*Spring-8/JASRI*)

Diffracted X-Ray Tracking (DXT) has been developed for obtaining the information of the 3D internal motions of single protein molecules with both high time-resolution and high precision. The weak point in DXT is to use the large facility of synchrotron orbital radiation source. DXT using a small light source is very effective. When using a laboratory X-ray source, it is very clear that the signal to background ratio is better for the monochromatic X-ray diffraction than Laue diffraction using white x-rays. As experimental results using Rigaku FR-D (Cu anode, 50kV, 60mA), we confirmed that correlation between the motions of diffraction spots and time-resolved fluctuations of the diffracted x-ray intensity in each detector pixel.

1Pos304 MALDI 法におけるマトリクスの分光学的解析

Spectroscopic analysis of matrices in ionization process of matrix-assisted laser desorption/ionization

Noritaka Masaki, Shigetoshi Okazaki (*Dept. Med. Spec., Hamamatsu Univ. Sch. Med.*)

Matrix-assisted laser desorption/ionization (MALDI) is a major soft ionization method in mass spectrometry. Chemicals called matrix is a key in MALDI assisting ionization of molecules. Various chemicals are used as matrix, e.g. 2,5-dihydroxybenzoic acid. Recently, improvements of ionization efficiency in MALDI is a big analytical demand to analyze wider variety of molecules and to increase sensitivity for higher spatial resolution analysis. To that end, pretreatment of sample gathers attention. On the other hand, optical characteristics of matrix are almost unknown as they were usually used in an empirical manner. Aiming to transfer laser energy more effectively to improve ionization, we will discuss optical features of matrices and their contribution in ionization.

1Pos305 HbA1c の蛍光相関分析に及ぼすヘモグロビン光吸収の影響

Effects of hemoglobin absorption on fluorescence correlation analysis for HbA1c

Atsushi Matsuo, Yasutomo Nomura, Mayuka Chiba, Misaki Naraoka (*Maebashi Institute of technology*)

FCCS was proposed as a method for diagnosing diabetes with HbA1c. In the clinical examination of HbA1c, light absorption by Hb in the tested solution is inevitable if separation steps are omitted. Thus we examined the effect of Hb on FCCS measurements for HbA1c. As for the green fluorescent antibody, we have previously labeled anti-HbA1c with Alexa Flour 488. The red fluorescent antibody was formed by mixing 100 nM avidin Q-dot 655 conjugate and 100 nM biotinylated anti-Hb. Number of molecules and diffusion constant of both fluorescent antibodies were independent of the coexistence with Hb up to 50 uM which the hemolysate in clinical examination often contained. The immunocomplex formation of two color antibodies to HbA1c antigen was evaluated by FCCS in the hemolysate.

1Pos306* **高速原子間力顕微鏡を用いた癌細胞の核膜孔動態の可視化**
High-speed atomic force microscopy visualization of the nuclear pores dynamics in cancer cells

Mahmoud Shaaban Mohamed[1,2,3], Yosuke Kikuchi[4], Watanabe-Nakayama Takahiro[2], Azuma Taoka[4], Akiko Kobayashi[1,2,3], Masaharu Hazawa[1,2,3], Noriyuki Kodera[2], Takayuki Uchihashi[2], Yoshihiro Fukumori[4], Toshio Ando[2], Richard Wong[1,2,3] ([1]*Cell-Bionomics Research Unit, Kanazawa University*, [2]*Bio-AFM Frontier Research Center, Kanazawa University*, [3]*Lab of Mol. Cell Biol. Institute of Science and Engineering, Kanazawa University*, [4]*Institute of Science and Engineering, Kanazawa University*)

Morphological changes in the size and shape of the nucleus is prevalent in cancer metastasis. Remarkably, all these metastatic signaling pathways must enter the nucleus through a single gatekeeper, the nuclear pore complex (NPC). NPCs modulate nuclear shape and organization and have implicated in tumorigenesis. Visualization of nuclear dynamics in live cancer cells with nanometer resolution under physiological conditions is particularly desired, but puzzling in technical. Here, we show that high-speed atomic force microscopy can be used to visualize dynamic changes in nuclear pores in cancer cells. Our preliminary movies showed dynamic structural changes of the symmetric pores by morphometric analysis in colon cancer.

1Pos307 **高速 AFM による抗体分子の動的観察**
The dynamic behaviors of antibody molecules

Yoko Kawamoto-Ozaki, Norito Kotani, Kumaresan Ramanujam, Aya Murakami, Takashi Morii, Takao Okada (*Research Institute of Biomolecule Metrology Co.,Ltd.*)

Antibodies exhibit mechanisms which increase their affinity to antigens, viz. IgG has flexible hinge region and IgM forms oligomer. To understand the mechanisms in detail, we analyzed antibody in liquid by High-Speed Atomic Force Microscopy (HS-AFM). HS-AFM is the only method which can visualize the dynamic behavior of Ig in liquid.
We present three movies; 1) IgG was observed as Y shape where each two Fab regions were distinguished clearly and hinge regions swung flexibly. 2) In IgM pentamer, five Ig molecules were described. With adding 2-mercaptoethylamine to the pentamer in order to reduce, the pentamer was breaking into monomers gradually. 3) IgM monomer which had been already reduced was observed clearly. We discus about the different features between IgG and IgM.

1Pos308 **高速 AFM によるタンパク質の動的観察に向けた立体パターン基板の作製**
Fabrication of 3D-patterned Substrate as a Platform for HS-AFM Observation of Protein Dynamics

Akane Goto[1], Shin'nosuke Yamanaka[1], Mikihiro Shibata[1,2], Takayuki Uchihashi[1,2], Noriyuki Kodera[1,2], Toshio Ando[2] ([1]*Dept. of phys., Kanazawa Univ.*, [2]*Bio-AFM FRC*)

High-speed atomic force microscopy (HS-AFM) is a powerful tool to visualize dynamic behaviors of biological molecules in the solution. For the HS-AFM observation, flat substrates are usually used to be absorbed target molecules there. However, some biological molecules do not work with native activity on the flat surface due to steric disturbance between molecules and substrate. To expand biological application of the HS-AFM, we need a 3D structural substrate. Here, we made the substrates having a groove patterns in the width and the depth of a few dozen nm. These substrates will enable us to apply the HS-AFM to observe such proteins. In the presentation, we show fabrication process of this substrate and its application to imaging of protein dynamics.

1Pos309 **高速スイッチング蛍光タンパク質と改良された SPoD-ExPAN による超解像イメージング**
Superresolution imaging of live cells by fast photoswitching fluorescent protein and improved SPoD-ExPAN microscopy

Tetsuichi Wazawa[1,2], Yoshiyuki Arai[1,2], Tomoki Matsuda[1,2], Hiroki Takauchi[1], Yoshinobu Kawahara[1,2], Takashi Washio[1,2], Takeharu Nagai[1,2] ([1]*ISIR, Osaka Univ.*, [2]*CREST, JST*)

Previously we developed a highly-biocompatible superresolution microscopy technique using our fast photoswitching fluorescent protein Kohinoor and polarization demodulation/excitation angle narrowing (SPoD-ExPAN) that operated at an illumination power as low as ~1 W/cm^2 in contrast to conventional superresolution techniques (10^2–10^6 W/cm^2). However the image reconstruction algorithm hitherto used in SPoD-ExPAN often produced poorly-presented images. Thereby, an image reconstruction method with structural regularization has been devised so that superresolved images by SPoD-ExPAN are satisfactorily reconstructed. Accordingly, SPoD-ExPAN is able to be exploited to image functional probes such as genetically-encoded Ca^{2+} probe in live cells by time-lapse observation.

1Pos310 **生細胞核内における INO80 クロマチン再構成複合体の 1 分子イメージング**
Single-molecule imaging of the INO80 chromatin remodeling complex in the living cell nucleus

Yuma Ito[1], Masahiko Harata[2], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*Grad. Sch. Agr. Sci., Tohoku Univ.*)

The INO80 complex is an ATP-dependent chromatin remodeler that slides and evicts nucleosomes through its ATPase activity. We examined the dynamics by single molecule tracking analysis of a scaffold subunit Ino80 of the INO80 complex using HILO microscopy. Ino80 molecules were detected in both a nearly-immobile state and a mobile state, and they transited between the two states. This indicates the dynamic association of the INO80 complex with chromatin. The fraction of the immobile state was decreased in a mutant lacking Arp4 and Arp8 binding, but was almost the same in a mutant lacking ATP binding. The finding suggests that the interaction of the complex with chromatin is regulated not by ATP-biding ability but by Arp4- and Arp8-binding ability of the Ino80 subunit.

1Pos311* **アロディニア特異的な痛みに対する鎮痛薬評価系の確立に関する fMRI 研究**
An fMRI study to establish an evaluation system of analgesic agents on allodynia-specific pain

Naoya Yuzuriha[1], Sosuke Yoshinaga[1], Mitsuhiro Takeda[1], Hiroshi Sato[2], Hiroaki Terasawa[1] ([1]*Fac. Life Sci., Kumamoto Univ.*, [2]*Bruker Biospin K.K.*)

Fibromyalgia and neuropathic disorders characterized by chronic pain induce allodynia symptoms. The aim of this study is to evaluate the effects of analgesic agents on an animal model of fibromyalgia, using the fMRI technique. Before and after the administration of analgesic agents, functional data were acquired with an EPI sequence, during which a green laser was used to irradiate the hind paws. The T_2^*-weighted signal intensity analysis successfully revealed that the signal increases upon the laser stimulation were depressed by the analgesic agents. Comparisons of the activation-suppressing effects caused by other analgesic agents with different functional mechanisms will provide an evaluation system for new analgesic agents.

1Pos312 **CLIP-170 phosphorylation mediates repositioning of microtubule-organizing center during T cell activation**

Wei Ming Lim, Yuma Ito, Makio Tokunaga, Kumiko Sakata-Sogawa (*Sch. Life Sci. Tech., Tokyo Inst. Tech.*)

Microtubule-organizing center (MTOC) repositioned to the center of immunological synapse (IS) and drives T cell activation at the initial phase of immune response reaction. However, our understanding for the molecular mechanism of MTOC repositioning towards IS remains limited. We investigated how CLIP-170, a microtubule plus-end protein mediates MTOC repositioning using fluorescence imaging and quantitative analysis. To further unravel CLIP-170 significance, we generated mutants and carried out knockdown experiments. Our studies indicate that CLIP-170 plays a crucial role in MTOC repositioning.

1Pos313 **核小体構成タンパク質動態の１分子イメージング定量解析**
Single molecule imaging and quantitative analysis of Nucleolar-localized protein dynamics

Daiki Matsumoto[1], Yuma Ito[1], Noriko Saitoh[2], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*IMEG, Kumamoto Univ.*)

Nucleolus is the largest non-membrane-bound structure in the nucleus and contains both protein and RNA. In spite of its important role in ribosome synthesis, the mechanism how nucleolus keeps its dynamic structure without the menbrane still remains elusive. Nucleolus is consist of three compartments depending on each function; FC (rDNA transcription), DFC (rRNA processing) and GC (ribosome assembly). To investigate the relation between their structure and function, we established HeLa cell lines stably expressing fluorescence-tag-fusion proteins of the compartment markers. Single molecule tracking analysis showed notably different behabior of NPM1 (GC) and FBL (DFC). We will discuss the dynamics of the nucleolus compartments.

1Pos314 **高速超解像光学顕微鏡を用いた出芽酵母の膜交通の観察**
Observations of the membrane traffic in living yeast cells via the high-speed super-resolution optical microscope

Daisuke Miyashiro[1], Kazuo Kurokawa[1], Akihiko Nakano[1,2] ([1]*Riken, RAP*, [2]*University of Tokyo, Dep. Bio. Sci.*)

To elucidate fine and dynamic phenomena, such as membrane traffic, in living cells, we have developed a high-speed super-resolution optical microscope and a measurement method by a high spatiotemporal resolution.
Here we report the new system that can acquire a confocal optical microscope image of about 1 photon precision in 1000fps, and the new method of deconvolution based on the probability calculation. By using these new system and method, we report the detailed observation about the dynamics of COPII inner and outer coats in the living budding yeast.

1Pos315* **新規微分干渉顕微法を用いた生細胞ヘテロクロマチンにおける物質密度のイメージング**
"Density" imaging of heterochromatin in live mouse cells using OI-DIC microscopy

Ryosuke Imai[1,2], Tadasu Nozaki[1], Tomomi Tani[3], Kayo Hibino[1,2], Michael Shribak[3], Kazuhiro Maeshima[1,2] ([1]*Natl. Inst. of Genet.*, [2]*SOKENDAI*, [3]*MBL, Woods Hole, USA*)

It is an established view that heterochromatic regions in cell nuclei are highly dense. But how really dense are they? What are they like in live cells? To investigate the heterochromatic regions, we observed the mouse chromocenter, a good model heterochromatin, using the orientation-independent (OI)-DIC microscopy system, which is able to measure dry mass in live cells. Strikingly, we found that dry mass density of the chromocenters in live cells is only about 1.5-fold higher than that of surrounding euchromatin while their DNA density stained by fluorescent DNA dye was about 6-7-fold higher. This result suggests that the heterochromatic regions are not necessarily dense as we have ever expected. A possible heterochromatin environment will be discussed.

1Pos316 **線虫 *C. elegans* 胚発生における細胞動態の個体差定量解析**
Quantitative analysis of variability of cellular dynamics in *C. elegans* embryogenesis

Yusuke Azuma, Shuichi Onami (*RIKEN QBiC*)

The development of *C. elegans* proceeds through an invariant cell lineage. However, it is unclear to what extent the cellular dynamics is invariant. To address this question, we developed a method to evaluate the variability of cellular dynamics quantitatively. The method first detects positions of nuclei from 3D time-lapse images of embryos by a local maxima based image processing. The detected nuclei are subsequently used as markers for watershed segmentation of cell membrane. By applying the method, we segmented cell membrane in five embryos and identified variability in cell volume, cell division timing and cell-cell contacts. We will present the analysis and discuss regulation mechanism of the variability along the embryogenesis.

1Pos317 **神経分化時における神経細胞内温度イメージング**
Imaging of intracellular temperature in PC12 cell nerve differentiation

Masaki Kinoshita[1,2], Kohki Okabe[3,4], Hisashi Tadakuma[2], Yoshie Harada[1,2,5] ([1]*Grad. Sch. Bio., Kyoto Univ.*, [2]*iCeMS, Kyoto Univ.*, [3]*Grad. Sch. Pharm, Tokyo Univ.*, [4]*PRESTO, JST*, [5]*IPR, Osaka Univ.*)

Neural differentiation is an essential process in brain development. It has been reported that nerve differentiation is influenced by extracellular factors (e.g. cytokine) or intracellular factors (e.g. DNA methylation or histone modification). However, recent studies showed that neurite outgrowth ability was changed by extracellular heat shock, physical factors such as temperature has not been investigated until now. Here, we characterize the intracellular temperature in relation to nerve differentiation. Using fluorescent polymeric thermometer (FPT) and fluorescence-lifetime imaging microscopy (FLIM), we determined the intracellular temperature of PC12 pheochromocytoma cells and showed that temperature variation was involved in the differentiation of these cells.

1Pos318* **ヒト免疫応答の1細胞実時間イメージングによるアレルギー診断の可能性**

Potential allergy diagnosis by real-time single-cell secretion imaging of human immune response

Kaede Miyata[1], Yoshitaka Shirasaki[1,2], Nobutake Suzuki[1], Hiroki Kabata[3], Mai Yamagishi[1,2], Osamu Ohara[2], Koichi Fukunaga[3], Kazuyo Moro[2], Sotaro Uemura[1] ([1]*Department of Bological Sciences, Graduate school of Tokyo*, [2]*Institute of Physical and Chemical Research, IMS.*, [3]*Division of Pulmonary Medicine, Keio University*)

We developed a new tool for a parallel measurement platform for real-time single-cell secretion imaging with a multi-reservoir integrated nano litter-well array chip. It enables us to compare secretion responses dynamics on the multi specimens with various stimuli simultaneously.
Here, we applied this system to human Type 2 innate lymphoid (ILC2) cells, rarely obtained from 20 mL of peripheral blood (~$1x10^3$ cells), and monitored secretion response of allergic inducible proteins over 5 days. We found large difference in the onset and amplitude of secretion response between donors, which might reflect their body constitutions including allergic diathesis. Therefore, this new tool is expected to contribute to a more precise and classified diagnosis for allergy.

1Pos319 **ソフトウェア「閻魔」とEMCアルゴリズムを用いたタンパク質3次元電子密度分布の再構成：XFEL-CXDI実験を想定したシミュレーション**

Reconstruction of three-dimensional structures of a protein with software ENMA and EMC algorithm: A simulation for XFEL-CXDI experiment

Takashi Yoshidome[1], Yuki Sekiguchi[2,3], Tomotaka Oroguchi[2,3], Masayoshi Nakasako[2,3], Mitsunori Ikeguchi[4] ([1]*Dep. of Appl. Phys., Tohoku Univ.*, [2]*Fac. of Sci. and Tech., Keio Univ.*, [3]*RIKEN SPring-8 Center*, [4]*Grad. Sch. of Med. Life Sci. Yokohama City Univ.*)

Coherent X-ray diffraction imaging (CXDI) experiments enable us to visualize the structures of non-crystalline particles with micrometer to sub-micrometer dimensions. Using the X-ray free-electron laser (XFEL), two-dimensional diffraction patterns are collected in the diffraction-before-destruction scheme. To reconstruct three-dimensional electron density maps of a particle, it is necessary to classify the diffraction patterns in terms of structural polymorphism before the reconstruction. Here, we performed a simulation for XFEL-CXDI experiment of a protein predominantly adopting two states. After classifying the data using the custom-made software “ENMA”, we successfully reconstructed the three-dimensional electron density maps of the states using the EMC algorithm.

1Pos320 **X線自由電子レーザーを用いた低温コヒーレントX線回折イメージングによるシアノバクテリアの三次元構造解析**

Three-dimensional structure of a cyanobacterium visualized by cryogenic coherent X-ray diffraction imaging using X-ray free-electron laser

Amane Kobayashi[1,2], Yuki Sekiguchi[1,2], Koji Okajima[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Yayoi Inui[3], Takeshi Hirakawa[3], Sachihiro Matsunaga[3], Masaki Yamamoto[2] ([1]*Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*, [3]*Sci. Tech., Tokyo Univ. Sci.*)

Coherent X-ray diffraction imaging (CXDI) visualizes the structures of non-crystalline particles with the sizes of micrometer to sub-micrometer. In CXDI experiments, the projection electron density maps of the particles are reconstructed from the diffraction patterns of isolated sample particles under the irradiation of coherent X-rays. Furthermore, three-dimensional structures of a particle can be, in principle, reconstructed from a large number of the projection maps by using the single particle reconstruction method. Our experimental techniques allow us to collect a huge number of diffraction patterns in a short period of time at X-ray Free electron laser facility SACLA. Here, we report the reconstructed three-dimensional structure of a cyanobacterial cell.

2Pos001* カルシウム依存的な鞭毛運動の制御に関わるタンパク質カラクシンの構造解析
Structural analysis of calaxin, calcium-dependent flagellar movement regulator

Tomoki Shojima[1], Feng Hou[1], Yusuke Takahashi[1], Masahiko Okai[1], Katsutoshi Mizuno[2], Kazuo Inaba[2], Takuya Miyakawa[1], Masaru Tanokura[1] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*Shimoda Marine Research Center, Univ. Tsukuba*)

Calaxin is one of the neuronal calcium sensor proteins and regulates metazoan sperm flagellar movement in a Ca^{2+}-dependent manner. We determined the crystal structures of calaxin in the Ca^{2+}-bound and the Mg^{2+}-bound forms at 1.9 Å and 2.6 Å, respectively. The crystal structure is composed of the open state and the closed state. In the open state, the hydrophobic surface in the C-terminal domain is expanded compared to the closed state. Although the Ca^{2+}-bound and the Mg^{2+}-bound forms are similar, the fourth EF-hand in the closed state has a higher affinity for Ca^{2+} than Mg^{2+}, which may affect the conformational transition between the open and closed states.

2Pos002 Porphyromonas gingivalis の T9SS によって分泌される PGN_0123 の構造
Structure of PGN_0123, a Type IX secretion substrate of Porphyromonas gingivalis

Yusuke Handa[1], Keiko Sato[2], Koji Nakayama[2], Katsumi Imada[1] ([1]*Grad. Sch. Sci. Osaka Univ.*, [2]*Grad. Sch. Biomedical Sci., Univ. Nagasaki*)

Porphyromonas gingivalis, a gram-negative anaerobic bacterium, is a major periodontal pathogen. The virulence factors, such as gingipain proteases, are translocated through the Type IX secretion system (T9SS) to the cell surface, and are fixed on the surface by glycosylation. PGN_0123, one of the T9SS substrates, is expected to be involved in the glycosylation of virulence proteins, but the details are unclear. To elucidate the molecular mechanism of PGN_0123 function, we determined the crystal structure of the N-terminal domain of PGN_0123 at 1.4 Å resolution. The structure is similar to that of FimH, the Type 1 fimbrial adhesin. Compared with the structure of FimH, the putative sugar binding site of PGN_0123 adopts an open conformation.

2Pos003 時計タンパク質 KaiC のリン酸化状態と脱リン酸化状態における構造上の差異
Structural Differences between Phosphorylated and Dephosphorylated States of Clock Protein KaiC

Yoshihiko Furuike[1], Jun Abe[1], Eiki Yamashita[3], Takao Kondo[4], Shuji Akiyama[1,2] ([1]*Research Center of Integrative Molecular Systems (CIMoS), Institute for Molecular Science (IMS)*, [2]*Department of Functional Molecular Science, SOKENDAI (The Graduate University for Advanced Studies)*, [3]*Institute for Protein Research, Osaka University*, [4]*Graduate School of Science, Nagoya University*)

As a central oscillator in cyanobacterial circadian clock, KaiC provides time information to other clock-related proteins according to its own status of two phosphorylation sites, Ser431 (S) and Thr432 (T). KaiC experiences successive phosphorylation processes from dephosphorylated state (ST) to double-phosphorylated states (pSpT) in first half of a day, and dephosphorylation from pSpT to ST occurs in second half of a day. Crystal structures of KaiC in pSpT state have been reported so far, however, that of ST state was unknown. In the presentation, we will discuss the structural differences between ST and pSpT states on the bases of preliminary X-ray crystallographic analyses.

2Pos004 Structural characterization of Hsp104 from a thermophilic fungus, Chaetomium thermophilum

Yosuke Inoue (*Tokyo University of Agriculture and Technology*)

Hsp104 is a kind of molecular chaperone and AAA+ family proteins are widely conserved in eukaryotes. Hsp104 take the hexamer in the presence of ATP, and disaggregates protein. Detailed structural mechanism of disaggregation has remained unknown due to low structural stability of Hsp104. In this work, we have cloned Hsp104 from thermophilic fungi Chaetomium thermophilum which harbors structurally stable proteins. We have determined the crystal structure of hexamer of Hsp104, and the crystal structure suggests hexamer of Hsp104 forms a spiral structure. Further, to clarify structure under physiological condition, we have examined structure of Hsp104 in solution using a high-speed atomic force microscope (AFM).

2Pos005 圧力応答を示す YFP 挿入変異体の高圧下での結晶構造
Crystal structure of a pressure sensitive YFP mutant under high pressure

Mika Tsujii[1], Takayuki Nagae[2], Keiko Yoshizawa[3], Tomonobu Watanabe[3], Masahiro Nishiyama[4], Nobuhisa Watanabe[2], Tatsuya Kawaguchi[1], Katsumi Imada[1] ([1]*Grad. Sch. Sci., Univ. Osaka*, [2]*SRRC. Nagoya Univ.*, [3]*QBiC, Riken.*, [4]*Grad. Sch. Sci., Univ. Kyoto*)

Yellow fluorescent protein (YFP) based sensors are widely used to detect states of intracellular environment. We recently found that glycine insertion in β7 of YFP dramatically changes its fluorescence property dependent on hydrostatic pressure. Among the insertion mutants, YFP-3G, a mutant with insertion of three glycine residues, is most sensitive to the hydrostatic pressure. To understand the mechanism of the pressure dependent fluorescence change, we determined the crystal structures of YFP-3G and measured the fluorescence spectra of the crystals under high pressure. Interestingly, conformation of the chromophore is changed and the disordered part of β7 becomes structured at high pressure. These structural responses to the pressure may cause the fluorescence change.

2Pos006 Crystallization of Hepatitis B virus Core Protein in genotype C

Katsumi Omagari (*Nagoya City University*)

Hepatitis B virus (HBV) is a major human pathogen that causes serious liver disease. HBV has been classified into 8 geographically, genetically, and clinically diverse genotypes A to H. Genotype C, which is associated with more serious liver disease, is prevalent in Japan. HBV replicates through reverse transcription of an RNA intermediate, the pregenomic RNA (pg RNA). The replication occurs inside core protein (Cp). Knowledge of the structure of Cp would be valuable for understanding the molecular basis. No empirical structural data exist for Cp of genotype C. This work has established large expression system of Cp, and the Cp in genotyope C are crystallized. By using electronic microscope, the purified Cp assembled into spherical shells of 30nm in diameter.

2Pos007* 巨大タンパク質会合体ヘモシアニンの多孔質性結晶を用いた生体分子の包摂

Encapsulation of biomacromolecules into porous crystal of a huge protein complex hemocyanin

Asuka Matsuno[1], Ye Yuxin[2], Yuki Ohnishi[2], Akira Kitamura[1,2], Masataka Kinjo[1,2], Satoshi Abe[4], Takafumi Ueno[4], Yoshikazu Tanaka[1,2,3], Min Yao[1,2] ([1]*Graduate School of Life Science, Hokkaido University*, [2]*Faculty of Advanced Life Science, Hokkaido University*, [3]*JST, PRESTO*, [4]*Department of Life Science and Technology, Tokyo Institute of Technology*)

Molluscan hemocyanin is a huge hollow cylindrical protein complex. Recently, we determined crystal structure of squid hemocyanin, which revealed that the inner space of squid hemocyanin with a diameter of approximately 110Å is large enough for encapsulating biomacromolecules. Furthermore, hemocyanin, in the crystal, stacked as if straw with interacting through their outer wall region. This packing manner in combination with the presence of huge hollow space inside the cylinder suggests that hemocyanin is crystallized in the same condition even if it encapsulates biomacromolecules inside. In the present study, we confirmed, by using a confocal laser microscope, that several biomacromolecules with molecular mass between 3.8 - 250 kDa could be encapsulated.

2Pos008 蛋白質結晶中の分子間静電相互作用計算

Calculation of inter-molecular electrostatic interactions in protein crystals

Takuya Takahashi[1], Shigeru Endo[2], Masanori Ootaki[3], Yoko Sugawara[2] ([1]*Bioinfo., Coll. Life. Sci., Ritsumeikan Univ.*, [2]*Dept. Physics, Sch. Sci., Kitasato Univ.*, [3]*Dep. Pharmacology, St. Marianna Univ. School of Medicine*)

To obtain stable and desirable protein crystals, the prediction of the crystal habit from the single molecular structure will be important. Previous studies evaluated the molecular interactions in protein crystals and found good correlation between crystal habit and inter-molecular interaction by calculating the electrostatic free energy of protein crystals. In this study, we generalized the calculation algorithm because the previous calculation method was restricted to crystals with rectangular unit cells. The method was applied to ribonuclease A crystals. In the poster, detailed comparison with the experiments and another calculation method (macro-bond analysis) is shown as well as the algorithm and the effect of the revision.

2Pos009 X 線自由電子レーザー（XFEL）回折像からの生体分子三次元構造の復元プログラムの開発

Development of 3D reconstruction program for coherent diffraction patterns obtained by XFEL

Miki Nakano[1], Osamu Miyashita[1], Slavica Jonic[2], Atsushi Tokuhisa[1], Daewoon Nam[3], Yasumasa Joti[4], Changyong Song[3], Florence Tama[1,5] ([1]*RIKEN AICS*, [2]*IMPMC, Sorbonne University - CNRS UMR 7590, UPMC Univ. Paris 6, MNHN, IRD UMR 206*, [3]*POSTECH, Korea*, [4]*JASRI XFEL*, [5]*Grad. Sch. Science, Nagoya Univ.*)

3D structural analysis for single particles using X-ray free electron laser (XFEL) is a new structural biology technique. It enables us to observe molecules, which are hard to crystallize such as biological molecules and living tissues, in a state close to nature. In order to restore the 3D structure of the molecule from the diffraction patterns obtained by XFEL experiments, computational algorithms are necessary as one needs to estimate the laser beam incidence angles to the molecule and retrieve the phase information in Fourier space. We are developing a program package for XFEL analysis based on XMIPP, which is commonly used for image processing of single-particle 3D cryo electron microscopy. Our program shows success in restoring 3D structure of molecules.

2Pos010 Hsp90 の構造変化に関する理論的研究

Theoretical study of a conformational change in Hsp90

Kazutomo Kawaguchi, Hidemi Nagao (*Inst. Sci. Eng., Kanazawa Univ.*)

Heat Shock Protein 90 (Hsp90) is one of a group of molecular chaperones required for protein folding. A large conformational change between open and closed states of dimeric Hsp90 is observed in the functional cycle of Hsp90. Although intermediates are suggested by FRET, the molecular detail of the intermediates is not elucidated. In our previous studies, we have discussed the conformational change of the N-terminal domain of Hsp90 by all-atom molecular dynamics simulations [CPL 2013, JPS Conf. Proc. 2014, Mol. Sim. 2016]. In this study, we present the detail structure of the intermediates in the functional cycle of Hsp90 by Langevin dynamics simulations with the Go-like model. We also discuss the conformational transition in dimeric Hsp90.

2Pos011 Building a database of 3D biological shapes for the interpretation of XFEL diffraction patterns

Sandhya Tiwari[1], Osamu Miyashita[1], Florence Tama[1,2] ([1]*Riken Advanced Institute for Computational Science*, [2]*Nagoya University*)

X-ray free electron laser (XFEL) scattering experiments has been described as the future of structural biology. We aim to provide efficient interpretation of XFEL data by searching them against a database of biological shapes to obtain an initial model. We first assemble various 3D structures in existing structure databanks. Then, we reduce the dataset to a minimal set of hypothetical biological shapes. Here, we present the analysis of single particle cryo-EM structures. We analysed the variation in the shapes that exists within the 3D models, and within their simulated 2D projection images. This provides us with the extent to which 2D images of two distinct 3D shapes can be similar, giving us an idea of the expected degeneracy when searching for initial models.

2Pos012 胆汁酸輸送体の分子動力学シミュレーション

Molecular dynamics simulation of the bile acid transporter

Shin-ichiro Tasaki, Ryunosuke Yoshino, Yoshitaka Moriwaki, Kentaro Shimizu, Tohru Terada (*Grad. Sch. of Agri. Life Sci., Univ. of Tokyo*)

The apical sodium-dependent bile acid transporter (ASBT) plays a key role in reabsorption of bile acids in small intestinal epithelial cells. Because inhibition of the reabsorption can lower the cholesterol level in blood, it is receiving increased attention as a drug target. To understand its molecular mechanism of the bile acid transport, we constructed docking models between ASBT and a bile acid, taurocholic acid, using the structure of an ASBT homolog from Yersinia frederiksenii, and performed all-atom molecular dynamics simulations for the models in a lipid bilayer environment for more than 1 μs. We compared the stability between the models, analyzed the protein-ligand interactions, and examined the effect of the ligand binding on the protein structure.

2Pos013* フレキシィボタンパク質ータンパク質ドクイン： PaCS−MD の応用

Flexible-Body Protein-Protein Docking: an Application of Parallel Cascade Selection Molecular Dynamics

Duy P. Tran[1], Akio Kitao[1,2] ([1]*UTokyo, GSFS*, [2]*UTokyo, IMCB*)

Protein-protein docking is now considered to be useful for predicting structure of the complexes, although successful predictions are mainly limited to rigid protein pairs. In this presentation, we introduce the procedure of flexible docking based on Parallel Cascade Selection Molecular Dynamics (PaCS-MD) [R. Harada, JCTC 2013] by performing di/association simulations of the ligand toward/from the receptor to generate the bound conformations, taking into account all the dynamics and solvation of proteins. Currently, MDM2 protein in complex with part of p53 case can yield the best conformation which is 0.46 nm backbone RMSD from the X-ray structure (1YCQ). We further construct the Markov State Model from the PaCS-MD results for the kinetic properties of the complexes.

2Pos014 分子動力学シミュレーションを用いた Hras-GTP/GDP 複合体と溶媒水との水素結合の動きの解析

Analysis of dynamics of hydrogen bond between the solvent water and the Hras-GTP/GDP complexes by molecular dynamics simulations

Takeshi Miyakawa[1], Ryota Morikawa[1], Masako Takasu[1], Kimikazu Sugimori[2], Kazutomo Kawaguchi[2], Hidemi Nagao[2] ([1]*Tokyo Univ. of Pharm. & Life Sci.*, [2]*Kanazawa Univ.*)

In order to understand the mechanism of hydrolysis of GTP in the Hras-GTP complex, we study the structures of the Hras-GTP/GDP complexes in water solvent by molecular dynamics (MD) simulations.
We evaluated the potential parameters around Mg2+ in Hras-GTP/GDP complexes by quantum chemical calculations. We performed MD simulations of the Hras-GTP/GDP complexes in water solvent using parameters of AMBER03 and our parameters around Mg2+. We found that the positions and orientations of water molecules near GTP are different from those near GDP.
In this study, we analyze the dynamics of the hydrogen bond between the solvent water and the GTP/GDP in the Hras-GTP/GDP complexes.

2Pos015 分子動力学シミュレーションによる抗 HIV 中和抗体 PG16 の CDR-H3 における構造剛性の解析

Molecular dynamics study of the structural rigidity of CDR-H3 of anti-HIV neutralizing antibody PG16

Ryo Kiribayashi[1], Hiroko Kondo[1], Daisuke Kuroda[2], Toru Saito[1], Jiro Kohda[1], Akimitsu Kugimiya[1], Yasuhisa Nakano[1], Yu Takano[1] ([1]*Hiroshima City Univ.*, [2]*Showa Univ.*)

PG16 is a broadly neutralizing antibody to HIV-1. A crystal structure of the antigen-binding fragment (Fab) of PG16 shows that the long 28-residue complementarity determining region (CDR) H3 forms a unique subdomain referred to as “hammerhead.” However, despite the structural diversity of CDR H3s in general, a large conformational difference is not observed in the crystal structure of the CDR-H3 of PG16 in the antigen-bound and unbound (Cα-RMSD: 0.44Å).
In this study, using molecular dynamics simulations, we have examined the structural rigidity of CDR-H3 of antibody PG16. During the simulations, the CDR-H3 of PG16 kept the hammerhead structure, and motions of the whole subdomain were observed.

2Pos016 Molecular dynamics simulations of the basic amyloidogenic unit of IAPP

Richa Tambi[1], Satoshi Kosuda[1], Gentaro Morimoto[2], Makoto Taiji[2], Yutaka Kuroda[1] ([1]*Tokyo University of Agriculture and Technology*, [2]*Quantitative Biology Center, RIKEN*)

Amyloid aggregates of Islet amyloid polypeptide (IAPP) are associated with type II diabetes. Similarity was observed between amyloid fibers formed by IAPP and its short peptide fragment (NFGAILSS). In this work, we investigate the amyloid forming mechanism of this octapeptide sequence using an all-atom molecular dynamics (MD) approach. We simulated the wild type and seven alanine-scanned mutants of NFGAILSS for 100ns. Each MD simulations consisted of 27 peptides and approximately 30000 water molecules, and was performed using AMBER8 on a special purpose MD-GRAPE3 computer. Peptide clusters were observed for wild type as well as all the alanine scanned mutants, but the wild-type had the highest β structure content. This result is in line with experimental observations.

2Pos017 レプリカ交換モンテカルロ SAAP3D 法による C-ペプチドと Trp ケージの分子シミュレーション

Molecular simulation of C-peptide and Trp-cage by SAAP3D-REMC method

Michio Iwaoka, Natsuki Babe, Yuya Shoji (*Tokai University, Department of Chemistry*)

We recently developed an original molecular simulation program, SAAP3D-REMC, which combines the 3-dimensional single amino acid potential force field and the replica-exchange Monte Carlo algorism, and reported that the conformational potential of 10AA chignolin can be nicely reproduced by using the program. Here, SAAP3D-REMC has been applied to longer peptides, i.e., 13AA C-peptide and 20AA Trp-cage, to check performance of our program. After clustering of the obtained structures into several classes based on the main-chain rmsd, they were compared with the native ones in solution. In both cases, reasonable simulation results were obtained, but it was simultaneously suggested that optimization of the simulation conditions is necessary for these long peptide chains.

2Pos018 REST 法による TRP-cage のフォールディングシミュレーション

In silico folding simulation of Trp-cage using the REST method and its variants

Motoshi Kamiya[1], Yuji Sugita[1,2,3] ([1]*AICS, RIKEN*, [2]*RIKEN*, [3]*QBiC, RIKEN*)

Conformational search is still one of the critical problems in all-atom molecular dynamics (MD) simulations of biomolecules. Recently, the Replica Exchange with Solute Tempering (REST) method was proposed to enhance the conformational search. The method was reported to be efficient for various biological simulations with a reasonable additional computational cost compared to the conventional MD simulations. Recently, we have implemented REST and two-dimensional REST/REUS methods into the GENESIS software package. Folding simulations of the terminal capped Trp-cage variant known as TC5b using these methods shows that REST can accelerate the folding rate and a further improvement can be achieved by the two-dimensional REST/Rg-REUS method.

2Pos019 タンパク質のフォールディング過程における階層性と不均一性の分子論的起源

Molecular origin of heterogeneity and hierarchy behind protein folding

Toshifumi Mori[1,2], Shinji Saito[1,2] ([1]*IMS*, [2]*SOKENDAI*)

Proteins involve motions over a wide range of timescales. While conformational transitions such as folding and functioning are slow and appear to occur cooperatively, how the fast and often local dynamics proceed behind these slow motions remain unclear. In order to reveal these higerarchichal dynamics at molecular level, here we study the folding mechanisms of two prototypical fast-folding proteins. The ultra-long molecular dynamics trajectories from Anton are analyzed, and the folding/unfolding transition events are examined in detail. Although the proteins studied here are small, the results show that the transitions occur in a highly heterogeneous manner, thus suggests that heterogeneity in folding/unfolding transitions may be found more generally.

2Pos020 プロリン型人工アミノ酸を含むペプチドの分子動力学計算

Molecular dynamics simulation of peptide oligomers bearing the proline-type artificial amino acid

Yuko Otani[1], Satoshi Watanabe[1], Akio Kitao[2], Tomohiko Ohwada[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*IMCB, Univ. Tokyo*)

We reported synthesis of homooligomers of conformationally constrained proline-type β-amino acid. The presence of amide cis-trans isomerization hampered detailed structure analysis in solution. In this study, the solution structure of peptide oligomers is studied by molecular dynamics (MD) simulation. The umbrella sampling is conducted to sample conformational space of oligomers including trans-cis isomerization of tertiary amides. The optimum parameter values for reweighting were determined to best reproduce the experimental results of short oligomers. It is suggested that extended structures, which contain more trans-amide bonds, are stabilized as oligomer is elongated. MD simulation results of hetero-peptides will also be shown.

2Pos021 Theoretical study of diffusion of plastocyanin with Langevin equation

Makoto Wada, Satoshi Nakagawa, Shogo Kinoshita, Kurniawan Isman, Kouichi Kodama, Kazutomo Kawaguchi, Hidemi Nagao (*Nat. Sci. Kanazawa Univ.*)

Plastocyanin(PC) is one of a metal protein in the chloroplast. PC is received one electron by cytochrome f complex and passes thylakoid lumen, after the electron receiving PC gives electron photosystem I. Helmut and his coworkers said that thylakoid lumen is expanded because of light emission and have investigated the electron transport rate depend on the size of this area. By analyzing diffusion of PC, it may be possible to discuss the change in this effect. In this work we present a simple model to analyzed electron transport rate. Our simple model describes PC as one particle and we solve the langevin equation using a simple model to discuss the diffusion. As a result of the simulation, we confirm the distribution of oxidized and reduced molecules.

2Pos022* HIV-1 protease の触媒的加水分解反応に関する理論的研究

Theoretical study on catalytic hydrolysis of HIV-1 protease

Masahiro Kaneso, Shigehiko Hayashi (*Grad. Sch. Sci., Kyoto. Univ.*)

HIV-1 protease is one of the most important targets for treatment of AIDS. It catalyzes hydrolysis of a long peptide, Gag or Pol polyprotein, for virus growth. The protease is a homodimer and two aspartic acids involved in its catalysis reside in the center of the dimer. The protonation states of the two aspartic acids are essential for the binding affinity and the catalytic reaction of the protease. In this research, we performed MD simulations of the proteins with different protonation states of the aspartic acids and examined their conformations and behavior of water molecules near the active site. Based on the structures obtained by the MD simulations, we also carried out QM/MM calculations to investigate reaction profile of the first step of the hydrolysis.

2Pos023 ホタルルシフェラーゼの全原子を考慮した発光基質オキシルシフェリンの光吸収のｐＨ依存性の定量解析

Quantitative Analysis of pH Effect on Absorption Peaks of Oxyluciferin by Considering All Atoms of Firefly Luciferase

Hironori Sakai[1], Itsuki Kaji[2], **Naohisa Wada**[2] ([1]*Insti. of Fluid Science, Tohoku Univ.*, [2]*Food Life Sciences, Toyo Univ.*)

The color tuning mechanism of firefly bioluminescence depending on pH has been debated for a long time. It is suggested that protonation state change either in the active center of the enzyme (Luc) or in its substrate (OxyLn), or in both, makes the absorption peak of OxyLn shiftable. Previously, we showed that in the active center of Luc, only Asp424 within 5Å surrounding OxyLn depends on pH. In this report, protonation state of Luc active center was only considered according to the previous report, and calculations of absorption peaks performed. The predicted peaks were found to be 609 nm at pH 6.8, and 602 nm at pH 7.8 showing a very slight shift to short wavelength at high pH. Thus present model is hard to reproduce the large color-shift of firefly bioluminescence.

2Pos024 糖転移酵素の糖選択性とタンパク質認識に関わるアミノ酸の解析

Sequence and structure analysis of glycosyltransferases for understanding the sugar selectivity and target recognition mechanisms

Go Miyasaka[1], Kenji Etchuya[2], Yuri Mukai[1,2] ([1]*Sch. Sci. & Tech., Meiji Univ.*, [2]*Grad. Sch. Sci. & Tech., Meiji Univ.*)

The purpose of this study is to clarify the mechanisms of the sugar selectivity and protein recognition in protein glycosylation based on the three-dimensional structures of glycosyltransferases. Glycosyltransferases which directly modify the sugars to the proteins were analyzed in this study because the specificities of the sugar selectivity and protein recognition mechanisms were considered to depend on the sequences and structures of glycosyltransferases. Through the investigation of the amino acid residues which are related to the interaction between glycosyltransferases and sugars/proteins, a sugar-type specificity was especially found in the propensities of the aromatic and hydrophobic residues in the region close to the glycosylation sites.

2Pos025 アルカン合成関連酵素の機能発現における保存部位の役割

Alanine scanning mutagenesis reveals functional roles of conserved residues in an enzyme for alkane biosynthesis

Masashi Nomura, Hisashi Kudo, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

Cyanobacteria produce alka(e)nes using two proteins, an acyl-ACP reductase (AAR) and an aldehyde deformylating oxygenase (ADO). AAR catalyzes reduction of fatty acyl-ACP to aldehyde, while ADO converts the aldehyde into hydrocarbon. However, little is known about which residues of AAR are essential for the aldehyde producing activity. To clarify this issue, here we performed alanine scanning mutagenesis on the conserved residues of AAR. We revealed that most alanine substitutions decreased the AAR activity, indicating that most of the conserved residues are essential for the aldehyde producing activity of AAR. Thus, amino acid substitutions should be introduced at other than the conserved residues, to improve the AAR activity for bioenergy production.

2Pos026 示差走査型蛍光定量法を用いたアルドケト還元酵素の化合物選択性の評価

Evaluation of compound selectivity of aldo-keto reductases using differential scanning fluorimetry

Kabir Aurangazeb[1], Satoshi Endo[2], Naoki Toyooka[3], Mayuko Fukuoka[1], Kazuo Kuwata[1,4], **Yuji Kamatari**[5] ([1]*United Grad. Sch. Drug Dis. Med. Inf. Sci., Gifu Univ.*, [2]*Lab. Biochem., Gifu Pharm. Univ.*, [3]*Grad. Sch. Sci. Tech. Res., Univ. Toyama*, [4]*Grad. Sch. Med., Gifu Univ., Gifu Univ.*, [5]*Life Sci. Res. Ctr.*)

Inhibitors of AKR1B10 belonging to the aldo-keto reductase (AKR) superfamily are considered promising candidates for anti-cancer drugs. AKR1B1, a structurally similar isoform of AKR1B10, is involved in glucose metabolism. Thus, selective inhibition of AKR1B10 is required for the development of anti-cancer drugs. In this study, we first compared correlations between melting temperature (T_m) and the 50% inhibition concentration (IC_{50}) obtained from differential scanning fluorimetry (DSF) and an enzyme inhibitory experiment, respectively, and a good correlation was found, except for compounds with low solubility. We then evaluated their selectivity as inhibitors against all seven major human AKR1 family proteins and found that C18 is most specific for AKR1B10.

2Pos027 Unique mechanism for broad substrate specificity of human MTH1

Shaimaa Ali, **Teruya Nakamura**, Keisuke Hirata, Mami Chirifu, Shinji Ikemizu, Yuriko Yamagata (*Grad. Sch. Pharmaceut. Sci., Kumamoto Univ.*)

Human MutT homolog 1 (hMTH1) hydrolyzes oxidatively damaged nucleoside triphosphates, and prevents replicational errors caused by their misincorporation into a newly synthesized DNA strand. In contrast to *Escherichia coli* MutT, which hydrolyzes 8-oxo-dGTP to 8-oxo-dGMP with high substrate specificity, hMTH1 has broad substrate specificity for oxidized nucleotides including 8-oxo-dGTP and 2-oxo-dATP. In this work, we performed kinetic and structural analyses on the wild type and the substrate-binding site mutants of hMTH1. The relation between the structure and the enzymatic activity revealed that hMTH1 recognizes different types of the ligands via interchange of the protonation state at the neighboring aspartate residues in the substrate-binding site.

2Pos028* ラン藻由来アルカン合成関連酵素の構造機能解析

Structural and functional analysis of a cyanobacterial enzyme for alkane biosynthesis

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Acyl-(acyl carrier protein) reductase (AAR) is a key enzyme involved in the cyanobacterial synthesis of alkanes that can be used as biofuels. However, AAR has low activity, and little is known about the structure and function of AAR. Previously, we found two AAR strains with high and low activities, respectively. Here, we introduced single amino acid substitutions into the AAR with low activity, to make its sequence close to that of highly active AAR. We succeeded in increasing the solubility and activity of AAR and identified the residues determining the AAR properties. In addition, X-ray scattering measurements show that AAR has a heart-shaped structure with three globular domains. Our data are useful for improving the AAR activity for biofuel production.

2Pos029 クジラミオグロビンの分子進化

Tracing evolution of whale myoglobin by resurrecting ancient proteins

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The marine adaptation of whale myoglobin (Mb) was investigated by syntheses and analyses of ancestral globins. Amino-acid sequences of Mb from extinct genera of cetacean dated back to the Eocene epoch were inferred by the maximum likelihood method using all the available globin sequences at present. Three ancient globins were synthesized, two of which are from common ancestors between toothed and baleen whales, and another is a further common between whale and hippopotamuses. Their 3D structures were determined at high resolution and showed significant increase of net positive charges on the protein surface during the evolution. The differences in their thermodynamic stability, solubility and intermolecular interaction will be discussed in the light of whale evolution.

2Pos030 MEK1 リン酸化に伴う構造変化

Structual dynamics of MEK1 activation through phosphorylation

Minami Ando, Kei Moritsugu, Akinori Kidera (*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*)

MEK1 is a protein kinase that participates in MAPK signal pathway. Phosphorylation of two serine residues regulates MEK1 activity. To understand the structural basis of the activation through phosphorylation, we performed molecular dynamics (MD) simulations of MEK1 for both inactive/active structures and with/without the phosphorylation at the two serine residues. The analyses of the simulated trajectories revealed that the phosphorylation loosened the activation loop to open the binding cleft, and yielded a partially activated structure. We attempted to identify the energy barrier separating the fully activated form from the partially activated form using a targeted MD, and to elucidate the fundamental structure of MEK1 activation via phosphorylation.

2Pos031 アロステリーの概念拡張に向けて：トロンビンのアロステリック制御・再訪

Toward Expanding the Concept of Allostery: Thrombin Allosteric Regulation, Revisited

Ikuo Kurisaki[1,2], Masayoshi Takayanagi[1,2], Barberot Chantal[1,2], Masataka Nagaoka[1,2] ([1]*Grad. Sch. Info. Sci. Univ. Nagoya*, [2]*JST-CREST*)

In the last 20 years, it has been supposed that thrombin activation is allosterically regulated by Na^+-binding, while it still remains elusive whether it actually promotes the activation. We addressed the problem by employing molecular dynamics simulations and clarified that Na^+-binding in contrary prevents thrombin-substrate Michaelis complex formation. Instead, we found that the Na^+-binding cavity is involved in dewetting of S1-pocket upon thrombin-substrate complex formation. Furthermore, we observed that unbound Na^+ molecules are differently distributed around thrombin, suggesting promotion of complex formation. According to the sequential studies on thrombin activation, we propose the alternative mechanism of allosteric regulation.

2Pos032 サルコシン酸化酵素の反応生成物は４つの水チャネルの１つを選択的に移動する：平均力ポテンシャルによる検証

Potential of mean force shows that the reaction product of sarcosine oxidase selectively exits from one of four water channels

Takami Saito[1], Go Watanabe[2], Daisuke Nakajima[2], Haruo Suzuki[2], Shigetaka Yoneda[2] ([1]*Grad. Sch. Sci., Kitasato Univ.*, [2]*Sch. Sci., Kitasato Univ.*)

The X-ray structure of heterotetrameric sarcosine oxidase (HSO) revealed that the active site is situated deep inside of the enzyme protein near the FAD cofactor. Several water channels from the outside to the active site were proposed to be the pathways for the migration of substrates and products. Here, we show from the molecular dynamics (MD) simulation that one of the product (imino intermediate, 5-oxazolidinone: 5-OXA) selectively uses one of four channels in HSO. The details of the MD simulation of each channels will be presented, including the steered MD simulation of 5-OXA in each channel, followed by the umbrella sampling simulation and the calculation of the potential of mean force by using the Weighed Histogram Analysis Method.

2Pos033 Characterizing NO diffusion in nitrite reductase: nitric oxide reductase complex

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Nitrite reductase (NiR) and Nitric oxide reductase (NOR) are two important enzymes involving in the microbial denitrification. This process sequentially reduces nitrogen-containing compounds: $NO_3^- \rightarrow NO_2^- \rightarrow NO \rightarrow N_2O \rightarrow N_2$. In this process, NiR catalyzes the reaction forming nitric oxide (NO) while NOR catalyzes the reaction using NO as a substrate. How NO molecules can diffuse from periplasmic NiR to membrane-bound NOR remains a question. In this study, we characterize NO diffusion in this system using a molecular dynamics simulation approach. A NO diffusion pathway connecting NiR and NOR was found and the time of NO in water was only a few nanoseconds. This gives a clear insight into the function of the complex.

2Pos034* 二段階緩和モード解析による蛋白質シミュレーションの動的解析

Dynamical analysis of protein simulations by using two-step relaxation mode analysis

Naoyuki Karasawa[1], Ayori Mitsutake[1,2], Hiroshi Takano[1] ([1]*Grad. Sch. Sci. Technol., Keio Univ.*, [2]*JST, PRESTO*)

Relaxation mode analysis (RMA) based on relaxation phenomenon extracts slow dynamics of a system. In its previous application to a protein system, however, the long-time behavior was not described well. Two-step RMA has recently been developed in a homo-polymer system in order to improve description of long-time behavior. We firstly applied this method to a hetero-polymer system, protein. A 2 μs all-atom molecular dynamics simulation of hen egg-white lysozyme in explicit water at constant temperature 298.15 K and constant pressure 1atm was performed. The obtained trajectory was analyzed by two-step RMA. We found that two-step RMA not only describes long-time behavior well, but also extracts slow structural transitions of proteins more effectively.

2Pos035 MSES 法による EGFR キナーゼドメイン活性化の全原子構造解析

Structural basis for activation of EGFR kinase domain at atomistic resolution revealed by multiscale enhanced sampling

Kei Moritsugu[1], Tohru Terada[2], Akinori Kidera[1] ([1]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*, [2]*Grad. Sch. of Agri. and Life Sci., Univ. of Tokyo*)

Multiscale enhanced sampling (MSES) allows an enhanced sampling of the all-atom structure by coupling with the accelerated dynamics of the coarse-grained model. Here, we propose an extension via adiabatic separation to obtain further improvement in sampling efficiency, and have applied this to the structural change of EGFR kinase domain at atomistic resolution and including explicit solvent. Free energy surfaces calculated from the sampled structural ensembles both with and without an ATP have revealed large structural changes to the active form in activation loop, C-helix and N-lobe relative to C-lobe that are really induced by ATP binding. Structural pathways between the inactive and active forms have been examined to show an allosteric mechanism for EGFR activation.

2Pos036 生物学的レアイベントを再現する効率的構造サンプリング手法の開発

Developments of conformational sampling methods for reproducing biologically rare events of proteins

Ryuhei Harada, Yasuteru Shigeta (*Center for Computational Sciences, University of Tsukuba*)

Biological functions are strongly related to conformational transitions of proteins and induced as biologically rare events in long timescales. To reproduce the rare events, we introduce several efficient conformational sampling techniques of proteins newly developed by us. In our methods, multiple short-time MD simulations are utilized instead of long-time MD simulations. Our basic strategy is to restart multiple short-time MD simulations from appropriate initial structures that have high potential to transit. A cycle of the selecting and restating initial structures might drastically promote the structural transitions of proteins. We also introduce applications of our methods to biological targets.

2Pos037 タンパク質の長時間シミュレーションに関する緩和モード解析

Relaxation mode analysis for long time simulations of proteins

Ayori Mitsutake[1,2], Hiroshi Takano[1] ([1]*Dept. Physics, Keio Univ.*, [2]*JST, PRESTO*)

Relaxation mode analysis (RMA) was developed to investigate “dynamic” properties of polymer, homo-polymer, systems. In RMA, slow relaxation modes are extracted from molecular dynamics simulations. Recently, RMA has been applied to proteins, hetero-polymer systems to investigate dynamic properties of structural fluctuations [1,2,3]. Here, we apply RMA to long time simulations of proteins and show some results.

[1] J. Chem. Phys.135, 164102 (2011).[2] J. Phys. Soc. Jpn. 82, 023803 (2013); Seibutsu Butsuri (Biophysics), 49, Supplement S75, (Abstracts for the 47st annual meeting, The Biophysical Society of Japan) (2009)[3] J. Chem. Phys. 143, 124111 (2015).

2Pos038 Molecular basis for Hsp104-mediated prion propagation in yeast

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Neurodegenerative diseases, including Alzheimer’s disease and prion disease, are caused by aggregation of misfolded protein. In yeast prion system, Hsp104, in concert with Ssa1 and Sis1, is suggested to play crucial roles in prion propagation, acceleration of amyloid formation and disaggregation of amyloid. Yet, Hsp104-mediated prion propagation is poorly understood. Here we attempt to reveal how Hsp104/Ssa1/Sis1 chaperon machinery accelerates Sup35NM, a fragment of yeast prion Sup35, and disaggregates Sup35NM amyloid. Sup35NM amyloid formation is significantly accelerated in an ATP-dependent manner. I will further discuss Sup35NM amyloid disaggregation in the poster session. Our future studies will provide profound insights into the basic prion propagation mechanism.

2Pos039* HSP70 のフタの構造動態は基質結合にいかに関与するか？

How the lid exploits its structure dynamics in grasping the substrate in HSP70

Kohei Umehara[1], Naoya Tochio[2], Miho Hoshikawa[1], Shoji Ueki[3], Shin-ichi Tate[1,2] ([1]*Dept. Math. and Life Sci., Hiroshima Univ.*, [2]*RcMcD, Hiroshima Univ.*, [3]*Kagawa Sch. Pharm. Sci., Tokushima bunri Univ.*)

HSP70, a chaperon, captures unfolded protein segment in its substrate binding domain (SBD) to prevent their aggregation in cells. There is no structural report on the apo-SBD of HSP70, so that there is no clear evidence to describe how the ‘lid’ works in the absence of the substrate. In attempting to explore how the lid plays in capturing substrate, we solved the solution structure of the apo-SBD. Besides the structure, we further elucidated the lid orientation and dynamics relative to the SBD core part, by the use of various NMR techniques. The derived insights into the lid role are apparently controversial to those assumptively raised from the crystal structures. We will discuss the functional significance of the newly identified structure dynamics of the lid.

2Pos040 New insights into high molecular weight complex formation of 2-Cys peroxiredoxin and its chaperone function

Takamitsu Haruyama[1], Takayuki Uchihashi[1,2], Noriyuki Kodera[1], Toshio Ando[1], Hiroki Konno[1] ([1]*Bio-AFM FRC, Coll. Sci. & Eng., Kanazawa Univ.*, [2]*Coll. Sci. & Eng., Kanazawa Univ.*)

Peroxiredoxin (Prx) is a ubiquitous peroxidase that reduces reactive oxygen species in the cell. Prx acts as not only a peroxidase but also a molecular chaperone, and the functional change is due to the formation of high molecular weight (HMW) complex by overoxidation. Here, we studied the formation of oligomers and HMW complexes under several conditions of human 2-Cys PrxII (hPrxII) and their chaperon activity. Contrary to previous studies, overoxidation of hPrxII resulted in neither formation of spherical HMW complexes nor functional switching of hPrxII. However, the addition of a negatively charged lipid, e.g., phosphatidylserine, to overoxidized hPrxII produced spherical HMW complexes with a high chaperon activity.

2Pos041 Spectral characteristics of chimeric channelrhodopsins implicate the molecular identity involved in desensitization

Alemeh Zamani, Toru Ishizuka, Hiromu Yawo (*Tohoku University*)

Channelrhodopsin (ChR)-1 and ChR2 are the first-identified members of ChRs. Light absorption drives a generation of a photocurrent in the cell membranes expressing ChR2. However, the photocurrent amplitude attenuates to become a steady-state during prolonged irradiation. This process, desensitization, has been attributed to the accumulation of intermediates less conductive to cations. Here we compared the spectral sensitivity between desensitized and non-desensitized fractions of the chimeric ChRs between ChR1 and -2. It is suggested that the change of positional relationship of the polar residues in TMD5 and 7 and/or their protonation-deprotonation would affect the electron distribution of the retinal-Schiff base.

2Pos042 タンパク質の構造と機能の相関を利用した Channelrhodopsin と MtrF の戦略的立体構造モデリング

Strategic modeling of channelrhodopsins and MtrF based on the correlation between protein structures and functions

Hiroshi C. Watanabe[1,2], Yuki Yamashita[2], Marcus Elstner[3], Hiroshi Ishikita[1,2] ([1]*UTokyo, RCAST*, [2]*UTokyo, School of Engineering*, [3]*Karlsruhe Institute of Technology*)

Molecular simulation has become an essential approach for molecular biology, which provides us valuable intuition about protein structure and dynamics. One of the ultimate destinations of the molecular simulation in biology may be prediction of protein structures and functions without conducting any experiments. Regardless of a constant development of computational technology, however, it still remains auxiliary just to confirm and explain experiments and far from prediction. Here, we address an alternative modeling approach by molecular simulations focusing on the close correlation between protein structures and functions. As successful cases, we present two studies of channelrhdopsin (ChR), and MtrF, which mainly consist of α-helices and β-sheets, respectively.

2Pos043 **機械受容チャネル MscL のゲーティングにおいてメカノセンサーとゲートは密接に連動する**

Mechanosensor and the gate are tightly coupled in the mechano-gating of the bacterial mechanosensitive channel MscL

Yasuyuki Sawada[1], Takeshi Nomura[2], Masahiro Sokabe[1] ([1]*Mechanobiology Lab Nagoya Univ. Grad. Sch. Med.*, [2]*Physical Therapy Grad. Sch. Health Sciences Kyushu Nutrition Welfare Univ.*)

The bacterial mechanosensitive channel MscL is constituted of homopentamer of a subunit with TM1 inner and TM2 outer transmembrane helix. The major issue on MscL is to understand the gating mechanism driven by membrane tension. Upon membrane stretch, the helices are dragged by lipids at the tension sensor F78 and tilted, accompanied by outward sliding, leading to a gate expansion. To get insights into the relationship between F78 and the gate including G22, we performed MD simulations of G22N GOF MscL and spontaneous opening was shown during closed. Furthermore, the closed MscL can transmit resting tension to the gate via the interaction between F78 in TM2 and I32-L36-I40 in the neighboring TM1 and the transmitted force can lead to slight opening of the pore in G22N.

2Pos044 **電位依存性プロトンチャネル VSOP/Hv1 における亜鉛イオンの結合様式の解析**

A detailed analysis of the binding mode of a zinc ion to the voltage-gated proton channel VSOP/Hv1

Hiroko X. Kondo[1,2], Masayo Iwaki[3], Yusuke Kanematsu[1], Matsuyuki Shirota[2,4,5], Yasushige Yonezawa[6], Kengo Kinoshita[2,5,7], Hideki Kandori[3], Yu Takano[1] ([1]*GSIS, Hiroshima City Univ.*, [2]*GSIS, Tohoku Univ.*, [3]*Grad. Sch. Eng, Nagoya Inst tech*, [4]*Grad. Sch. Med., Tohoku Univ.*, [5]*ToMMo, Tohoku Univ.*, [6]*IAT, Kinki Univ.*, [7]*IDAC, Tohoku Univ.*)

VSOP/Hv1 is the voltage-gated proton channel and is sensitive to the pH gradient and electric potential across the membrane. Zinc ions also regulate its activation. The experimental results suggest that four residues: E115, D119, H136, and H189 are significant for the binding of zinc ions. However detailed mechanisms of the regulation by zinc ions still remain unclear. Here we analyzed the binding mode of a zinc ion to VSOP/Hv1 in detail by a combination of experimental and computational methods. We sampled several conformations of the protein in zinc-bound states by using the molecular dynamics simulation, and computed their vibrational modes by using quantum mechanics/molecular mechanics calculations.

2Pos045 **Molecular dynamics investigation of the full maltose transporter with and without the maltose binding protein MalE**

WeiLin Hsu, Tadaomi Furuta, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech.*)

In study, we performed MD simulations to examine the maltose transporter's fluctuations and investigated the correlation between each subdomain. Our results indicated that only binding Mg^{2+}-ATP to the NBDs is not sufficient to facilitate the maltose transporter from the resting (open) state to closed state. On the other hand, the maltose binding protein (MalE) may be the key subdomain to mediate the closure of this transporter. With MalE, the motion of $MalK_2$ showed obvious negative correlations with the MalF-P_2 region; however, without MalE, the correlations between MalF-P_2 region and $MalK_2$ decreased significantly. This observation gave us a new point of view to understand how a binding protein mediates the conformational change of an ABC importer.

2Pos046* **Binding and conformational dynamics of TOM20 and mitochondrial targeting signals using computational methods**

Arpita Srivastava[1], Osamu Miyashita[2], Florence Tama[1,3] ([1]*Dept. Phys., Sch. Sci., Nagoya Univ.*, [2]*RIKEN Adv. Inst. Comp. Sci.*, [3]*ITbM, Nagoya Univ.*)

Most mitochondrial proteins are imported from the cytosol through the translocase complex of the outer mitochondrial membrane (TOM). One of the components of TOM, Tom20, recognizes the targeting signals in N-terminal mitochondrial precursor proteins, which are then delivered to mitochondria. X-ray structures of Tom20/presequence complex can be classified into different conformations; A, Y and M based on the binding mode of Tom20 to presequence. This complex exists in dynamic-equilibrium between these multiple conformations. The co-crystallization of Tom20/presequence requires disulfide tethering. We are using methods like molecular dynamics (MD) and replica-exchange molecular dynamics (REMD) simulations of tethered Tom20/presequence complex to study its dynamics.

2Pos047 **細胞間接着結合における α カテニン分子の力-構造-機能ダイナミクス**

Mechanical, structural and functional dynamics of α-catenin molecule at intercellular adherens junctions

Koichiro Maki[1,2], Taiji Adachi[1,2] ([1]*Institute for Frontier Medical Sciences, Kyoto University*, [2]*Department of Micro Engineering, Graduate School of Engineering, Kyoto University*)

At intercellular adherens junctions, α-catenin molecule plays as a tension-sensor under intercellular tension by changing its conformation to recruit vinculin. In this study, we explored dynamical behaviors of the α-catenin molecule from mechanical, structural and functional aspects by employing 1) nanofishing and 2) structural imaging using AFM. As a result of AFM nanofishing, we found that the unfolding force of α-catenin is increased by "holding" to wait for its conformational change under tension. In addition, the conformational change was determined by AFM structural imaging. This mechano-adaptive conformational change is critical for α-catenin to play as a tension-sensor by keeping its functional structure under intercellular tension after mechanical activation.

2Pos048 **高速 AFM によるバクテリアコンデンシン複合体 MukBEF の構造動態の研究**

Structural dynamics of bacterial condensin complex MukBEF studied by HS-AFM

Hironori Yoneda[1], Kouichi Yano[2], Noriyuki Kodera[3,4], Kenta Yagi[1], Hironori Niki[2], Toshio Ando[1,3,5] ([1]*Div. of Math & Phys. Sci., Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ.*, [2]*Natl. Inst. of Genet.*, [3]*Bio-AFM FRC, Inst. of Sci. & Eng., Kanazawa Univ.*, [4]*PRESTO, JST*, [5]*CREST, JST*)

Using HS-AFM, we have been analyzing the structural dynamics of bacterial condensin complex MukBEF in the presence of various nucleotides and/or DNAs. HS-AFM images showed that MukB takes two structural states, closed- and open-forms, in which the two heads of MukB are attached and detached, respectively. The population of closed-form increases slightly in the presence of ATP or ATP analogs that induce putative ATP- or ADP.Pi-bound states. This tendency was observed even in the presence of MukE and MukF, although the mass of MukB head increases by the binding of MukE and MukF. MukBEF does not bind dsDNA, whereas solo MukB bind dsDNA and slide along it while keeping attached to dsDNA with the heads. We will also report the study of interaction between MukBEF and ssDNA.

2Pos049 高速 AFM によるダイナミン 1ーアンフィファイジン複合体の動態観察

High-Speed AFM imaging of dynamics of Dynamin1-Amphiphysin1 complexes

Daiki Ishikuro[1], Tetsuya Takeda[3], Toshiya Kozai[1], Yusuke Kumagai[1], Kaho Seyama[3], Huiran Yang[3], Hiroshi Yamada[3], Takayuki Uchihashi[1,2], Toshio Ando[1,2], Kohji Takei[3] ([1]*College of Science and Engineering, Kanazawa Univ.*, [2]*Bio-AFM FRC, Kanazawa Univ.*, [3]*Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama Univ.*)

Dynamin is a mechanochemical GTPase required for membrane fission in endocytosis. Dynamin forms a higher ordered "helical" structure which cuts membrane in a GTP hydrolysis-dependent manner. Previous studies proposed two possible mechanisms of the Dynamin-mediated membrane fission: "twist and cut" (pinchase) or "tear and cut" (poppase). However, precise mechanism of membrane fission by Dynamin remains to be elucidated.
In this study, we applied High-Speed AFM to analyze the Dynamin-mediated membrane fission. Using cell-free system consisting of liposomes and proteins, we have successfully visualized Dynamin dynamics coupled with GTP hydrolysis. In the presentation, we will discuss possible mechanisms of the Dynamin-mediated membrane fission.

2Pos050* 過飽和条件下におけるタンパク質分子内二次構造変化と芳香族アミノ酸立体配置

Internal Secondary Structural Changes and Aromatic Rings Conformation of Protein under Supersaturated Condition

Yufuku Matsushita[1], Hiroshi Sekiguchi[2], Jae-won Chang[1], Keigo Ikezaki[1], Masaki Nishijima[3], Daizo Hamada[4], Yuji Goto[5], Yuji Sasaki[1,2] ([1]*Grad. Sch. Front. Sci., Univ. Tokyo*, [2]*JASRI/SPring-8*, [3]*UIC, Osaka Univ.*, [4]*Int. Res. Cent., Kobe Univ.*, [5]*IPR, Osaka Univ.*)

Protein equilibrium clusters formation in supersaturated solution had been reported by several approaches. However, the driving force of this molecular conformation still has not been figured out in detail. Here, we report the structural changes of lysozyme within the equilibrium clusters at the supersaturated condition by Circular Dichroism spectroscopy. The molar ellipticity in specific wave length regions clearly demonstrated that the secondary structure ratio (200- 240 nm) and aromatic rings conformation (270- 300 nm) of lysozyme were drastically changed in supersaturated crystal nucleation phase condition (4, 15 ℃) compared with the control (25 ℃). The results were supported by high-speed Diffracted X-ray Tracking (DXT) and nano-scale MD-simulation study.

2Pos051 二次元蛍光寿命相関分光法による脂質膜上シトクロム *c* の環境に依存した構造ゆらぎの研究

Study of environment dependent dynamics of cytochrome *c* on a lipid membrane by 2D fluorescence lifetime correlation spectroscopy

Miyuki Sakaguchi[1], Masaru Yamanaka[2], Shun Hirota[2], Kunihiko Ishii[1,3], Tahei Tahara[1,3] ([1]*Molecular Spectroscopy Lab. RIKEN*, [2]*NAIST Graduate School of Materials Science*, [3]*RIKEN Center for Advanced Photonics*)

Conformational fluctuation of cytochrome *c* (cyt*c*) is induced on a cardiolipin (CL)-containing lipid membrane, which mimics an inner mitochondrial membrane. This fluctuation has been proposed to trigger the initial step of apoptosis. In this study, the dynamics of dye-labeled horse cyt*c* was investigated by 2D fluorescence lifetime correlation spectroscopy. Fluctuation between a partially unfolded conformational ensemble and unfolded state was detected in the microsecond region. In addition, the ensemble averaged fluorescence decay curves indicated that the equilibrium between the partially unfolded ensemble and unfolded state was affected by the CL concentration and cyt*c*-lipid ratio. It reflects the environment-dependent plasticity of cyt*c* on the membrane.

2Pos052* 一分子蛍光分光測定により明らかになったユビキチンの変性状態の不均一性とゆっくりとしたダイナミクス

Significant Heterogeneity and Slow dynamics of the Unfolded Ubiquitin Detected by Single-Molecule Fluorescence Spectroscopy

Masataka Saito[1,2], Kamonprasertsuk Supawich[1], Keiichiro Kushiro[3], Madoka Takai[3], Eric H.-L. Chen[4], Po-Ting Chen[4], Rita P.-Y. Chen[4], Hiroyuki Oikawa[1,2], Satoshi Takahashi[1,2] ([1]*Grad. Sch. Sci., Tohoku Univ.*, [2]*IMRAM, Tohoku Univ.*, [3]*Sch. Eng., Univ. Tokyo.*, [4]*IBC, Academia Sinica*)

We investigated the equilibrium folding of ubiquitin by line confocal method of single molecule fluorescence spectroscopy. We obtained time series of sm-FRET efficiency from the labeled ubiquitin at the temporal resolution of 100 microseconds. At low concentrations of denaturant, we obtained traces assignable to the native state. In contrast, traces with low FRET efficiencies were detected at the higher denaturant concentrations, corresponding to the unfolded state. Our data suggested that the denatured ubiquitin are heterogeneous and that the transitions among the heterogeneous conformations occur slower than several milliseconds. We hypothesize that the broadening in the unfolded state was caused by the local structural heterogeneity around the labeled fluorophores.

2Pos053 高活性型 DHFR 変異体の構造揺らぎの NMR 解析

NMR analysis of structural fluctuation of a highly active mutant of *Escherichia coli* dihydrofolate reductase

Takuro Nobe, Yuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

Escherichia coli dihydrofolate reductase (DHFR) is well known as a model protein for the study of the relationship between enzymatic function and protein dynamics. Previous studies have shown that the transition rate for each step in the catalytic cycle of DHFR is consistent with the rate of structural fluctuation, suggesting that protein structural fluctuation determines the rates of enzymatic reactions. To clarify whether protein fluctuation is fast for highly active mutants, here we study the dynamics of a highly active mutant of DHFR by NMR relaxation measurements. We have purified a mutant protein of DHFR that has higher kcat than the wild type. The NMR results will be presented at the meeting.

2Pos054 C-ペプチド領域をもたないウシ膵臓インスリンの二本鎖酸化的フォールディング経路

Double-chain oxidative folding pathways of bovine pancreatic insulin without C-peptide region

Kenta Arai[1], Toshiki Takei[1,2], Reina Shinozaki[1], Yuya Asahina[2], Hironobu Hojo[2], Michio Iwaoka[1] ([1]*Tokai Univ.*, [2]*Inst. for Prot. Res., Osaka Univ.*)

Insulin, which comprises two peptide chains, A-chain and B-chain, is stabilized by two interchain disulfide (SS) bonds, Cys^{A7}-Cys^{B7} and Cys^{A20}-Cys^{B19}, and one intrachain SS bond, Cys^{A6}-Cys^{A11}. The time course of SS intermediates during the double-chain folding showed that $[Cys^{A6}\text{-}Cys^{A11}]^{A}$, which is the most populated along 1SS species of A-chain, is first accumulated, and subsequently dimerize by oxidative combination with reduced B-chain to form metastable intermediate, 2SS* ($[Cys^{A6}\text{-}Cys^{A11}, Cys^{A20}\text{-}Cys^{B19}]^{AB}$), as a precursor of the native state. Finally, the major folding pathways of double-chain insulin were clearly characterized here by isolation and structural identification of key SS intermediates for the first time.

2Pos055* 圧力ジャンプ FTIR 法を用いた β ラクトグロブリンのフォールディング反応解析

A pressure-jump FTIR study of the folding reaction of β-Lactoglobulin

Satoshi Hayakawa, Tsubasa Yamamoto, Minoru Kato (*Dept. Applied Chemistry, Ritsumeikan Univ.*)

β-Lactoglobulin has a high α-helical propensity predicted from the amino acid sequence, although its native form is predominantly composed of β-structure. Some research groups suggested that this protein has an intermediate with non-native α-helical structure in the refolding process. In this study, we have carried out pressure-jump kinetic studies to analyze the secondary structural change in the folding and unfolding reactions of βLG using FTIR spectroscopy. Based on these results, we have calculated reaction rates and activation volumes. Finally, we discuss whether the intermediate with non-native α-helical structure exists in refolding process.

2Pos056 700 MPa 高圧下で観測された蛍光蛋白質 Akane families の特異な蛍光挙動

Unique properties of a GFP-like protein Akane families, observed under 700 MPa high-pressure

Akihiro Maeno[1], Yuko Kato[2], Mitsuru Jimbo[3], Kei Amada[4], Kazuyuki Akasaka[5] ([1]*Chem., Kansai Med. Univ.*, [2]*Electro. Res. Lab., Fukuoka Inst. Tech.*, [3]*Marine Biosci., Kitasato Univ.*, [4]*Life, Environ. and Mat. Sci., Fukuoka Inst. Tech.*, [5]*Grad. Sch. Life and Environ. Sci., Kyoto pref. Univ.*)

We carried out high-pressure fluorescence spectroscopy up to 700 MPa to study the physicochemical property of a GFP-like protein named Akane families isolated from the octocoral Scleronephthya gracillima. We found some unique features; [1] Akane families consist of two forms emitting green fluorescence at 510 nm and red fluorescence at 629 nm, and they are neither interconvertible by photo-irradiation or thermally at native type. [2] The red fluorescence emitting form consists of two substates in thermal equilibrium. [3] Pressure unfolding takes place beyond 500 MPa. These are the first report concerning to the physicochemical properties of multicolor fluorescent protein. The overall relationship including the biological and structural properties will be discussed.

2Pos057 タンパク質ジスルフィドイソメラーゼ様触媒活性を有する新規低分子ジセレニド化合物を用いた酸化的フォールディングに関する研究

Study on the oxidative folding by using a novel small-molecular diselenide compound having protein disulfide isomerase-like activity

Yuki Asano, Haruhito Ueno, Michio Iwaoka, Kenta Arai (*Tokai Univ.*)

A nascent polypeptide chain efficiently folds into the native state through the formation of disulfide (SS) bonds between the cysteinyl thiols. This oxidative folding process is generally assisted by protein disulfide isomerase (PDI) having a CXXC motif as the active center. In this study, a new class of water-soluble cyclic diselenide (SeSe) reagents, which have a low redox potential (< -370 mV), were applied as a small-molecular PDI-like catalyst to the oxidative folding experiments of bovine pancreatic ribonuclease A (RNase A) and hen egg white lysozyme (HEL). As the results, SeSe reagents catalyzed the in vitro oxidative folding of both reduced forms of RNase A and HEL. In the poster session, the details for the PDI-like catalytic mechanisms will be discussed.

2Pos058* 分子内ジスルフィド結合がリポカリン型プロスタグランジン D 合成酵素の熱安定性及び構造安定性に与える影響

Effects of an intramolecular disulfide bond on the thermal and conformational stability of lipocalin-type prostaglandin D synthase

Shogo Atsuji[1], Yoshiaki Teraoka[1], Young-Ho Lee[2], Yuji Goto[2], Takashi Inui[1] ([1]*Grad. Sch. Life & Envi. Sci., Osaka Pref. Univ.*, [2]*Inst. Prot. Res., Osaka Univ.*)

Lipocalin-type prostaglandin D synthase (L-PGDS) is an extracellular transporter for lipophilic molecules and a novel drug delivery vehicle for poorly water-soluble drugs. L-PGDS has a typical lipocalin fold that consists of an eight-stranded anti-parallel β-barrel and a highly conserved internal disulfide (SS) bond.
In the present study, we investigated the role of SS bond of L-PGDS in the thermal and conformational stability using CD and NMR spectroscopy with MD simulation. CD measurements showed that the T_m values of L-PGDS and SS bond-deleted mutant were 68.8°C and 57.8°C, respectively. The ^{1}H-^{15}N HSQC measurements and MD simulations revealed that the removal of SS bond influenced the dynamics of L-PGDS, especially affecting the β-hairpin structure close to SS bond.

2Pos059 タンパク質の翻訳時フォールディングにおけるリボソーム効果の粗視化分子動力学シミュレーション研究

Ribosomal effects on protein cotranslatoinal folding studied by coarse-grained molecular dynamics simulation

Kazushi Mochizuki, Shoji Takada (*Graduate School of Science, Kyoto Univ.*)

Many, if not all, proteins are known to fold while they are being synthesized by the ribosome. But, mechanisms that such cotranslational folding enhances the efficiency of successful protein folding have not been understood. Using coarse-grained molecular dynamics simulation, we study the effect of ribosomal surface. A recent report suggested that the ribosomal surface can work as a kind of chaperone in the folding of the first nucleotide-binding domain (NBD1) of the cystic fibrosis transmembrane conductance regulator (CFTR). We conduct cotranslational folding simulations of that protein, where the system contains a ribosome tunnel and surface represented by simple geometry and interactions.

2Pos060 蛋白質の相互作用とダイナミクスに及ぼす高分子混み合いの影響：全原子分子動力学法による研究

Influence of Macromolecular Crowding on the Dynamics and Interactions of proteins: All-atom Molecular Dynamics Study

Isseki Yu[1], Tadashi Ando[2], Takaharu Mori[1], Jaewoon Jung[3], Ryuhei Harada[3], Yuji Sugita[1,2,3], Michael Feig[4] ([1]*RIKEN*, [2]*RIKEN QBIC*, [3]*RIKEN AICS*, [4]*Michigan State Univ.*)

Inside of a cell is highly crowded with a large number of macromolecules together with solvents and metabolites. How variable interactions within dense cellular environments may affect the structure and dynamics, and ultimately function is one of the most fundamental questions in life science. We constructed full atomistic model of the cytoplasm of bacteria and various levels of protein crowding models. Using these model, we performed MD simulation with the highly parallelized MD program GENESIS. Influence of crowding level on the translational/rotational motion of macromolecules are analyzed combined with NMR data. The corrective motions between macromolecules and their molecular mechanisms are also discussed.

2Pos061* 高温条件下で変性したDEN4 ED3の可逆的なオリゴマー形成の一残基置換による阻害

Unusual reversible oligomerization of unfolded Dengue envelope protein domain 3 at high temperature and its abolition by a point mutation

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We report DSC experiments between 10 and 120°C of Dengue 4 envelope protein domain 3 (DEN4 ED3), a small 107 residue monomeric globular protein domain. The thermal unfolding of DEN4 ED3 was fully reversible but exhibited two very peculiar endothermic peaks, which separated away with increasing protein concentrations. This strongly suggested the presence of unfolded tetramers at temperatures around 80-90°C, which dissociated to unfolded monomers at even higher temperature. Furthermore, we constructed a DEN4 ED3 V380K variant that underwent a fully reversible two-state thermal unfolding indicating that the formation of temperature oligomer was successfully abolished by a single mutation.

2Pos062 球殻状超分子集合における局所的相互作用の役割

The role of local interactions on the spherical shell-shaped supermoleculer assembly

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Escherichia coli ferritin is a spherical shell-shaped supermoleculer consisting of 24 identical subunits arranged with 4/3/2 symmetry. Previously, we have studied the assembly mechanism of this supermolecule using time-resolved small-angle X-ray scattering. To elucidate the role of individual interfaces in the assembly, we prepared six point mutants (H106A, H106P, F117A, H128A, H128F and H128V) and two deletion mutants (139Δ and 146Δ), in which mutation sites were selected for perturbing interfaces around 3- and 4-fold symmetry axes, respectively. The structures of these mutants were analyzed by circular dichroism, small-angle X-ray scattering and ultracentrifugation. Although H106P and 139Δ were not able to form 24-mer, other mutants could form 24-mer.

2Pos063 赤外分光法によるカルシウム結合タンパク質の金属配位構造解析 - 合成ペプチドアナログの凝集による問題

Infrared study of the Ca^{2+}-coordination structures of Ca^{2+}-binding proteins: the problem of aggregation of synthetic peptide analogues

Masayuki Nara[1], Hisayuki Morii[2], Takuya Miyakawa[3], Masaru Tanokura[3] ([1]*TMDU*, [2]*AIST*, [3]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*)

Infrared spectroscopy has been applied to study the synthetic peptide analogues of the Ca^{2+}-binding sites of Ca^{2+}-binding proteins. We investigated the 17-residue peptide analogues corresponding to the site I-IV of the three Nicotiana tabacum calmodulin (NtCaM) isoforms: NtCaM1, NtCaM3 and NtCaM13. The bands at about 1553 cm^{-1}, which are due to the bidentate coordination mode, were observed for most peptide analogues for each NtCaM isoform in the Ca^{2+}-bound state. However, some 17-residue peptide analogues aggregated in the apo and/or Ca^{2+}-loaded states and the Ca^{2+}-bound coordination structure was not formed due to sample aggregation. We discuss the problem of aggregation for the model peptide analogues on the basis of infrared spectral profiles.

2Pos064* 競争的凝集形成機構に基づいた蛋白質異常凝集の理解

Understanding of aberrant protein aggregation based on the competitive aggregation mechanism

Masayuki Adachi, Masatomo So, Yuji Goto (*Inst. Protein Res., Osaka Univ.*)

Although many diseases have been found to be associated with protein aggregates (amyloid fibril and amorphous aggregate), the mechanism is unclear. Here, we focused on heating rate to elucidate the aggregation mechanism. At pH 2.0, upon heating the solution, the transition to fibrils occurred at a certain temperature. Then, fibrils were degraded at higher temperatures. At pH 4.8, amorphous aggregation occurred at fast heating rate. Subsequently, fibrils formed accompanied by dissolution of amorphous aggregates. From these results, we suggest that (1) amorphous aggregates form at fast heating rate by increasing the protein-protein interaction, (2) fibrils form coupled with dissolution of preformed amorphous aggregates, and (3) fibrils degrade at higher temperatures.

2Pos065 ヘパリンによるアミロイド線維形成の促進と抑制の分子機構

Molecular mechanism underlying the heparin-induced acceleration and inhibition of amyloid fibrillation

Ayame Nitani, Hiroya Muta, Masayuki Adachi, Masatomo So, Yuji Goto (*Inst. Protein res., osaka Univ.*)

It is known that there are two types of aggregates; i.e. amyloid fibrils and amorphous aggregates. It has been suggested that the distribution of these species depends on the concentrations of additives. Here, we focus on the effect of heparin on aggregation of hen egg white lysozyme at acidic pH.
At low heparin concentrations, amyloid fibrils did not form. At medium heparin concentrations, only amorphous aggregates formed immediately, but amyloid fibril did not. Interestingly, at high heparin concentrations, amorphous aggregation did not occur, whereas amyloid fibrillation occurred after several hours under ultrasonication. These results indicate the concentration-dependent complicated effects of heparin on the acceleration and inhibition of fibrillation.

2Pos066 キメラカルシトニンによるヒトカルシトニンアミロイド凝集阻害機構の解明

Analysis of amyloid formation and inhibition mechanisms of human calcitonin by chimera calcitonin

Chiaki Ota[1], Hiroko Tanaka[1], Tomoyasu Aizawa[2], Yoichi Yamazaki[1], Mikio Kataoka[1], Hironari Kamikubo[1] ([1]*Grad. Sch. Mat. Sci., NAIST*, [2]*Grad. Sch. Life Sci., Hokkaido Univ.*)

Human Calcitonin (hCT) is a peptide hormone used as medication. However, it easily forms amyloid fibrils. Salmon calcitonin (sCT) had been known to suppress amyloid formation of hCT, kinetically. In the previous study, we revealed that a chimera peptide (shCT) derived from sCT and hCT strongly inhibits the amyloid formation of hCT. In order to elucidate the inhibition mechanisms, we investigated amyloid formation reaction of hCT when adding amyloid fibrils of hCT treated by shCT as amyloid seeds. In the results, we observed obvious lag phase, as if there were no amyloid seeds. The fact suggests, that shCT can be tightly bound to the terminal extension of amyloid fibrils to inhibit the amyloid fibril elongation.

2Pos067* **プリオンタンパク質とプリオンタンパク質を標的とする RNA 分子の Aβ 線維化への影響**

The effects of prion protein and a RNA molecule that binds to prion protein on Aβ fibrillation

Mamiko Iida[1,2], Tsukasa Mashima[1,2], Yudai Yamaoki[2], Takashi Nagata[1,2], Masato Katahira[1,2] ([1]*Grad. Sch. of Energy Sci., Kyoto Univ.*, [2]*Inst. of Adv. Energy, Kyoto Univ.*)

Amyloid β protein (Aβ) forms oligomers and fibrils. Recently, it was proposed that prion protein (PrP) anchored on a cell membrane acts as a receptor of Aβ oligomers, and their binding triggers synaptic dysfunction related with Alzheimer's disease. A RNA molecule, R12, tightly binds to a lysine rich region of PrP that is also a known binding-site for Aβ oligomers. Therefore, R12 was thought to compete with Aβ oligomers. Here, the time-course of Aβ fibrillization was monitored by fluorescence of thioflavin S in the absence or presence of PrP and/or R12. PrP effectively inhibited the Aβ fibrillization in the absence of R12, however, the fibrillization restarted upon addition of R12. This suggests that R12 hinders the inhibitory effect of PrP by competitive binding to PrP.

2Pos068 **High-level expression, purification and characterization of the plant antimicrobial peptide snakin-1 in *Pichia pastoris***

Md. Ruhul Kuddus[1,2], Farhana Rumi[1], Motosuke Tsutsumi[1], Megumi Yamano[1], Masakatsu Kamiya[1], Takashi Kikukawa[1], Makoto Demura[1], Tomoyasu Aizawa[1] ([1]*Grad. Sch. Life Sci., Hokkaido Univ.*, [2]*Dhaka Univ.*)

Snakin-1 (SN-1) is a cysteine-rich plant antimicrobial peptide with strong antimicrobial activity which was isolated from potato (*Solanum tuberosum*). Here, we carried out the expression of a recombinant SN-1 in the methylotrophic yeast *Pichia pastoris*. A large amount of pure recombinant SN-1 (about 40 mg/1L culture) was obtained after purification. The identity of the recombinant SN-1 was verified by MALDI-TOF MS, CD and ^{1}H NMR experiments. These results indicated that the recombinant SN-1 had a folding with six disulfide bonds that was identical to the native SN-1. The membrane permeability assay indicated that SN-1 can disrupt both outer and inner membrane of bacterial cell. This is the first report on the recombinant expression of bioactive SN-1 in *Pichia pastoris*.

2Pos069 **A study on tryptophan-dependent translation termination arrest of TnaC**

Tomoki Shinozawa, Ryo Iizuka, Zhuohao Yang, Takashi Funatsu (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

The *Escherichia coli* TnaC, a 24-amino acid peptide, regulates the expression of the downstream genes (*tnaA* and *tnaB*) via its tryptophan-dependent translation termination arrest. In brief, in response to elevated tryptophan concentration in the cell, translation termination of TnaC is inhibited, allowing transcription and translation of *tnaA* and *tnaB*. In order to investigate the molecular mechanism of the tryptophan-dependent translation termination arrest, we performed *in vitro* translation assays using TnaC with a tetra-cysteine tag to analyze the lifetimes of translation termination arrest at different concentrations of tryptophan. In the meeting, the details of the results will be discussed.

2Pos070* **1 分子不凍タンパク質の温度依存ダイナミクスと AgI との吸着関係**

Temperature Dynamics of Single Molecular AntiFreeze Proteins and adsorption to AgI interaction

Rio Okada[1], Tatsuya Arai[2], Yuhuku Matushita[1], Jae-won Chang[1], Hiroshi Sekiguchi[3], Tadashi Mori[4], Masaki Nishijima[4], Keigo Ikezaki[1], Sakae Tsuda[2], Yuji Sasaki[1,3] ([1]*Frontier Science., Adv. Material Science., Tokyo Univ.*, [2]*AIST / Grad. Schl. of Life Sci., Hokkaido Univ.*, [3]*JASRI*, [4]*Grad. Schl. of Eng., Div. of Ap. Chem., Osaka Univ.*)

AntiFreeze Proteins (= AFPs) attach to a surface of ice crystals and inhibit their growth. So far the adsorption property of AFPs is not investigated with single molecular level. Here, we performed single molecule lpAFP observations using Diffracted X-ray Tracking (= DXT) at SPring-8 BL40XU. Using DXT, we observed the molecular dynamics of lpAFP and its adsorption property to AgI, which has a similar crystal structure to ice, under several temperature conditions. From these experiments, we revealed that the single molecular lpAFP dynamics became larger at 5℃ and lpAFP adsorption to AgI was increased at 2℃. These results suggest that there is a strong correlation between lpAFP’s molecular motion and adsorption property.

2Pos071 **青色光センサータンパク BlrP1 の反応の光強度依存性**

Light intensity determines photoreaction of blue-light sensor protein BlrP1

Kosei Shibata, Yusuke Nakasone, Masahide Terazima (*Grad. Sch. Sci. Unv. Kyoto*)

BlrP1 is a bacterial blue-light sensor protein consisting of a BLUF domain and an EAL domain. The BLUF absorbs blue light and enhances an enzymatic activity of the EAL. Previously, we reported that BlrP1 is in an equilibrium between the monomer and the dimer in solution, and only the dimer undergoes a conformational change in the EAL region upon photoexcitation. In this study, we examined the excitation power dependence on the photoreaction by the transient grating method and found that reaction yield strongly depended on the energy of the excitation pulse. From the detailed analysis, we concluded that the conformational change occurs only when both two monomer units in the dimer are excited. This indicates that the BlrP1 works as a light intensity sensor in nature.

2Pos072 **反転膜小胞を用いたべん毛軸構造蛋白質の輸送順序の解析**

Secretion order of the Class-II flagellar axial proteins analyzed by inverted membrane vesicles (IMV)

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Many bacteria swim in aqueous environment by rotating a filamentous organelle, the flagellum. The flagellar axial structure is composed of the rod, the hook and the filament. The flagellar axial component proteins are exported through the flagellar type III protein export apparatus to the distal end for self-assembly. The export apparatus transports 20 to 30 thousands of protein subunits of more than 12 different types of proteins. Thus protein secretion control is essential to construct the flagellum. Here we investigated the secretion order of the class-II flagellar axial proteins, which is needed for construction of the rod and the hook, using IMV. The results indicated that the secretion order of the class-II proteins related to the proximal rod formation.

2Pos073 Alp7/TACC-Alp14/TOG protein complex promotes assembly of *S. pombe* microtubules

Douglas Drummond[1], Frauke Hussmann[2], Daniel Peet[2], Douglas Martin[3], Robert Cross[2] ([1]*Faculty of Agriculture, Kyushu Univ.*, [2]*Warwick Medical Sch., UK*, [3]*Lawrence Univ., USA*)

Purified tubulins will self-assemble to form dynamic microtubules (MTs), however MT dynamics are normally modulated by MT associated proteins in vivo. We have used darkfield microscopy to measure the dynamics of MTs assembled from purified *S. pombe* yeast tubulin in vitro and found MT growth rates were reduced compared to those we previously observed in live *S. pombe* cells. However, Alp14 a TOG-family MT plus end tracking protein can accelerate *S. pombe* MT assembly both in vivo and in vitro. We also found that the Alp14 binding protein Alp7/TACC was essential for this function in vivo. TIRF microscopy in vitro showed that Alp7 increases the occupancy of Alp14 on MT ends. Together Alp7 and Alp14 promote MT formation at the low concentrations of tubulin found in vivo.

2Pos074 タウタンパク質に対する Pin1 由来のプロテアーゼの活性
Activity of a protease derived from Pin1 for tau protein

Teikichi Ikura, Nobutoshi Ito (*Med. Res. Inst., Tokyo Med. Dent. Univ.*)

The Alzheimer's disease-related protein, tau, aggregates into neurofibrillary tangles when it is hyperphosphorylated. A peptidyl-prolyl isomerase (PPIase), Pin1, targets a specific pS/T-P motif of the hyperphosphorylated tau, and restores the function of tau. The function of Pin1 for tau, however, is not effective enough to prevent progress of dementia. Recently we succeeded in converting the PPIase activity of Pin1 into the proteolytic activity by a single mutation. In the present study, we investigated how efficiently the proteolytic activity of the Pin1 mutant worked on tau protein and its aggregate. The availability of this activity of the mutant protein will be discussed on the basis of these results.

2Pos075* アミロイド線維形成の圧力依存性を解明するための計算研究
Computational Research to Reveal the Pressure Dependency of the Formation of Amyloid Fibrils

Naohiro Nishikawa[1,2], Yoshiharu Mori[2], Yuko Okamoto[1,3,4,5], Hisashi Okumura[2,6] ([1]*Grad. Sch. of Sci., Nagoya Univ.*, [2]*IMS*, [3]*Str. Biol. Res. Cent., Grad. Sch. of Sci., Nagoya Univ.*, [4]*Cent. for Comp. Sci., Grad. Sch. of Eng., Nagoya Univ.*, [5]*Info. Tech. Cent., Nagoya Univ.*, [6]*SOKENDAI*)

Misfolding and self-assembly of proteins are considered to be the causes of various diseases. A disease group named Amyloidosis is thought to be caused by protein misfolding into insoluble aggregates referred to as amyloid fibrils.
There are some reports that the atomic structures of the amyloid fibrils may be changed at high pressures, but the detailed structures are not known yet. In order to reveal the atomic structures of amyloid fibrils under several pressures, we performed molecular dynamics simulations. We applied simulated tempering method for isobaric-isothermal ensemble, which is a simulation technique for efficient sampling, to the system consisting of 5 fragments of Amyloid-beta peptide to discuss the pressure dependency of amyloid fibril formations.

2Pos076* ESI-QTOF MS 法を用いたアミロイドベータペプチドのオリゴマー形成機構の解析
ESI-QTOF MS analyses of the oligomerization mechanism of amyloid β peptides

Shintaro Yoshida[1], Sosuke Yoshinaga[1], Mitsuhiro Takeda[1], Ayumi Tanaka[1], Takashi Hamaguchi[1], Hitomi Yamaguchi[1], Shigeto Iwamoto[1], Takashi Saito[2], Yoshihiko Takinami[3], Toshiyuki Kohno[4], Takaomi C. Saido[2], Hiroaki Terasawa[1] ([1]*Fac. Life Sci., Kumamoto Univ.*, [2]*RIKEN BSI*, [3]*Bruker Daltonics*, [4]*Kitasato Univ. Sch. Med.*)

The deposition of senile plaques is observed in the brains of Alzheimer's disease (AD) patients. Amyloid β peptide (Aβ) oligomers, formed in the process of senile plaque production, are thought to be neurotoxic in AD. Although the structures of Aβ fibrils have been elucidated by NMR, the structures of Aβ oligomers and the details of the Aβ oligomerization process remain to be elucidated. To reveal the intermolecular binding pattern of Aβ oligomers, we utilized ESI-QTOF MS to analyze Aβ dimers, which were generated by applying the PICUP (Photo-Induced Cross-linking of Unmodified Proteins) method to a mixture of stable isotope-labeled and unlabeled Aβ. Our results indicated that Aβ dimers mainly adopt a parallel β-sheet structure, in the same manner as Aβ fibrils.

2Pos077 Processing of XFEL still images with a reference oscillation data set for crystal structural analyses of Cytochrome c Oxidase

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X-ray free electron laser, which with the enormous energy, can easily disrupt the crystal. Serial Femtosecond Rotation Crystallography (SF-ROX) has been developed for a diffraction experiment method in SACLA. In this method a crystal mounted on a goniometer-head is exposed by the X-ray pulse and translates of the next exposure to obtain serial diffraction images with an equal rotation interval.
To obtain high quality data set from SF-ROX diffraction images we have initiated development of data processing procedure. A reference intensity data set obtained by the oscillation method using crystals grown in the same batch as those used in SF-ROX method will be used into the post-refine procedure instead of the reference generated by raw data set.

2Pos078 中性子タンパク質結晶構造解析での水素高感度検出のための動的核偏極法の予備的結果
The preliminary result of Dynamic Nuclear Polarization method for more sensitive detection of hydrogen in Neutron Protein Crystallography

Naoya Komatsuzaki[1], Ichiro Tanaka[1,2], Takahiro Iwata[3], Daisuke Miura[3], Yoshiyuki Miyachi[3], Genki Nukazuka[3], Hiroki Matsuda[3], Toshiyuki Chatake[4], Katsuhiro Kusaka[2], Nobuo Niimura[2] ([1]*Coll. of Eng., Ibaraki Univ.*, [2]*Frontier Res. Ctr., Ibaraki Univ.*, [3]*Faculty of Sci., Yamagata Univ.*, [4]*RRI, Kyoto Univ.*)

In order to improve the hydrogen sensitivity for neutron protein crystallography (NPC), dynamic nuclear polarization (DNP) of hydrogen in protein is expected to gain it about eight times larger than the normal NPC. Several technical difficulties, however, should be overcome to realize the DNP method in NPC; freezing a large protein single crystal and obtaining high polarization ratio of a sample crystal.
Previously, optimal concentration of radical in a protein single crystal for the DNP experiment could be determined by ESR method.
In the present research, a preliminary DNP experiment of protein single crystal was performed, and the proton polarization arrived at the maximum value of 22%.

2Pos079 Elucidating the mechanisms of proton pumping in cytochrome c oxidase by time resolved IR spectroscopy

Chen Li, Tatsuhito Nishiguchi, Shun Yamauchi, Kyoko Shinzawa-Itoh, Shinya Yoshikawa, Satoru Nakashima, Takashi Ogura (*Grad. Sch. Sci., Univ. Hyogo*)

Cytochrome c oxidase (CcO) is the terminal enzyme in the cellular respiration chain, which reduces molecular oxygen (O2) to water molecule. This reaction is coupled with proton pumping across the membrane. We developed a new time-resolved IR spectroscopy with flow cell system that can detect the enzyme reactions in water solution under physiological conditions. By using the new system, we succeeded to detect different spectra of CcO during reacting with oxygen under buffer solution. The low frequency peak shift in carboxyl group region could reveal the mechanism of hydrogen network change inside the protein during proton pumping. Changes of protonated state residues during the reaction intermediate could be clarified by our experiment.

2Pos080 ストップトフロー装置と過渡回折格子法を組み合わせたタンパク質反応検出手法の開発
Time-resolved detection of the transient grating signal using stopped flow system

Shunki Takaramoto, Yusuke Nakasone, Masahide Terazima (*Dept. of chem, Univ. Kyoto*)

We have developed a novel technique combining the transient grating (TG) method and stopped-flow (SF) apparatus to investigate the reaction dynamics of protein molecules. The TG method is a powerful technique to trace overall protein reactions including conformational changes and intermolecular interaction changes induced by photoexcitation. However, the reaction has to be triggered by light. In the new method, protein reaction is triggered by mixing of two solutions using an SF device, and the subsequent reactions are monitored by the TG method. This technique can be applied to proteins which do not react by light irradiation. Now, we are measuring the protein folding/unfolding kinetics by this newly developed technique.

2Pos081 顕微ラマン分光法による微小液滴内での酵素活性検出系の開発
Detection of enzymatic activity in femtliter droplets using micro-Raman spectroscopy

Hironobu Yamashita[1], Kazuhito Tabata V.[1,2], Hiroshi Ueno[1], Hiroyuki Noji[1,3] ([1]*Department of Applied Chemistry, the University of Tokyo*, [2]*PRESTO, JST*, [3]*ImPACT, CAO, Govt*)

Chitinase is an enzyme that degradates chitin fiber. Several methods for measuring chitinase activity are known these days; however, most of them require boiling or heating steps for color development. Fluorescence microscopy is one possible approach for high-throughput and highly sensitive detection, however, there is no commercially available fluorogenic substrate which has strong fluorescence in acidic conditions optimal for the chitinase activity. To detect chitinase activity in optimal conditions, we developed a new method for detecting enzymatic activity in small droplet arrays based on micro-Raman spectroscopy. By combining surface-enhanced Raman spectroscopy (SERS) and computational clustering analysis, we can monitor enzymatic reactions in femtliter droplets.

2Pos082 蛋白質周囲の水分子運動の測定を目的とした蛍光アップコンバージョン測定装置の開発
Development of fluorescence up-conversion apparatus to investigate hydration dynamics around the protein surface

Asahi Fukuda, Tomotaka Oroguchi, Masayoshi Nakasako (*Grad. Sci. Tech., Keio Univ.*)

Recent structural and molecular dynamics studies on hydration structures of an enzyme have revealed that domain motions of the enzyme are induced by the rearrangement of hydration water molecules in its active-site cleft. Dynamical motions of hydration water molecules can be investigated by the time-dependent Stokes shifts (TDSS), which is a spectral shift of dye fluorescence caused by the reorientation of water molecules around dye molecule. Because TDSS occurs in the time scale of a few to several hundred ps. We have been developing an up-conversion apparatus to observe sub-ps-resolved fluorescence from a tryptophan residue located in the ligand-binding site in an enzyme. Here we report our custom-made apparatus and its performance.

2Pos083 Grid inhomogeneous solvation theory を用いた血液凝固因子 Xa の水和解析
Hydration analysis of Coagulation Factor Xa using grid inhomogeneous solvation theory

Hiroyuki Sato, Azuma Matsuura (*Fujitsu Laboratories Ltd.*)

We investigated the hydration structure in the active site of human coagulation factor Xa (fXa). Water distribution in the active site of fXa was calculated using partly constrained MD simulation. Then free energy of the hydration structure was evaluated using grid inhomogeneous solvation theory (GIST). We found that the side chain fluctuation of the averaged structure of the MD trajectory has an important role for identifying the appropriate hydration structure. As a result, GIST in combination with the partly constrained MD provides useful information for assessing the binding affinity of known ligands.

2Pos084 キナーゼと ATP 競争阻害剤との結合自由エネルギー計算における、薬剤結合サイトの立体構造柔軟性がもたらす影響
The effect of conformational flexibility on the binding free energy calculation between kinases and their ATP-competitive inhibitors

Mitsugu Araki[1], Narutoshi Kamiya[1,2], Miwa Sato[3], Masahiko Nakatsui[1,5], Takatsugu Hirokawa[4], Yasushi Okuno[1,5] ([1]*RIKEN, AICS*, [2]*Grad. Sch. Sim. St., Univ. Hyogo*, [3]*Mitsui Knowledge Industry Co., Ltd.*, [4]*AIST, Molecular Profiling Research Center for Drug Discovery*, [5]*Grad. Sch. Med., Kyoto Univ.*)

Accurate prediction of protein-ligand binding affinities remains challenging because of protein flexibility in solution. Conformational flexibility of the ATP-binding site in the CDK2 and ERK2 kinases was identified by analyzing molecular dynamics simulations. The binding free energy (DG) of 24 inhibitors toward these kinases was assessed using an alchemical free energy perturbation method, MP-CAFEE. When the free energy simulation started from a single equilibrated conformation, large calculation errors were observed. However, when the starting structure was set to multi-conformations to cover flexibility, the calculation errors meaningfully decreased, demonstrating that conformational flexibility of protein-compound interactions is a key factor in DG estimation.

2Pos085* 溶質構造エントロピー計算法の理論的研究

Theoretical study for solute configurational entropy calculation methods

Simon Hikiri[1], Takashi Yoshidome[1,2], Mitsunori Ikeguchi[1] ([1]*Grad. Sch. of Med. Life Sci. Yokohama city Univ.*, [2]*Dept. of Appl. Phys., Tohoku Univ.*)

Configurational entropy (S_c) is an important component of free energies in biomolecular processes. We have assessed the accuracy of six methods, which are based on quasi-harmonic approximation, for S_c calculation from molecular dynamics simulation by comparison with results of theoretically (but conditionally) rigorous method. The Boltzmann-quasi-harmonic (BQH) method using internal coordinates outperformed the other methods. The introduction of improper torsions in the BQH method improves its performance, and anharmonicity of proper torsions in proteins is identified to be the origin of the superior performance of the BQH method.
In the presentation, we will present an application study with BQH method for biomolecular processes.

2Pos086 分子シミュレーションにおける静電相互作用計算法：零多重極子法の理論と実際

Computational method for electrostatic interactions in molecular simulation: theory and practice in the zero-multipole summation

Ikuo Fukuda[1], Han Wang[2], Narutoshi Kamiya[3], Kota Kasahara[4], Tohru Terada[5], Shun Sakuraba[6], Haruki Nakamura[1] ([1]*IPR, Osaka Univ.*, [2]*Freie Universitaet Berlin*, [3]*Univ. of Hyogo*, [4]*Col. Life Sci., Ritsumeikan Univ.*, [5]*Grad. Sch. Agr. Life Sci. Univ. Tokyo*, [6]*Grad. Sch. Frontier Sci., Univ. Tokyo*)

Almost all computation time in molecular simulation is spent for calculating the long-range interactions. The electrostatic long-range interactions require special cares, since the straight truncation does not serve due to the slow decay of the coulombic function. We have succeeded in computing the electrostatic interactions with low cost and high accuracy, by developing the zero-multipole summation method (ZMM). The ZMM is based on a simple physical assumption—electrostatic cancellation—and derived mathematically. It realizes a pairwise cutoff sum of a deformed potential function and requires neither the periodic boundary condition nor the reciprocal-space computation. We will present the theory, accuracy, speed, applications, and program implementation for the ZMM.

2Pos087 天然状態と似たそして異なるアポミオグロビン折り畳み中間体の構造

Native-like and non-native structures in the folding intermediate of apomyoglobin

Chiaki Nishimura (*Fac. Pharm. Sci., Teikyo Heisei Univ.*)

Protein folding is an important issue for its function. The folding kinetics and non-native structure in the intermediate are reported in the poster. On the basis of the combination studies of the pH-pulse labeling and NMR, the folding of the mutants of apomyoglobin was examined. 1) B-helix stabilized mutants indicated the faster folding than WT. Not only the region of B-helix but also that of G-H turn was stabilized in the intermediate. It was concluded that the structure in the intermediate for these mutants is native-like. 2) F-helix, which was not folded at the native state of WT, was folded in the intermediate of P88K/A90L/S92K/A94L. Slower folding was monitored for the mutant than for WT, suggesting that non-native structure exists in the intermediate.

2Pos088 理想タンパク質の安定性のオリジンを探る

Stability for de novo designed ideal proteins revisited

Mami Yamamoto[1,2], Rie Koga[1], Takahiro Kosugi[1,2], Nobuyasu Koga[1,2,3] ([1]*CIMoS, IMS.*, [2]*SOKENDAI*, [3]*JST, PRESTO*)

Previously, we designed ideal protein structures for various topologies completely from scratch, in which secondary structure and loop lengths are optimized for folding. Interestingly, most of the designed proteins are very stable and their melting temperatures are above 100C. To elucidate the factors for such high stability, we are conducting multiple mutations of the designed proteins. At first, hypothesizing that the number of carbon atoms in the core is a key to the stability, we mutate residues from small hydrophobic amino acids to large ones or from large to small and then investigate how the melting temperature changes. We discuss the experimental results together with the computational analyses for the stability of mutated structures.

2Pos089 c-Myb- KIX 間相互作用を阻害するペプチドの合理的設計

Rational design of a peptide inhibitor of the c-Myb-KIX interaction

Shunji Suetaka[1], Yoshiki Oka[2], Yuuki Hayashi[1,2], Munehito Arai[1,2] ([1]*Dept. Integrated Sci., Univ. Tokyo*, [2]*Dept. Life Sci., Univ. Tokyo*)

The transcriptional activator c-Myb interacts with the KIX domain of CBP/p300 and regulates hematopoiesis. However, aberrant expression of c-Myb causes human tumors, such as leukemia. Therefore, an inhibitor of the c-Myb-KIX interaction is potentially useful as an anti-tumor drug. c-Myb is intrinsically disordered and interacts with KIX by the conformational selection mechanism, suggesting that stabilization of helical structure of c-Myb strengthens the interaction with KIX. Here, we designed peptide inhibitors of the c-Myb-KIX interaction by introducing mutations into the transactivation domain of the wild-type c-Myb to stabilize its helical structure. We will compare the binding affinity with KIX between the wild-type c-Myb and the designed peptides.

2Pos090 β シートモデルタンパク質 OspA への αB クリスタリンのアミロイド形成配列移植と評価

Grafting of a short amyloid forming sequence from αB crystalline into β-rich model protein, OspA

Yuki Hori[1], Kenta Hongo[2], Norio Yoshida[3], **Koki Makabe**[1] ([1]*Graduate School of Science and Engineering, Yamagata University*, [2]*School of Information Science, JAIST*, [3]*Department of Chemistry, Graduate School of Sciences, Kyushu University*)

The atomic details of amyloid are still elusive because of their solid-state aggregate nature. To overcome this limitation, we have developed a model system based on the single-layer β-sheet of Borrelia Outer surface protein A (OspA). In this work, we extend our model system approach to an amyloid forming peptide sequence from αB crystalline. We choose “VLGDV” sequence as a grafting sequence. The peptide shows amyloid-like fibril formation. Then we grafted this peptide sequence into the OspA SLB portion. The resulting mutants were expressed as soluble protein and monomeric. We successfully determined the crystal structures of the mutants. We will discuss the details of the structures with MD simulation results.

2Pos091 レアメタルに特異的に結合するペプチドの設計

Design of peptides that specifically bind to the rare metal

Yoshihiro Iida, Atsuo Tamura (*Grad. Sci., Univ. Kobe*)

Since the rare metal has been used in various high-tech industries these days, they will likely to be depleted in the near future. Thus we have to recycle rare metals in urban mines. Here we try to design peptides that have capability of selectively binding and recycling the rare metal complex ions. The secondary structures of the designed peptides were found to be changed when specific metal ions were added. One of the designed peptide, M2SAP7, specifically bound to gold, and the other, M2SAP5, to platinum. Furthermore the gold binding peptide gradually interconverted the gold ion into nano colloid concomitant with the change in color to magenta. This study will serve as a stepping stone to further development of recycling and sensing rare metals.

2Pos092* 進化分子工学によるフィチン酸塩加水分解酵素の活性向上

Improving activity of a phytate-hydrolyzing enzyme by directed evolution

Manami Wada, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

Phytase is the only enzyme to release inorganic phosphate (PO_4^{3-}) from phytates. Phytates are salt forms of phytic acid and tend to accumulate in soil as forms unavailable to plants. However, phytases secreted from plant roots have low activity with optimal temperature higher than soil temperature. Hence, we aimed at modifying phytase properties to achieve higher activity at soil temperature by directed evolution. We constructed a plasmid containing *E. coli appA* phytase gene and confirmed over-expression of the enzyme in *E. coli* by SDS-PAGE and activity assay. Mutations in *appA* will be introduced by error-prone PCR and the mutants with improved properties will be screened using high-throughput methods. Highly active phytase will be useful for improving plant growth.

2Pos093 Toward Directed Evolution of Bacterial Biosensor for Arsenite Detection by Compartmentalized Partnered Replication

Seaim Lwin Aye, Asuka Ueki, Kei Fujiwara, Nobuhide Doi (*Grad. Sch. Sci. Tech., Keio Univ.*)

Arsenic is a well-known toxin that holds a constant threat to a large population worldwide. Our ultimate goal is to develop an arsenite-inducible biosensor with the best possible sensitivity and specificity. In order to evolve the *ars* operon of *E. coli*, we intend to apply compartmentalized partnered replication (Nat. Biotechnol. 32:97, 2014), in which the *ars* operon is coupled with a DNA polymerase expression. However, our DNA polymerase has poor activity in a water-in-oil emulsion. To overcome this problem, in this study, we performed directed evolution of DNA polymerase with improved efficiency by using compartmentalized self-replication (PNAS 98:4552, 2001). We have successfully obtained DNA polymerase mutants with practically useful properties for emulsion PCR.

2Pos094 フェムトリットルチャンバーアレイを用いたスクリーニングシステムによるアルカリフォスファターゼの進化分子工学

Directed evolution of alkaline phosphatase by femtoliter chamber array screening system

Makoto Kato, Yi Zhang, Kazuhito Tabata, Hiroyuki Noji (*Grad. Sch. Eng., Univ. Tokyo*)

Directed evolution is an attractive concept for improvement of protein functions. By combining a femtoliter chamber array and a micro-manipulator, we have established an integrated and versatile screening platform to conduct cell-free protein synthesis from a single DNA molecule in an individual chamber (i.e., single clone), and enabled the recovery of DNA of interest from the chamber. The massively parallel chambers with uniform volume enable both quantitative and high-throughput screening. As a proof-of-concept, we are trying to improve the activity of alkaline phosphatase, a widely-used reporter enzyme for diagnostic assays, and we accomplished the simultaneous protein synthesis and the activity quantification through a coupled fluorogenic assay.

2Pos095* ファージ選別実験への応用を目指したスプリットGFPによる蛍光ファージの作製

Construction of fluorescent phages based on split GFP for the phage sorting technique

Naoki Mikoshiba[1,2], Yuki Shimizu[1,3], Rie Kiriguchi[1,2], Seiji Sakamoto[1,3], Hiroyuki Oikawa[1,2,3], Kiyoto Kamagata[1,2,3], Takehiko Wada[1,3], Satoshi Takahashi[1,2,3] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Life Sci., Tohoku Univ.*, [3]*Grad. Sch. Sci., Tohoku Univ.*)

Our goal is to establish a new technique of protein design using combination of the phage display method and the single-molecule sorting system. For this purpose, fluorescent phages emitting a strong fluorescence is required. We first prepared a phage displaying split green fluorescent protein (split GFP) at the N-terminus of g3p. However, the fluorescence intensity was insufficient for our sorting device. Accordingly, we next attempted prepared a new fluorescent phage by displaying split GFP on g8p, which emits fluorescence more than 10 times larger than the previous phage. We also succeeded in displaying yellow fluorescent protein and cyan fluorescent protein on the phage. We propose that the developed fluorescent phages can be a potential tool for other applications.

2Pos096 Toward design of diverse all-α protein structures

Kouya Sakuma[1,2], Rie Koga[1], Takahiro Kosugi[1,2], Nobuyasu Koga[1,2,3] ([1]*CIMoS, IMS.*, [2]*SOKENDAI*, [3]*JST, PRESTO*)

All-α protein structures have recently been design targets, but all of the designs were bundle or coiled-coil. The structural diversity of all-α structures still remains to be explored. Our goal is to comprehensively design and reach more complex all-α structures like Globin from scratch. We computationally generated myriad all-α backbone conformations by combining ideal α-helices ranging from 1 to 35-residue lengths and typical 18 helix-loop-helix motifs we had previously identified. Selecting compact backbones with “core-to-be” cavities, we designed amino-acid sequences that show funnel-like energy landscape *in-silico*. We report experimental validations for the foldability of the design sequences.

2Pos097 エクソンペプチドの構造予測と解析
Prediction of Exon-Peptide Structures and Analysis

Michiko Nosaka (*National Institute of Technology, Sasebo College*)

We have studied the tertiary structures of exon-peptide of retinol binding protein by a structure prediction-site. Since the prediction is performed with the known structure of the homologous sequence, the predicted structures of these exon-peptides are expected to have similar to the parts of the known structure. However, the prediction of these peptides presents an interesting result: All predictions of some exon-peptides are as expected but four of five outputs of an exon are different from the expected structure. This result suggests us the relationship of a unique characteristics of the similar proteins; the change of secondary structure observed during the refolding process after chemical denaturation.

2Pos098* 粗視化分子動力学計算によるサブヌクレオソームの構造解析
Structural modeling of the subnucleosome using coarse-grained molecular dynamics simulations

Masahiro Shimizu, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

Tetrasome is a subnucleosome complex composed of DNA and histone tetramer $(H3\text{-}H4)_2$. Since nucleosomes are assembled through a tetrasome, the structural information of the tetrasome is crucial to understand the mechanisms of chromatin assembly. However, the structures of the tetrasome have not been solved.
In this research, we studied the structures of the tetrasome by coarse-grained molecular dynamics simulations. In simulations, we found two types of structures. One is "open" structure, in which $(H3\text{-}H4)_2$ binds a hairpin-shaped DNA. The other type is "closed" structure, in which the $(H3\text{-}H4)_2$ is wrapped by DNA. The latter type is a novel nucleosome-like structure, where we observed both left-handed and right-handed closed tetrasomes in the simulations.

2Pos099 Holliday junction DNA facilitates RuvA-RuvB complex formation

Yong-Woon Han[1], Reiko Yamamoto[1], Kimiko Nakao[1], Hisashi Tadakuma[1], Yoshie Harada[1,2,3] ([1]*Kyoto Univ., iCeMS*, [2]*Grad. Sch. Biostudies, Kyoto Univ.*, [3]*Inst. for Protein Res., Osaka Univ.*)

The Escherichia coli RuvA-RuvB protein complex promotes branch migration of Holliday junction DNA, which is the central intermediate of homologous recombination. RuvA is a Holliday junction DNA specific DNA binding protein and forms a stable tetramer. RuvA interacts with RuvB and facilitates RuvB binding to DNA. RuvB is an ATPase and forms hexameric ring in an ATP-dependent manner. RuvB hexameric rings pump out dsDNA through the RuvA-Holliday junction DNA core to promote branch migration of Holliday junction DNA. To characterize RuvA-RuvB-Holliday junction DNA complex in more detail, in this study, we constructed fluorescently labeled RuvA and characterized RuvA interaction with RuvB and/or Holliday junction DNA with spectroscopic analysis.

2Pos100 定量的イメージング法を用いた単一細胞由来のグルココルチコイド受容体のホモ二量体と転写活性の関連解析
Quantification of homodimeric glucocorticoid receptor and transcriptional activity from single cell using quantitative imaging techniques

Sho Oasa[1], Akira Sasaki[2], Shintaro Mikuni[1], Johtaro Yamamoto[1], Masataka Kinjo[1] ([1]*Fac. Adv. Life Sci., Hokkaido Univ.*, [2]*AIST*)

The aim of this study is to reveal the relationship between a homodimer of glucocorticoid receptor (GR) and transcriptional activity in single-cell level by single-cell method combining fluorescence correlation spectroscopy and microwell (FCS-microwell system). It is well-known that the homodimeric GR binds to glucocorticoid response elements (GRE) in a promoter region of target genes and regulates their gene expressions upon ligand binding. However, the relationship between the homodimeric GR and transcriptional activity remains unclear, yet. In this study, the relationship between a concentration of the homodimeric GR and transcriptional activity from single cell were determined simultaneously at single-cell lysate in microwell by FCS and fluorescent reporter assay.

2Pos101 (6-4)光回復酵素の T(6-4)T と T(6-4)C の光活性及び光修復における 赤外分光解析の比較
Comparative FTIR study of photoactivation and photorepair of T(6-4)T and T(6-4)C photoproducts by *Xenopus* (6-4) photolyase

Mai Kumagai[1], Daichi Yamada[1,2], Tatsuya Iwata[1], Elizabeth D Getzoff[4], Junpei Yamamoto[3], Shigenori Iwai[3], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*Ochanomizu University*, [3]*Osaka University*, [4]*The Scripps Research Institute*)

(6-4) photolyase ((6-4) PHR) is a flavoprotein that reverts UV-induced (6-4) photoproduct ((6-4) PP) into normal bases. (6-4) PHR can repair both T(6-4)T and T(6-4)C, where the OH and NH_2 groups are bound at C5 position of 5' side, respectively. We have measured the photoactivation and photorepair processes of T(6-4)T by *Xenopus* (6-4) PHR using light-induced difference Fourier-transform infrared (FTIR) spectroscopy. Here, to investigate the structural perturbation of the enzyme by (6-4) PP binding and the transfer of different functional groups, photoactivation and photorepair processes of T(6-4)T and T(6-4)C were compared by FTIR spectroscopy. The molecular mechanisms of (6-4) PHR will be discussed, based on the present spectroscopic observations.

2Pos102 3 本鎖 DNA 結合蛋白質が 3 本鎖 DNA を認識する分子機構
Molecular mechanism of the triplex DNA-binding protein to recognize triplex DNA

Kazuki Kiuchi[1], Kohta Sugiyama[1], Ryotaro Kishi[1], Satoru Unzai[2,3], **Hidetaka Torigoe**[1] ([1]*Dept. Applied Chem., Fac. Sci., Tokyo Univ. Sci.*, [2]*Grad. Sch. Medical Life Sci., Yokohama City Univ.*, [3]*Fac. Biosci. Applied Chem., Hosei Univ.*)

Stm1 is a triplex DNA-binding protein. We found that an intrinsically disordered region of the triplex DNA-binding domain of Stm1 (Stm1TBD) may recognize the shape rather than the sequence of the triplex. Here, we studied the triplex recognition mechanism of Stm1TBD by isothermal titration calorimetry. The dissociation constant was 10 μM, which was significantly larger than that of sequence-specific duplex DNA-binding domain (dsDBD) and agreed with sequence-nonspecific binding of Stm1TBD. Though ΔS of dsDBD-dsDNA binding was positive due to positive dehydration ΔS, ΔS of Stm1TBD-triplex binding was negative. Negative conformational ΔS due to the conformational restraint of intrinsically disordered region of Stm1TBD upon triplex binding may contribute to the negative ΔS.

2Pos103* **Characterization of the deamination activity of APOBEC3B by real-time NMR, which is distinct from that of APOBEC3G**

Li Wan[1], Takashi Nagata[1,2], Ryo Morishita[3], Akifumi Takaori[4], Masato Katahira[1,2] ([1]*Grad. Sch. of Energy Sci., Kyoto Univ.*, [2]*Inst. of Adv. Energy, Kyoto Univ.*, [3]*CellFree Sciences Co., Ltd.*, [4]*Dept. of Hematology and Oncology, Grad. Sch. of Med., Kyoto Uni.*)

Human APOBEC3 (A3) members deaminate cytosine to uracil in single-stranded (ss) DNA. Here, we studied the length and position dependent characteristics of the A3B C-terminal domain (CTD). A3B CTD turned out to deaminate its target 5'-TC-3' sequence in rather short ssDNA and the target sequences that are located less close to either the 5'- or 3'-ends. These features are quite different from those of A3G, which shows low activity to short ssDNA and prefers to deaminate its target sequence located close to the 5'- end. The distinct features of A3B CTD can be rationally interpreted that A3B CTD slides along ssDNA with short distance and then dissociates from ssDNA rapidly and diffuses into the bulk solution during the process of the target searching.

2Pos104 **Sequence-Specific Protein-DNA Interactions for Molecular Simulations Modeled by Position Weight Matrix**

Cheng Tan, Shoji Takada (*Dept. Biophysics, Graduate School of Science, Kyoto University*)

Experiments have revealed that most DNA-binding proteins possess unique specificity, meanwhile many proteins each recognizes distinct sequence motifs. The comprehensive information much more than single consensus sequence is represented in the form of Position Weight Matrix (PWM). Here we developed a method to model the specific protein-DNA interactions in coarse-grained molecular dynamics simulations based on PWM and the native structure. By applying this model to several protein-DNA complexes, we test how our model is capable of modelling the subtle specificity of protein-DNA recognition. This model can also be extended to computational investigations of the weak-sequence-specific protein-DNA binding, such as the searching process of transcription factors.

2Pos105 **Mechanistic studies of transcriptional regulation by non-CpG methylated DNA**

Jianshi Jin[1,2,4], Tengfei Lian[1,2], Chan Gu[1,3], Kai Yu[1,2], Yi Qin Gao[1,3], Xiao-Dong Su[1,2] ([1]*Biodynamic Optical Imaging Center(BIOPIC), Peking University*, [2]*School of Life Sciences, Peking University*, [3]*College of Chemistry and Molecular Engineering, Peking University*, [4]*RIKEN Quantitative Biology Center (QBiC), Laboratory for Integrative Omics*)

DNA in a non-CpG context (mCH) was found to occur widely on genomic DNA recently, and its level may vary during cellular development as a highly controlled process. Using single-molecule and ITC assays, we studied the mCH effects on DNA/protein interactions. We found mCH may have different effects on DNA/protein interactions. The effects of mCH on transcriptional regulation were also validated by cell-based luciferase assay. Furthermore, the mechanisms of mCH influencing DNA/protein interactions were investigated by crystallographic analyses and MD simulation. With BisChIP-seq assays, we found that GR can recognize highly methylated sites in cells. Therefore, we conclude that mCH provides a novel mechanism for gene regulation through directly affecting the TF binding.

2Pos106 **転写調節機構の統計的解釈〜コンセンサス配列でわかること、わからないこと〜**
Global Statistical Control of Transcriptional Pausing by Repetitive Genomic Sequences

Masahiko Imashimizu[1,2,3], Ariel Afek[4], Hiroki Takahashi[2], David Lukatsky[4] ([1]*IMSUT*, [2]*MMRC, Chiba Univ.*, [3]*NIH/NCI*, [4]*Ben-Gurion Univ. of the Negev*)

RNA polymerase (RNAP) pauses at highly nonrandom positions across genomic DNA, broadly regulating transcription, however, molecular mechanisms responsible for the recognition of such pausing positions remain poorly understood. Here, using a combination of statistical mechanical modeling and high-throughput sequencing and biochemical data, we evaluate the effect of thermal fluctuations on the regulation of RNAP pausing. We demonstrate that a nonrandom landscape of repetitive DNA sequence elements encoded in the genome, statistically affects diffusive RNAP backtracking. This entropy-dominated effect stems from the increased microscopic heterogeneity of an elongation complex.

2Pos107 **Molecular dynamics of transcription factor-nucleosome interactions**

Giovanni Brandani, Shoji Takada (*Kyoto University*)

In Eukaryotic cells, the DNA wraps around histone octamers to assemble into nucleosomes, and the precise positioning of nucleosomes on the DNA sequence is one of the key mechanism employed to regulate genome expression. Nucleosome association controls the spacial organisation of the genome, which in turn will affect the search mechanism of transcription factors (TF) for their target sites. Moreover, the positioning of DNA binding sites within nucleosomes is known to have a large effect on the kinetics of TF association and dissociation. However, the molecular details of these processes are still unclear. We performed coarse-grained molecular dynamics simulations to investigate the key features of TF-nucleosome interactions.

2Pos108 **Structured RNAs Induce Intersubunit Rolling and Codon-Anticodon Weakening During Ribosomal Frameshifting**

Kaichun Chang[1], Emmanuel Salawu[2,3], Jin-Der Wen[1], **Lee-Wei Yang**[2,3,4] ([1]*National Taiwan University*, [2]*National Tsing Hua University*, [3]*Bioinformatics Program, Academia Sinica*, [4]*National Center of Theoretical Science*)

Programmed ribosomal frameshifting (PRF) is a non-conventional translational mechanism allowing viruses and bacteria to produce different gene products with a single mRNA template. Despite abundant structural biology and single molecule data, a mechanical understanding in how it occurs remains elusive. To understand the perturbed dynamics of the ribosome upon encountering a structured mRNA, we applied resolution-exchanged models to combine elastic network model, steered/targeted MD, linear response theory and explicit solvent MD simulations to reveal a newly identified “rolling” motion of the 30S subunit, whereby PRF-promoting pseudoknot (PK), but not the base-triple-weakened PK, distorts tRNAs and in turn disrupts mRNA-tRNA base-pairings and thereby promoting PRF.

2Pos109 リンカー DNA により繋がっているダイヌクレオソーム構造のサンプリング

Sampling di-nucleosome structures connected by linker DNA of various lengths

Hiroo Kenzaki[1], Shoji Takada[2] ([1]*ACCC, RIKEN*, [2]*Grad. Sch. Sci., Univ. Kyoto*)

To take hierarchical structure of chromatin, interaction between nucleosomes is a primary element. Interaction between neighboring nucleosomes is limited by connecting linker DNA. We performed coarse-grained simulation of di-nucleosome that connected linker DNA, where AICG2+ model is used for protein and 3SPN.2C model used for DNA. The simulations were repeated by systematically changing the lengths of linker DNA. The observed di-nucleosome structures are critically dependent on the length of the linker DNA, where interactions between nucleosomes are largely mediated by certain histone tails.

2Pos110 酵母間期染色体の力学モデルと核内構造・動態の解析

Analysis of interface chromosome dynamics of yeast by course-grained models

Takamasa Yamamoto[1], Hiraku Nishimori[1,2], Akinori Awazu[1,2] ([1]*Dept. Math and Life Sci. Hiroshima Univ.*, [2]*RcMcD*)

The eukaryotic chromosomes involve highly folded chromatin structures. The motions of such chromatin structures are physically limited by their own hierarchical structures. On the other hand, the chromatin structures transit dynamically in all cell stage that regulates appropriate transcriptions, DNA replications, and repairs. Then, we constructed a coarse-grained model of chromosomes in fission yeast nucleus by spring beads model to investigate the dynamics of the interphase intra-nuclear structures. We performed molecular dynamics simulation of this model to discuss the influences on the positioning of chromosomes and nucleolus by the presence of the interactions between centromere domain and spindle pole body, and that between telomere and nuclear envelope.

2Pos111 細胞分裂およびアポトーシス過程におけるクロマチンダイナミクスの 1 分子解析

Chromatin dynamics in mitosis and apoptosis

Kayo Hibino[1,2], Kazuhiro Maeshima[1,2] ([1]*NIG*, [2]*SOKENDAI*)

Chromatin structures and dynamics vary during cell cycle and also in response to cell signaling. For instance, chromatin condensation occurs in mitosis, and also in apoptosis, which is induced by DNA damage or extracellular signal. Although local chromatin morphologies in mitosis and apoptosis appear similar, their dynamic aspects, which can reflect their functional differences, remain unclear. Here we have measured local fluctuation of condensed chromatin in mitotic and apoptotic cells by single nucleosome imaging. Fluorescently labeled nucleosomes in mitotic and UV-induced apoptotic HeLa cells were observed by an oblique illumination microscopy. In this meeting, we discuss difference in local chromatin fluctuation between them and its functional relevance.

2Pos112 Grab & Watch: Correlative optical Tweezers-Fluorescence Microscopy (CTFM) as a versatile tool for chromatin studies

Andrea Candelli[1,2], Gerrit Sitters[1,2], **Rosalie Driessen**[1,2], Willem Peutz[1,2], Olivier Heyning[1,2], Gijs Wuite[1,2], Erwin Peterman[1,2] ([1]*LUMICKS*, [2]*VU University, Amsterdam*)

The combination of optical tweezers with confocal fluorescence microscopy and microfluidics is a promising approach to investigate biological systems because of its unique ability to simultaneously manipulate, sense and visualize individual biomolecules with exquisite sensitivity and resolution. Using Correlative optical Tweezers-Fluorscence Mircoscopy (CTFM) , we carry out a series of demonstration experiments, exploring applications in the fields of chromatin structure and function, DNA repair and molecular motors. These experiments show that the technological advances in hybrid single-molecule methods can open up new venues in the field of nucleic acids research.

2Pos113 クロマチンのエピジェネティック状態を用いたヒト間期核内における染色体三次元構造のシミュレーション

Simulating three-dimensional organization of chromosomes in human interphase nucleus using epigenetic state of chromatin

Shin Fujishiro, Masaki Sasai (*Nagoya Univ.*)

Experimental studies suggested that chromosomes of higher eukaryotes are folded and positioned in interphase nuclei in a way related to epigenetic regulation. Although chromosome structures were characterized by various simulation studies, their relation to the epigenetic regulation is still unclear. We develop a computational model of human genome to clarify how the epigenome establishes and maintains the chromosome organization. We model each chromosome as a block copolymer comprising euchromatin and heterochromatin regions and equilibrate them with the Langevin dynamics method. Our preliminary results showed that entropic effects of copolymers induce phase separation in a nucleus, which accumulates the heterochromatin regions near nuclear and nucleolar peripheries.

2Pos114* 蛍光相互相関分光法を用いたグルココルチコイド受容体-DNA間相互作用の定量化

The different interaction affinity of monomeric or dimeric GR on DNA determined by fluorescence cross-correlation spectroscopy

Daisuke Yamashita, Mari Saito, Sho Oasa, Shintaro Mikuni, Masataka Kinjo (*Grad. Sch. Life Sci., Univ. Hokkaido*)

Glucocorticoid receptor (GR) is a member of the nuclear receptor superfamily. It is well-known that upon ligand binding, GR binds to glucocorticoid response elements (GRE) as a dimer and regulates a transcription of target genes. It was reported that a monomeric GR could also regulate but the different genes from the dimeric GR binding sites. The aim of this study is to reveal the differences of transcriptional mechanism between the monomeric and dimeric GR through GR-DNA interaction. In this study, EGFP-fused wild type GR (WT) and dimerization-deficient mutant (A458T) were purified from sf9 insect cells. The dissociation constant of the interaction between the purified EGFP-GR and different types of Alexa-labeled GRE using fluorescence cross-correlation spectroscopy.

2Pos115 Effects of cumulative acetylation in histone H3 tail studied by an enhanced conformational sampling MD simulation

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Eukaryotic genome DNA forms a protein-DNA complex referred to nucleosome and the nucleosomes condense into a higher structure, chromatin. The condensed chromatin prevents transcription proteins from accessing the DNA and inactivates transcription. Progressive lysine acetylation of histone tails activates transcription in general, however, the molecular view is not well understood.
To investigate the structural effect of the cumulative acetylation, we performed enhanced conformational sampling MD simulations on H3 histone tails in a context of nucleosome structure with various acetylation states. In this poster, we show the structural effects of cumulative acetylation and discuss the impact on nucleosome stability and further relaxation of condensed chromatin structure.

2Pos116 Photo-control of the ribosome movement along mRNA using a reversible photo-cross-linking probe

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Here, we present a method for controlling the movement of the ribosome along mRNA using a DNA oligonucleotide containing a 3-cyanovinylcarbazole nucleoside, which serves as a reversible photo-crosslinker. The oligonucleotide is covalently cross-linked to mRNA with its complementary sequence through irradiation at around 366 nm, thereby preventing movement of the ribosome. The ribosome would start to move when the cross-linking is cleaved upon irradiation at around 312 nm. As a proof-of-concept, we have demonstrated that the movement of the ribosome can be controlled in a reconstituted cell-free translation system. Using this method, we are now trying to probe the dynamic movement of the ribosome during translation at a single-molecule level.

2Pos117 転写によって誘起されるクロマチンブラシの相分離
Transcription driven phase separation in chromatin brush

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In a eukaryotic cell, DNA is packed in a nucleus by stabilizing chromatin. Recent experiments show that embryonic stem cells show the fluctuations of nucleosome density in a relatively long time and length scales, analogous to critical fluctuations. In contrast, differentiated cells show regions of relatively large nucleosome density that coexists with regions of smaller nucleosome density, analogous to phase separation. Here we analyze the dynamics of nucleosome assembly and disassembly, by taking into account the interactions between RNA polymerase and nucleosomes during transcription and passive diffusion, to predict the physical mechanism involved in the phase separation during differentiation.

2Pos118 蛍光消光現象を利用した単層カーボンナノチューブ表面上での一本鎖 DNA 吸着過程の観察
Monitoring adsorption process of single stranded DNA to single-walled carbon nanotubes surfaces by fluorescence quenching

Shizuma Sato[1], Gilbert Bustamante[2], Jing Yong Ye[2], Kazuo Umemura[1] ([1]*Tokyo Univ. of Sci.*, [2]*UTSA*)

Single-walled carbon nanotubes (SWNTs) and single stranded DNA (ssDNA) hybrids by physisorption are one of the promising nanobiodevices for biomedical applications. However, quantitative analysis of amount of ssDNA adsorbed on SWNT surfaces have not been demonstrated yet. Here we developed a new method of monitoring adsorption process of ssDNA to SWNTs. It is well known that fluorescent emission is quenched when a dye is adsorbed on SWNTs. Using a fluorescein labelled ssDNA, we could estimate amount of DNA adsorbed on SWNT surfaces by measuring decrease of fluorescent emission. We measured the fluorescence quenching as a function of time. Quenching ratio was 98% in the standard hybridization condition. These results indicate that most of ssDNA adsorbed on SWNT surfaces.

2Pos119 DNA の網目構造が微粒子の拡散に及ぼす影響
Effect of a mesh structure of DNA on diffusion of a small particle

Masaya Tanoguchi, Yoshihiro Murayama (*Tokyo Univ. of Agri. and Tech.*)

In dense DNA solution, DNAs are entangled and a mesh structure is formed. To reveal the effect of the mesh structure of DNA on the diffusion of a small particle, we observed the diffusion of a bead whose diameter was 0.5, 1, or 3 µm. To vary the mesh size, concentration of lambda phage DNA (48,502 bp) was varied 0.05 - 1.00 mg/mL. The bead images were captured at 1000 fps, and for a given time interval t, mean values of variance of the bead position were calculated. For $t < 0.1$ s, the mean value is proportional to t^{α} and the exponent α decreases from 1 which means normal diffusion to about 0.6 which means a restricted diffusion with the increase of the DNA concentration for any sizes of beads.

2Pos120 イオン液体ー水混合溶液中の β-ラクトグロブリンの会合構造
Aggregation of β-lactoglobulin in alkylammonium-based nitrate ionic liquid-water mixture

Koji Yoshida, Ayako Fujiyoshi, Toshio Yamaguchi (*Fukuoka Univ.*)

Aggregation state of β-lactoglobulin was investigated in aqueous solutions of three alkylammonium-based nitrate ionic liquids (RAN-ILs): methylammonium nitrate (MAN), ethylammonium nitrate (EAN), and propylammonium nitrate (PAN) as a function of IL concentration and temperature by small-angle X-ray scattering and differential scanning calorimetry. The solvent property changes from electrolyte-solution-like to molten salt-like property depending on the concentration of IL. In MAN-water mixtures, the protein formed irregular aggregates after thermal denaturation. It is similar to the case in $NaNO_3$ and NaCl solutions. The addition of EAN and PAN to the protein aqueous solution decreased the denaturation temperature and resulted in a fractal aggregate of the protein.

2Pos121 OH 伸縮振動のラマン分光によるアミノ酸水和層の解析
Hydration analysis of amino acids by Raman OH-stretching spectroscopy

Yasutaka Naito[1], Yuki Ochiai[2], George Mogami[2], Makoto Suzuki[2] ([1]*Sch. Eng., Univ. Tohoku*, [2]*Grad. Sch. Eng., Univ. Tohoku*)

To assign the Raman bands of protein hydration water from the solution spectrum, we have analyzed the Raman bands of hydration water of amino acid aqueous solutions in the wave-number range from 2700 to 4000 cm^{-1}. Firstly, each amino acid dissolved in D_2O/H_2O mixed solvent was measured to assign the Raman bands such as CH-stretching (2900 - 3100 cm^{-1}) and fluorescence bands. After removing the non-water bands from the raw Raman spectrum in H_2O, the spectrum was decomposed into the bulk water component and the hydration layer component. The hydration layer spectrum was further decomposed into 4-site hydrogen bonded, 2-site bonded and non-bonded water, and compared with the results by the previous dielectric relaxation results.

2Pos122 広帯域誘電緩和分光を用いた球状および膜タンパク質における水和と熱活性効果の研究
Effects of hydration and thermal excitation of globular and membrane proteins studied by broadband dielectric spectroscopy

Naoki Yamamoto[1], Shota Ito[2], Eri Chatani[1], Hideki Kandori[2], Keisuke Tominaga[1,3] ([1]*Graduate School of Science, Kobe University*, [2]*Graduate School of Engineering, Nagoya Institute of Technology*, [3]*Molecular Photoscience Research Center, Kobe University*)

Protein is always fluctuating by the thermal fluctuation of solvent, water. The fluctuation is thought to be important for proteins to exhibit their functions. The dynamics of thermal fluctuation are usually observed in picosecond or nanosecond region as low-frequency vibrations or rotational relaxations. The dynamics of these time regions, which correspond to gigahertz (GHz; 109 Hz) to terahertz (THz; 1012 Hz), can be monitored by using broadband dielectric spectroscopy. Dielectric spectroscopy enables us to monitor the dynamics of protein and hydration water thorough modulation of the total dipole moment of the system. In this study we studied temperature and hydration dependence of the complex dielectric spectra of lysozyme and purple membrane.

2Pos123* 分子動力学法を用いた蛋白質周囲の水和ダイナミクスの検討：溶媒条件と蛋白質構造の影響について
Effects of solvent pH and protein conformations on water dynamics around a denatured protein with molecular dynamics simulation

Takafumi Fujiyoshi[1], Naoki Ogasawara[1], Yuji Ezaki[2], Yuta Nonaka[2], Kota Kasahara[2], Takuya Takahashi[2] ([1]*Graduate School of Life Sciences, Ritsumeikan University*, [2]*College of Life Sciences, Ritsumeikan University*)

Molecular dynamics (MD) studies have revealed that the dynamics of hydration water around intrinsically disordered proteins (IDPs) shows qualitatively different behavior compared with globular proteins. Here, to examine the effect of protein conformation on the water dynamics, we performed MD simulations of poly-glutamic acids (PGA) in two different protonation states (i.e., different pH) and three water models (i.e., TIP3P, TIP5P, and SPC/E) with denatured and folded conformations. In the first hydration layer, the water dynamics in the neutral condition, the denatured conformation and TIP5P model is relatively fast, although there were no significant differences in the hydrogen bond number normalized by the bulk value among these conditions.

2Pos124 ミオシン周囲の局所誘電環境
Local dielectric environment around myosin

Takato Sato, Tohru Sasaki, Jun Ohnuki, Mitsunori Takano (*Dept. of Pure. & Appl. Phys., Waseda Univ.*)

In force-generating cycle of actomyosin, ATP-binding to myosin forces myosin to unbind from actin filament, which is crucial step in this cycle. In our previous simulation study, it is shown that ATP-binding induces dielectric response of myosin and moves high charge-density actin-binding loop away from actin filament allosterically (Biophys. Soc. Annu. Meeting 2014). By conducting molecular dynamics simulation, we explore the role of local dielectric environment around myosin which could alter the actin-myosin electrostatic interaction. We also discuss the role of the (seemingly) hydrophobic interaction through calculating compressibility around myosin.

2Pos125 水溶液中における芳香環間相互作用の統計熱力学
Statistical thermodynamics of aromatic-aromatic interactions in aqueous solution

Tomohiko Hayashi, Masahiro Kinoshita (*Inst. Adv. Energy, Kyoto Univ.*)

We analyze the potential of mean force (PMF) between toluene molecules in water along face-to-face stacked (FF) and point-to-face T-shaped (TS) paths using a statistical-mechanical theory of molecular liquids. The contribution to the PMF from hydration is decomposed into energetic and entropic components. In water, the FF stacking is considerably more stable than the TS contact. For the former, by the energetic hydration effect the London dispersion interaction is somewhat cancelled but the electrostatic repulsive interaction is significantly screened. Further, a large gain of water entropy occurs. We also discuss the relevance to the interactions between aromatic rings which are believed to play a pivotal role in a variety of biological processes.

2Pos126* Lennard-Jones ポテンシャルのパラメータの変更による溶質周囲の水分子のダイナミクスの探求
Effects of Lennard-Jones potentials on the dynamics of water molecules around a solute

Yuki Takimoto[1], Kou Sakuma[2], Nana Okita[2], Kota Kasahara[2], Takuya Takahashi[2] ([1]*Grad. Sch. Life. Sci., Ritsumeikan Univ.*, [2]*Col. Life. Sci., Ritsumeikan Univ.*)

To analyze the dynamics of water molecules at the atomic level, the molecular dynamics (MD) method has been extensively applied. However, conventional water models were not suitable to reproduce the “fast water” around an ion. Here, we examined various combinations of the repulsion and attraction terms in the Lennard-Jones potential of H atom, based on the TIP5P model. We analyzed the dynamics of water molecules in terms of the rotational relaxation time (tr) and the translational diffusion coefficient (Dt). As a result, although Dt showed a certain trend regardless of the repulsion term, tr showed some different trends that depend on the strength of the repulsion term. In addition, there was a very high correlation between the tr and radial distribution.

2Pos127 親水性アミノ酸残基周辺の水和構造における酸素原子ローンペアの影響

Effects of lone pairs of oxygen atoms on hydration structures around polar amino-acid residues: MD simulation study

Tomotaka Oroguchi, Masayoshi Nakasako (*Sci. Tech., Keio Univ.*)

The physical properties of hydration structures of proteins have been often investigated by molecular dynamics (MD) simulation. However, potential functions using in these simulation studies do not take explicitly into account electron lone-pairs (LPs) of oxygen atoms. In this study, we have developed a parameter set of potential function, in which LPs are introduced as extra-charge points of oxygen atoms. Then we calculated hydration structures around polar amino-acid residues by MD simulations with the developed parameter set. The comparison of the calculated hydration structures with those obtained from high-resolution cryogenic crystallography indicate that LPs are necessary to account hydrogen-bond interaction modes between polar residues and water molecules.

2Pos128 機械学習によるタンパク質親水／疎水表面における水分子の動的振る舞いの解析

Analysis of water behavior near the protein hydrophobic/ hydrophilic surface by machine learning techniques

Taku Mizukami[1], Nguyen Viet Cuong[3], Ho Tu Bao[2], Dam Hieu Chi[2] ([1]*JAIST, Materials Science*, [2]*JAIST, Knowledge Science*, [3]*HPC SYSTEMS Inc.*)

Water plays an important role in molecular expression of functions. The dynamical behavior of water molecules and its correlation for protein function are still in mystery. Recently, we developed an integration of molecular dynamics simulation and data-mining method to characterize a water dynamical behavior from the output data of MD simulation. In this study, the Gaussian Mixture Model method for classification of water behavior was expanded to operate the multiple time-constant features that enable to identify the fast- and slow-diffusional behavior and to investigate the correlation between them. By means of the methodology, we found the classified behavior of water and their correlations that are characteristic for hydrophobic or hydrophilic surface of protein.

2Pos129* MM/3D-RISM 法を用いた Pim1-リガンド系における結合自由エネルギーの予測

Estimation of binding free energy based on the MM/3D-RISM method for the Pim1-ligand system

Takeshi Hasegawa[1], Masatake Sugita[1], Takeshi Kikuchi[1], Fumio Hiata[2] ([1]*Dept. of Bioinfo., Col. Life Sci., Ritsumeikan Univ.*, [2]*Toyota Physical and Chemical Research Institute*)

To seek the drug candidate molecules, it is necessary to assess the binding affinity of a protein with its inhibitor. Although many computational methods for estimation the binding free energy have been proposed, these methods have some disadvantages, especially, in evaluating the solvation structures at an active of protein. In this regards, the MM/3D-RISM method, which combines the 3D-RISM theory with an MD simulation, has a great advantage. The method samples the structural fluctuation of protein with MD, and calculates the solvation structure and free energy along the conformational space of protein by means of 3D-RISM. We apply this method to estimate the binding free energy between Pim1 kinase and its inhibitors.

2Pos130 ROXS によって mCherry の安定性と明るさが向上する

in-vivo ROXS improve the brightness and photostability of a red fluorescent protein, mCherry in *C. elegans* embryos

Yukinobu Arata, Yasushi Sako (*Cell. Info. Lab. RIKEN*)

ROXS (Reducing and OXidizing System) is shown to improve the photochemical performance of chemical fluorescent dyes by using both a reducing and an oxidizing agent. In ROXS, the triplet and ionized state of fluorescent dyes is considered to quickly recover into the ground state by balancing the redox reaction (Vogelsang et al 2008 and Cordes et al 2009). However, it remains unknown whether ROXS is effective for fluorescent proteins. We found that oxidizing agent, Methyl viologen (MV) and reducing agent Ascorbic acid (AS) cooperatively improved the stability and brightness of mCherry but not GFP in C. elegans embryos. In this poster, we will discuss how ROXS is useful in a live imaging during animal development.

2Pos131 Axon bundle regulates cortical tissue stiffness in the developing brain

Misato Iwashita, Yoichi Kosodo (*Korea Brain Research Institute*)

We previously reported that cortical tissue, especially Intermediate Zone (IZ), exhibits the remarkable shift in stiffness during brain development. However, little is known what defines the stiffness of the IZ. Axons gradually fasciculate and form a tangential bundle in the IZ. Our histological data suggested that the pattern of fasciculation of axons correlated to the shift in stiffness in the IZ. Thus, we examined that the axon bundle affects the stiffness of the IZ. First, we ablated the axon bundle in the IZ partially by laser using two-photon microscope, then measured stiffness of axon-disrupted region using Atomic Force Microscopy. We found that the stiffness of the IZ decreased significantly, suggesting that the axon bundle contribute to the stiffness of the IZ.

2Pos132* Mouse-ferret differences in the mechanical property of the developing cerebral cortex: tissue-level and single cell-level assessments

Arata Nagasaka, Tomoyasu Shinoda, Takaki Miyata (*Grad. Sch. Med., Univ. Nagoya*)

In the developing cerebral cortex, the proliferative zone (called VZ) is filled with neural progenitors. We previously found that cell density in VZ was greater in ferrets than in mice. To ask possible involvements of mechanical factors in neural progenitors' behaviors, we comparatively assessed the physical property of VZ between mice and ferrets. Our AFM analysis revealed that ferret VZ was stiffer than mouse VZ at the inner surface, while contractility assessed by laser ablation on the VZ surface was similar between mice and ferrets. Interestingly, single-dissociated VZ cells' stiffness was smaller in ferrets than in mice, suggesting that tissue-level mechanical properties may be yielded through balancing cellular densification and single cells' physical property.

2Pos133 Macroscopic dynamics of vascular endothelial cells in angiogenesis

Naoko Takubo[1], Kazuaki Naekura[1], Ryo Yoshida[2], Terumasa Tokunaga[3], Osamu Hirose[4], Yasunobu Uchijima[1], Yukiko Kurihara[1], Hiroki Kurihara[1] ([1]*Graduate School of Medicine, The University of Tokyo*, [2]*The Institute of Statistical Mathematics, Research Organization of Information and Systems*, [3]*Faculty of Computer Science and Systems Engineering, Kyushu Institute of Technology*, [4]*College of Science and Engineering, Kanazawa University*)

It has been shown that collective vascular endothelial cells (ECs) in angiogenesis show complex movement called cell mixing. However, how ECs collectively form complex vessels remains unknown. In this study, to quantitatively delineate macroscopic EC dynamics in the whole new vessel, we demonstrated time-lapse live imaging for in vitro angiogenic model based on mouse aortic ring assay. As a result, regularity of macroscopic EC dynamics including U-turn behavior, in which a cell moves forward, then turns and backs to the basal vessel, was observed by newly-developed automated cell-tracking system. Moreover, it was suggested that macroscopic EC dynamics based on cell-cell interaction were regulated by supply of cells from the basal vessel.

2Pos134 ヒトの原腸形成時の細胞運動を in vitro で一細胞解析
Single cell tracking of migration during human gastrulation in vitro

Shota Miyazaki[1], Yuta Yamamoto[1], Kohei Nakazono[1], Minh N. T. Le[1], Shuji Fuji[1,2], Kiyoshi Ohnuma[1,3] ([1]*Department of Bioengineering, Nagaoka University of Technology*, [2]*Department of Materials Science and Technology, Nagaoka University of Technology*, [3]*Department of Science of Technology Innovation, Nagaoka University of Technology*)

Gastrulation is the first dynamic deformation of embryo to form a complex body. Although human gastrulation is known to follow the pattern observed in birds, the details are unknown because of ethical and technical limitations. Here we utilize human induced pluripotent stem cells (hiPSCs) to study the detail of human gastrulation in vitro. Time-lapse imaging showed that the differentiated cells started to disperse individually and move significantly faster than the undifferentiated cells suggesting that hiPSCs differentiated to be in the gastrulation state. The mean square displacements analysis showed that the migrations are random both undifferentiated and differentiated cells suggesting that some external factors are needed for coordinated movement as shown in vivo.

2Pos135 多細胞運動におけるソリトン様運動関連遺伝子の探索
Searching the responsible genes for biological soliton in multicellular movement

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Soliton is a physical phenomenon observed in wave motion. We found the distinct characteristics of solitons in the mass cell movement of non-chemotactic mutants of the cellular slime mould, Dictyostelium discoideum (Kuwayama and Ishida 2013). These mutant cells do not form multicellular aggregation. However, they exhibit a characteristic self-reinforcing solitary wave named Soliton-Like-Structure (SLS) under starvation conditions. It was also found that SLS induction is mediated by adhesive cell-cell interactions. By NGS, we identified 55 genes which have SNP in the coding regions of the mutant strains. In this meeting, we report and discuss the possibilities of some of those as the responsible genes.

2Pos136 A model for analyzing phenomena in multicellular organisms with multivariable polynomials

Hiroshi Yoshida (*Dep. Math., Kyushu Univ.*)

Most of life maintains itself through turnover, namely cell proliferation, movement, and elimination. Inspired by such a biological fact, and together with various operations of polynomials, I here propose “polynomial-life” model towards analysis of some phenomena in multicellular organisms. Polynomial-life is multicells that are expressed as multivariable-polynomials. A cell is expressed as a term of polynomial, in which point (m,n) is described as a term $x^m y^n$ and the condition is as its coefficient. Starting with a single term and following reductions by set of polynomials, I simulate development from a cell to a multicell. In this framework I present various patterns through the polynomial-life model and discuss patterns maintained through turnover.

2Pos137 *C. elegans* の受精における二相性カルシウム波の分子基盤
The molecular underpinnings of the biphasic calcium wave during fertilization in *C. elegans*

Jun Takayama, Shuichi Onami (*RIKEN QBiC*)

Calcium waves during fertilization trigger animal embryogenesis. By using high-speed *in vivo* imaging and image processing, we have recently demonstrated that the calcium response in *C. elegans* consists of a rapid local rise and a following propagating wave. Moreover, genetic analysis and simulation revealed that the sperm calcium channel TRP-3 induces the local calcium rise and suggested that TRP-3 acts as a calcium conduit (Takayama and Onami, 2016). Germline-specific knock-down experiments suggested that the IP3 receptor calcium channel is not required for the calcium wave propagation. To identify channels responsible for the calcium wave propagation, we are conducting an RNAi screen targeted to all predicted cation channels expressed in the germline.

2Pos138 機能する昆虫飛翔筋-ウサギ骨格筋ハイブリッド筋線維の再構成
Reconstitution of functional insect flight- and rabbit skeletal hybrid muscle fibers as monitored by X-ray diffraction

Hiroyuki Iwamoto (*SPring-8, JASRI*)

Stretch activation is essential for the action of asynchronous flight muscle. To explore its molecular mechanism, we have been trying to create insect flight muscle fibers in which endogenous actin is replaced by that from rabbit skeletal muscle. Because the reconstituted thin filaments are not Ca2+-regulated, we need to add a myosin inhibitor to prevent contraction during actin exchange. Previously we used blebbistatin as an inhibitor and successfully reconstituted the thin filaments, but the inhibition was irreversible. Here we used BDM instead, at a high concentration, and found that the thin filaments can be reconstituted in the presence of BDM. As monitored by X-ray diffraction, rigor actomyosin complexes are formed, indicating that the myosin remains functional.

2Pos139 アクチン骨格阻害剤 Latrunculin A はアクチンを脱重合させ重合を阻害する

Latrunculin-A; a drug to inhibit actin cytoskeleton, depolymerizes and inhibits actin polymerization under TIRF microscopy

Ikuko Fujiwara[1], Thomas Pollard[2] ([1]*URA, NITech.*, [2]*MCDB, Yale Univ.*)

Latrunculin A (LatA) binds actin monomers to inhibit polymerization. However this is not enough to explain the rapid actin disassembly. Our TIRF assay visualized that LatA accelerated the rate of actin depolymerization in ATP containing buffer (Kd=6.0 uM). The saturated depolymerization rate was same as ADP-actin. The severing event was observed, indicating that LatA binds to actin filaments. Depolymerization rates of ADP- or ADP-Pi-actin filaments were not affected by LatA. Solution assays showed that this rapid actin depolymerization was Pi release independent. TIRF assay confirmed that LatA slows actin elongation, consistent with the stoichiometrical sequestering (Kd = 0.094 uM). This indicates that LatA binds ATP-actin monomer 60-fold tighter than filamentous actin.

2Pos140 バキュロウイルス-昆虫細胞を用いた、組換え β-アクチンの発現精製系の構築

Expression and Purification of Recombinant Human β-Actin in Insect Cells

Mizuki Matsuzaki[1], Sae Kashima[1], Kayo Maeda[1], Mahito Kikumoto[1], Tomoharu Matsumoto[1], Kotaro Tanaka[1], Motonori Ota[2], Akihiro Narita[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Grad. Sch. Info. Sci., Nagoya Univ.*)

We constructed the expression system for recombinant Human β-actin by using a baculovirus-based system in insect cells. The transfer vector for the expression of the humanβ-actin has a Strep-Tag II affinity tag (WSHPQFEK) at the N terminus. The recombinant actin after cleaving the tag has polymerization and depolymerization activities. To determine whether the recombinant actin has the native fold, the actin filaments were observed by EM. The diffraction pattern shows that the recombinant actin forms normal actin filament.

2Pos141 結合した HMM 濃度の違いによる F-アクチンの状態変化

HMM induced structural changes of actin monitored by in vitro fluorescence along single filaments

Marie Kobayashi, Satoshi Nakamura, Takahiro Hayashizaki, Hajime Honda (*Dept. Bioeng., Nagaoka Univ. Tech.*)

To know the cooperative structural changes of actin protomers during in vitro assay, it is important to know the spatiotemporal distributions of HMM along single filaments. The number of S1 estimated at about 4/micrometer along a filament by electron microscopy should alter on conditions. Here, we have labeled actin by fluorescent dye, Prodan, which intensity increased as actin polymerization and again as binding to HMM. The extent was proportion to the amount of HMM added when measured by spectrophotometer. In contrast, the fluorescence from single actin filament was found to increase in sigmoidal manner to the density of HMM on the glass. These result indicate that the density of HMM along F-actin in an in vitro assay may differs from those in solution.

2Pos142* 超解像イメージング法を用いた骨格筋ミオシン分子動態の直接計測

Direct measurement of dynamics of individual skeletal myosin by using a super-resolution localization method

ZhiYuan Zhang, Yuto Ashida, Masahito Ueda, Hideo Higuchi, Motoshi Kaya (*Department of Physics, Graduate School of Science, The University of Tokyo*)

Our previous works suggest that cooperative mechanism may exist among skeletal myosin molecules during force generating process-myosin molecules synchronize to work together as load increase. To verify this cooperativity, we need to directly observe the dynamics of multiple individual myosin molecules embedded in a myofilament, whose area is smaller than the diffraction limit. Here we report success in direct displacement measurements of 2-3 myosin molecules labelled by quantum dots independently beyond the size of diffraction limit.

2Pos143* HS-AFM で一方向的な運動が観測されたローターレス Enterococus hirae V_1-ATPase の結晶構造解析

Crystal structures of rotorless Enterococus hirae V_1-ATPase which shows unidirectional dynamics by HS-AFM

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Vacuolar ATPases (V-ATPases) function as ATP-dependent ion pumps, and hydrophilic V_1 portion is known as rotary motor. The A_3B_3 complex, which is the catalytic domain of V_1, forms a hexagonal ring by the three catalytic A subunits and the three non-catalytic B. the unidirectional cooperative rotational motion of the A_3B_3 complex of Enterococus hirae has been observed by high speed atomic force microscope (HS-AFM) method. In this study, we obtained crystal structures of nucleotide-free and nucleotides bound the A3B3. In our poster, We would like to discuss molecular mechanism of the unidirectional coorperative rotation of the rotorless V_1-ATPase bases on the crystal structures and HS-AFM observation.

2Pos144 F_1-ATPase の中心軸回転を駆動する触媒サブユニットの１分子構造変化観察

Single-molecule detection of conformational changes in the catalytic subunit of F_1-ATPase that correlate with rotation of the shaft

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F_1-ATPase is a rotary molecular motor which converts chemical energy into mechanical work. Single-molecule rotation assay of F_1-ATPase has enabled studies on the mechanical work, but a mechanism by which this energy transduction is mediated has not been extensively studied at the single-molecule level. Our purpose is to identify key conformational changes of the catalytic subunit that are required to correlate ATP hydrolysis at the catalytic site with rotation of the central shaft. Secondary structures of the catalytic subunit that are expected to represent motions of each domain were labeled with bifunctional fluorescent probes. We found that these single fluorophores on some molecules showed changes in their orientation depending on direction of the central shaft.

2Pos145* V_1 回転分子モーターでの外来タンパク質の回転

Rotation of endogenous proteins in V_1 rotary motor

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V_1 moiety of V-ATPase is a rotary motor, in which a rotor D subunit rotates relative to a stator A_3B_3 using ATP hydrolysis energy. The FliJ of bacterial flagella and V_1-D share a coiled coil structure albeit there is no apparent sequence homology between them. In this study, we inquire whether the FliJ is able to function as a rotor in A_3B_3 or not, by producing a series of chimera rotors consisting of both FliJ and V_1-D. These reconstituted complexes including the xenogeneic rotor composed of FliJ exhibited torque 2/3 of the WT. Our results indicate that none of residue specific interaction between a rotor and stator hexamer is strictly needed for producing the torque. In addition, we found that right-handed coiled coil protein can work as a rotor axis in A_3B_3.

2Pos146* 1分子観察とドッキングシミュレーションで蛍光基質を通じて明らかになった化学反応に伴う酵素の構造変化

Conformational change of the rotary motor F_1-ATPase revealed by single-molecule imaging and docking simulation

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F_1-ATPase is an ATP-driven rotary motor in which the central shaft rotates against the catalytic stator ring $\alpha_3\beta_3$. To probe the structure of the catalytic core, we observed the fluorescent ATPs bound to the catalytic site using the polarization-modulation microscopy which enables to quantify the intensity, mobility and orientation of single fluorophore. After ATP hydrolysis, the intensity and polarization factor decreased. Furthermore, the docking models of α and β subunits with Cy3-nucleotide indicated that the Cy3 fluorophore was more exposed to the bulk. These results indicates that structural changes between intermediates can be indirectly observed through hydrolyzable fluorescent nucleotide.

2Pos147 光渦トラップを使った力学測定システムの開発：DNA オリガミを使った F_1-ATPase の回転可視化

Development of optical vortex trapping system with DNA origami for the precise measurements of torque generated by F_1-ATPase

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To analyze mechanical property of F_1-ATPase (F1), we have been developing an optical trap system with an optical vortex (OV), which induces constant torque on an object along ring-shaped intensity profile. Previously we have succeeded in applying torque to F1 via a polystyrene particle by an OV. For precise measurement, the particle has to be along to the ring-shaped profile, whose size is predetermined. To adjust their radii, we used a DNA origami nanostructure as a linker between the particle and a rotor of F1. With this system, we achieved to visualize rotational motion of F1 hydrolyzing ATP. The radius of rotation was evaluated to be the same length as designed. We will optimize a length of a linker to measure torque generated by F1.

2Pos148 *Enterococcus hirae* 由来 V_1-ATPase の回転とヌクレオチド結合解離の１分子同時観察

Single-molecule simultaneous observation of rotation and nucleotide binding/release of *Enterococcus hirae* V_1-ATPase

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V_1-ATPase (V_1) is a rotary molecular motor driven by ATP hydrolysis. To understand chemo-mechanical coupling scheme of *Enterococcus hirae* V_1, we simultaneously observed mechanical rotation of the rotor subunit and binding/release of Cy3-labeled ATP/ADP on the catalytic site. Binding of Cy3-ATP always coincided with timing of rotational steps within time resolution of the observation up to 6.7 ms, indicating that ATP binding triggers rotation. After binding, release of the product Cy3-ADP occurred around 240° to 360° rotational steps. Interestingly, detailed analysis revealed that Cy3-ADP sometimes remained bound even after the completion of the 360° rotation, suggesting that ADP release is not always tightly coupled with the mechanical rotation.

2Pos149 高熱菌 *Bacillus* PS3 由来 F_oF_1-ATP 合成酵素のプロトン輸送活性および H^+/ATP の決定

Determination of the proton pump activity and the H^+/ATP ratio of thermophilic *Bacillus* PS3 F_oF_1-ATP synthase

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The F_oF_1-ATP synthase synthesizes ATP using proton electrochemical potential across a membrane. The reaction is reversible, therefore it can hydrolyze ATP and pump protons. To observe the proton pump activity, we reconstituted the enzyme into liposome membrane labeled with a pH-sensitive fluorescent dye. Uptaking protons induced internal acidification and fluorescence change. The proton pump activity was calculated from fluorescence change, which obeyed Michaelis-Menten kinetics with the V_{max} of 434±17 H^+/s. V_{max} of the ATPase activity measured under the identical condition was 128±18 ATP/s. These values yielded the H^+/ATP ratio of 3.4±0.5, in agreement with the value of 3.3 predicted from structural studies.

2Pos150 細菌べん毛モーターの回転方向変換制御機構の解明

Elucidation of the directional switching mechanism of the bacterial flagellar motor by electron cryomicroscopy

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Many bacteria swim by reversibly rotating flagella. The three switch proteins, FliG, FliM and FliN, form the C-ring on the cytoplasmic face of the MS ring and control counterclockwise-clockwise (CCW/CW) switching of the motor rotation. To understand the switching mechanisms in detail, we analyzed the C ring structures of wild type (CCW form) and CW-locked mutants by electron cryomicroscopy (cryoEM). Although 3D map was calcutated without imposing symmetry, the C ring struture showed 34-fold symmetry. By docking crystal structures of the switch proteins into the 3D map, we built a partial swich complex model. We will report the structural change between the CCW and CW forms and the switching mechanism of flagellar motor rotation.

2Pos151 Molecular dynamics study of pressure effects on unbinding of the CheY-FliM complex

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The rotational switching of the bacteria flagella motor is controlled by binding of the signaling molecule CheY onto FliM which is a part of motor basal body. Recently, it was found that a high hydrostatic pressure can induce the rotational switching in the absence of CheY. To gain more insights into pressure effects on the motor switching, we studied the binding stability of the CheY-FliM complex under different pressure conditions using molecular dynamics simulations. The dissociation of CheY-FliM complex under a high/low pressure was observed using an efficient sampling method (PaCS-MD) that enhances the conformational transitions. In this presentation, we report differences in the binding stability and discuss the mechanism describing the pressure effect.

2Pos152 海洋性ビブリオ菌 FliG の EHPQR-motif 周辺構造によるべん毛の回転方向決定
Direction of flagellar motor rotation determined by the structure around EHPQR-motif of FliG in marine *Vibrio*

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Bacterial flagellar motor can rotate to either Clockwise (CW) or Counter Clockwise (CCW). The torque of motor is generated by interaction between FliG and PomA proteins and the interaction state determines the rotational direction. To understand how to determine the direction, we made many mutants of *Vibrio* FliG. We found that E144D gives tumbling and Q147H gives CW-bias phenotypes. Those mutation sites belong to EHPQR-motif, which is assumed to act as a hinge of conformational change in FliG. We also found that G214S and G215A, inferred to be close to the hinge region, give CCW-bias and CW-bias phenotype, respectively. We are examining the structural changes in the EHPQR-motif mutants by MD simulations. We'd like to discuss the rotational state of the hinge region.

2Pos153* 2 種イオン駆動型べん毛モーターの入力と出力の関係
Input-output relationship of dual ion driven flagellar motor

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The bacterial flagellar motor converts the free energy that gained from the ion current across the membrane to high-speed rotation. The motor consists of a rotor surrounded by multiple ion-conducting stator units. Coupling ions are known to be H^+, Na^+ or K^+. *Bacillus alcalophilus* uses Na^+ and K^+ conducting MotPMotS (BA-MotPS) as a stator. Interestingly, the motor of *E. coli* expressing BA-MotPS is driven by both Na^+ and K^+, although natural *E. coli* motor runs by H^+ flux.
In this study, we measured both ion-motive force and motor speed of MotPMotS motor in *E. coli*. The speed varies linearly to the sodium- and potassium ion-motive force similar to the original *E. coli* H^+-driven motor, indicating that the energy conversion mechanism underlying the motor is common.

2Pos154 高度高塩菌ハロバクテリウムサリナラムのべん毛の回転とステップ運動の直接観察
Direct observation of rotation and steps of the archaellum in the swimming halophilic archaeon Halobacterium salinarum

Yoshiaki Kinosita[1], Nariya Uchida[2], Daisuke Nakane[1], Takayuki Nishizaka[1] ([1]*Department of Physics, Gakushuin University*, [2]*Department of Physics, Tohoku University*)

Archaea swim by rotating archaeal flagellum termed "archaellum". Here, to clarify the motility mechanism, we applied our microscope techniques to *Halobacterium salinarum*. By 3-D tracking of QDs, the left-handed corkscrewing of cell body was detected. Under TIRF illumination, a right-handed helical structure of archaella with rotation speed at 23 Hz were revealed. Using these structural and kinetic parameters, we computationally reproduced the swimming motility with a hydrodynamic model and estimated the archaellar motor torque to be 50 pN nm. Finally, in tethered-cell assay, we observed intermittent pauses during rotation with ~36°or 60° intervals. From the estimation of the energy input as 10 or 6 ATPs per revolution, the energy efficiency is estimated as ~6-10%.

2Pos155 キネシンネックリンカーの構造変化に伴う自由エネルギー変化
Measuring of energy at neck linker docking of single kinesin molecule

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Kinesin is a molecular motor which carries cargo such as organelles moving along microtubules unidirectionally utilizing ATP hydrolysis energy in a cell. It is generally postulated that the neck linker docking contributes to the force generation of kinesin. However, the contribution was not quantitatively determined. Here we determined the size of conformational change of kinesin neck linker and force dependency of docking-undocking reaction rate by optical tweezers. From the force dependency, we determined the energy gap between two conformations of neck linker.

2Pos156 LZMW を利用した高濃度蛍光 ATP 存在下でのキネシン運動と ATP 結合の同時蛍光 1 分子計測
Simultaneous fluorescent observation of kinesin motility and ATP occupancy with high concentration of fluorescent labeled ATP using LZMW

Kazuya Fujimoto[1], Yuki Morita[1], Hirofumi Shintaku[1], Michio Tomishige[2], Ryota Iino[3], Hidetoshi Kotera[1], Ryuji Yokokawa[1] ([1]*Kyoto University Department of Micro Engineering*, [2]*The University of Tokyo Department of Applied Physics*, [3]*National Institute of Natural Science Institute for Molecular Science*)

Kinesin moves along a microtubule toward its plus end by repeating 8-nm steps. Although the coupling between mechanical step and hydrolysis of a ATP molecule has been extensively studied, the timing of attachment or detachment of ATP to kinesin motor domain is still controversial. In this study, we tried to establish a simultaneous observation of kinesin motility and ATP attachment/detachment to a kinesin using Linear Zero Mode Waveguide (LZMW), which has a potential to elevate the concentration of fluorescent ATP (fATP) to micromolar level. We realized simultaneous observation of kinesin motility and attachment of ATP at 500 nM, which is 5 times higher than the limit in conventional methods.

2Pos157 CYK-4 による kinesin-6 の回転運動揺らぎ
CYK-4 induces the large fluctuations of the left-handed rotational movement of dimeric kinesin-6

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Centralspindlin, kinesin-6 (motor)/CYK-4 (Rho-GAP) complex, is essential for assembly of the central spindle. CYK-4 binding constrains the configuration of the two motor domains in kinesin-6. In this work, using 3D tracking microscopy, 3D movements of centralspindlin, dimeric kinesin-6 and monomeric kinesin-6 along a suspended microtubule were quantified. We found that the three kinesins displayed a left-handed spiraling movements around the microtubule. The large fluctuations of lateral displacement and the reduction in the rotational velocity were observed with centralspindlin and monomeric kinesin-6. These findings suggest that kinesin-6 subunits of centralspindlin may use a single motor domain by the constraints on the configurations of the two motor domains.

2Pos158 微小管と光応答性 DNA による物質輸送システムの構築
Construction of a nano-transportation system by using microtubules and photoresponsive DNA

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Biomolecular motor systems, such as microtubule-kinesin, are the smallest natural machines which perform mechanical works by consuming chemical energy. Nowadays reconstructed microtubule-kinesin system is being used for different nanotechnological applications, for example nano-transportaters and active probes. However, regulated nano-transportation through controlled loading and unloading of cargos at a desired place and time has not been realized yet. In this study, by employing photoresponsive DNA, we aim to spatiotemporally regulate the microtubule-kinesin based transportation by controlling the loading and unloading of cargo materials. This work will open a new door to employ biomolecular motor systems for targeted transportation of nanomaterials.

2Pos159 キネシンの非平衡熱散逸
Non-equilibrium dissipation of kinesin

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Kinesin is a molecular motor that carries cellular cargos along microtubules. Although its working mechanism has been understood in terms of the molecular structure, little is known on the thermodynamic energetics. Recently, an equality is proposed, in which violation of fluctuation-response relation (i.e. the difference between the fluctuation of the motor velocity and the linear response to a small perturbation) provides the non-equilibrium dissipation consumed by the system. Here, on the basis of the equality, we measured the dissipation by single kinesin motor using optical tweezers. Combining to the numerical simulation with a phenomenological 2-state model, we discuss the origin of non-equilibrium energetics in working kinesins in quantitative detail.

2Pos160 生細胞内における微小管へのキネシン結合速度定数の直接計測
Direct measurement of the binding rate constant of kinesin to microtubules in living cells

Taketoshi Kambara, Yasushi Okada (*RIKEN, QBiC*)

It has been established that conventional kinesin, KIF5, selectively moves along a specific subset of microtubules in living cells. To understand the mechanism of the selective binding, it would be important to examine whether kinesin binding to specific subsets of microtubules is enhanced, inhibited or both. Here, we established a method for direct measurement of the binding rate constant of kinesin to microtubules in living cells using TIRF microscopy. To our surprise, there are four populations of microtubules that show different binding rate constant. These data suggest the existence of several mechanisms as the guidance cues for KIF5 by accelerating its binding to some specific subset of microtubules.

2Pos161 *Flavobacterium johnsoniae* の滑走に関与するマルチレール構造
The multi-rail structure contributes to gliding motility of *Flavobacterium johnsoniae*

Satoshi Shibata, Koji Nakayama (*Graduate Sch. of Biomedical Sciencces, Nagasaki Univ.*)

F. johnsoniae exhibits gliding motility on surfaces by using the adhesin SprB, which propels along a left-handed helical loop on the cell surface. Our detail analysis of SprB movement showed that SprB overtook and passed another SprB during movement, suggesting the presence of multiple lanes for SprB in a helical track. In support of this idea, electron microscopic analysis revealed that the multi-rail structure, which formed a complex with SprB and required gliding proteins (Glds), was present at the periplasmic side of outer membrane. These results suggest that the multi-rail structure is a part of gliding machinery and enables SprB to move smoothly, resulting in efficient gliding of the cell.

2Pos162* マイコプラズマ・モービレ滑走運動におけるシングルユニットが発生する力
Force generated by single unit in *Mycoplasma mobile* gliding

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Mycoplasma mobile, a fish pathogen glides on solid surfaces. The cell has 450 units of gliding machinery, composed of intracellular motors and surface proteins. In this study, we focused on stall force. As many units contribute to the stall force, we limited the working unit number by addition of a free sialylated oligosaccharide, a binding target of gliding machinery. In this condition, we detected force increments as repeated small steps. Each step reflects single unit movement, and the force was calculated to be 1.6 pN. This force is smaller than those of conventional motor proteins, suggesting that the gliding machinery contains large gears, probably Gli521. We will discuss the mechanism of gliding in more concrete way from this study.

2Pos163 Dynamics of Type IV pili controlled by light direction in unicellular cyanobacteria

Daisuke Nakane, Takayuki Nishizaka (*Dept. of Phys., Gakushuin Univ.*)

Type IV pilus filaments (T4PFs) are cell surface appendage observed in various bacteria, and used as one of motility machineries. They repeat the cycle of extension, attach to surfaces and retraction, to propel the cell forward. Here we directly observed T4PFs in a model cyanobacteria *Synecosystis* sp. PCC6803 under optical microscopy at single-cell level. The lateral blue light stimulated the extension of T4PFs specifically at the forward side of optical axis resulting in negative phototaxis. However, the partial blue light positively controlled the extension specifically at the stimulated area. These apparent inconsistency providing a new insight into the signal processing strategies in tiny organisms which have comparative size to the wavelength of visible light.

2Pos164 マイコプラズマモービレ由来滑走タンパク質 Gli349 の構造ドメインの探索とその構造解析
Determination of domain boundaries and analysis of domain structures of the gliding protein Gli349 from Mycoplasma mobile

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Mycoplasma mobile glides on a glass surface using a Gli349 protein, but the mechanism remains unsolved. To reveal the mechanism, it is required to determine detailed structures of Gli349. Because Gli349 is a huge protein (349 kDa) and has multiple, extended conformations, however, it is necessary to dissect it into fragments for structural analysis. Here, we predicted domain boundaries of Gli349 by bioinformatics and constructed 33 fragments with a solubility-enhancement tag. Among them, we could purify two soluble fragments, I(HD) and O(HD), and measured their structural properties by circular dichroism and X-ray scattering. We also performed limited proteolysis of full-length Gli349 to determine the domain boundaries. The results will be discussed in the meeting.

2Pos165 高圧力顕微鏡法による深海微生物の遊泳運動観察
Swimming motility of deep-sea bacteria measured by high-pressure microscopy

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Schwanella benthica strain DB21MT-2 is an absolutely piezophilic bacterium. This strain was isolated from the world's deepest sediment (Mariana Trench, Challenger Deep at a depth of ~11,000 m). Here, we studied the swimming motility of DB21MT-2 cells by high-pressure microscopy. Most cells did not swim in solution at 0.1 MPa. In contrast, the fraction of the swimming cells increased with increases of pressures, and then reached to a maximum at 50 MPa. The fraction and speed of the swimming cells were ~0.2 and ~15 μm s^{-1}, respectively. Our results showed that DB21MT-2 is equipped with a motility machinery suitable for high hydrostatic pressure environments.

2Pos166* *Mycoplasma mobile* の滑走装置に局在するペアになった F 型 ATP アーゼのパラログ
Paired F-type ATPase paralog in gliding machinery of Mycoplasma mobile

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M. mobile, a fish pathogen, glides on solid surfaces with a unique mechanism based on ATP hydrolysis. The internal structure of the gliding machinery has about 28 filaments covered with about 17 particles, including paralogs of F-type ATPase α and β subunits. In this study, we isolated and analyzed the particles by negative-staining electron microscopy and image averaging. Surprisingly, two hexamers similar to F-type ATPase formed a pair connected by two arms. The isolated particles showed ATPase activity of 0.1 ATP molecule/s. High-speed atomic force microscopy (AFM) suggested triangle steps of rotation. These results suggest that the motor for *M. mobile* gliding has evolved from F-type ATPase.

2Pos167* Dynamics and heterogeneity of ATP production and consumption in single C2C12 myotubes

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Adenosine 5' -triphosphate (ATP) is consumed as energy source in many biological processes, such as muscle contraction, membrane transport, and metabolic reactions. ATP is produced by the cytosolic glycolysis and mitochondrial electron transport system. It remains unclear how ATP level is coordinately controlled in cytosol and mitochondria during muscle contraction. In this study, we will report dynamics and heterogeneity of electrical pulse stimulated-cytosolic and mitochondrial ATP levels in single C2C12 myotube by using FRET-based fluorescent ATP probe called ATeam (Imamura et al, PNAS, 2009).

2Pos168 C 型インフルエンザウイルスの運動機構
Motile mechanism of influenza C virus

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An influenza C virus (ICV) membrane glycoprotein HEF is composed of three functional domains: hemagglutinin, esterase and fusion domains. Hemagglutinin domain associates with a viral receptor and esterase destroys the receptor. Fusion domain mediates fusion between virus and endosome membranes. Here, we report a novel function of hemagglutinin and esterase as virus motile machinery for virus infection. Using total internal reflection fluorescence microscopy, we examined ICV behavior on glass surfaces with immobilized receptors that mimicked cell surfaces. On the surface ICV moved straight at the same rate. We think that ICV moves by exchanging hemagglutinin-receptor bindings and esterase controls orientation and rate of virus movement by destroying receptors.

2Pos169 鞭毛・繊毛の表面運動：現象の普遍性と膜タンパク質のダイナミクス

Surface motility in eukaryote cilia/flagella: Generality and membrane protein dynamics

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Chlamydomonas flagella display surface motility such that small plastic beads attached to the surface actively move toward tip and base. This motility enables cells to glide on solid surfaces. Previous studies suggested that it is driven by the intra-flagella transport system (IFT) based on kinesin and dynein, possibly through the movement of transmembrane proteins. However, it is not understood how a cell can glide beyond the flagellar length. Here we show that some flagellar glycoproteins tend to detach from the membrane, and that their turnover is crucial for long-distance gliding. Surface motility is likely common to cilia/flagella of various organisms, since a similar bead movement is observed in the apical ciliary tuft in sea urchin embryos.

2Pos170 微小管ネットワークによって引き起こされる細胞質回転流動

Spatial confinement of active microtubule networks induces large-scale rotational cytoplasmic flow

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The cytoplasm is an active matter that generates directed motion such as cytoplasmic streaming. Although the cytoplasmic flow driven by actin networks has been extensively studied, that by microtubule networks remains unclear. Here, we examined whether and how microtubule networks induce cytoplasmic flow in Xenopus egg extracts. In bulk extracts, the microtubule network continually emerged vortex flow on the length-scale of 10 μm, which was not sustained over 1 min. Meanwhile, encapsulation of the extracts in droplets produced rotational cytoplasmic flow, on the length-scale of 100 μm, persisting for 1 hr in one direction. These suggest that the physical boundary arranges microtubule networks to induce cytoplasmic flow on the larger length- and time- scales.

2Pos171 軸糸直径サイズ変化による鞭毛繊毛の屈曲運動の制御

Regulation of cilia and flagella bending movements through the change of axoneme diameter

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Cilia and flagella movements are based on microtubule (MT) sliding powered by dynein. *Chlamydomonas* mutants lacking a central pair (CP) or radial spokes (RS), are non-motile, while double mutants between the mutants and suppressor mutants partially lacking MT-crosslink structure (Nexin) are motile. Here we examined the structural bases for the mutant motility. X-ray fiber diffraction studies showed that the size of axoneme diameter is different among mutants; ~180 nm in the CP/RS mutants, ~185 nm in the Nexin mutants, and ~183 nm in the double mutants and wt, indicating that the lack of nexin restores normal size of diameter in CP/RS mutants. We suggest that the balance between CP/RS and nexin regulates axoneme diameter size for the effective dynein-MT interactions.

2Pos172 中心子の普遍的 9 回対称性構造の構築機構

Assembly mechanisms of the nine-fold symmetry of the centriole structure

Masafumi Hirono (*Frontier Biosci. Hosei Univ.*)

Centrioles have a conserved structure with nine triplet microtubules arranged in rotational symmetry. The cartwheel, a subcentriolar structure consisting of a hub and nine spokes, is critically important for establishment of the centriole's 9-fold symmetry. Here, we examined effects of changing its spoke number and connection property to triplets by engineering two cartwheel components. The results showed that the spoke number does not solely determine the triplet number, and the connection between the cartwheel and triplet is crucial for establishment of the centriole structure. We thus surmise that cartwheels and centriolar microtubules assemble independently of each other, and their dynamic interaction stabilizes only the centrioles with 9-fold symmetry.

2Pos173 X 線繊維回折を用いた真核生物鞭毛軸糸の構造ダイナミクス解析

X-ray fiber diffraction study on structural dynamics of flagellar axonemes of Chlamydomonas

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The bending mechanism of eukaryotic flagellum has remained a long-standing unresolved issue. We investigate the mechanism using X-ray fiber diffraction. Using Chlamydomonas, we explored the spatial arrangement and dynamics of axonemal components under physiological conditions by small angle X-ray fiber diffraction. The axonemes were oriented in a physiological solution by continuous shear-flow and were exposed to intense and stable X-rays generated in the synchrotron radiation facility SPring-8 BL45XU or BL40XU. Diffraction patterns were obtained in different Ca ion concentrations. In the high Ca ion concentrations, the 48-nm and 24-nm meridional reflections changed their intensity profiles, suggesting changes in the helical nature of the whole axonemes.

2Pos174* 3-D measurement of bending dependency of the maximum force of the single tracheal cilium

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Highly coordinated dyneins generate ciliary asymmetric beating. We here measured, using optical tweezers with 3-D tracking, the force of surface-immobilized cilium. A bead was attached to the tip of the cilium, and trapped at various points of beating stroke to estimate the maximum force from the time course of 3-D displacements. In representative two examples, forces driven by same cilia decreased as the trapping positions displaced closer to ends of effective stroke. For three representative data in which contributions of the cilium stiffness were subtracted from the apparent maximum forces, similar dependency was observed. These observations suggest that the active bending force from dyneins in the power-stroke state depends on the bending deformation of the axoneme.

2Pos175 アルファシヌクレインタンパク質による輸送性微小管の制御機構

Alpha-synuclein binds unconventional microtubules that have a unique function

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The neuronal protein α-synuclein has been linked to Parkinson's disease, but how synucleins play a causative role is not clear. Here we show that α-synuclein is required for the creation of unconventional microtubule named as transportable microtubules (tMTs), which function as carriers for anterograde cytoplasmic dynein transport. Live-cell imaging indicated the co-transport of synuclein with cytoplasmic dynein to the plus ends of MTs. We further observed α-synuclein surrounded the MT in a pattern similar to that of bamboo nodes and preferentially bound to 14-protofilaments MTs by electron microscopy. Indeed, we found rare MTs with 14-protofilaments in the rat femoral nerve which increased in quantity upon nerve ligation, suggesting they are transported.

2Pos176 マウス Tppp (Tubulin Polymerization Promoting Protein)の機能解析

Analysis of Tppp (Tubulin Polymerization Promoting Protein)

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Tppp/p25 is a microtubule protein that is detected in protein inclusions in various neurodegenerative diseases. The physiological role of Tppp/p25 has remained obscured, however. In this work, to clarify the role of Tppp/p25 in vivo, we have examined phenotype of Tppp and Tppp3 knockout mice. Further, we constructed purified recombinant mouse Tppp and Tppp3 proteins to examine functions of these proteins on microtubule polymerization.

2Pos177 マウス繊毛細胞における Dpcd の機能解析

Role of Dpcd in motile cilia of mice

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Motile cilia plays important role on transport of fluid in our body. Disruption of function of motile cilia leads to hydrocephalus, bronchitis, and infertility. Thus, it is important to understand mechanism of motility of cilia. Here we examine role of Dpcd on function of motile cilia. Expressions of Dpcd are found only in motile ciliated cells of mice. By analyzing phenotype of knockout mice and transgenic mice, protein, we have addressed physiological role and subcellular localization of Dpcd in motile ciliated cells of mouse airway.

2Pos178 IFT（繊毛内輸送）に関与する基底小体微小管の機能に関する研究

The function of the basal body microtubules associated with intraflagellar transport (IFT)

Yurika Koiso, Shin Yamaguchi, Mitsuhiro Sugawa, Takuya Kobayashi, Yoko Y. Toyoshima, Junichiro Yajima (*Department of Life Sciences Graduate School of Art & Sciences, The University of Tokyo*)

IFT is essential for ciliogenesis. Also, basal bodies at cilia base play an important role in organizing ciliary microtubules structure. However, the spatiotemporal mechanism that IFT particles assemble at basal bodies of cilia base is unknown. Here, we examined microtubules orientation and function of basal bodies *in vitro*. We isolated basal bodies and found that kinesin move toward the distal end of basal bodies, indicating that basal bodies have uniform polarity orientation of microtubules with plus-ends distal to the cilia. Furthermore, kinesin was able to pass through a CPC (physical gate for IFT) of basal bodies. Our result suggests that basal bodies may assemble kinesins carrying IFT particles at the cilia base.

2Pos179 Distinctive structured radial spoke of mouse sperm underlies wave propagation of flagella

Kaoru Horiuchi[1], Hironori Ueno[2], Akihiro Narita[3], Hiroshi Hamada[4], Kyosuke Shinohara[1] ([1]*Tokyo University of Agriculture and Technology*, [2]*Aichi University of Education*, [3]*Nagoya University*, [4]*RIKEN Center for Developmental Biology*)

Understanding of principle on sperm swimming is important for fertility treatment. Previous works suggest that axonemal dyneins and radial spokes cooperatively generates the wave propagation of sea urchin sperm. How mammalian sperms achieve stable wave propagation has remained unknown, however. Here we report radial spoke is essential for generation of the wave propagation of mouse sperm. We have generated Rsph4a knockout mice and examined motion and ultrastructure of mouse sperm. In Rsph4a KO mice, sperms can generate bending but the bending failed to travel. Further, we have carried out cryoelectron tomography of mouse sperms. Unexpectedly, the wild type mouse sperm harbors distinctive structured radial spoke compared with that in the other eukaryotes.

2Pos180 クラミドモナス鞭毛から精製したラジアルスポークの特性

Properties of the Purified Radial Spoke of Chlamydomonas Flagella

Hitoshi Sakakibara[1], Yosuke Shimizu[1], Pinfen Yang[2], Hiroaki Kojima[1] ([1]*Protein Biophys. Gr., NICT*, [2]*Dept. Biol. Sci., Marquette Univ.*)

Radial spokes of flagella are the only structure to connect the peripheral microtubules and central-pair apparatus, and thought to play important roles for generating well-ordered flagellar waveforms. Using purified Chlamydomonas radial spokes, we examine the properties of them in vitro. By negative-staining EM, we observed its structure consisting of a base, a shaft, a neck, and a head. The joints of the neck-shaft, and the shaft-base were indicated to be flexible. From the distribution of neck-shaft angles (s.d.~15 deg), the stiffness of the joint was estimated to be ~6.6 x 101 (pN nm/rad). Combine with in vivo parameters, we can estimate the statical friction between radial-spoke head and central pair apparatus up to 1.3 pN.

2Pos181 ハプトネマの微小管系急速コイリング運動メカニズムを探る

Unveiling a mechanism for rapid microtubule coiling movement of haptonema

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The haptonema is a microtubule-based motile machinery uniquely present in haptophytes, the roles of which include prey capture and gliding migration. It is composed of six or seven singlet microtubules, and is as long as ten times of the cell body in some haptophyte species. The most notable movement of haptonema is "coiling", which occurs in several milliseconds by mechanical stimuli. To reveal the mechanism of haptonematal coiling, we carried out detailed observations of haptonematal structures. We found that haptonematal microtubules were cross-linked with each other by certain filamentous structures. Interestingly, some haptonemata showed one of the microtubules winding around the others in a helical path. Possible meanings of the observations will be discussed.

2Pos182 cAMP シグナルリレーにおける膜電位変化の計測と制御

Measurement and control of membrane potential changes in cAMP signal relay

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The social amoebae *Dictyostelium discoideum* is a model organism for studies on cell motility, chemotaxis and differentiation. Chemotactic stimulation by cAMP is known to elicit an influx of Ca^{2+} into the cell cytoplasm. This suggests that the membrane potential is changed by cAMP stimulation. However it remains unknown how the membrane potential works in the cell motility and signal transduction. To investigate the role of membrane potential, we measured periodic membrane potential changes depending on the spontaneous cAMP oscillations and controlled the membrane potential using optogenetic tools in *Dictyostelium* cells. We will discuss the role of membrane potential in the cAMP signal relay.

2Pos183 動的な場における時間空間知覚メカニズムの解析：走化性パラドクスの克服と細胞の整流作用

Delineating temporal and spatial sensing in migrating cells: chemotactic wave paradox and rectification of the leading edge response

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The importance of temporal sensing in chemotaxis of crawling cells has been debated for decades. In Dictyoselium, cell aggregation is dictated by chemotaxis to traveling waves of chemoattractant cAMP. Because the gradient direction reverses during the wave passage, how unidirectional migration is achieved remains unresolved. This is the so-called 'chemotactic wave paradox'. Though microfluidic emulation of dynamic gradients we found that the chemotactic response is rectified; the response is suppressed when the chemoattractant concentration is decreasing over time. The timescale dependence of the wave chemotaxis together with our theoretical analysis suggests an underlying reaction-diffusion mechanism.

2Pos184 細胞か接着により誘導される細胞の集団運動

Cell-Cell Adhesion guiding Collective Cell Migration

Katsuyoshi Matsushita (*Department of Biological Sciences, Graduate School of Science, Osaka University*)

Collective cell migration is frequently observed in development of organisms. Guidance mechanism of collective cell migration has been experimentally investigated and a lot of its hypothesis were proposed. However, the guidance mechanism based on cell-cell adhesion for collective cell migration were not sufficiently examined to date. To theoretically examine the possibility of the guidance using cell-cell adhesion, we develop a model using Cellular Potts model and examine the efficiency of cell-cell adhesion in guidance of a certain collective cell migration. We find that the cell-cell adhesion can guide the motion of cells in the same direction and through it efficiently induces the collective cell migration.

2Pos185 力測定で明かす神経幹細胞の集団遊走

Measuring the Forces in Neural Stem Cell Monolayer

Masahito Uwamichi, Masaki Sano (*Dept. of Phys., The Univ. of Tokyo*)

Experimental methods to measure cellular forces have been rapidly developed, mainly regarding single cell. Meanwhile, few experiments on forces within a cell monolayer also revealed the scenario to realize the jamming-like state in a confluent system. However, some cell types keep moving even in a high density state, and the mechanism for this behavior has not yet been unveiled. The explanation from the mechanical perspective on such a behavior is important to predict and control cell dynamics in some situations like after transplantation of stem cells.
We conducted an experiment to measure traction force from a neural stem cell monolayer. Then, we can reconstruct the stress inside the cells using the Newton's law of inertia. We will explain the obtained results.

2Pos186 細胞性粘菌における集団的回転運動の定量解析

Quantitative analysis of collective rotational motion of Dictyostelium cells

Taihei Fujimori[1], Akihiko Nakajima[1,2], Ryo Yokota[3], Ryo Nakabayashi[4], Gen Honda[1], Tetsuya Kobayashi[3], Satoshi Sawai[1,2] ([1]*Grad. Sch. of Arts & Sci., Univ. Tokyo*, [2]*Res. Ctr. Complex Syst. Biol., Univ. Tokyo*, [3]*Inst. of Ind. Sci., Univ. Tokyo*, [4]*Univ. Tokyo*)

Rotational motion is one of the characteristic mode of collective cell migration. Although it is often observed in multicellular development, quantitative information at the single cell level is still lacking. We traced all individual cells in Dictyostelium cells' aggregate rotating in the microfluidic chamber which provides confined environment. We revealed that cell population can be divided in two by calculating cell-cell correlation. One group is in outer region; cells maintain constant distance from the center of aggregate. The other is in inner region; cells frequently change position between inside and outside. We will discuss about the mechanism that accounts for the characteristics of rotational motion and how it contributes to Dictyostelium development.

2Pos187 Spatial heterogeneous and transient dynamics during collective cell migration in a monolayer of MDCK epithelial cells

Preetom Nag[1], Helal Khalifa[1,2], Hiroshi Teramoto[3], Naoya Yamaguchi[4], Chun-Biu Li[1], Hisashi Haga[2], Tamiki Komatsuzaki[1,2] ([1]*Research Institute for Electronic Science, Hokkaido University*, [2]*Graduate School of Life Science, Hokkaido University*, [3]*Hitachi, Ltd. Research & Development Group*, [4]*Skirball Institute of Biomolecular Medicine, New York University Langone Medical Center, USA*)

Collective cell migration plays an important role in many biological processes such as wound healing, embryonic development, cancer progression etc. To understand the mechanism of how individually migrating cells contribute to a collective migration, here we focus on collectively migrating Madin-Darby Canine Kidney (MDCK) epithelial monolayer. To obtain the flow field within the monolayer, here we adopt particle image velocimetry (PIV) technique which is typically used in fluid mechanics. The vector field obtained from PIV analysis is used to measure finite-time Lyapunov exponent (FTLE) field that computes the separation rate between nearby two points in a flow field. Indeed, FTLE field is used to elucidate different types of motion within the migrating monolayer.

2Pos188 細胞の協調運動における接着結合タンパク質の役割
Roles of adherence junction proteins in the collective cell movement in vitro and vivo

Takeomi Mizutani, Kazushige Kawabata (*Department of Advanced Transdisciplinary Sciences, Faculty of Advanced Life Science, Hokkaido University*)

Collective cell movement plays important roles in embryogenesis. However, the detailed mechanism is obscure. We focused proteins related to cell-cell adhesion and examined the effect of their deletion on collectiveness of cell movement using a cell line (MDCK cell). Deletion of tight junction proteins little affected the collectiveness. On the other hand, deletion of adherence junction proteins decreased the collectiveness. Besides, depletion of adherence junction proteins in Zebrafish embryo decreased the collectiveness and resulted in the fatality. These results suggest that adherence junction proteins are essential component for collective migration in vitro and vivo.

2Pos189* 筋分化 C2C12 におけるインスリン刺激時 S6K 活性のダイナミクスと不均一性
Dynamics and Heterogeneity of S6K activity in insulin stimulated-C2C12 myotubes

Haruki Inoue[1], Katsuyuki Kunida[2], Daisuke Hoshino[2], Takumi Wada[2], Shinya Kuroda[1,2] ([1]*Grad. Frontier Sci., Univ. Tokyo*, [2]*Grad. Sch. Sci., Univ. Tokyo*)

The insulin regulates various signal transduction and metabolisms. Especially, insulin regulates protein synthesis through p70 S6 kinase (S6K). We have previously shown that S6K exhibits adaptive response to insulin stimulation (Kubota et al., Molecular Cell, 2012). However, the dynamics and heterogeneity at single cell level resolution remain to be explored. In this study, we acquire the time series of insulin stimulated-S6K activity in single C2C12 myotube using live cell imaging. Furthermore, we will uncover the principal of dynamics and heterogeneity based on quantitative analysis and model analysis.

2Pos190 Beating rate changes of isolated cardiomyocyte clusters in different thermal environments

Wei Wang, Tomoyuki Kaneko (*LaRC, Dept. Frontier Biosci., Hosei Univ.*)

It's known that heart rate will slow down or speed up depending on the temperature. To understand the behavior of isolated cardiomyocyte clusters in the same condition, we measured the beating rate of isolated cardiomyocyte clusters by repeating two different environmental temperatures (room temperature and living body temperature). Our finding shows the heart beating at room temperature was slower and more unstable than living body temperature. And even if we repeated the experiment, the heart beating returned to the original pace. The results of this study demonstrate that isolated cardiomyocyte clusters can sense and respond to temperature change. And this capacity can be considered to contribute to helping heart to change its heart rate at different temperatures.

2Pos191 改良型蛍光 ATP センサーを用いた一細胞及び細胞内局所 ATP 濃度の測定
Quantification of single-cell and subcellular ATP concentrations using an improved fluorescent ATP indicator in mammalian cells

Hideyuki Yaginuma, Yasushi Okada (*QBiC, RIKEN*)

Adenosine triphosphate (ATP) provides energy to intracellular reactions. How synthesis and consumption of ATP is balanced inside cells is not clear. Recent studies propose a non-uniform heterogeneous ATP distribution inside a cell but the evidence is limited. We previously developed "QUEEN", a fluorescent ATP indicator protein with an improved quantitative property for measurement of ATP inside bacterial cells at 25°C. To quantify ATP in mammalian cells, here we developed an improved version of QUEEN suitable for measurement at 37°C. We used the new indicator to measure the ATP concentrations both in entire cytosol and in subcellular structures inside the cell. We propose that our improved QUEEN will be an important tool for studying intracellular energy balance.

2Pos192* がん細胞の損傷回復過程の定量評価
Quantitative evaluation of recovery from damage in cancer cells

Morito Sakuma[1,2], Kazuhito Tabata[1,2], Hiroyuki Noji[2], Hideo Higuchi[1] ([1]*Department of Physics, Graduate School of Science, The University of Tokyo*, [2]*Department of Applied Chemistry, Graduate School of Engineering, The University of Tokyo*, [3]*JSPS Research Fellow*)

Cancer cells show genetic heterogeneity, and the response to a therapy was different on each cells. Thus, cancer cells that survive therapy have a high probability of forming secondary tumors. Therefore, the effects of a therapy should be precisely evaluated at single cell level. In this experiment, the effects of therapy were evaluated by vesicle motility analyzed by the fluctuation of intensity in phase-contrast images. Higher vesicle motility was observed near nuclear before damage. At 2 min after damage, vesicle motility was decreased, and then increased. These results indicated that this method could be applied to evaluate recovery rate from damage. The results of single cell analysis of recovery rate of cancer cells will be discussed in the symposium.

2Pos193* ナノ秒パルス電場による細胞内応答の顕微ラマン・蛍光分光法を用いたその場観測

In situ observation of the intracellular responses to nanosecond pulsed electric fields by Raman and fluorescence spectroscopy

Yusuke Horii, Hirotsugu Hiramatsu, Takakazu Nakabayashi (*Graduate School of Pharmaceutical Sciences, Tohoku University*)

Application of nanosecond pulsed electric field (nsPEF) to cells is known to induce changes in cell conditions such as a change in intracellular ion concentration and apoptosis, which is expected to be applied as new therapies. In the present study, we investigated the change in intracellular environments due to nsPEF by Raman and fluorescence spectroscopy to clarify the mechanism of the effect due to nsPEF.
We developed a new electrode for applying nsPEF to cells, which enables us to perform the in situ observation of the cell response to nsPEF. We observed the increase in intracellular Ca2+ concentration and apoptosis due to nsPEF by fluorescence microscopy. We also measured Raman spectra of biological molecules in a cell in the presence of nsPEF.

2Pos194* ミトコンドリア輸送・膜電位・ATP と神経伸展の相関解析

Correlation analysis of transport, membrane potential, and ATP levels of mitochondria and neurite extension

Rika Suzuki, Kohji Hotta, Kotaro Oka (*Keio Univ.*)

In neurites, it is known that mitochondria (MTs) transported to growth cone (GC) have high membrane potential (MP), and locally anchored MTs contribute to morphogenesis via ATP production. However, no direct relation between transport, MP, and ATP levels of MTs and neurite morphogenesis has been demonstrated, we analyzed it with fluorescent imaging.
MPs and ATP levels had a positive correlation. ATP levels were high in anterogradelly-transported MTs and MPs are low in retrogradelly-transported MTs. Also in GC, MP and gross ATP production were larger than in axon. Moreover, artificial decrease in ATP of MTs caused neurite retraction. From above, active MTs are transported to GC, and ATP production at the edge of neurites is related to neurite elongation.

2Pos195* 細胞内局所発熱がストレス顆粒形成を開始する

Intracellular local thermogenesis initiates stress granule formation

Beini Shi[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*PRESTO, JST*)

Temperature governs a wide variety of cellular processes. However, the detail and significance of intracellular temperature are still unclear. In this study, we focused on the intracellular temperature during stress. In eukaryotic cells, cytoplasmic mRNAs assemble stress granule (SG), responsible for translation regulation under adverse environment. Given the fact that arsenite stress, SG inducer, targeted mitochondria, we hypothesized that the temperature change concerning mitochondria in the cell under stress might involve in this stress response signaling. Here, we demonstrated that SG formation had intrinsic relationship with local temperature change, which may be a novel principle of temperature signaling in cell biology.

2Pos196 Investigating the contribution of cytoskeletons on intracellular temperature variation

Takashi Yanagi[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharmac. Sci., Univ. Tokyo*, [2]*JST, PRESTO*)

Recent studies have shown that intracellular temperature distributes inhomogeneously, which might significantly influence intracellular biochemical reactions. However, the mechanism of this phenomenon is not fully understood. Because cytoskeletons are the most principal organelle in cells and distributes throughout the cell, we speculated that cytoskeletons might contribute to the intracellular temperature change and/or the cellular response to heat. In this study, we disrupted cytoskeletons, and performed temperature measurement in both steady state and mitochondria-stimulated cells by fluorescence thermometer and fluorescence microscopy and found cytoskeleton-dependent temperature distribution.

2Pos197 Attempt to Detect Functional Interaction between FoF1-ATPase and Adenine Nucleotide Translocator

Saki Yamashita, Takahiro Shibata, Yoshihiro Ohta (*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*)

Mitochondria are organelles that produce most of ATP required by cells. For ATP synthesis in the mitochondrial matrix, FoF1-ATPase, phosphate carrier, adenine nucleotide translocator form a complex known as ATP synthasome. The aim of this study is to examine the functional interaction among proteins constituting ATP synthasome. For this purpose, FRET-based ATP sensor, GO-ATeam was expressed in mitochondria of C6 cells. When ADP was added to the cells permeabilized with streptolysin O, ATP concentration was decreased in mitochondria. In the presence of oligomycin, however, ATP concentration in mitochondria was not significantly decreased. These results suggest the functional interaction between adenine nucleotide translocator and FoF1-ATPase.

2Pos198 Effects of mitochondrial volume on the generation of reactive oxygen species

Sawako Kimura[1], Satoshi Honda[2], Norihiro Umeda[2], Yoshihiro Ohta[1] ([1]*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*, [2]*Dept. of Mech. Sys. Eng., Tokyo Univ. of Agr. and Tech.*)

Reactive oxygen species (ROS) are mainly generated in mitochondria and act as signal molecules. Once excessively generated, ROS are known to induce cellular damages. Therefore, adequate control of ROS generation is necessary for cells. The aim of this study is to examine the effect of mitochondrial volume change on the ROS generation. For this purpose, we isolated mitochondria from porcine hearts and adsorbed on a cover slip. To observe ROS generation by each mitochondrion, mitochondria were stained with MitoSOX Red, a fluorescent indicator of superoxide anion. When we pressed a mitochondrion with AFM, ROS generation by the mitochondrion was significantly increased. This suggests that the decrease in mitochondrial volume leads to the enhancement of ROS generation.

2Pos199 Partial contribution of mitochondrial permeability transition to t-butyl hydroperoxide-induced cell death

Naoko Ashida, Xiaolei Shi, Hyonjin Choi, Yoshihiro Ohta (*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*)

Mitochondrial permeability transition (MPT) is thought to determine cell death under oxidative stress. However, MPT inhibitors only partially suppress oxidative stress-induced cell death. Here, we demonstrate that cells in which MPT is inhibited undergo cell death under oxidative stress. When C6 cells were exposed to 250 μM t-butyl hydroperoxide (t-BuOOH), mitochondrial depolarization lead to cell death. The fluorescence of calcein entrapped in mitochondria prior to addition of t-BuOOH was significantly decreased to 70% after mitochondrial depolarization. Cyclosporin A suppressed the decrease in mitochondrial calcein fluorescence, but not mitochondrial depolarization. These results show that t-BuOOH induced cell death even when it did not induce MPT.

2Pos200 Monitoring of mitochondrial activity during cell division

Kyunghak Cho, Kotoe Hirusaki, Yoshihiro Ohta (*Div. of Biotech. And Life Sci., Inst. of Eng., Tokyo Univ. of Agr. and Tech.*)

During cell division, cells drastically change their morphology and reorganize intracellular components. Since these are energy-requiring, the energy supply in cells could also change. The aim of this study is to examine the mitochondrial activity changes during cell division. As mitochondrial activity, we measured mitochondrial membrane potential, the generation of reactive oxygen species and ATP concentration in mitochondria. We also measured the effect of mitochondrial division on mitochondrial activity changes. During cell division, mitochondria were transiently depolarized and the generation of reactive oxygen species was decreased. These results indicate that mitochondrial activities changes during cell division. The details will be discussed.

2Pos201* パターン化人工膜を用いて脂質ラフトによる光シグナル伝達の制御機構を解明する
Regulation of phototransduction by lipid rafts studied with a micropatterned model membrane

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Phototransduction is believed to be regulated by lipid rafts. We quantitatively evaluated the affinity of the membrane proteins involved in the phototransduction (rhodopsin (Rh), transducin (Gt), phosphodiesterase (PDE6), and S-modulin) to lipid rafts (raftophilicity) by using a model membrane having patterned liquid ordered (Lo) (raft model) and liquid disordered (Ld) (non-raft model) bilayer domains. Raftophilicity of Rh increased upon dimerization, whereas that of Gt decreased after photo-activation. These results are consistent with the results in the disk membrane. Raftophilicity of PDE6 was very low, suggesting that photo-activated Gt have a higher chance to interact with PDE6, enhancing the phototransduction.

2Pos202 全反射照明蛍光顕微鏡を用いたRalGDS分子のEGF依存的な膜局在化メカニズムの解明
Elucidation of the EGF dependent localization mechanism of RalGDS molecule to plasma membrane using TIRF microscopy

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RalGDS is a guanine nucleotide exchange factor specific for small G-protein Ral. It is thought that the translocation of RalGDS from cytoplasm to plasma membrane is required for Ral activation. In this study, to understand the localization mechanism of RalGDS, we performed single molecule imaging of full-length RalGDS and its truncated constructs, RBD and REMCDC in living HeLa cells. Our result revealed that the association rate of RalGDS to plasma membrane was increased by interaction of RBD and Ras-GTP. While, the dissociation rate from plasma membrane was decreased by the interaction of REMCDC domain and Tyr-64 residue of Ras. We are currently examining the correlation of these kinetics and Ral activation.

2Pos203 分子動力学シミュレーションによる膜の細孔形成自由エネルギー解析
Free energy analysis of membrane pore formation by molecular dynamics simulations

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The battle against bacteria infection draws many attentions in the present. In particular, super bacteria, which are resistant to existing antibiotics, are a threat to the human race. Therefore, development of antimicrobial agents, which can target bacterial membranes and kill the bacteria, is attracting attention. The agents can self-assemble and form pores on the membrane, causing leakage of cytoplasm. The pore formation process by the agents has been widely studied. However, it is still not clear about the relation between membrane structure and free energy of pore formation. In this work, we analyze the process of membrane pore formation by molecular dynamics simulation and explore crucial factors of antimicrobial agents which control activity of pore formation.

2Pos204* Min反応拡散波の油中水滴内再構成
Reconstitution of Min Reaction-Diffusion Waves in Water-in-Oil Microdroplets

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Min system is a critical machinery for division of bacterial cells at the accurate position. The components of Min system, MinC, MinD, and MinE, oscillate from a pole to another pole of bacterial cells in an ATP dependent reaction-diffusion manner and determine the position of the division septum called Z-ring at the center of the cell. The Min system has been reconstituted in only open environments such as on planer lipid bilayers or outside surface of liposomes. However, reconstitution of Min system in confined environments like living cells has not been accomplished. Here, we found critical conditions to drive the Min system in confined environments and successfully reconstituted Min reaction-diffusion waves in water-in-oil microdroplets coated with polar lipids.

2Pos205 ラクトフェリシン B フラグメントと大腸菌および GUV との相互作用

Interactions of a fragment of lactoferricin B with *E.coli* and single GUVs

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Lactoferricin B (Lfcin B) and its fragments are antimicrobial peptides (AMPs). We investigated interactions of Lfcin B (4-9) with *E.coli* and single GUVs. Lfcin B (4-9) did not induce the influx of SYTOX green from the outside of *E.coli* into its cytoplasm up to 10 min, indicating that Lfcin B (4-9) did not damage the plasma membrane rapidly. Moreover, Lfcin B (4-9) did not induce any leakage of calcein from single DOPG/DOPC-GUVs. These results are different from those of Lfcin B (1). Using the single GUV method for CPPs (2), we found that lissamine rhodamine B-labeled Lfcin B (4-9) entered the lumen of DOPG/DOPC-GUVs without leakage of AF647 from the GUVs, indicating that it is a CPP-type AMP.

(1) Biochemistry, 54, 5802, 2015, (2) Phys. Chem. Chem. Phys., 16, 15752, 2014

2Pos206 フォトクロミック脂質を用いた蛍光脂質のダイナミクス

Intermembrane transfer of fluorescent lipid analogs using photochromic lipid analogs as FRET acceptors

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We synthesized photochromic phospholipid analog in which spiropyran is conjugated to the headgroup of phosphatidylglycerol. Liposomes containing the new lipid (nitro-BIPS-PG) exhibit absorbance at 545 nm and fluorescence at 600 nm (excitation at 543 nm) in UV-dependent manner. Dithionite quenched more than 80% of the fluorescence of nitro-BIPS-PG in small unilamellaer vesicles whereas NBD-labeled phosphatidylethanolamine in the same membrane was quenched 67 %, suggesting rapid flip-flop of nitro-BIPS-PG. Using nitro-BIPS-PG as FRET acceptor, it became possible to quantitate the intermembrane transfer of NBD-lipids without using detergent.

2Pos207 脂質膜の力学的特性は細胞透過ペプチド・トランスポータン 10(TP10)の単一ベシクルへの侵入に影響を与える

Mechanical Properties of Lipid Bilayers Affect the Entry of Cell-Penetrating Peptide Transportan 10 (TP10) into Single Vesicles

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To reveal the mechanism of the entry of TP10 into cells, we investigated the effects of lateral tension in lipid membranes on the entry of CF-TP10 into single DOPG/DOPC (2/8)-GUVs using the single GUV method for CPPs (1). CF-TP10 entered the GUV lumen before leakage of AF647 from the GUV. The fraction of entry of CF-TP10 before pore formation and also the rate of pore formation increased with tension. The CF-TP10-induced fractional area change of GUV membranes increased with time more slowly than the CF-TP10 concentration in the GUV membrane. This indicates the imbalance of CF-TP10 concentration in both the monolayers, inducing stretching of the inner monolayer. We discuss the mechanism of the entry of CF-TP10 into the single GUVs.

(1) Biochemistry, 53, 386, 2014.

2Pos208 一定張力が誘起する膜破壊の活性化エネルギーを用いた解析

Analysis of Constant Tension-Induced Rupture of Lipid Membranes Using Activation Energy

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The stretching of biomembranes plays important roles in various physiological and physicochemical phenomena. Here we analyzed the rate constant k_p of constant tension σ-induced rupture of GUVs using its activation energy U_a. First, we investigated the temperature dependence of k_p for charged DOPG/DOPC-GUVs and obtained the values of U_a of σ-induced rupture of the GUVs. A theoretical equation of U_a including electrostatic interaction effects well fit the data of the σ dependence of U_a. The Arrhenius equations for k_p using experimentally determined U_a fit well to the data of the σ dependence of k_p, indicating the usefulness of this equation.

(1) J. Chem. Phys., 143, 081103, 2015, (2) Phys. Chem. Chem. Phys, 18, 13487, 2016

2Pos209 抗菌ペプチド・マガイニン２が脂質膜中に誘起するポア形成のメカニズム

A Mechanism of Antimicrobial Peptide, Magainin 2-Induced Pore Formation in Lipid Membranes

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Antimicrobial peptide magainin 2 (Mag) forms pores in lipid membranes, which is the main cause of its bactericidal activity, but the mechanism of pore formation remains poorly understood. In our previous paper, we demonstrated that a Mag-induced pore is a stretch-activated pore and the stretch of the inner monolayer is a main driving force of the pore formation.[1] Here we made a theory of Mag-induced pore formation and obtained the rate constants of Mag-induced pore formation theoretically and compared with the experimental data quantitatively. The results of other experimental data indicate the validity of the theory. On the basis of these results, we discuss the mechanism of Mag-induced pore formation.

(1) Langmuir, 31, 3391, 2015

2Pos210 引張下コレステロール含有リン脂質二重膜における指組み構造の形成：分子動力学シミュレーション

Stretch-Induced Interdigitated Phase Formation in Phospholipid/Cholesterol Bilayer: Molecular Dynamics Simulation

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Interdigitated (Li) phase of phospholipids is known to be formed when exposed to high hydrostatic pressure or small alcohols. Recent molecular dynamics (MD) simulation studies reported that the Li phase is also formed in phospholipid bilayers under mechanical stresses, but its mechanism is still unclear. To clarify this, we performed MD simulations of stretched DPPC/cholesterol bilayers under various constant areal strains. We found that a Li phase domain was formed in the stretched bilayer when the areal strain exceeded a critical value. Additionally, we explained the mechanism of the Li phase formation based on the balance of the tension from the bilayer deformation, the line tension of the domain boundary, and the surface tension between Li phase bilayer and water.

2Pos211 細胞サイズ液滴内における高分子溶液の拡散とその空間閉じ込めの影響

Diffusion in polymer solutions confined in cell-sized droplets: effect of confinement size

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Molecular diffusion in intracellular environment is important since it rules cellular metabolism through molecular transportation. Intracellular environment is crowded with various polymers such as proteins, nucleic acids and other small molecules that are confined in lipid membranes. We aimed to elucidate how the confinement by lipid membrane affects polymer diffusion in molecularly crowded environment. The cellular environment was modeled using cell-sized water in oil (w/o) microdroplets, which lipid monolayer confines polymer solution. We found that there is a trend of diffusion coefficient of polymers increase with decrease in the confinement size. The results pave a way to understand confinement effect on transportation phenomena in vivo.

2Pos212 Investigating interactions and dynamics of pleckstrin homology domains on a lipid membrane surface

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Pleckstrin homology (PH) domains are lipid recognition modules in peripheral membrane proteins (PMPs). Their major function is to localize PMPs to cell membranes via interactions with phosphatidyl-inositol phosphates (PIPs). Here, we present a molecular dynamics simulation study that provides insights into the molecular details of interaction and dynamics of PH domains on lipid membranes. We show that the PH domain associates with PIP molecules in the membrane via a positively charged loop. Moreover, we show heterogenious anomalous diffusion of a PH domain, where the diffusivity of the PH domain fluctuates substantially depending on the number of bound PIPs.

E. Yamamoto et al., Structure (doi: 10.1016/j.str.2016.06.002).
E. Yamamoto et al., Sci. Rep. 5, 18245 (2015).

2Pos213* D体フェニルアラニンを含む抗菌ペプチドPhenylseptinの脂質膜との相互作用解析

Analysis of interaction between antimicrobial peptide phenylseptin containing a D-phenylalanine and membrane

Yuta Matsuo, Izuru Kawamura (*Grad. Sch. Eng., Yokohama Natl. Univ.*)

L-phenylseptin (L-Phes) and its diastereomer D-phenylseptin (D-Phes) are antimicrobial peptides isolated from the frog skin secretion of Hypsiboas punctatus. D-Phes containing a D-Phe at the 2nd amino acid position, which is generated by post-translational modification of L-Phes, has a phe-phe-phe sequence at N-terminus. They show the significant difference on antimicrobial activity to X. axonopodis pv. glycines. Here, we showed the QCM, CD and 31P solid-state NMR results to investigate how difference of interaction between each peptide and DMPC lipid bilayers. In CD experiments, both peptides showed the similar α-helical secondary structure. Binding affinity of D-Phe to DMPC lipid bilayer is higher than that of L-Phes. It may be the difference of peptide structure.

2Pos214* 人工生体膜とナノ空間を利用した1分子計測技術の開発

Single-molecule observation technique based on a model membrane and a nanometric gap structure

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We developed a novel platform for single-molecule observation by combining a model biological membrane and a nanometric structure (nanogap-junction). A micropatterned membrane composed of polymeric and fluid bilayers was formed on a glass substrate. Lipid vesicles were bounded onto the polymeric bilayer, and an elastomer (PDMS: polydimethylsiloxan) sheet was put onto the vesicles, forming a nanogap-junction between the fluid bilayer and PDMS. Vesicles acted as an adhesion layer with a defined thickness. Target molecules were transported into the nanogap junction by specific interaction and lateral diffusion of the fluid bilayer. Single molecules could be observed owing to the selective transport and reduced background noise.

2Pos215* カチオン性抗菌ペプチドボンビニンH2およびH4のリーシュマニア原虫模倣膜との特異的な相互作用

Specific interaction of cationic antimicrobial peptides bombinin H2 and H4 with *Leishmania* protozoa mimetic membrane

Shiho Kaneda, Akira Naito, Izuru Kawamura (*Grad. Sch. Eng., Yokohama Natl. Univ.*)

Bombinin H2 and its diastereomer H4 are antimicrobial peptides from frog skin secretions of the *Bombina variegata*. In this study, we used two kinds of negative charged membranes like *Leishmania* protozoa (DOPC/DOPE/DOPI/DOPS/ergosterol) and *Staphylococcus aureus* (DMPG/DMPC/cardiolipin) to investigate the interaction with the peptide. ^{31}P solid-state NMR spectra of *Leishmania* model membrane showed clearly an isotropic signal at 0.0 ppm deduced from strong membrane disruption by bombinin peptides. Meanwhile, in the *S. aureus* model, the isotropic signal did not appear even though similarly negative charged membrane, although rather changes in the lipid motion were observed. It is indicated that bombinin H2 or H4 specifically interacts with membrane of *Leishmania* protozoa.

2Pos216 水の脂質膜透過に対する膜の張力の効果

Effect of Lateral Tension on Membrane Permeability of Water in Lipid Membranes

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Tension in biomembranes plays important roles in various physiological and physicochemical phenomena.(1) Here we investigated the effect of tension on membrane permeability of water in DOPC membrane. For this purpose, we measured the volume change of single DOPC-GUVs upon transfer into a hypotonic solution in the presence of various tensions in the membrane using the micropipette method. The analysis of the time courses of the volume change provided the values of the membrane permeability of water in DOPC membranes, which increased greatly with tension. On the basis of the results, we discuss the mechanism of the results.

(1) Phys. Chem. Chem. Phys, 18, 13487, 2016

2Pos217 部分フッ素化リン脂質からなる人工膜内色素分子の発光挙動

Investigation of fluorescence emission from dye-lipid in partially fluorinated lipid bilayer

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Partially fluorinated lipids, which have both self-organization properties of lipids and unique properties of fluorocarbons, draw much attention as a matrix for membrane protein crystallization. In this study, the relation between structure of partially fluorinated lipid membrane and increased availability as a new class of biological reagent were investigated by temperature dependent fluorescence spectrum measurements. Red-shifted fluorescence emission from dye-lipids(Rh-DOPE) in partially fluorinated lipid membrane was observed below the phase transition temperature and considered to be caused by the increased interaction between surrounding lipids, such as a dipole-dipole interaction. The fluorescence life-time measurement was also performed for more details.

2Pos218 パターン化人工膜を利用した NAP-22 の膜結合と凝集挙動解析

Membrane binding and aggregation of neuronal acidic protein of 22kDa (NAP-22) studied with a patterned model membrane

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Neuronal acidic protein of 22kDa (NAP-22) is a neuron-enriched membrane protein whose function remains unknown. NAP-22 is associates with the detergent resistant membrane (DRM), suggesting its affinity to lipid rafts. We studied the interaction between NAP-22 and membrane lipids by using a patterned model membrane composed of polymeric and fluid bilayers. We found that phosphatidylserine (PS) enhanced membrane binding of NAP-22 and modulated its affinity to lipid raft. On the other hand, cholesterol did not enhance membrane binding. We discuss the mechanisms of localization to DRM and the aggregation formation and their implications to the functional roles of NAP-22.

2Pos219 Phospholipase C and D induced defects in POPC and POPC:POPG lipid bilayers: A simulation study

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Phospholipid headgroup are cleaved by the enzymes phospholipase C and D (PLC and PLD) and leave diacylglycerol and phosphatidic acid defects in lipid bilayer. In surface plasmon resonance experiments, phospholipase reactions with the bilayer of POPC and POPC:POPG liposomes leads their detachment from Biacore L1 chips with rates PLC>PLD and PLD>>PLC respectively. Understanding the role of defects formed from phospholipase reactions with liposomes is essential for designing liposomal containers for rapid, point-of-care detection of disease biomarkers. We employed MD simulations of lipid defects in POPC and POPC:POPG bilayers, which enable us to observe the effects of phospholipase-induced defects including fluidity and bilayer thickness.

2Pos220 固体 NMR と MD シミュレーションによる抗菌ペプチドアラメチシンとメリチンの膜結合構造と配向の解明

Structure and orientation of antimicrobial peptides alamethicin and melittin in membrane revealed by solid-state NMR and MD simulation

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The structure, topology and orientation of membrane-bound antimicrobial peptide alamethicin and melittin were studied using solid state NMR and MD simulation. ^{13}C chemical shift interaction was observed in [1-^{13}C]-labeled alamethicin and melittin in the membrane bound states under rapidly rotating peptide motions about the bilayer-normal. Alamethicin bound to DMPC bilayers showed that the transmembrane N- and C-terminal α-helical and 3_{10}-helical axes were tilted 17 and 32° to the bilayer normal, respectively. On the other hand, melittin bound to DMPG bilayers indicate that the pseudo transmembrane N- and C-terminal α-helical axes were tilted 32 and 30° to the bilayer normal, respectively. Structures obtained by MD simulation were in good agreements with NMR structures.

2Pos222 巨大一枚膜ベシクルに内包された DNA コンピュータ基盤遺伝子発現制御システム

Development of a DNA computer-based gene-regulatory system encapsulated in a giant unilamellar vesicle

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We developed a molecular system consisting of a DNA computer-based gene-regulatory module, a cell-free protein synthesis system, and a cell-sized giant unilamellar vesicle (GUV). The DNA computer-based gene-regulatory module in the GUV was directly controlled with a small molecule added from exterior of the GUV. The addition of small molecule finally produced GFP in the GUV. Compared with usual *in vitro* experiments, an environment in the system is close to that of living cell. However the system never receive interferences from the genetic network of living cells. Therefore the system is suitable to examine intrinsic functions of artificial genetic networks designed in silico. The system would be a useful tool for synthetic biology research.

2Pos223 Local pressure tensor calculation for molecular simulations and its application to lipid membranes

Koh Nakagawa, Hiroshi Noguchi (*ISSP, Univ. of Tokyo*)

Bridging discrete molecular system and continuum system is a challenging task. Many efforts have been paid to mapping the stress in molecular simulations to the continuum space. Recently, calculation method of local stress field in molecular system whose interactions consist of many-body potential was intensively debated. Many-body potentials such as angle (three-body) and dihedral (four-body) potentials are widely used in the MD simulation of biomolecules. Stimulated by the recent Admal and Tadmor's works, we propose the new force decomposition method for three-body potentials and discuss non-uniqueness of the local stress fields. We applied it to lipid bilayer membranes and demonstrated that its pressure profile is largely varied by the decomposition methods.

2Pos224 Formation of vesicles using self-reproducing oil droplet system

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Mixing an aqueous solution of an aldehyde containing an imidazole hydrochloride functionality with octylaniline led to the spontaneous formation of autocatalytic oil droplets. Thereafter, non-catalytic aldehyde molecules that do not react well with octylaniline were added to this autocatalytic system. As a result, the catalytic molecules that formed within the oil droplets promoted the condensation between octylaniline and non-catalytic aldehyde, which ultimately led to the synthesis of vesicular membrane molecules with imine functionality within the molecular aggregates. We proposed a protocell model that construct boundaries using a new process, owing to the formation of robust vesicles using an existing autocatalytic, self-reproduction oil drop system as scafold.

2Pos225* アクチンフィラメントの細胞膜シート上への結合

The binding of actin filament on the cell membrane flat sheet

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Filamentous actin (F-actin) networks on cell membrane play an essential role in the regulation of cell membrane shapes. A contact of F-actin to the cell membrane has been focused for a long time. However, it is still difficult to elucidate the interaction between F-actin and cell membrane surface. Recently, we have developed cell membrane model (cell membrane flat sheet: CMFS) for analyzing the interaction. The CMFS exposing inner leaflet can be prepared from GPMVs adsorbed on glass surface. We found that F-actin filaments stained by Rho-phalloidin bind on the CMFS. Moreover, F-actin seems to bind on the CMFS rather than glass surface. The CMFS may be appropriate as the cell membrane model to analyze the physical property of F-actin on cell membrane surface.

2Pos226* アクチン線維を封入した巨大リポソームの形態変化

Shape change of giant liposomes encapsulating actin filaments

Shunsuke Tanaka, Masahito Hayashi, Kingo Takiguchi (*Grad. Sch. Sci., Nagoya Univ.*)

Our previous studies demonstrated that actin-encapsulating giant liposomes (GLs) prepared by the natural swelling change to various shapes, such as disk- and semi-dumbbell-likes, as actin polymerizes inside. In this report, we find that actin-encapsulating GLs prepared by the method of centrifugation of water-in-oil emulsion usually change to spindle shape. In the spindle-shaped GLs, the direction of actin filaments tends to be aligned. The aspect ratio of spindle-shaped GLs could be further changed by osmotic pressure. The shape of GLs would be caused by spontaneous alignment of crowding actin filaments. We will observe the effect of actin-binding proteins such as myosin on the morphogenesis of actin-encapsulating GLs.

2Pos227 全反射赤外分光法を用いたウシオプシンと匂い分子の相互作用研究

ATR-FTIR study on the interactions between bovine opsin and odorants

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Animal visual rhodopsins are photoreceptor proteins which covalently bind an 11-cis retinal, and are often regarded as a model of GPCRs. Animal olfactory receptors, which bind various odorant molecules, also belong to the GPCR superfamily. Therefore, although an animal rhodopsin works as a photoreceptor, it may have a capability to bind odorant molecules. In this study, we are investigating the binding ability of bovine opsin (an apo protein of bovine rhodopsin) for three odorant molecules (geraniol, nerol, and citral) by using ATR-FTIR spectroscopy. Preliminary results showed odorant-induced changes in the amide I region, implying these odorants can indeed interact with opsin. Furthermore, using mutant opsins, we will discuss how they make the specific interactions.

2Pos228 Glu381Lys 点変異体を用いたニワトリクリプトクロム 4 の光反応メカニズムの解析

Photoreaction mechanism of chicken cryptochrome4 studied by using a Glu381Lys mutant

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Cryptochromes (CRYs) are thought to function as photoreceptors or light-driven magnetoreceptors in many organisms including vertebrates, though their photoreception mechanisms are still unclear. Previously, we established an expression and purification system for chicken CRY4 (cCRY4) and analyzed its chromophore redox cycle. In this study we examined the importance of Glu381 presumably locating near the chromophore-binding domain of cCRY4 in the photocycle. We expressed Glu381Lys mutant and compared the light-induced chromophore-reduction and protein conformational change with wildtype (WT). We found that the redox cycle of chromophore of WT and mutant is very similar, but protein conformation change was different from WT and mutant.

2Pos229* ニワトリクリプトクロム４の光依存的な相互作用分子とその分子メカニズム

Identification of chicken CRY4-interacting molecules and the interaction mechanism

Ayano Orii, Shingo Kondo, Keiko Okano, Toshiyuki Okano (*Dept. Eng. and Biosci., Grad. Sch. Adv. Sci. and Eng., Waseda Univ.*)

Cryptochrome (CRY) is assumed to be a circadian clock oscillator or photoreceptor or light-driven magnetoreceptor. Previously, we found that chicken CRY4 (cCRY4) changes its structure including C-terminal region (CRY C-terminal extension: CCE) after photoreduction of the FAD chromophore. In this study, we identified CRY-Interacting proteins (CRIPs) by Yeast Two-Hybrid (Y2H) screening and found that CRIPs interact with cCRY4 in a light-dependent manner. We analyzed CCE mutants of cCRY4 by Y2H assay to reveal correlation between the conformational change and the light-dependent interaction. Some cCRY4 mutants bind to CRIPs stronger than wild type, leading us to propose a molecular model for the light-dependent interaction between cCRY4 CCE and CRIPs.

2Pos230 桿体視細胞に発現する視物質の熱活性化頻度

Thermal activation rates of visual pigments expressed in rods

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Most vertebrates have two types of photoreceptor cells, rods and cones, which are responsible for scotopic and photopic vision, respectively. Rods show a low threshold of photon detection and higher sensitivity than cones. Rod visual pigment, rhodopsin, was optimized for the function of rods by the acquisition of the low thermal activation rate (kth). Some species of amphibians and reptiles exceptionally have rods containing not rhodopsin but cone visual pigments. However, it remains unknown whether or not they acquired the low kth like rhodopsin. Here, we compared the kth of amphibian and reptile visual pigments by biochemical and spectroscopic methods. Based on our results, we discuss the molecular mechanism to optimize the visual pigments for scotopic vision.

2Pos231 トランスデューシンは PDE を"間接的に"活性化する

Transducin activates cGMP phosphodiesterase indirectly

Teizo Asano, **Shuji Tachibanaki**, Satoru Kawamura (*Grad. Sch. Frontier Biosci., Osaka Univ.*)

The activation reaction of cGMP phosphodiesterase (PDE) by activated trimeric G protein, transducin (Tr*), is a key reaction to produce a photoresponse of vertebrate photoreceptor cells. So far, it is thought that, when Tr* activates PDE, it directly binds to a inhibitory subunit of PDE (PDEγ) in a holo-PDE (PDEαβγγ) and relieve the catalytic activity of PDE. In this study, we determined the kinetic parameters of the interaction reaction between PDE and Tr*. As the result, we found that the binding affinity of Tr* to holo-PDE is notably lower than that to free PDEγ. This result suggests the possibility that Tr* indirectly activates PDE by shifting the equilibrium between active and inactive PDE by controlling the concentration of free PDEγ.

2Pos232* 低温赤外分光法によるサル緑感受性視物質がもつ塩化物イオン結合部位の構造解析

Structural analysis of chloride binding site of monkey green studied by light-induced difference FTIR spectroscopy

Shunta Nakamura[1], Kota Katayama[2], Hiroo Imai[3], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*Dept. Pharm CWRU, USA*, [3]*Primate Res Inst., Kyoto Univ.*)

Primate green visual pigment is a G-protein-coupled receptor, which binds 11-cis retinal as a chromophore. The absorption maximum of the wild-type monkey green is red-shifted by the binding of chloride ion to the protein. In contrast to chloride ion, which functions as the physiological cofactor, nitrate ion does not affect color change upon binding. It has been proposed that His197 and Lys200 are the key residues for chloride binding, whereas structure of the binding site remains unclear. In this study, we identified additional mutations that affect chloride-dependent color change, to which we applied light-induced difference FTIR spectroscopy. Structure of the binding site of monkey green will be discussed based on the FTIR results.

2Pos233 新口動物の光受容タンパク質 Opn5 の多様性

Diversity of the photoreceptor protein Opn5 found in deuterostomes

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Opsins are the universal photoreceptive molecules for visual and non-visual photoreceptions in animals and are classified into several distinct groups. Opn5 forms an independent group whose members in vertebrates are diversified into four subgroups. Our analysis of the molecular properties showed that vertebrate Opn5 subgroups share G protein coupling property and are diversified based on their spectral sensitivities and their binding preference for retinal isomers. In this study, to get insight into the evolutional origin of diversified vertebrate Opn5 subgroups, we analyzed Opn5 genes found from several other deuterostome genomes. We would like to discuss the diversity of the molecular property of Opn5 widely found in deuterostomes.

2Pos234 Light-dependent association and dissociation of arrestin with bistable opsins

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Our previous studies suggested that most non-visual opsins have bistable nature, which means an interconvertible photoreaction between stable inactive and active states and could be an advantage for optogenetic applications. Several bistable opsins reach different levels of photoequilibrium between the two states under spectrally different light conditions, resulting in different G protein activation levels in cultured cells. To understand details about such photoequilibrium-dependent G protein activation for optogenetic applications, it is important to characterize interaction of arrestin with the two states of bistable opsins. Here, we investigated binding of arrestin to the active state and releasing from the inactive state in mammalian cultured cells.

2Pos235 桿体アレスチンのスプライスバリアント・p44 の自己会合の解析

Self-association of p44, a splice variant of visual rod arrestin

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Arrestin quenches the signal transduction by binding to phosphorylated GPCRs. Visual rod arrestin is self-associated at the physiologic concentration, and arrestin oligomer is likely to be a reservoir of an active monomer. While binding of phosphorylated C-tail of Meta-II induces the exposure of the finger loop of arrestin, which tightly binds to Meta-II, finger loop of p44 is exposed without binding of phosphorylated C-tail. To study the relationship between oligomerization and binding activity of arrestin, self-association of p44 was investigated by small-angle X-ray scattering. While arrestin monomer is an active form, p44 is oligomerized in the concentration-dependent manner. The physiologic relevance of self-association of arrestin will be discussed.

2Pos236 疾患に関わるロドプシン変異体の FTIR 研究
FTIR study of disease-causing mutations of rhodopsin

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Rhodopsin belongs to a family of G-protein-coupled receptors, which binds 11-cis retinal through a Schiff base linkage at K296. It is known that several rhodopsin mutations lead to serious diseases such as retinitis pigmentosa and congenital stationary night blindness, whose mechanisms have been extensively studied. Single amino acid mutations at G90 or T94, locating close to K296, impair visual function. We like to know how structural dynamics differ in the disease-causing mutants of rhodopsin. For this aim, light-induced difference FTIR spectroscopy is a powerful method, and sample preparation is now in progress. We like to discuss molecular mechanism of disease-causing mutations of rhodopsin from structural basis.

2Pos237 N-terminal region of modified Volvox channel rhodopsin-1(mVChR1) enchances Na+ Influx by drowing hydrogen ion

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The absorption spectrum of VChR1 (550 nm) derived from chlorophyte green algae Volvox is longer wavelength than that of ChR2 (450 nm). In our previous study, we developed modified VChR1 (mVChR1) which has broad absorption spectrum (450-550 nm) from VChR1 and N-terminal of Chlamydomonas ChR1. We succeed in using mVChR1 to restore vision includes the transduction of mVChR1 gene into retinal ganglion cells or ON-bipoloar cells in genetically blind mice and rats. However, it was still unknown the 3D structure of mVChR1 and VChR1. In this study, to reveal the region that has the function of the enhancing cation influx, we made comparison between the structure and the function of mVChR1 and native-VChR1 by using of ab initio structure modeling technique of Rosetta model.

2Pos238 フグ眼球由来の細胞株における広範囲な光波長応答性
A wide-range spectral photosensitivity in the puffer fish ocular cells

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Light triggers the transcription of clock genes in fish cultured cells, and this is likely linked to the photic entrainment of the circadian clock. To better understand the light-responsive transcription mechanisms, we focused on Fugu Eye cell derived from the eye ball of Takifugu rubripes. We searched for the light-inducted and/or clock-controlled genes by both microarray analysis and qRT-PCR, and identified 15 genes including the clock (-related) genes. Messenger RNA levels of cryptochrome and photolyase genes are controlled by light. The photic regulation are dependent on not only blue light but also green and red light, indicating the contribution of multiple photoreceptors or a novel photoreceptor having a wide spectral range of spectral sensitivity.

2Pos239 *Guillardia theta* 由来ロドプシン様タンパク質の分子機能解明
Molecular functions of rhodopsin-like proteins from *Guillardia theta*

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Microbial rhodopsins are membrane proteins having various kinds of functions, such as light-driven ion transporters, light sensors and light-activated enzymes. *Guillardia theta (G.theta)* possesses 44 genes encoding putative microbial rhodopsins in their genome. Functional studies have been performed for some proteins, but functions of most of others remain unknown.
In this study, we intended to investigate the gene expression and molecular functions of microbial rhodopsin-like proteins from *G. theta*. Gene expression analysis revealed that 11 rhodopsin-like genes were expressed in native cells under normal growth condition. We tried to express these proteins in yeast, among which we confirmed expression of 6 proteins. We would like to discuss their molecular functions.

2Pos240* 海洋性細菌のもつ光駆動イオンポンプ
Light-driven ion-pump activity of native marine bacteria

Yuichi Hashimoto[1], Rei Abe-Yoshizumi[1], Yoshitaka Kato[1], Keiichi Inoue[1,2], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*JST PRESTO*)

In 2000, light-driven eubacterial proton pump proteorhodopsin (PR) was found in the ocean. Since then, PR function has been mainly analyzed by *E. coli* expression system, because culture of native marine bacteria is not easy. We previously succeeded measuring proton pump activity of PRs in native cells. Here we found that the pump activities of flavobacterium *Nonlabens dokdonesis* DSW6 depend on the growth-phase, which was not proportional to the expression level of rhodopsin. This suggests the pump activity of rhodopsin being somehow modified under physiological conditions. Various possibilities can be considered such as different photocycle in native cell membrane and influence of secondary transport, whose experimental examination will be presented.

2Pos241 好熱性紅色光合成細菌 *Alc. tepidum* 由来の光捕集複合体の単離精製と分光学的特性評価
Purification and spectroscopic study of the light-harvesting complexes from thermophilic purple bacterium *Allochromatium tepidum*

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Allochromatium (A.) tepidum is a new species of thermophilic purple sulfur bacteria isolated from a sulfidic New Zealand microbial mat. It grows optimally near 45 C, slightly lower than another thermophilic purple sulfur bacterium *Thermochromatium (T.) tepidum*, but distinctly higher than the mesophilic bacterium *A. vinosum*. Unlike *T. tepidum*, the core antenna complex (LH1) of *A. tepidum* exhibits an absorption maximum at 890 nm, a feature similar to that of *A. vinosum* and the majority of purple bacteria. In this work, we describe isolation and purification processes of the LH1-RC and LH2 complexes from *A. tepidum* strain NZ. Comparisons of the biochemical, spectroscopic and calorimetric properties between the three purple sulfur bacteria will be presented.

2Pos242 Thermochromatium tepidum 由来光捕集１複合体における部位特異的変異体の分光学的解析

Spectroscopic characterization of site-directed mutants in Light-Harvesting 1 complex from Thermochromatium tepidum

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The light-harvesting 1 reaction center (LH1-RC) complex from Thermochromatium (Tch.) tepidum exhibits a largely red-shifted Qy absorption at 915 nm and enables the uphill energy transfer from LH1 to RC complexes. The unusual electronic transition is caused by the binding of Ca2+ to the LH1 C-terminal domain. Although the electron density map at 3.0 Å demonstrated the presence of 16 Ca2+ tightly bound to the LH1, their coordination involving putative ligands and contribution to the Qy transition are not fully understood. In study, we developed a mutagenic system to yield functional chimeric LH1-RC complexes, comprised of Tch. tepidum-derived LH1 and Rhodobacter sphaerodes-derived RC. Spectroscopic characterization of these chimeric LH1-RC complexes are presented.

2Pos243 光合成光捕集複合体における金属イオン認識の構造基盤

Structural basis for the metal-ion recognition of the bacterial core light-harvesting complex

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The structure of core light-harvesting complex (LH1) from thermophilic purple bacterium *Thermochromatium (Tch.) tepidum* has been determined (*Nature* 508, 288; 2014), in which 16 Ca-binding sites have been identified. The *Tch. tepidum* LH1 shows an enhanced thermostability and red-shifted Qy band at 915 nm compared to their counterparts. These unique properties are regulated by the Ca ions bound to LH1. The Ca ions can be replaced by other divalent metal ions, resulting in a blue-shifted LH1-Qy at ~ 890 nm with reduced thermostability. In this study, we show the crystal structures of Sr- and Ba-substituted LH1-RCs, and discuss the relationships between the structural features and the metal-induced changes in spectroscopic and thermodynamic properties.

2Pos244 NMR study of the interaction on the two ferredoxin isoforms with ferredoxin-NADP$^+$ reductase

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Ferredoxin (Fd) is reduced by Photosystem I and oxidized by Fd-NADP+ reductase (FNR). Green alga possesses six Fd isoforms, indicating that Fd-dependent pathways depend not only on PetF, which corresponds to Fd1, but also on other isoforms to be optimized for Fd partners. Among six Fds, the amino acid sequence and the crystal structure of Fd2 shows high similarity to that of Fd1. In this study, we performed NMR analysis to determine the interaction sites on Fd1 and Fd2 with FNR. They interacted with FNR through almost the same amino acid residues. We show that FNR distinguishes between Fd1 and Fd2 even in the presence of both Fds although the structures and interacting sites on Fd1 and Fd2 are similar, implying the precise molecular recognition in the side chain level.

2Pos245* 同一メナキノン分子の酸化還元電位が２種の光合成反応中心蛋白質で 500mV も異なる理由

Redox potential difference of 500 mV for menaquinones in two types of photosynthetic reaction centers

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Purple bacteria use bacterial reaction centers (bRC) and higher green plants use photosystem I (PSI) as photosynthetic reaction centers (RC). Both RC have similar electron transfer pathways, using the same quinone molecule, menaquinone. It is known that the Em values of menaquinone is -150 mV in bRC, whereas ≤ -700 mV in PSI, a remarkable difference of ~500 mV (1).

In the present study, we calculated the Em values of menaquinones using an electrostatic approach based on the crystal structures of bRC and PSI. We reproduced the difference in the Em value ~500 mV. We clarified the factor that differentiates the Em values of the menaquinones in the protein environment of the two reaction centers.

(1) Brettel and Leibl, 2001, Biochim. Biophys. Acta, 1507, 100-114

2Pos246 ヘリオバクテリア光合成反応中心の過渡吸収変化と低温蛍光解析

Analyses of transient absorption changes and low-temperature fluorescence in the photosynthetic reaction center of heliobacteria

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The reaction center of heliobacteria (hRC) is the homodimeric type 1 RC comprising two identical PshA polypeptides. We measured Xe-flash-induced absorption changes of the hRC core protein (hRCc) to reinvestigate the effects caused by moderately reducing conditions containing dithiothreitol (DTT) at alkali pH. The intensity of absorption changes immediately after excitation was somehow changed. In addition, the absorption spectrum at 77K showed the disappearance of the shoulder at 810 nm and some heterogeneity of Chl a-670. We, therefore, measured fluorescence emission spectra at 77K and found that two major peaks at 815 and 825 nm behaved differently dependent upon redox conditions. This might indicate two independent energy transfer pathways linked to the P800.

2Pos247 Magnetic structure of reduced [2Fe-2S] Rieske cluster from green sulfur bacteria Chlorobaculum tepidum studied by ESEEM

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Magnetic properties of [2Fe-2S] in Rieske protein cluster isolated from C. tepidum were investigated using EPR spectroscopy. [2Fe-2S] EPR signals were observed in the presence of dithionite. The g-values were g_x=1.81, g_y=1.90, g_z=2.03, respectively. The obtained $g_{x,y}$-values are consistent with previously observed result (only g_x=1.815, g_y=1.90), which are slightly different isolated cytochrome in other species. To investigate the detail magnetic structure, HYSCORE measurements have been performed. HYSCORE results show some magnetic couplings of N and H round 2Fe-2S center. By isotope exchange of ^{14}N /^{15}N, these signals were identified.

2Pos248 好熱性紅色光合成細菌 Thermochromatium tepidum 由来反応中心複合体におけるカルシウムイオンの機能的、構造的役割

Functional and structural roles of calcium ion in the reaction center from thermophilic purple bacterium, Thermochromatium tepidum

Michie Imanishi[1], Masayuki Kobayashi[2], Manami Kobayashi[1], Mari Matsuzaki[3], Yuki Yura[3], Takashi Ohno[3], Seiu Otomo[4], Yukihiro Kimura[3] ([1]*Faculty of Agriculture, Kobe University*, [2]*Ariake National College of Technology*, [3]*Guraduate school of Agriculture, Kobe university*, [4]*Faculty of Science, Ibaraki University*)

An X-ray crystal structure of the light-harvesting 1 reaction center (LH1-RC) complex from Thermochromatium (Tch.) tepidum at 3.0Å resolution revealed that the LH1 ring is composed of 16-subunits, each of which binds one Ca2+ to enhance thermal stability and enable the unusually red-shifted Qy absorption at 915 nm. Intriguingly, the structural information provided an evidence that the RC also binds one Ca2+ at the interface of C- and M-subunits and near the special pair. However, roles of the Ca2+ bound to the RC are largely unknown. In the present study, we investigated functional and structural roles of the RC-bound Ca2+ and discussed the physiological meanings of the Ca2+ based on the results from resonance Raman and FT-IR spectroscopic analyses.

2Pos249 Initial formation of the radical pair in reaction center complex of *Heliobacterium modesticaldum* detected by transient ESR

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Heliobacteria has the type I reaction centers (RC), composed of the complete homodimer proteins. The electron transfer in heliobacteria RC is the key for the evolution of RC for regulating quantum reaction process.
We measured the initial charge separation and recombination in *H. modesticaldum* RC core by transient electron spin resonance (ESR). The sample was initially illuminated at 210 K and cooled down to 14 K under light. Typical Electron Spin Polarization (ESP) pattern was obtained by laser flash. In the range of 50 mT, the E/A ESP signal was observed, known as P800T. Before the formation of $P800^T$, another ESP signal was detected in the range of 3 mT, assigned to the radical pair of$P800^+A_0^-$. The kinetics and electric structure of $P800^+A_0^-$state will be discussed.

2Pos250* フラビンタンパク質で目指す人工光合成

Artificial photosynthesis based on the engineered flavoprotein LOV

Nozomi Ueda, Yukiko Ono, Tatsuya Iwata, Masayo Iwaki, Hideki Kandori (*Nagoya Institute of Technology*)

Our aim is to create light-driven redox catalysts utilizing genetically modified flavoproteins. To achieve it, the LOV domain, which binds FMN, is taken as a plausible candidate. In the WT LOV, light irradiation generates a triplet excited state of FMN, followed by the adduct between FMN and the nearby Cys. Last year, we reported that in the Cys-to-Ala mutant, light-excited FMN could oxidise external electron donors, resulting in FMNH, confirmed by UV-Vis spectroscopy. In this presentation, the following reductive reaction was investigated, in which FMNH could be reoxidised by the external electron acceptors. The reoxidation kinetics of FMNH was shown to depend on the redox potential of acceptors. The applications and advantages of flavocatalysts will be discussed.

2Pos251* 分子動力学シミュレーションによる光捕集複合体の自己組織化過程に関する理論的研究

Theoretical study on the self-organization process of the light-harvesting complexes with molecular dynamics simulation

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The light-harvesting (LH) complexes such as LH1 and LH2 have high self-organization ability. It is experimentally known that the LH complexes can be reversibly dissociated into subunits (B820), and the B820 subunit can be further reversibly dissociated into its fundamental components (B777), i.e., one polypeptide and one pigment. However, the detailed structure and formation process are unknown. In this study, we investigate the detailed structure and formation process of the B820 subunits with molecular dynamics simulations. We find that the N-terminal regions of both the polypeptides contribute significantly to stabilizing the B820 subunit. This result is in good agreement with the experimental one. We also present a new method for calculating the binding free energy.

2Pos252 クリプト藻由来のカチオンチャネルロドプシンのイオン透過メカニズムの電気生理学による研究

Electrophysiological study of cation channelrhodopsins from cryptophyte algae

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Guillardia theta (G.theta) possesses 44 genes encoding putative microbial rhodopsins in their genome. Among those 18 genes retain a bacteriorhodopsin-homologous motif of which two aspartate (D85 and D96) in a close proximity of the chromophore are served as a proton acceptor and a donor of retinal Schiff base respectively. These two residues are critical for the vectorial proton transport. Recent study shows that the three gene products out of 18 genes function as light-gated cation channels but not proton pumps.
Here we present another variant of a cation channelrhodopsin from *G.theta*. We observed light sensitive channel activity, when expressed in mammalian cells. Characteristics such as ion selectivity and kinetics properties will be discussed.

2Pos253 アニオンチャネルロドプシン 2 の光開閉型 Cl^-/H^+対向輸送活性

A light-dependent Cl^-/H^+ antiport activity in anion channelrhodopsin-2

Satoko Doi[1], Takashi Tsukamoto[1], Srikanta Chowdhury[2], Susumu Yoshizawa[3], Akihiro Yamanaka[2], Yuki Sudo[1] ([1]*Grad. Sch. of Med. Dent. & Pharm. Sci., Okayama Univ.*, [2]*RIEM, Nagoya Univ.*, [3]*AORI, Univ. of Tokyo*)

Anion channelrhodopsin-2, ACR2, was recently identified from the cryptophyte algae *Guillardia theta* as a light-gated anion channel showing an extremely high neural silencing activity. By using orexin neurons, the strong neural silencing activity of ACR2 in mammalian cells was also confirmed in this study. However, the reason for the strong activity of ACR2 was still unclear. Then we developed a functional expression system for ACR2 in *Escherichia coli* cells. Utilizing that system, we estimated that ACR2 functions as a light-dependent Cl^-/H^+ antiporter (inward Cl^- and outward H^+) and that the positive charge of R84 inhibits its antiport activity. Thus we propose that the reason for the hypersensitivity of ACR2 is the cooperative role of dual ion transportation.

2Pos254 サーモフィリックロドプシンの耐熱性・高光遺伝学活性の構造基盤

Structural basis for high thermal stability and efficient optogenetic function of thermophilic rhodopsin

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Thermophilic rhodopsin (TR) is a light-driven proton pump derived from the extreme thermophile Thermus thermophilus JL-18 [1] and it showed irreversible transition from trimer to monomer upon thermal irradiation [2]. In this study, we demonstrated its high thermal stability compared with other rhodopsins and also report the potential availability for optogenetics as a neural silencer [3]. In addition, we determined its crystal structure at 2.8 Å resolution and the structural changes upon thermal irradiation were investigated by molecular dynamics simulations [3]. Their implications for high thermal stability and efficient optogenetic function would be discussed.

Tsukamoto et al., [1] (2013) JBC, 288, 21581, [2] (2014) JPCB, 118, 12383, [3] (2016) JBC, 291, 12223.

2Pos255 新規酵素ロドプシンの機能解析

Characterization of a novel enzyme rhodopsin

Kazuho Yoshida[1], Satoshi Tsunoda[1], Leonid S. Brown[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*Univ. Guelph*)

Function of microbial rhodopsins involve ion pump, ion channel, light sensor for phototaxis response and several enzymatic activities like kinase and cyclase. As these molecules are genetically targeted into various types of cells, they are widely applied as optogenetics tools for optical control of biological functions. In this poster, we introduce a novel type of rhodopsin which is tied via the C terminus to an enzyme domain. The full length gene could be expressed in mammalian cells with ubiquitously present retinal and exhibited an enzymatic reaction in light-dependent manner. Because of the high repeatability and the rapid reactivity, it would be a candidate of a new tool kit for optogenetics study.

2Pos256 ユニークな光反応を示す微生物型ロドプシンの研究

Microbial rhodopsins with unique photoreaction

Yoshitaka Kato, Keiichi Inoue, Shota Ito, Satoshi Tsunoda, Yurika Nomura, Hideki Kandori (*Grad. Sch. Eng., Nagoya Inst. Tech.*)

Microbial rhodopsins contain all-*trans* or 13-*cis*, 15-*syn* retinal in the resting state. Only the former is functional, where all-*trans* to 13-*cis*, 15-*anti* photoisomerization initiates each function, and 13-*cis*, 15-*anti* to all-*trans* thermal isomerization returns to the initial state. An exceptional case can be seen for *Anabaena* sensory rhodopsin (ASR), a photochromic sensor between all-*trans* and 13-*cis*, 15-*syn* states. In ASR, all-*trans* to 13-*cis*, 15-*anti* photoisomerization finally leads to the 13-*cis*, 15-*syn* stable photoproduct. Recently discovered inward proton pump *Po*XeR is classified into the same family of ASR, and the present flash photolysis and HPLC analysis suggest similar photoreactions between *Po*XeR and ASR.

2Pos257* マイクロデバイスと大腸菌の融合を基とした人工細胞の開発

A step towards creating life: Development of the hybrid cell based on the fusion of micron-scaled device and *E. coli*

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The author has constructed 'a hybrid cell system' *via* membrane fusion between an *E. coli* protoplast and a micron-scaled reactor 'ALBiC', of which orifice is sealed with lipid bilayer. In this work, to investigate the viability of the hybrid cell with *E. coli* cell, we introduced DNA molecules coding β-galactosidase (β-gal) into the ALBiC prior to the fusion. After fused with *E. coli* cells, 20% of the ALBiC chambers showed the significant activity of β-gal. This result verified that the central dogma in the hybrid cell is still active after fusion, suggesting the hybrid cell system retains variability. This hybrid cell system is expected to be a cellular size plat form to build new artificial cell systems.

2Pos258* 翻訳と共役した再帰的 DNA 複製システムの確立

The establishment of translation-coupled recursive DNA replication system

Yoshihiro Sakatani[1], Norikazu Ichihashi[1,2] ([1]*Grad. Sch. Info., Osaka Univ.*, [2]*Grad. Sch. Bio., Osaka Univ.*)

Various biological functions have been constituted in vitro to understand the design principle of these functions. However, genomic DNA replication system has not been constituted yet. We are attempting to construct a transcription-and translation-coupled DNA replication (TTcDR) system. We first assembled a circular DNA encoding phi29 DNA polymerase gene and a cell-free translation system. In this TTcDR system, phi29 DNA polymerase was translated and replicate the genomic circular DNA more than 10-fold in 12 h.

One of the problem of this system is recursivity; the initial template DNA is circular, whereas the product is linear. To overcome this problem, we next combined Cre recombinase with TTcDR system to circularize the product linear DNA.

2Pos259 Characterization on *Escherichia coli* L-Form

Shino Toe[1], Kazuhito V. Tabata[2,3,4], Yoshiki Moriizumi[2,3], Hiroyuki Noji[2,3] ([1]*Dept. Appl. Chem., UG. Sch. Eng, Univ. Tokyo*, [2]*Dept. Appl. Chem., Grad. Sch. Eng, Univ. Tokyo*, [3]*ImPACT, Cab. Office, Gov. Japan*, [4]*PRESTO, JST*)

Some bacteria are known to grow and undergo cell division even after cell wall depletion. This unique cell division phenomenon, called L-form attracts large attention as a model system to study how protocells may divide into daughter cells. In the present study, we characterized the colony forming efficiency (CFE) of L-form cells from E. coli on a culture plate with a cell wall synthesis inhibitor (L-form plate). We found while CFE on L-form plate was significantly low, 0.05%, that was remarkably improved to 54% after picked out from L-form plate. This finding indicates that colony forming ability on L-form plate is inherited. Further characterization on L-form will be discussed.

2Pos260 対称性の自発的破れによる遺伝子の起源
The origin of genes through spontaneous symmetry breaking

Nobuto Takeuchi (*Univ. Tokyo, Grad. Sch. of Arts and Sciences*)

The heredity of the modern cell is provided by a small number of non-catalytic template molecules, the gene. How did genes originate? Here, we demonstrate the possibility that gene-like molecules emerge in protocells through spontaneous symmetry breaking between the complementary strands of replicating molecules. The model assumes a population of protocells, each containing a population of replicating catalytic molecules. Protocells are selected towards maximizing the catalytic activity of internal molecules, whereas molecules tend to evolve towards minimizing it. These conflicting tendencies induce symmetry breaking, whereby one strand of replicating molecules completely loses catalytic activity and decreases its copy number, like genes.

2Pos261* 円偏光によるL型アミノ酸過剰生成機構の理論的探求
Theoretical investigation of the generation of L-form amino acid excess by the CPL irradiation

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Homochirality is one of the main topics that have been considered for the controversial problems in the origin of life. Since the discoveries of amino acids and their significant enantiomeric excess (ee) values in meteorites, the chiral selection of biomolecules is now considered to originate in space. Circularly polarized light (CPL) may produce the asymmetric photo-dissociation of chiral molecule that may become a trigger of the ee generation. Here, We present the typical astronomical radiation efficiently works to create the ee of amino acids in interstellar space. We perform ab-initio calculations to evaluate the CPL absorptivities of the three key amino acids found in meteorites and identify the important wavelength to generate ee in space.

2Pos262 3D ゲノム構造の集団ベースモデリング
Population-based framework of 3D genome modeling

Takeshi Sugawara (*RcMcD, Hiroshima University*)

Progress of Hi-C technologies opened the way to the identification of genome-wide chromatin interactions and 3D modeling of genome structures. However, it remains challenging to extract information about structural variability and dynamics of chromosomes from ensemble-averaged Hi-C data. Here, we show a population-based framework for dividing averaged Hi-C data into an ensemble of distinct chromosome structures. In this poster, we will discuss how such a data-driven approach can attack issues of cell-to-cell variability as well as dynamics of 3D genome structures.

2Pos263 Dynamic chromatin domains revealed by super-resolution live-cell imaging

Tadasu Nozaki[1], Sachiko Tamura[1], Ryosuke Imai[1], Tomomi Tani[2], Masaru Tomita[3], Takeharu Nagai[4], Yasushi Okada[5], Kazuhiro Maeshima[1] ([1]*Natl. Inst. Genet.*, [2]*MBL*, [3]*Inst. Adv. Biosci., Keio Univ.*, [4]*ISIR, Osaka Univ.*, [5]*QBiC, RIKEN*)

Recent studies have suggested that chromatin forms numerous domains as functional units of the genome. However, questions remain how they form and behave in living cells. Here, by combining super-resolution imaging and single nucleosome tracking, we developed a novel nuclear imaging system that allowed us to visualize the spatial organization of chromatin domains along with their dynamics in living mammalian cells. We have clearly demonstrated the quantitative relations between the epigenetic state and dynamics: more heterochromatic regions show less movement. Furthermore, we observed the chromatin domains during mitosis, suggesting that they act as “Lego blocks” of chromosomes to retain epigenetic information throughout the cell cycle.

2Pos264 分裂酵母クロマチン動態の網羅的解析
Comprehensive Analysis of Chromatin Dynamics in Fission Yeast

Toshinori Namba[1], Sayaka Suzuki[2], Takeshi Sugawara[1], Da-Qiao Ding[3], Yasushi Hiraoka[4], Yuichi Togashi[1], Masaru Ueno[5], Shin-ichi Tate[1] ([1]*RcMcD, Hiroshima Univ.*, [2]*Dept. of Math. and Life Sci., Hiroshima Univ.*, [3]*Adv. ICT Res. Inst., NICT*, [4]*Dept. of Biol. Sci., Osaka Univ.*, [5]*Grad. Sch. of Adv. Sci. of Matter, Hiroshima Univ.*)

One of the central challenges in biology is to reveal the relationship between genome structure and functions such as transcription and replication. Recent progress of Hi-C technologies enabled the detection of genome-wide chromatin interactions and the 3D organization of chromosomes. However, Hi-C cannot evaluate structural fluctuations; chromatin loci diffuse around the nucleus and chromosomes show structural variability. In order to examine genome-wide chromatin structure and dynamics, we tracked chromatin loci of the fission yeast S. pombe during the interphase, and analyzed time-lapse data of the 3D coordinates throughout the genome. In this poster, we will discuss how our quantitative analysis can provide a clue to understand chromosome structure and dynamics.

2Pos265 長さの異なる塩基配列組み合わせの頻度・分布を用いたヌクレオソーム配置推定
Predict nucleosome positioning by incorporating the frequencies and distributions of three length-different nucleotide segments

Akinori Awazu[1,2] ([1]*Dept. of Math. and Life Sciences, Hiroshima Univ.*, [2]*RcMcD , Hiroshima Univ.*)

Nucleosome plays important roles in transcription and chromatin structure formation in Eukaryotes. The studies of nucleosome positioning rules provide a deeper understanding of these processes. Nucleosome positioning prediction was performed using a model consists of following variables characterizing a DNA sequence: the number of 5-nucleotide sequences, the number of 3-nucleotide combinations in 1 period of a helix, and mono- and di-nucleotide distributions in DNA fragments. Using benchmark datasets for yeast, human, C. elegans, and D. melanogaster, the present model was shown to have a better prediction performance than recent predictors. This model might also display the common and organism-dependent factors that affect the nucleosome forming abilities of sequences.

2Pos266 New rules of protein structures

Shunsuke Nishiyama[1], Shintaro Minami[2], **George Chikenji**[1] ([1]*Dept. of Comp. Sci. & Eng., Nagoya Univ.*, [2]*Grad. Sch. of Inf. Sci., Nagoya Univ.*)

It is widely accepted that the set of observed protein topologies is highly limited. These limitations have been explained as the consequence of several rules, such as a strong preference for right handedness of beta-alpha-beta connections and prohibiting loop crossing. These rules, however, are insufficient for explaining highly skewed frequency distribution of the observed beta sheet topologies in the protein structure database. Here, we report some new rules for limiting protein topology.We show that these new rules are usefull for understanding the distribution of the observed beta sheet topology.

2Pos267 全原子 Motion Tree による構造変化の解析
Description of protein structural changes by full-atom Motion Tree

Ryotaro Koike (*Grad. Sch. of Info. Sci., Nagoya Univ.*)

The PDB provides multiple structures of a protein under different conditions, e.g. ligand-bound and ligand-free states. The comparison of structures clarifies the structural change and the molecular mechanism upon ligand binding. We developed the tool to compare structures using Ca atom coordinates and to describe structural changes in a tree diagram. The diagram, named ‘Motion Tree’, illustrates various protein motions, from a small loop motion to a large domain motion. In this study, I have extended the Motion Tree to compare structures using all atoms except for hydrogen. The full-atom Motion Tree was applied to a set of structure pairs. A number of motions were newly identified such as movements of side-chain atoms, but the calculation is naturally slow.

2Pos268 分子動力学法を用いた、ポリグルタミン酸のアンフォールドダイナミクス
Unfolding dynamics of poly-glutamic acid in using molecular dynamics method

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Protein function is much related to its structure, and hence to understand process of protein structural changes is of paramount importance. In order to achieve this purpose, dynamics of poly-glutamic acid (PGA) has been studied from both experiments and theoretical approaches. The two unfolding mechanism of a helical PGA have been proposed: (i) the helix is broken from the terminal and (ii) the helix bends from center. However, the atomic details are not well understood. In this study, we performed all-atom molecular dynamics (MD) simulations during 7.5μs in total. As a result, several unfolding processes of the PGA in both (i) and (ii) mechanisms were observed. This means the unfolding process of PGA undergoes in multiple ways rather than a single specific mechanism.

2Pos269 MEGADOCK-Azure: Microsoft Azure クラウド環境での並列タンパク質間相互作用予測計算
MEGADOCK-Azure: High-performance protein-protein interaction predictions on Microsoft Azure HPC

Masahito Ohue[1], Yuki Yamamoto[1,2], Hiroyuki Sato[3], Takashi Matsushita[3], Yutaka Akiyama[1,2] ([1]*Sch. of Computing, Tokyo Tech.*, [2]*ACLS, Tokyo Tech.*, [3]*IMSBIO Co., Ltd.*)

Cloud computing environment, such as Amazon AWS, Microsoft Azure, Google Cloud Platform, etc., achieves performance improvement remarkably in recent years, and is also useful in parallel computing (high-performance computing, HPC) fields. Cloud is able to use thousands of CPU cores and GPU accelerators casually, and several software are used very easy by cloud images. We have transplanted an original protein-protein interaction prediction (protein-protein docking) software, MEGADOCK, to the Microsoft Azure HPC environment. We have obtained strong scaling value of 84% with virtual machines of Azure A9 instance.

2Pos270 非エバルト静電ポテンシャル計算法“零多重極子和法”の開発と検証
Development and Evaluations of a Fast and Accurate Non-Ewald Electrostatic Potential Scheme, the Zero-Multipole Summation Method

Kota Kasahara[1], Shun Sakuraba[2], Ikuo Fukuda[3], Jinzen Ikebe[4], Ryuhei Harada[5] ([1]*Col. Life Sci., Ritsumeikan Univ.*, [2]*Grad. Sch. Frontier Sci., Univ. Tokyo*, [3]*IPR, Osaka Univ.*, [4]*QST, MMS*, [5]*CCS, Univ. Tsukuba*)

In molecular dynamics (MD) simulations, development of fast and accurate non-Ewald methods is required. We have developed a non-Ewald method, named “zero-multipole summation method” (ZMM). Here, we performed all-atom replica exchange MD (REMD) simulations with the two systems (poly-alanine 8mer and Trp-cage) to elucidate the nature of the conformational ensembles generated by the ZMM. As a result, there were no significant differences between the conformational ensembles of poly-alanine generated by the PME and the ZMM, and the ZMM was ca. 1.5-fold faster than the PME. On the other hand, although the energy landscapes of Trp-cage calculated by the PME and the ZMM were well agreed qualitatively, ZMM tends to stabilize the native conformation relative to the PME.

2Pos271 Local structures around protein phosphorylation sites

Hafumi Nishi, Kengo Kinoshita (*Grad. Sch. Info. Sci., Tohoku Univ.*)

Protein phosphorylation plays a crucial role in almost all cellular signaling. As phosphorylation sites should be accessible and recognized by kinases, the local structures of phosphorylation sites might have specific patterns as compared to non-phosphorylation sites. Here we explored possible local structures of phosphorylation sites by mapping phosphorylation sites onto protein 3D structures. In the dataset, about 14% of the sites were found in the middle of alpha helices or beta strands, which were more than six residues long. Analyses of corresponding kinases indicated that several phosphorylation motifs may be able to be present within alpha helices, whereas others may prefer to be disordered due to the amino acid composition of phosphorylation motifs.

2Pos272　膜タンパク質の構造分類：93 フォールドの同定
We found at least 93 membrane protein folds in structure classification

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Fold recognition (FR) method has been contributing to predict folds for soluble proteins, while the shortage of structure numbers had prevented us from improving FR methods for membrane proteins (MPs). However, resent increase numbers of the MPs structure help us to propose a comprehensive classification of MP folds.
We performed "all-against-all" sequence/structure alignment for known MP structures. In the dispersion diagram between sequence similarity (%) and structure similarity (RMSD: Å) we obtained the area (>30% and <4.5Å) of the same folds and using these thresholds we classified 234 MPs to 93 fold structures. Based on these datasets we developed a new FR strategy specialized for MP sequences by using physicochemical parameters assigned on transmembrane helices.

2Pos273　Culture-independent identification of genes encoding agarase from environmental bacteria using agarose gel microdroplets

Eiji Shigihara[1], Ryo Iizuka[1], Takashi Sakurai[1], Yuji Hatada[2], Dong Hyun Yoon[3], Tetsushi Sekiguchi[4], Shuichi Shoji[3], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., Univ. Tokyo.*, [2]*Dept. of Life Sci. and Green Chem., Saitama Inst. of Technol.*, [3]*Dept. of Nanosci. and Nanoeng., Waseda Univ.*, [4]*Res. Org. for Nano&Life Innov., Waseda Univ.*)

Environmental microbes are a great source of industrially valuable enzymes with potent and unique catalytic activities. However, the majority of microbes remain unculturable and thus are not accessible by culture-based methods. Recently, we envisioned a culture-independent method for identifying microbial enzyme-encoding genes using water-in-oil microdroplets (Nakamura et al.(2016) Sci.Rep.6,22259). Now, we attempt to obtain agarase-encoding genes from environmental bacteria using agarose gel microdroplets. In the meeting, details of the system will be discussed.

2Pos274　人工平面脂質二重膜を用いた抗菌性ペプチドの分子進化研究
Molecular evolution of antimicrobial peptides using artificial planar lipid bilayers

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Living system has changed their characteristics evolutionary to give rise to biodiversity including the antimicrobial peptides (AMPs). AMPs play a role of biological defense mechanisms, and induce the bacterial cell lysis by pore formation in the cell membranes. We have studied the molecular mechanisms of the pore formation of several AMPs using planar lipid bilayers. Here, we observed channel current of three different AMPs from three different species in an evolutionary tree: ascidiacea (urochordata), frog (amphibia) and human (mammalian). As the results of pore forming properties using bacterial model membrane, we found that the AMPs pores become larger with following the evolution of the species due to increase the hydrophilic region in the structure.

2Pos275　タンパク質コーパスによる分散表現：ランダム配列の意味空間マッピングによる偽タンパク質の探索
Distributed representation analysis of a protein corpus: Can we identify fake proteins by mapping random sequences on a semantic space?

Hiroshi Imamura, Shinya Honda (*AIST*)

According to the distributional hypothesis in linguistics, the semantics of a word is determined by its context. We prepared a protein corpus consisted of n-gram of natural protein sequences. Reducing the dimension of a co-occurrence matrix of the n-gram gave distributed representation of the n-gram, i.e., the vectors that represent "semantics" therein. We, for an experiment, represented natural and artificial random sequences as the sum of the n-gram's vectors. The natural- and random-derived vectors were discriminated on a semantic space, which was depicted in terms of a 2D projection map. Machine learning was used to classify random sequences into natural or random. We identified a few percent of random sequence as natural, fake protein.

2Pos276　蛋白質構造安定性の平衡淘汰
Selection maintaining protein stability at equilibrium

Sanzo Miyazawa

Recently it was indicated that fitness costs due to misfolded proteins are a determinant of evolutionary rate and selection originating in protein stability is a driving force of protein evolution. Here we examine the effects of protein abundance/indispensability and structural constraint on substitution rate under the selection maintaining protein stability. The probability distribution of nonsynonymous to synonymous substitution rate per site over fixed mutants indicates that nearly neutral selection is predominant only in low-abundant, non-essential proteins, and in the other proteins, positive selection on stabilizing mutations is significant to maintain protein stability at equilibrium as well as random drift on slightly negative mutations.

2Pos277*　原子間力顕微鏡による初期発生胚の弾性率のタイムラプスイメージング
Time-lapse imaging of elastic modulus of ascidian embryo during early development by atomic force microscopy

Yuki Fujii[1], Wataru Koizumi[2], Taichi Imai[2], Kohji Hotta[2], Kotaro Oka[2], Takaharu Okajima[1] ([1]*Grad. Schl. Inform. Sci. and Tech. Hokkaido Univ.*, [2]*Grad. Schl. Biosci. and Bioinfo. Keio Univ.*)

The process of cell division and cellular differentiation of ascidian embryo is highly organized during the developmental stage. Atomic force microscopy (AFM) revealed that the elastic modulus, E, of cells in embryo was oscillated during a developmental stage [1]. However, it is less known about the universal features of single cell mechanics in developmental process. In this study, we succeeded in capturing time-lapse E images of embryonic cells in the developmental periods from the fertilization to the morula stages by AFM. We found that the spatial-temporal behaviors of E were much different between animal and vegetal poles, and characteristic heterogeneous structures of E emerged in the vegetal pole.
[1] Y. fujii, et al. The 53th Annual Meeting of the BSJ. (2015)

2Pos278 信号処理蛋白質 Raf の生細胞内 ALEX 計測
In-cell ALEX measurement of cytosolic signaling protein Raf

Kenji Okamoto[1], Kayo Hibino[2], Yasushi Sako[1] ([1]*RIKEN*, [2]*NIG*)

We have investigated the structural distribution of cytosolic signaling protein Raf in living cells by using single-molecule FRET (smFRET) measurement based on fluorescence burst detection. One of problems in smFRET measurement is photobleached acceptor dyes that imitate zero-FRET population in FRET distribution. We introduced the alternative laser excitation (ALEX), for which donor and acceptor excitation light are switched alternatively at high repetition rate (10kHz in our setup), and have succeeded to obtain smFRET distribution in a living cell while the molecules with photobleached dyes are excluded. The system is applied to investigate the intracellular structural distribution of Raf and its change upon EGF stimulation of the cell.

2Pos279 細胞内環境におけるアンチセンス分子自己相補形成の mRNA に対する親和性への寄与
Contribution of self-complementarity of antisense molecule to the affinity for mRNA in intracellular environment

Shunsuke Takeda[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*PRESTO, JST*)

Investigating gene-specific and dynamic behaviors of mRNA is important in understanding cell functions. To reveal those mRNA behaviors in living cells, we adopted fluorescence imaging of endogenous mRNA using antisense oligonucleotide probes. However, the problem is that there is no methodology for obtaining probes with high binding rate. Here, we investigated the relationship between probe sequences and their affinities to mRNA in living cells. As a result, we found some probes with self-complementarity showed high affinity. This indicates the effect of entropy change might be more predominant than that of enthalpy change in cells. We speculate this might be caused by the heterogeneity of intracellular environment, such as inhomogeneous temperature distribution.

2Pos280 顕微ラマン分光法によるバクテリア一細胞の代謝活性測定
Measuring metabolic activities in single bacterial cells by Raman microspectroscopy

Yota Kato[1], Hiroshi Ueno[1], Hiroyuki Noji[1,2] ([1]*Grad. Sch. Eng., Univ. Tokyo*, [2]*ImPACT, JST*)

The analysis of the individuality of the bacterial metabolic activity is essential for the understanding of their adaptation to environmental conditions, such as a persister cell phenomenon. We measured the metabolic activities in single *E. coli* cells by using D_2O labeling and Raman microspectroscopy. Cultured in a medium containing D_2O, active cells incorporate the deuterium and generate C-D bonds, mainly in lipids and proteins, through the metabolic pathways. The C-D bonds produce a detectable peak in Raman spectra. After incubation with D_2O, the dead cells treated with antibiotics showed lower C-D peak intensities than the untreated cells. This result suggests the C-D peak can work as an indicator of the metabolic activity of individual cells.

2Pos281 上皮細胞シートの頂端膜揺らぎ：走査型イオンコンダクタンス顕微鏡
Scanning ion conductance microscopy (SICM) measurement of apical membrane fluctuation in epithelial cell monolayer

Kenta Aoki[1], Ryosuke Tanaka[1], Cho Nam-Joon[2], Takaharu Okajima[1] ([1]*Grad. Schl. Inform. Sci. and Technol., Hokkaido Univ.*, [2]*NanyangTechnol Univ.*)

Epithelial cell monolayer is a basic tissue that is usually observed in organs. Each epithelial cell tightly interacts the neighboring cells via cell-cell bonds, and the apical membrane is an important role in mediating cell signaling. The dynamics of the apical membrane has been measured in the intracellular regions [1]. However, little is known about how the apical membrane is fluctuated in the intercellular regions, that is, at the cell-cell boundary. Here, we investigated the detailed spatial distribution of cell membrane fluctuations by scanning ion conductance microscopy (SICM) and observed that membrane fluctuations of epithelial cells are heterogeneous over a wide intercellular region. [1] Mizutani et al. Appl. Phys. Lett. 102, 173703(2013).

2Pos282* 蛍光偏光相関分光法により明らかになった生細胞内での分子混雑と回転拡散の関係
The relationship between rotational diffusion and crowding in living cell revealed by polarized fluorescence correlation spectroscopy

Makoto Oura[1], Johtaro Yamamoto[2], Takahiro Matsuda[1], Jian Ping Gong[2], Masataka Kinjo[2] ([1]*Hokkaido Univ. Grad. Sch. Life Sci.*, [2]*Hokkaido Univ. Fac. Adv. Life Sci.*)

Rotational diffusion in cells is an important process for molecular interaction in a tiny volume, because molecules randomly rotate in nm or sub-nm scale, it corresponds to the size of biomolecules and also environment. To evaluate the rotational diffusion, we combined fluorescence correlation spectroscopy and polarized optical system (pol-FCS). This system can detect the fluctuation of fluorescent signal in nano-sec range. Through the measurement using several kinds of cell and gel, the relationship of molecular translational and rotational diffusion was clarified in cytosol of living cells. We assume that this technique can be applied to investigate molecular nano-dynamic properties in cells, such as molecular crowding effect.

2Pos283 細胞イメージングシステムを用いたナノバイオプローブの生体適合性評価
Evaluation of　biocompatibility of nano-bio probes by using Cell imaging system

Yuko Nakane[1,2], Takashi Jin[2] ([1]*Tomy Digital Biology Co., Ltd.*, [2]*RIKEN QBiC*)

Near infrared fluorescence at 1000- 1400 nm is expected for the non-invasive in vivo imaging. There are some nano-bio probes for the 2nd NIR fluorescence imaging including our developed quantum dots, but little is known about their biocompatibility.

We examined the cell proliferation test to evaluate the biocompatibility of the 2nd NIR probes by using an imaging cell cytometer. Single walled carbon nano tube, rare-earth nano-particles and quantum dots were used as the probes. HeLa cells were plated in 96-well plates. After culturing the cells overnight, the probes were added to each well at different concentration. Then the number of cells in each well was counted after 0 , 7, 24, 48 and 60 hrs and the effect of each probe on the biocompatibility was evaluated.

2Pos284 金ナノ粒子を用いた加熱による細胞内局所温度の制御

Manipulating the local temperature in a single cell with gold nanoparticles

Takaaki Honda[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharma., Univ. Tokyo*, [2]*JST PRESTO*)

Recent study has shown that intracellular temperature is heterogeneous, and suggested that local temperature might regulate various biochemical reactions in a cell. Here, we aimed to develop the method to control the intracellular local temperature for investigation of cell functions. We heated a single cell by gold nanoparticles (GNPs) which efficiently convert light to heat, and measured the intracellular temperature by fluorescent polymeric thermometer. Irradiation of GNPs which were attached to the cell with a laser showed an increase of the intracellular average temperature by about 10℃. Moreover, we succeeded in inducing a cell response by this method. These results indicate that controlling the intracellular local temperature can manipulate cell functions.

2Pos285 人工細胞開発に向けた細胞内 ATP 濃度の定量計測系開発

Development of Quantitative ATP Concentration Measurement Method in Single Cells for Artificial Cell System

Hiroki Ashikawa[1], Kazuhito V. Tabata[1,2,3], Hiromi Imamura[4], Rikiya Watanabe[1,3], Hideyuki Yaginuma[5], Hiroyuki Noji[1,2] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Toyko*, [2]*ImPACT, CAO, Govt. Japan*, [3]*PRESTO, JST*, [4]*Grad. Sch. Bio., Univ. Kyoto*, [5]*QBiC, RIKEN*)

We have been trying to develop artificial cell system by fusing a protoplast of E.coli to a micro chamber sealed with lipid bilayer. Although we have succeeded in observing the fusion, there is no established method to measure the energy state of the hybrid cells quantitatively. In this study, we developed methods of observing the activity in the hybrid cells by measuring ATP concentration ([ATP]) with Queen (ratiometric fluorescent ATP indicator). We encapsulated Queen to the micro chambers in advance, and fused protoplasts to the chambers. As a result, we have been able to measure [ATP] of the hybrid cells. We report the results about [ATP] changes of hybrid cells, and discuss better conditions which hybrid cells maintain the ability to synthesize ATP.

2Pos286 バイオセンサシステムのための水素化アモルファスシリコン薄膜上のアミノ酸含有ゲルの電圧電流特性解析および蛍光性分子薄膜に関する研究

Voltage current property of amino acid containing hydrogel and molecular film on hydrogenated amorphous silicon film for biosensor system

Makoto Horigane[1], Shotaro Minato[1], Hiroshi Masumoto[2], Takashi Goto[3], **Yutaka Tsujiuchi**[1] ([1]*Mat. Sci. & Eng., Akita Univ.*, [2]*Front. Res. Inst., Tohoku Univ.*, [3]*Inst. Mat. Res., Tohoku Univ.*)

On an attempt for fabrication of biosensor using photo-controlled film system, we have been researching, by using ionic conduction in laminated gels on hydrogenated amorphous silicon film. In this study, voltage current analysis of amino acid containing hydrogel photo-controlled on hydrogenated amorphous silicon film, are done using cottrell equation system, using several amino acids that are the elementally elements of bio molecule and has potential of diversity to electro chemical device. Furthermore, fluorescent molecular film photo-controlled on hydrogenated amorphous silicon film was fabricated and analyzed its property for constructing biosensor system. Multi-dimensional analysis method is to be contributed for a high presision biosensor system.

2Pos287 オンチップ１細胞計測系によるマクロファージの複数貪食の制御解析

Studies on regulation mechanism of multiple phagocytosis of macrophage by single cell on-chip measurement assay

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For understanding regulation of simultaneous polarity of phagocytosis on a surface of single macrophage, we examined the correlation of phagocytosis and feeding time of a plurality of Zymosans on single macrophage and observed. Zymosans were guided to macrophage by optical tweezers and were contacted physically on the surface of different positions of a macrophage simultaneously or with intervals. Multiple phagocytosis did not occur simultaneously when we attached Zymosans to macrophage with more than 6 s intervals, whereas simultaneous phagocytosis occurred in less than 6 s intervals. The results indicate that phagocytosis is not independent local phenomenon on cell membrane, and that existence of internal unit time of cell recognition, e.g., 6 s.

2Pos288 Development of Novel Scanning Microscope for Measurement of Emission and Excitation Spectra Simultaneously

Sankar Jana, Yutaka Shibata (*Tohoku University*)

We have developed a novel scanning microscope equipped with a function to measure emission, excitation spectra and corresponding images at the same time. White light continuum generated by a fs laser-pumped photonic-crystal fiber is dispersed by a prism and line focused on a sample. The excitation wavelength depends on the position along the line focus. The emissions excited by different wavelengths are collected at different heights of the polychrometer entrance slit and detected as a 2D image on EMCCD, where vertical and horizontal axes correspond to the excitation and emission wavelengths, respectively. 3D imaging is achieved with a combination of a 2D piezo stage and a supersonic motor. We will present fluorescence images of plant specimen with the developed system.

2Pos289 高速 AFM によるアクチン様細胞骨格タンパク質 MamK 繊維の直接観察

Direct observation of actin-like MamK cytoskeletal filaments by high-speed AFM

Yousuke Kikuchi[1], Marina Inagawa[2], Zachery Oestreicher[1], Azuma Taoka[1,3], Yoshihiro Fukumori[1] ([1]*Sch. of Nat. Sys., Col. of Sci. and Eng., Kanazawa Univ.*, [2]*Grad. Sch. of Nat. Sci. and Tech., Kanazawa Univ.*, [3]*Bio-AFM Center, Col. of Sci. and Eng., Kanazawa Univ.*)

Magnetospirillum magneticum AMB-1 has intracellular membranous organelles called magnetosomes that provide them the capability of sensing the geomagnetic field. MamK cytoskeletal filaments are associated with the magnetosomes, and are thought to act as a scaffold to keep positioning of the magnetosomes. This study aim to characterize the nature of the MamK cytoskeletal filaments allowing magnetosomes in chain-like arrangement. To characterize dynamic nature of the MamK filament, we attempted to visualize the MamK polymerization by using high-speed AFM. However, the purified MamK formed aggregate or oligomer that prevented AFM observation. Therefore, we established the purification method to obtain monomeric MamK, then we imaged polymerization process of MamK filaments.

2Pos290 高速 AFM による古細菌 *S. solfataricus* 由来ミニ染色体維持 (ssoMCM)タンパク質複合体の観察

Observation of *S. solfataricus* archaeal minichromosome maintenance (ssoMCM) protein complex by high-speed AFM

Daisuke Noshiro[1], Noriyuki Kodera[1,2], Toshio Ando[1,3] ([1]*Bio-AFM FRC, Inst. of Sci. & Eng., Kanazawa Univ.*, [2]*PRESTO, JST*, [3]*CREST, JST*)

Minichromosome maintenance (MCM) proteins are members of the AAA+ superfamily that contain a highly conserved ATPase module, and play an essential role in DNA replication initiation as a hexameric DNA helicase. The archaeal MCM protein from *S. solfataricus* (ssoMCM) is known to form a homohexamer and has been studied as a simple model system for understanding how MCM proteins unwind dsDNA coupled with ATP hydrolysis. In this study, we used high-speed AFM to visualize the formation of oligomers of ssoMCM protein. We observed that the protein forms ladder-like structures with varying sizes as well as ring or partial ring structures. These structures are not of the well-known hexameric complex, suggesting that ssoMCM protein can form variants of complexes.

2Pos291 ストレプトリジン O による膜孔形成の高速 AFM 観察

High-speed AFM Observation of Membrane Pore Formation by Streptolysin O

Hirotaka Ariyama[1], Noriyuki Kodera[1], Toshio Ando[1,2] ([1]*Bio-AFM Frontier Research Center, Kanazawa Univ.*, [2]*Dept. Phys., Kanazawa Univ.*)

Streptolysin O (SLO) is one of pathogenic, membrane pore-forming proteins and is produced by group A streptococci, whose infection causes scarlet fever, rheumatic fever, and pharyngitis. SLO monomers assemble on membranes to form membrane pores of 20-30 nm in diameter. In this study, we used high-speed AFM (HS-AFM) to visualize the molecular process of pore formation by SLO at submolecular spatial and sub-100 ms time resolution. The HS-AFM images showed the formation of SLO oligomers such as arc-shaped and ring-shaped ones on the membrane containing 60% Cholesterol. Oligomers in the pore state were observed to fuse to form a ring within 300 ms. In the prepore-to-pore state transition, the height reduction was propagated over the length of oligomers.

2Pos292 ストレス顆粒内内在性 mRNA のナノスケール蛍光イメージング

Nanoscale Fluorescence Imaging of Endogenous mRNAs in Stress Granules

Ko Sugawara[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*JST, PRESTO*)

Stress granules (SGs) are cytoplasmic mRNA granules formed under stress conditions. Although it is known that SGs are responsible for the dynamic translation regulations in stressed cells, the detailed mechanism by which mRNAs are spatiotemporally organized in SGs remains unclear. Here, we investigated the nanoscale localization and dynamics of endogenous mRNAs in SGs by single-molecule localization microscopy and single particle tracking combined with the antisense oligonucleotide probes. We observed that endogenous mRNAs form high-density domains with a diameter of ~70 nm in SGs and found that more than half of mRNAs are stationary inside SGs. Our results suggest that SGs involve the mRNA anchoring domains that would be key sites for the dynamic stress responses.

2Pos293 人知を超える超高速・高精度蛍光形態サイトメトリー

Ghost Cytometry: fluorescence "imaging" cytometry beyond human's limit

Sadao Ota[1,2], Hiroyuki Noji[1,3] ([1]*Sch. Eng., Univ. Tokyo*, [2]*JST, PRESTO*, [3]*JST, ImPACT*)

In this talk, I will report development of next-generation "imaging" flow cytometry that is high throughput (>> 10,000 cells/sec) and accurate beyond humans's limit. The invention integrates a novel ultrafast and highly sensitive imager and machine learning. By fully utilizing the power of huge data rapidly generated by a statistical number of single cells, our invention takes a field of high content cytometry to the next stage.

2Pos294 線形ゼロモード導波路を用いたアクチン重合メカニズムの 1 分子解析

Single molecule observation of actin polymerization using linear zero-mode waveguides

Soichiro Fujii[1], Ryo Iizuka[1], Masamichi Yamamoto[1], Makoto Tsunoda[1], Takashi Tanii[2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*Fac. Sci. Eng., Waseda Univ.*)

Actin is a ubiquitous cytoskeletal protein, which is essential for the structure and function of eukaryotic cells. Actin polymerization occurs through three phases termed nucleation, elongation, and steady-state phases. A previous study showed that the ends of actin filaments grow and shorten more rapidly than would be predicted from measured rate constants for monomer association and dissociation. In this study, we observed actin polymerization using single-molecule fluorescence imaging with linear zero-mode waveguides to examine the oligomeric states of actin incorporated into filament ends. We found that monomer associates with filament ends in elongation phase, whereas small oligomers (dimer to trimmer) are also added to filaments in steady-state phase.

2Pos295 T 細胞活性化における微小管動態の超解像解析

Super-resolution analysis of microtubule dynamics on T cell activation

Hengyu Shi, Yuma Ito, Wei Ming Lim, Kumiko Sakata-Sogawa, Makio Tokunaga (*Sch. Life Sci. Tech., Tokyo Inst. Tech.*)

Elucidation nanometer-scale structures and dynamics of molecules and complexes in living cells is a subject of increasing interest in the life sciences. Thanks to the invention of super-resolution microscopy, the limitations of diffraction-limited optical microscopy, about half of the wavelength, are becoming clear. During response of T cell activation, the microtubule-organizing center (MTOC) is known to be translocated toward the center of the immunological synapse. However, functional relationships between MTOC translocation and microtubule dynamics remain elusive. We visualized microtubules with fluorescently labeled MAP4 at a few tens of nanometer resolution using PALM. We will discuss structural changes in activation of T cells.

2Pos296 自発的光スイッチング蛍光タンパク質による簡便超解像イメージング

Simple and easy way for superresolution imaging by spontaneously switching-on fluorescent protein

Yoshiyuki Arai, Hiroki Takauchi, Takeharu Nagai (*ISIR, Osaka Univ.*)

Photoswitchable fluorescent proteins (PSFPs) play an important role for superresolution imaging such as single molecule localization microscopy (SMLM). For SMLM, optimal timing control of instruments is required to acquire data. However, such controls complicate the microscopy system. Here, we developed new PSFP, named SSPF, from Dreikalng that is categorized as a decoupled type among PSPFs. SSFP can be spontaneously recovered from off-state to on-state. The rate constant of the switching-on is so quick that we could apply this phenomenon for SMLM. Once SSFP is switched off, what we should do is only stream acquisition with continuous illumination of excitation light. Our system enables to lower the hurdle for researchers who want to utilize superresolution techniques.

2Pos297* 細胞内グルタチオンの求核付加・解離平衡に基づく超解像蛍光イメージングプローブの開発

Development of spontaneously blinking fluorophores based on nucleophilic addition of intracellular glutathione for superresolution imaging

Akihico Morozumi[1,4], Mako Kamiya[2,5], Shinnosuke Uno[1], Keitaro Umezawa[1], Toshitada Yoshihara[3], Seiji Tobita[3], Yasuteru Urano[1,2,4] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Grad. Sch. of Med., The Univ. of Tokyo*, [3]*Grad. Sch. Sci. Tech., Gunma Univ.*, [4]*AMED CREST*, [5]*JST PRESTO*)

Single-molecule localization microscopy (SMLM) provides super-resolution images by repeated precise localization of individual fluorophores. However, in order to induce blinking of conventional fluorophores, intense laser irradiation and additives are usually required, limiting the applications to live cells under physiological conditions. Here, we propose a novel mechanism of blinking based on nucleophilic addition and dissociation of intracellular glutathione (GSH). Based on this strategy, we succeeded in developing two fluorophores, with which two-color SMLM has been achieved in fixed cells at physiological concentrations of GSH without intense laser irradiation. Further, we recently succeeded in live-cell SMLM without any additives by using one of the fluorophores.

2Pos298* 自由行動マウスの脳活動計測を可能にする化学発光膜電位センサーの開発

Development of a chemiluminescent voltage indicator applicable to brain activity recording in freely moving mice

Shigenori Inagaki[1], Masakazu Agetsuma[2], Hidekazu Tsutsui[3,4], Yoshiyuki Arai[2], Kazushi Suzuki[5], Yuka Jinno[4], Yasushi Okamura[1,4], Tomoki Matsuda[2], Takeharu Nagai[1,2,5] ([1]*FBS., Univ. Osaka*, [2]*ISIR., Univ. Osaka*, [3]*Sch. Mat. Sci., JAIST*, [4]*Grad. Sch. Med., Univ. Osaka*, [5]*Dep of Biotech, Univ. Osaka*)

Fluorescence imaging using genetically encoded voltage indicators (GEVIs) are practically useful for decoding electrical information from defined cell populations. However, its complex optics regarding fluorescence excitation sometimes hinder the use of GEVIs.
Here, we present a world's first chemiluminescent GEVI named LOTUS-V applicable to high-speed neural activity recording (approx. 100 Hz) in freely moving mice. Since the excitation light is absent, LOTUS-V allows the investigation of mouse cerebral cortex during locomotion without the need for complex optics and insertion of an optical fiber. Thus, LOTUS-V opens a door to an easy and minimally invasive method for brain activity imaging that is applicable to a wide range of neuroscience research.

2Pos299 自動１分子イメージング装置の開発

Development of automatic single molecular imaging system

Masato Yasui[1], Jun Kozuka[1], Michio Hiroshima[1], Taku Tsuzuki[2], Yasushi Sao[3] ([1]*RIKEN QBiC*, [2]*Osaka University*, [3]*RIKEN Cellular Informatics Laboratory*)

We have developed the automatic single molecule imaging system for imaging based drug screening. By using our developed focusing technique, our microscope can automatically focus on the glass surface. Because our microscope mounts deep learning, if we teach the appropriate cells for the observation to the microscope, the cells in 96 well is automatically found. Multi-color single molecule imaging is possible. By using dispenser, we can add the drug, and check the effect of drug from the single molecule imaging data. Automatic analysis system is included, which shows the basic statistics and the diffusion states of single molecules on the membrane. Our microscope will change the way of research and drug screening.

2Pos300 Fluorescence recovery after photobleaching (FRAP) analysis of INO80 chromatin remodeling complex

Tsubasa Isogaki[1], Yuma Ito[1], Shota Ichikawa[1], Hiroshi Kimura[2], Masahiko Harata[3], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*Inst. Innov. Res., Tokyo Inst. Tech.*, [3]*Grad. Agr. Sci., Tohoku Univ.*)

The INO80 chromatin remodeling complex has important roles in transcription, DNA replication and repair by regulating chromatin structure. INO80 contains actin-related proteins Arp4, Arp5 and Arp8. However, the roles of Arps in the mechanisms of chromatin regulation are not fully understood. Fluorescence recovery after photobleaching (FRAP) analysis of Ino80, a scaffold subunit of the INO80 complex, and Arps in the nucleus showed different FRAP curves and suggested the dynamic formation of the INO80 complex. To investigate the interaction mechanism of Arps, we performed FRAP analysis of Ino80 under the condition of knocking down of Arp4, Arp5, or Arp8. We will discuss the dynamic mechanisms of Arps for chromatin remodeling.

2Pos301* The Correspondence between Raman Microspectroscopy and Omics Data

Koseki Kobayashi-Kirschvink[1], Hidenori Nakaoka[1], Arisa Oda[2], Kunihiro Ohta[2,3], Yuichi Wakamoto[1] ([1]*Dep. Bas. Sci., Univ. Tokyo*, [2]*Dep. Life. Sci., Univ. Tokyo*, [3]*Dep. Bio. Sci., Univ. Tokyo*)

Raman microspectroscopy has the potential to report the molecular compositions of a label-free living cell at the sub-micron spatial resolution. However, the complexity of the spectrums obtained from biological samples have hampered their intuitive interpretations regarding the molecular compositions. Here, we propose a systematic method to reveal the molecules contributing to the spectrums by employing the linear relations between the spectrums and omics data. We give preliminary results with Raman and RNA-seq data of S. Pombe under various stress conditions, and show the validity of the method. This approach provides a systematic method in interpreting a Raman spectrum, and may also be the first non-invasive omics-like data acquisition scheme to be realized.

2Pos302 電子顕微鏡法のための画像処理パッケージ Eos/PIONE の更なる発展

Further progress of Eos/PIONE for image analysis packages for electron microscopy

Takuo Yasunaga, Takafumi Tsukamoto, Ayaka Iwasaki (*Dept. of Biosci. Bioinfo., School of Comp. Sci. Systems, Eng., Kyushu Inst. Tech.*)

We have been developing a novel tool, Eos/PIONE for image analysis of electron micrographs since 1996. The current version of the packages supplies automatically-generated GUI for each of more than four hundred of programs such as 3D-reconstruction, 2D and 3D image analysis. Segmentation, feature extraction, and classification of 3D objects or maps are also provided. Some programs can use GPGPU to improve the speed of analysis. Besides, we offer some integration sets of applications such as single particle analysis, electron tomography, and so on. The package is an open-access one on OSDN or GITHUB, which can be freely downloaded. If you hope, we readily collaborate with making new tools.

2Pos303 X 線自由電子レーザーを用いたコヒーレント回折イメージング実験におけるデータ解析の自動化と酵母細胞核の三次元構造解析への応用

Automated data analyses for 3D structural reconstruction of yeast nuclei in coherent diffraction imaging using X-ray free-electron laser

Yuki Sekiguchi[1,2], Amane Kobayashi[1,2], Koji Okajima[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Masaki Yamamoto[2] ([1]*Grad. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*)

Coherent X-ray Diffraction Imaging (CXDI) visualizes electron density distribution inside micrometer-sized biological particles without sectioning or staining. In CXDI experiments, electron density maps of particles projected along the direction of incident coherent X-rays are retrieved from diffraction patterns. 3D structures of particles can be reconstructed from a large number of projected maps. By utilizing X-ray free-electron laser at SACLA, 2,000,000 diffraction patterns can be recorded in single beamtime. However, data analyses, especially identification of worthwhile data and retrieval of projected maps, largely depend on users' experience and are time-consuming. Here, we propose automated data analyses and apply it to 3D structural analyses of yeast nuclei.

2Pos304 フォトクロミック分子を利用した低分子量 G タンパク質の光可逆的制御

Photo-regulation of Small G protein RhoA using Photochromic Molecules

Kaori Masuhara[1], Masahiro Kuboyama[1], Nobuyuki Nishibe[2], Shinsaku Maruta[1,2] ([1]*Grad. Sch. Bioinfo., Univ. SOKA*, [2]*Dept. Bioinfo., Fac. Engineer., Univ. SOKA*)

RhoA is one of small G-protein Rho family, which plays important role of regulation of cellular processes such as cell motility, adhesion, and apoptosis. In this study, we performed to control the GTPase activity of small G protein RhoA using photochromic molecules photo-reversibly. We prepared RhoA Q63L mutant then introduced cysteine residues into the functional site as switch 1 and 2 regions V38, E40, and Y66. Mutant RhoA modified with isomerization of photochromic molecules 4-phenylazophenyl maleimide (PAM). Photo-regulations of the GDP-GTP exchange on the PAM modified RhoA mutants were monitored with fluorescence-labeled GTP derivative Mant-GTP. Cis-PAM-V38, E40, or Y66 showed faster GDP-GTP exchange than Trans-PAM-V38, E40, or Y66 in the without Mg buffer.

2Pos305 Structure and mechanism of the multimerization of small GTPase protein Ras induced by chamical modification at HVR domain

Takashi Hashimoto[1], Shinsaku Maruta[1], Yasunobu Sugimoto[2] ([1]*Soka University*, [2]*Nagoya University*)

We have previously shown that chemical modification of the cysteine residues in the HVR of small GTPase protein Ras with bulky hydrophobic SH group specific reagentsmultimer induced multimerization of Ras. The HVR is believed to perform physiologically important roles. Therefore, the multimerization phenomenon may reflect the function of Ras. In this study, we analyzed conformation of the Ras multimer by synchrotron X ray scattering and fluorescent probes. Small angle X ray scattering data suggested that Ras modified with fluorescent probes forms pentamer. We also estimated the possible conformation of the pentamer. Interfaces between the Ras pentamer was examined by the fluorescence quenching method. The resutlts suggested that HVR region is involved in the interface.

2Pos306 環状型サイトカインの安定性を向上させる結合末端ループ長の選択

Selection of the loop length about circularized cytokines

Risa Shibuya[1], Takamitsu Miyafusa[2], Wataru Nishima[2], Shinya Honda[2] ([1]*Front. Sci. , Univ. of Tokyo*, [2]*BMRI, AIST*)

Granulocyte-colony stimulating factor (G-CSF) is a kind of cytokine, and plays an important role in the treatment of neutropenia. However the low stability of G-CSF require its improvement. To improve stability of G-CSF, we circularized a main chain of G-CSF using split intein. In this study, we expected that the loop length that connects the terminal ends of G-CSF would be involved in stability and reaction efficiency of G-CSF, so prepared circularized G-CSF of the various mutant. Actually, we have found that thermal stability and reaction efficiency of circularized G-CSF vary with the loop length. These results indicates that it is possible to optimize the loop length for improving protein stability. Here, we mainly report on results of the most stable mutant.

2Pos307 Interaction between ring or linear DNA vs. nanopore/nanoslit

Takayuki Nakayama[1], Yoshiaki Iitsuka[1], Seiya Minato[1], Surat Wangwarunyoo[2], Naoto Sakashita[1], Kentaro Ishida[1], Toshiyuki Mitsui[1] ([1]*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*, [2]*Chulalongkorn Univ.*)

Although solid-state nanopore devices have demonstrated the capability of single DNA molecule sensing, there are certain issues to deal with relatively longer DNA molecules, such as lambda or t4. Particularly, these molecules clog at nanopores and generate electric noise on the ionic current measurements for sensing. However, the mechanism of the DNA clogging remains unknown. In order to elucidate the clogging mechanism, we have directly observed the interactions between DNA and pores near the pores by optical microscopy, and found that the electroosmosis by the DNA itself was essential. In this presentation, we will show the clogging probabilities of linear or ring DNA molecules with various length and their pore size or shape dependence and discuss the results.

2Pos308 Photo-regulation of Small G-proteins Ras Using Photochromic Molecules

Masahiro Kuboyama, Kaori Masuhara, **Shinsaku Maruta** (*Soka University*)

Small G-protein Ras is activated in GTP binding state and inactivated in GDP binding state. The switching of Ras is regulated by Guanine nucleotide Exchange Factor (GEF) and GTPase Activating Protein (GAP). In this study, it is focused on the interaction between Ras and αH helix which is Ras interface region of SOS (Ras-GEF) to photo-regulate Ras activity artificially with photochromic molecules. We prepared SOS mimicking peptide crosslinked with bifunctional azobenzene derivative ABDM intramolecularly. The ABDM-SOS peptide showed reversible secondary structural change by light irradiation. The inhibition effect of the ABDM-SOS peptide was also examined by nucleotide exchange reaction and fluorescent polarization.

2Pos309 脂質膜上チャネル形成のための 10 ナノメートルスケールのポアを持つ DNA オリガミナノ構造

DNA origami nanostruture with pore of ten nanometer scale for forming channel on lipid membrane

Koichiro Katayama[1], Ibuki Kawamata[1], Yuki Suzuki[1,2], Satoshi Murata[1], Shin-ichiro Nomura[1] ([1]*Graduate school of Engineering, Tohoku University*, [2]*Frontier Research Institute for Interdisciplinary Sciences, Tohoku University*)

We propose a DNA nanostructure with 12 nanometer size pore as an equipment of molecular robot. The purpose of synthesizing such nanostructure is mimicking the functionalities of biological channel that form pore for transporting molecules between inside and outside of lipid membrane. Unlike conventional artificial channels constructed by DNA with pore size of a few nanometers for transporting ions, our structure has a potential to pass larger molecules. Our nanostructure is designed using a method called DNA origami in order to build a rigid molecule with precise geometry. In the poster presentation, results of atomic force microscopy observation and other experiments to assay the channel activity will be discussed.

2Pos310* 撹拌操作が引き起こすゲノム DNA の二本鎖切断：新規実験手法の提案

How to keep genome-sized DNA safe against stirring stress: Quantitative analysis through single DNA observation

Hayato Kikuchi, Yuko Yoshikawa, Rinko Kubota, Kenichi Yoshikawa (*Lab. Biol. Phys., Facul. Life Med. Sci., Doshisha Univ.*)

Currently, it has been difficult to eliminate and analyze giant DNA molecules above the size of hundreds kilo base pairs (kbp). Here, by use of fluorescence microscopy we report our results on the quantitative evaluation of double-strand breaks, DSB, on 100kbp-sized DNA molecules under mechanical agitation, such a mixing procedure in a sample tube. It has become evident that the probability of DSB critically dependent on the size of DNA, i.e., the DSB probability per kbp for 100kbp-sized DNA decreases almost two-orders for 10kbp-sized DNA. We will propose a novel method of mixing of DNA solution to decrease the DSB probability in a significant manner.

2Pos311* Four-way junction DNA 形成による癌特異的 microRNA 発現パターンの自律的検出

Programmable system for recognition of microRNA expression pattern using four-way junction DNA formation

Moe Hiratani, Masayuki Ohara, Ryuji Kawano (*The Dep. of Biotech. and Life Sci., Tokyo Univ. of Agr. and Tech.*)

MicroRNA (miRNA) has been receiving an attention as a next-generation of an early diagnostic marker of cancers. In the case of small cell lung cancer (SCLC), it has been reported that miRNA-20a and miRNA-17-5p are overexpressed, and then they are simultaneously released into blood. To recognize these expression pattern, we propose to use DNA computing and nanopore techniques. In this study, programmable DNAs form four-way junction structure in the case of the simultaneous expressed miRNA-20a and miRNA-17-5p. The four-way junction DNA can be rapidly detected by nanopore with real-time and label-free. Moreover, we aim at the recognition of miRNA expression pattern directly from SCLC patients' serum.

2Pos312 光による DNA ハイドロゲルのパターン形成

Patterning of DNA hydrogel using light

Suguru Shimomura, Takahiro Nishimura, Yusuke Ogura, Jun Tanida (*Grad. Sch. Info. Sci. & Tech., Osaka Univ.*)

This study proposes an optical DNA hydrogel patterning method for controlling the shape flexibly, remotely, and without a mold. The hydrogel is constructed by self-assembly of Y-shaped DNA (Y-DNA) and linker DNA (L-DNA). To achieve optical control, we introduce cap-DNA that holds quenchers functioning as photothermal convertors. The cap-DNA prevents Y-DNA from binding to L-DNA by preferential binding to Y-DNA. With excitation light irradiated, the cap-DNA is dissociated from the Y-DNA and then a DNA hydrogel is generated. The shape of the DNA hydrogel depends on the distribution of excitation light. We experimentally confirmed that excitation of the quenchers promoted DNA hydrogel generation, and the shape was changed according to the scanning trajectory of laser-beam.

2Pos313 キメラ受容体によるバクテリアバイオセンサーの特異性改変

Modification of ligand specificity in bacterial biosensor with hybrid chemoreceptors

Hana Satou[2], Nao Fujii[2], Takashi Sagawa[1], Hiroto Tanaka[1], Kazuhiro Oiwa[1,2], **Hiroaki Kojima**[1] ([1]*Frontier Lab.,KARC,NICT*, [2]*Sch. Sci., Univ. Hyogo*)

Recently, we developed a bacterial biosensor based on chemotaxis response of *Escherichia coli*. To increase the detection range of this sensor, we constructed several hybrid receptors composed by Tap and Tar fused at different position in the amino acids sequence. In contrast to the WT Tar, hybrid receptors fused at the HAMP domain, known as the target region of restriction enzyme *NdeI*, showed significant increase in CW bias (0.3 to 0.7) and showed weak response to the attractant. On the other hand, another hybrid receptor fused at border of transmembrane domain retains signaling property of WT Tar. From these results, we proposed the novel method to construct the hybrid chemoreceptor maintaining the signaling property of the original receptor.

2Pos314* 金ナノ粒子のデジタル計数法による標的DNAの高感度検出 High-sensitivity Homogeneous DNA hybridization assay by Digital Counting of Gold Nanoparticle Dimers

Takaha Mizuguchi, Keiko Esashika, Toshiharu Saiki (*Grad. Sch. Sci. Tech., Keio Univ.*)

In this paper, we present a high-sensitivity homogeneous DNA hybridization assay by digital counting of gold nanoparticle (AuNP) dimers. We prepared two kinds of DNA-modified AuNPs which couple with each other and form AuNP dimer when target DNA exists in a solution. This target DNA labeled by AuNP dimer can be detected by the confocal optical setup and analyzing the signals from individual AuNPs. By digital counting of the AuNP dimers, the target detection limit of our method can be estimated to be at single pM level.

2Pos315* A single integrated gene nano-chip functioning in an artificial cell

Takeya Masubuchi[1], Masayuki Endo[2], Ryo Iizuka[3], Ayaka Iguchi[4], Yoon Doung Hyun[4], Tetsushi Sekiguchi[5], Hao Qi[1,6], Ryosuke Iinuma[1], Yuya Miyazono[1], Shuichi Shoji[4], Takashi Funatsu[3], Hiroshi Sugiyama[2,7], Yoshie Harada[2], Takuya Ueda[1], Hisashi Tadakuma[1,2] ([1]*Grad. Sch. of Frontier Sci., The Univ. of Tokyo*, [2]*iCeMS, Kyoto Univ.*, [3]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [4]*Grad. Sch. of Adv. Sci. and Eng., Waseda Univ.*, [5]*Research Organization for Nano & Life Innovation, Waseda Univ.*, [6]*Dept. of Chem. Eng. and Tech., Tianjin Univ.*, [7]*Grad. Sch. of Sci., Kyoto Univ.*)

In living systems, genetic expression is highly ordered. Through modifying and utilizing genetic circuits, we can control or reconstruct living system. Currently, however, the circuit complexity has been limited, because of its compounds limitation. Also freely diffusing molecules used as the trigger has potential to cause unintended reaction with other molecules. Here, we create a molecular device consisted of transcription factors (T7-RNAP and target genes) and DNA origami tile scaffold. The molecular device had unique features such as orthogonality, where the device transcribes only its own gene, and rational designability, where the transcription activity can be designed by molecular layout.

2Pos316* アメーバ型分子ロボット：モータータンパク質とDNAデバイスを内包した巨大リポソームの形状変化とその制御 Amoeba type molecular robot: controlling shape change of giant liposome entrapping molecular motors and DNA circuits

Yusuke Sato[1], Yuichi Hiratsuka[2], Ibuki Kawamata[1], Satoshi Murata[1], Shin-ichiro Nomura[1] ([1]*Grad. Sho. Eng., Tohoku Univ.*, [2]*Sch. Mat. Sci., JAIST*)

Creating an artificial molecular system is one of the purposes in molecular robotics. To realize controllable motion of the molecular robot, integration of molecular devices such as actuators and controllers is essential. We aim to develop "amoeba type molecular robot" capable of controllable shape change. In our design, molecular motors (kinesin/microtubule) and DNA circuits are encapsulated into giant liposome. In experiment, the amoeba type robot showed continuous shape change by the motors over 2 hours. Moreover, we succeeded in controlling start and stop of the shape change by applying DNA signal. We anticipate that our results can contribute to construct artificial cells and molecular robots with chemotaxis.

2Pos317 心筋細胞集団同士を繋ぐ線維芽細胞の距離に対する同期 Synchronization of large clusters of cardiomyocytes connected with fibroblasts and its distance change

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To analyze the synchronization process of clusters of chicken embryo-derived cardiomyocytes, we used the on-chip multi electrode array system with agarose microchambers. Two sets of cardiomyocyte clusters were connected with fibroblasts by additional agarose micro fabrication during cell culture, and it was observed whether or not to synchronize when distance of fibroblasts changed. As a result, they were synchronized within the fibroblast distance of 300 μm, and pace making is not depended on the fast beating rate or the stability of fluctuation. It was suggested that another mechanism of synchronization between large size clusters of cardiomyocyte exists different from tissues or single cardiomyocytes. Our guess is that the pace making depend on the number of cells.

2Pos318 目的細胞の回収を目指した微小液滴内培養法の開発 Development of a single cell cultivating method using a microdroplets forming technique for sorting specific cells

Hideyuki Terazono[1], Masao Odaka[1], Akihiro Hattori[1], Kenji Matsuura[1], Kenji Yasuda[1,2] ([1]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*, [2]*Dept. Physics, Waseda Univ.*)

We developed a technique to enclose cells in alginate microdroplets using a microfluidic technique to sort specific cells from a mixture containing several types of cells. A cross-junction microfluidics formed uniform sizes of alginate solution-in-oil emulsions and cells were enclosed in the microdroplets. Furthermore, Alginate microdroplets were allowed to transfer from oil phase to water phase very easily and cultivate in the culture medium by using a surfactant in the microdroplets forming step. This technique provides us rapid formation of microdroplets containing single cell for sorting specific cells and identification of cell types.

3Pos001 Single Particle Analysis of *Eh*V-ATPase by Phase-contrast cryo-Electron Microscopy

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*Eh*V-ATPase is an ATP-driven Na^+-pump isolated from *Enterococcus hirae*, which consists of 24 subunits from 9 different proteins. In our previous study, a single particle 3D reconstruction of the Zernike-phase contrast cryo-electron microscopy images successfully revealed a whole structure of detergent-solubilized *Eh*V-ATPase at nano-meter scale after a selection and motion collection of the sub-frames. However, V-ATPase's flexibility made it difficult to improve spatial resolution. In this study, to reveal the whole structure at higher resolution, we inserted PA-tag into the dynamic subunit and bound the antibody to fix the flexible structure. Then, we analyzed the structure by single particle cryo-electron microscopy.

3Pos002 TEM と ASEM を用いたタンパク質複合体・細胞組織の親水環境での観察
Electromicroscopy of protein complexes, cells and tissues in hydrophilic environment

Chikara Sato[1], Nassirhadjy Memtily[1], Mari Sato[1], Toshiko Yamazawa[2], Masaaki Kawata[1] ([1]*AIST*, [2]*Dept. Mol Physiol, Jikei Univ. Sch. Med.*)

Visualization of biological structures in aqueous condition at electron microscopy (EM) resolution is still challenging due to the present limit of microscopy. To observe the dynamics activities of protein complexes, we have developed single particle reconstruction using cryo-TEM and ASEM. In ASEM, an inverted scanning electron microscope (SEM) observes the wet sample from beneath an open dish. Various activities in neurons and tissue was directly observed in a hydrophilic condition using TEM and ASEM. We have also observed protein complex formations yielded by bacterial activities and a dynamic gathering of signaling proteins in cultured cells.

3Pos003 クライオ電子顕微鏡による *Thermus thermophilus* V-ATPase の単粒子解析
Single-particle analysis of *Thermus thermophilus* V-ATPase by Cryo-EM

Atsuko Nakanishi[1], Jun-ichi Kishikawa[1], Kaoru Mitsuoka[2], Ken Yokoyama[1] ([1]*Dept. LifeSci. Kyoto Sangyo Univ.*, [2]*Res. Ctr. UVHEM. Univ. Osaka.*)

The V-ATPase from *Thermus thermophilus* V_oV_1 (*T. th*V_oV_1) is a rotary enzyme that mediates the coupling between ATP synthesis/hydrolysis in V_1 and proton translocation across membranes through V_o. To understand the molecular mechanism of *T. th*V_oV_1, the whole structural information of the enzyme at atomic resolution is required. Here we report a cryo-EM density map of *T. th*V_oV_1. The map showed dodecamer proteolipid ring and 8 membrane-embedded α helices of I subunit, which contained two tilted α helices forming α hairpin that contacted L-ring. Based on the structure, we discuss the proton pathway in the V_o domain.

3Pos004 クライオ電子顕微鏡単粒子解析によるマウスノロウイルス VLP の構造解析
Structural Analyses of Murine Norovirus VLPs by Cryo-Electron Microscopy Single Particle Analysis

Chihong Song[1], Motohiro Miki[2,3], Reiko Todaka[2], Kei Haga[2], Akira Fujimoto[2], Kazuhiko Katayama[2], Kazuyoshi Murata[1] ([1]*NIPS*, [2]*NIID*, [3]*Denka Seiken*)

Human noroviruses are the most common cause of epidemic nonbacterial gastroenteritis in winter, but the mechanism producing the antigenic diversity is almost unknown. We generated virus-like particles (VLPs) of murine norovirus S7 and GV1 as surrogates of human norovirus, by using a baculovirus expression system. We reconstructed and compared the 3D structures of both VLPs by cryo-electron microscopy single-particle analysis at sub-nanometer resolution. Although the difference of the amino acids is only 6% in the two genotypes, the whole structure of the capsids was drastically changed. Especially, the residues forming the interaction in the neighboring protrusion (P)-domains were switched between these genotypes.

3Pos005 2D ハイブリッド解析による電子顕微鏡平均画像の成分解析
Component analysis of averaged EM images by 2D hybrid analysis

Atsushi Matsumoto[1], Junichi Takagi[2], Atsushi Kawaguchi[3], Kenji Iwasaki[2] ([1]*National Institutes for Quantum and Radiological Science and Technology*, [2]*Osaka University*, [3]*Tsukuba University*)

We are developing a computational approach (2D hybrid analysis) to build a 3D atomic model from an electron microscope (EM) image of a biological molecule. In this approach, firstly a lot of atomic models are built by deforming the X-ray crystal structure. Then, the projections of each model are created. Finally, they are compared with the EM image to select the best-fitting atomic model.
We have recently introduced the realistic projection model to improve the accuracy of the computation. This has enabled us to decompose the averaged EM images. In principle, averaging of EM images should be performed using the raw images of molecules with the same conformation and orientation. In reality, mixture occurs. Now we can detect such mixture by the 2D hybrid analysis.

3Pos006 Structural analysis for V1-ATPase from a variety of prokaryotes

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V-ATPase is composed of two parts, hydrophilic V1 and membrane-embedded Vo. In bacterial enzymes, the V1 portion is known as a rotary motor, in which a central axis DF complex rotates relative to catalytic A3B3 stator driven by ATP hydrolysis energy.
To determine V1 structure and to investigate its diversity, we have tried to construct expression vectors of V1 from a variety of prokaryotes. Finally, we purified three kinds of bacteria V1 from Thermus thermophilus, Deinococcus radiodurans and Ruminiclostridium thermocellum. In this study, we report 3D reconstituted structures of these V1 determined by single particle analysis using cryo electron microscopy.

3Pos007 最新低温電子顕微鏡“CryoARM”の性能

Performance of State-of-the-art CryoEM, named “CryoARM”

Takayuki Kato[1], Naoki Hosogi[2], Takeshi Kaneko[2], Isamu Ishikawa[2], Keiichi Namba[1,3] ([1]*Grad. Sch. of Front. Biosci., Osaka Univ.*, [2]*JEOL*, [3]*RIKEN, QBiC*)

Cryo electron microscopy(cryo-EM) is becoming a very powerful approach to analyze the structure of individual biomolecular machines. We developed state-of-the-art CryoEM with JEOL, named “CryoARM”. It has an auto sample loader which can store 12 samples, an auto liquid nitrogen filling system which can keep the stage and sample loader temperature lower than 100 K during a week and K2 direct detector. We tried to observe the thon ring from Pt/Ir in the biological sample observation conditions, we were able to observe beyond 2 Å resolution signal. To evaluate of CryoARM, we are trying to analyze the structure of b-galactosidase, TMV and GDH by single particle image analysis. I will explain and discuss about the performance of the CryoARM.

3Pos008 電顕３次元密度マップからαヘリックスを認識する混合正規分布モデルの開発

Detection of alpha-helices from the 3D EM density map using Gaussian mixture model

Takeshi Kawabata, Haruki Nakamura (*IPR, Osaka U.*)

Recently, the electron microscopy (EM) provides high resolution 3D density maps with 3-10 Å, which enable us to de novo atomic modeling, without helps of any known 3D structures. For the first step for the de novo modeling, we developed the helix detection program using Gaussian mixture model (GMM). Different from the standard GMM, our program only uses a shape of Gaussian distribution functions (GDFs) stored in the GDF library, which contains GDFs corresponding to poly-Ala alpha helices with various lengths. Detected GDFs of helices are transformed to atomic models, and refined their positions. The results were quite good for test proteins. For the next step to assign amino acids, detailed atomic models with side chains are necessary.

3Pos009 Towards Understanding the Molecular Architecture of Human DNA Polymerase δ using Electron Microscopy and Computational Modeling

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DNA Polymerase δ is one of the main polymerases that carry out replication in eukaryotic cells and plays a crucial role in the DNA repair. In spite of its importance, structural information for the mammalian Pol δ complex is surprisingly limited. In this work, we have employed Electron Microscopy and computational modeling to determine the overall molecular architecture of human Pol δ complex. Low resolution map of the Pol δ shows two regions of high density, similar to yeast Pol δ SAXS envelope. The structures of the four individual subunits were either taken from PDB or modeled using homology-based and de-novo structure modeling methods. These were then fit into the map to obtain the molecular architecture of the Pol δ complex.

3Pos010 GPI アタッチメントシグナル領域の二次構造解析

Secondary structural analysis of GPI attachment regions

Keiya Inoue[1], Daiki Takahashi[2], Tatsuki Kikegawa[2], Kenji Etchuya[2], Yuri Mukai[1,2] ([1]*Sch. Sci. & Tech., Meiji Univ.*, [2]*Grad. Sch. Sci. & Tech., Meiji Univ.*)

GPI transamidase are considered to recognize the specific structures of the regions around ω-sites in the premature GPI-anchored proteins.
The purpose of this study is to uncover the recognition and digestion mechanisms of GPI transamidase based on analyzing the secondary structures. In this study, the secondary structures of GPI-ASs which were fused GFP proteins and were expressed in Escherichia Coli, were analyzed by circular dichroism (CD). One of the GPI-ASs in this study has been derived from human prion protein (PRIO_HUMAN), and the other one has been designed artificially by analyzing amino acid propensity. According to the CD spectra in far UV region, the α-helix content of GFP fused GPI-AS increased compared with GFP.

3Pos011 インターセクチン２のコンホメーション解析

Conformational Analysis of Multidomein Protein Intersectin 2

Kazutaka Murayama[1,2], Miyuki Murayama-Kato[2], Ryogo Akasaka[2], Daisuke Sugimori[3], Mikako Shirouzu[2] ([1]*Tohoku Univ. Biomed. Eng.*, [2]*RIKEN, CLST*, [3]*Fukushima Univ. Symbio. Sys. Sci.*)

Intersectin-2 (ITSN2) can be involved in clathrin-mediated endocytosis. ITSN2 is a multidomain protein, including five SH3 domains followed by the DH-PH module. Multidomain proteins are one of the difficult targets for conventional structural analysis methods, like an x-ray crystallography/NMR. To investigate global conformation of proteins, analytical ultracentrifugation, dynamic light scattering, or charge-state-distribution by Electron-Spray-Ionization mass spectrometry is useful. In this study, ITSN2(719-1543aa; (SH3)x5-DH-PH) were analyzed by various methods. The results suggest that ITSN2 can be elongated form, but not fully extended. The loop regions connecting SH3 domains can contribute to the global conformation in solution.

3Pos012 タンパク質の構造変性と回転拡散係数

Rotational diffusion coefficients of proteins along denaturation curve

Yoshitake Tomoyuki, Terazima Masahide (*Graduate School of Science, Kyoto University*)

Protein structure is certainly important information for understanding the protein reactions and functions. Translational diffusion coefficient (Dtr) is one of the physical parameter which reflects the structure of proteins and has been used to probe the structural change of proteins during reactions. On the other hand, there are few reports on the relation between the protein structures and rotational diffusion coefficients (Drot). We investigated the relation between rotational diffusion coefficients, which were measured by the fluorescence polarization decay method, and the secondary structure of proteins along the denaturation curves. Drot was compared with the secondary structure measured by the CD method.

3Pos013 シアノバクテリア時計タンパク質 KaiC の AFM 観察

AFM observation of a ring-shaped structure of KaiC

Jun Abe, Atsushi Mukaiyama, Yoshihiko Furuike, Shuji Akiyama (*Division of Trans-Hierarchical Molecular Systems, Research Center of Integrative Molecular Systems (CIMoS), Institute for Molecular Science (IMS)*)

The ATPase function of KaiC is usually kept extremely low and less dependent on temperature. Recently, on the basis of X-ray crystallographic analyses, we proposed a hypothesis that an asymmetrical deformation of a ring-shaped hexamer of KaiC is related to the regulatory mechanism of cyanobacterial circadian clock. In order to test this hypothesis in solution, we conducted AFM measurements of KaiC at the single-molecule level. Through the careful optimization of fixation conditions, the ring-shaped structures could be resolved finely enough to discuss the characteristics and variety of individual KaiC hexamers. In this presentation, we will discuss recent progress.

3Pos014 高速 AFM による 20S プロテアソーム関連タンパク質の動態観察

Dynamics observation of the 20S proteasome-related proteins using High-Speed AFM

Toshiya Kozai[1], Tadashi Satoh[2], Arunima Sikdar[3,4], Hirokazu Yagi[2], Maho Yagi-Utsumi[3], Takayuki Uchihashi[1], Toshio Ando[1], Koichi Kato[3,4] ([1]*Dept. of phys., Kanazawa Univ.*, [2]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [3]*Okazaki Inst. Integ. Biosci.*, [4]*Nat. Univ. SOKENDAI*)

Some assembly chaperones are involved in a complexation process of the eukaryotic 20S proteasome. Recent genomic analysis identified archaeal homologs of the assembly chaperones, PbaA and PbaB. PbaA does not have the proteasomal binding capacity, but it forms a complex of 10:10 stoichiometry with being combined with PF0014. Meanwhile, among human proteasome α subunits, α7 has a unique feature of self-assembly into a homo-tetradecamer with a double ring structure. In addition, the α7 homo-tetradecamer is disassembled upon the addition of α6, resulting in a 1:7 hetero-octameric α6-α7 complex. Here we applied HS-AFM to elucidate the structure of PbaA-PF0014 complex, its assembly process and also disassembly mechanism of α7 double ring due to the interaction with α6.

3Pos015 立体構造予測において疎水効果を評価するための新しい指標： 仮想原子の周りのコンタクト数

A new measure for hydrophobicity: Contact number around an imaginary atom

Yota Masuyama, George Chikenji (*Grad. Sch. Eng., Nagoya Univ.*)

In protein structure prediction, it is important to accurately estimate hydrophobic interaction free energies. So far, some measures that estimate hydrophobic interaction have been developed, such as accessible surface area (ASA) and contact number (CN). These measures, however, have difficulty: they cannot discriminate buried and exposed residues precisely. To overcome the difficulty, we recently proposed a new measure of hydrophobicity, the CN around an imaginary atom, and showed that it outperformed the currently existing scoring functions in the decoy/native discrimination test. This presentation reports a further improvement of our measure that appropriately deals with buried polar residues forming hydrogen bonds or exposed hydrophobic residues not forming cluster.

3Pos016 統合失調症疾患感受性遺伝子産物 G72 タンパク質の構造機能予測

Structure and function prediction of the G72 protein, the product of a susceptible gene for schizophrenia

Yusuke Kato, Kiyoshi Fukui (*Institute for Enzyme Research, Tokushima University*)

G72 is one of the most important susceptible genes for schizophrenia. Only primates have G72. Thus, G72 may be related to a higher-order mental activity although its function is elusive. No known protein structure is phylogenically related to G72. Thus, it is difficult to apply structure prediction strategies that use known structures as templates. G72 is prone to aggregate, which hampers crystallization. Thus, we predicted the structure of G72 with a novel ab initio approach that does not require a template structure. The average accuracy of the novel approach was 3.85 Å, corresponding to the size of a small amino acid, based on the tests with sequences of known structures. Intriguingly, the predicted structure of G72 was similar to transporters and adapter proteins.

3Pos017 残基間平均距離統計に基づくコンタクトマップによる天然変性領域の予測

Prediction of IDRs by a contact map based on inter residue average distance statistics

Takumi Shimomura, Takeshi Kikuchi (*Univ. Ritsumei*)

IDPs (Intrinsically Disordered Protein) account for about one third of proteins that function in human bodies. However, their 3D-structures cannot be determined with techniques such as X-ray crystallography and NMR analysis. This study aims to predict disorder residues in IDP from its sequence. We selected the target proteins as follows. That is, disorder proteins were collected from Disprot. Order proteins were collected from PDB. The target proteins are analyzed by ADM（Average Distance Map） analysis. The results indicate that the present technique shows the at least comparable accuracy to the several methods proposed so far. We are attempting to improve our method to provide more accurate result.

3Pos018 A new threading method based on the physical characteristics of sequence-structure compatibility

Kyosuke Tomoda, Yota Masuyama, George Chikenji (*Grad. Sch. Eng., Nagoya Univ.*)

Protein structure comparison studies have revealed that many evolutionary unrelated protein pairs showed surprisingly significant structure similarities. Unfortunately, such relationship cannot be detected by the majority of the current protein structure prediction methods, because they heavily rely on evolutionary information. To utilize protein structure database more effectively, it is highly desired to develop a protein structure prediction method that does not rely on evolutionary information. Here, we propose a new threading algorithm mainly based on the physical characteristics of sequence-structure compatibility. This presentation will describe the detail of our method and show the results of benchmark tests.

3Pos019 EMDB, PDB, SASBDB 中の多階層構造データを対象としたウェブベースのサービス
Web based services for multiscale structure data in EMDB, PDB and SASBDB

Hirofumi Suzuki[1,2], Takeshi Kawabata[1], Gert-Jan Bekker[1,2,3], Haruki Nakamura[1,2] ([1]*IPR, Osaka Univ.*, [2]*PDBj*, [3]*FBS, Osaka Univ.*)

Recent innovation in structure analysis methods, especially in cryo-EM, increase many values of structure data. Increasing data in different data types (atomic model, density map, or dummy-atom model) are stored in the various structure databanks, such as EMDB, PDB and SASBDB. Their scales are widening from molecules to cells. Typical search and browse systems are designed for a particular data type in a particular database. We have been developed web based services for EMDB, PDB and SASBDB data. ***EM Navigator*** is a browser for 3DEM data. ***Omokage search*** provide a shape similarity search. ***Yorodumi*** integrates structure viewers and the metadata. Recently, we have reformed these services in a unified and modern interface supporting PCs and mobile devices.

3Pos020 単独で構造を維持するドメインデータベース「IS-Dom」の他のデータベースに依存しない拡張
Standalone definition of putatively independent structural domain: IS-Dom

Soichiro Ide[1], Teppei Ebina[2], Richa Tanbi[1], Yutaka Kuroda[1] ([1]*Tokyo University of Agriculture and Technology*, [2]*Department of Physiology, Graduate School of Medicine, The University of Tokyo*)

Protein domains that can fold independently are significant targets of high through put proteomics analysis. IS-Dom is a dataset reporting Structural Domains that can presumably fold independently (ISD). Our initial IS-Dom dataset was constructed by quantitatively assessing SCOP and CATH defined domains for inter-domain hydrophobic clusters and hydrogen bonds using the full length protein's atomic coordinates. Here, we propose a novel algorithm that defines ISDs from atomic coordinates only and without using prior domain boundaries.

3Pos021 Attempts at CA-type formal analysis of fibrous assembly of particles

Takashi Konno (*Mol. Physiol., Med., Univ. Fukui*)

In the framework of 2D and 3D cellular automata (CA), transition rules leading to fibrously assembled “structures” were constructed and analyzed. The elements could represent proteins in an abstractive form. The analysis in high dimensional CA systems could naturally be unexhaustive, but careful choice of the CA transition rules gave valuable insights into the physical reality. The rules could also be translated into the “energy” term. “Fibrous” pattern of a state in the CA lattice could directly be regarded as “fibers”, but more abstractive definitions of “structure” were also challenged. This study is an initial step towards elucidating hidden logics unconsiously employed for recognizing “structures” in daily and/or scientific life.

3Pos022 生物の低温適応と蛋白質配列の進化
Cold adaptation of organisms and the evolution of protein sequences

Matsuyuki Shirota[1,2,3] ([1]*Grad. Sch. Med., Tohoku Univ.*, [2]*ToMMo, Tohoku Univ.*, [3]*Grad. Sch. Inform Sci., Tohoku Univ.*)

Organisms living in extremely cold environment face with various stresses for survival, such as low enzymatic activity, freezing and fluctuations on protein stability, but how they have evolved to adapt their environment is largely unknown. Here I examined the protein sequences of Belgica antarctica, the only insect endemic to Antarctica, in comparison to other insects including Drosophila melanogaster. B Antarctica had smaller fraction of Ala, Pro, Gly and Arg residues, which have G or C bases at the first and second positions of codon, reflecting decreased GC content in the coding region. B Antarctica have evolved to have smaller genome size and low GC content, and such requirements on the genome sequence thus restrict the protein sequence of the organism.

3Pos023 スライディングとストランド間移動を用いたヒト抗ウイルス因子 APOBEC3G の高効率な DNA 配列探索：実時間 NMR による新知見
Sliding and intersegmental transfer on DNA enhance target search of human anti-viral factor APOBEC3G: insight by the real-time NMR study

Keisuke Kamba[1], Takashi Nagata[1,2], Masato Katahira[1,2] ([1]*Inst. of Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. of Energy Sci., Kyoto Univ.*)

The human cytosine deaminase APOBEC3G (A3G) restricts HIV infection. A3G deaminates the cytidines that are located close to the 5’ end more effectively than ones that are less close to the 5’ end of the viral cDNA. This phenomena is called deamination polarity. However, the knowledge of deamination mechanism has been limited. In this study, we have investigated enzymatic nature of by using the real-time NMR monitoring method that we developed previously. Firstly, we found that sliding enhances deamination activity of A3G. Secondly, the electrostatic interactions between A3G and the phosphate backbone of an ssDNA was shown to be the key for sliding. Finally, we found that intersegmental transfer also enhances deaminase activity of A3G.

3Pos024 分子動力学ドッキング・シミュレーションによるスーパーコイル DNA 結合（SDR）ペプチドとクロスオーバー DNA の選択的結合メカニズムの解析
Molecular dynamics docking study on selective binding mechanisms of supercoiled-DNA recognition (SDR) peptide and spatially-crossover DNA

Hiroshi Nishigami[1], Kakeru Sakabe[1], Jiyoung Kang[1], Kuniaki Sano[2], Kimiko Tsutsui[2], Ken Tsutsui[2], Kazuhiko Yamasaki[3], Masaru Tateno[1] ([1]*Grad. Sch. Life Sci., Univ. Hyogo*, [2]*Dept. Neurogenomics, Grad. Sch. Med. Dent. Pharm. Sci., Okayama Univ.*, [3]*Biomedical Res. Inst., AIST*)

The supercoiled-DNA recognition (SDR) peptide, which was isolated from the HIV transcriptional factor LEDGF (p75/SBP75), was previously indicated to exhibit the selective binding to the negative supercoiled structures of double-stranded DNA (dsDNA). In the 2015 annual meeting of BPSJ, we reported our structural models of the complex of SDR and the spatially-crossover dsDNA, which are currently impossible to be experimentally obtained. In the present study, we performed molecular dynamics docking simulations to investigate the substantial effects of Mg2+ and the mechanisms of the selectivity and cooperativity that were experimentally found in the SDR-dsDNA recognition. In the session, we further discuss the thermodynamic free energy profiles to explain these features.

3Pos025 Unravelling the mechanism of (6-4) photolyase enzyme

Hisham Dokainish, AKio Kitao (*The University of Tokyo*)

Ultraviolet radiation cause harmful covalent modifications in DNA, of adjacent pyrimidine, forming either cyclobutane pyrimidine dimers (CPD) or pyrimidine (6-4) pyrimidone photoproduct ((6-4) PP). Photolyase enzymes (CPD PHR and (6-4) PHR) undo these modifications using photorepair mechanisms, respectively. Although the repair mechanism in CPD PHR is well understood, the repair pathway in (6-4) PHR remains elusive and controversial. Here we synergistically use (QM/MM) calculations, (QM/QM/MM) calculations, MD, and reaction rates calculations of electron transfer using Marcus theory to first define the radical position in reactive complex. Second, we propose a new mechanism for repair in wild type as well as in His365/Ala mutant complex.

3Pos026 暗視野顕微鏡を用いたタンパク質過飽和溶液中ナノスケールダイナミクス構造観察

Nano-scale Observations of Supersaturated Protein Dynamics using Dark-Field Microscopy

Kazuki Yoshimura[1], Yufuku Matsushita[1], Keigo Ikezaki[1], Hiroshi Sekiguchi[2], Yuji Goto[3], Yuji Sasaki[1,2] ([1]*Grad. Sch. Front. Sci., Univ. Tokyo*, [2]*JASRI/SPring-8*, [3]*IPR, Osaka Univ.*)

Protein crystallization process is based on the mechanism that the concentration fluctuations give rise to clusters that coalesce under certain condition to start nucleation step. The feature of the metastable state is considered as key roles for a crystal morphology controlling. In this study, we dedicated to observe local dynamics of the solution during nucleation by detecting gold nanoparticles undergoes Brownian motions using Dark-Field Microscopy (DFM). Our experiments were performed to the solutions with different pHs (4.0- 8.5), buffers (Tris, Acetate), and NaCl concentrations (0.5- 1.5 M). We present the DFM method in detail, and the relationship between the local dynamics of the solutions and each final crystals morphology.

3Pos027 Analysis and control of protein crystallization using short peptide tags without affecting structure, thermal stability and function

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Short peptide tags attached to proteins are emerging tools for protein research. Here we report the effects of ten peptide tags on the crystallization behavior of a BPTI variant. The tags did not affect the structure, thermodynamics and activities. Six of eight tagged variants crystallized under the condition of untagged BPTI diffracted at high resolution. Most tags were invisible, indicating high flexibility having no interactions with nearby residues. Variants with long-term solubility (LS) 1-6mg/mL produced well diffracted crystals, while variants with LS 6mg/mL did not crystallize or produced poorly diffracting crystals. Therefore, short peptide tags could be a generic tool for tuning protein solubility and improving protein crystallization.

3Pos028 滴定 X 線溶液散乱測定を用いたアダプター蛋白質 GGA-ユビキチン相互作用の解析

An analysis of the interaction of GGA with ubiquitin by using titration SAXS measurement

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The GGAs are a family of clathrin coat adaptor proteins involved in vesicular transport. GGA is composed of three domains, an N-terminal VHS domain, a GAT domain, and a C-terminal GAE domain. GAT acts as a platform for multiple interaction partners, such as Arf, Ub, and so on. Despite a long linker, VHS and GAT interact with each other to be a closed form of GGA. Upon binding of an M6PR peptide to VHS, VHS is released from GAT. Here we investigated the effect of the domain rearrangement on the Ub binding to GAT by using titration SAXS measurements. The Kd for Ub and GAT decreased in the presence of M6PR, suggesting that the binding of M6PR to VHS would regulates the interaction surface on GAT through the domain rearrangement.

3Pos029 K63 ジユビキチンと TAB2 複合体の拡張サンプリング

Large-scale configurational sampling of K63-linked di-ubiquitin complexed with TAB2

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Npl4 Zinc Finger (NZF) domain is a ubiquitin-binding domain involved in ubiquitin-dependent pathways. In the NF-κB signaling pathway, the complex formation of the NZF domain of TAK1 binding protein 2 (TAB2) with K63-linked polyubiquitin is the key process. In order to understand how TAB2 recognize the specific linkage of the polyubiquitin chain, we performed molecular dynamics simulations of K63-linked di-ubiquitin bound to TAB2. Multiscale enhanced sampling (MSES) simulation was also attempted for the large-scale configurational sampling of the protein-protein interactions of the complex to analyze how the ubiquitin-substrate binding is achieved against the large structural fluctuations of K63-linked di-ubiquitin.

3Pos030 Periodic Formation of the Cyanobacterial Circadian Clock Protein Complexes

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The cyanobacterial circadian clock can be reconstituted in vitro. The phosphorylation level of KaiC oscillates with a period of approximately 24 hours in the presence of only KaiA, KaiB, KaiC, and ATP. These three proteins could form various complexes during oscillation. We investigated oligomeric structural properties and temporal behavior of the circadian clock protein complexes by using Native-PAGE technique and found that four different KaiABC complexes are assembled and disassembled periodically. Furthermore, analysis of the KaiC mutants to mimic the KaiC phosphorylation state indicates that these complexes formation is coupled to the KaiC phosphorylation state. We will discuss the role of these complexes in the molecular mechanism of the KaiABC system.

3Pos031 広角溶液散乱測定のための環境整備と時計タンパク質への応用

Wide-angle x-ray scattering studies on circadian clock systems

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Wide-angle x-ray scattering (WAXS) is one of the powerful methods to characterize hierarchical structures of bio-molecular systems. In a typical experiment, a sample solution containing quasi-monodispersed bio-macromolecule is irradiated by a focused x-ray beam, and then the intensity of the scattered x-ray is recorded using a 2D-detector as a function of the momentum transfer (Q) up to 2.5 Å-1 (d=~2.5 Å). We developed a software to reduce scattering images into one-dimensional WAXS curves with exact geometrical- and polarization-corrections of each pixel on the detector. In this presentation, the fundamentals of the correcting procedures will be first overviewed, and then a recent application to cyanobacterial clock system will be shared with future perspectives.

3Pos032 X 線小角散乱法を用いた神経軸索伸長系関連蛋白質 shootin1 の動的な構造変化の解析

Structural alteration of shootin1 upon phosphorylation revealed by using small angle x-ray scattering

Shoki Nakata[1], Keito Yoshida[1], Kentarou Baba[2], Yohei Shibata[1], Yoichi Yamazaki[1], Naoyuki Inagaki[2], Hironari Kamikubo[1] ([1]*Grad. Sch. of Mater. Sci., Nara Inst. of Sci. & Tech.*, [2]*Grad. Sch. of Biol. Sci., Nara Inst. of Sci. & Tech.*)

Shootin1 involved in nerve axon-elongation system can bind to both of an F-actin binding protein and a cell adhesion protein, where the mechanical coupling mediates the signal-force transduction. It was reported that the binding affinities are influenced by phosphorylation of shootin1. However, because of the lack of the structural information of shootin1, it is still unclear how shootin1 can control the affinities. In this study, in order to understand the molecular mechanism, we revealed the molecular shape of shootin1 phosphorylated by Pak1 in addition to the non-phosphorylated form by using SAXS. In the results, shootin1 was swollen upon phosphorylation and partially unfolded. Comparing the structural models, we will discuss the mechanism of the affinity regulation.

3Pos033 Dynamical system of alpha-crystallin oligomers

Rintaro Inoue, Takumi Takata, Noriko Fujii, Masaaki Sugiyama, Nobuhiro Sato, Yojiro Oba (*Research Reactor Institute,Kyoto University*)

Alpha-crystallin possesses a dynamic quaternary structure triggered by its constituting subunit dynamics. It is considered that the elucidation of a mechanism of subunit dynamics in homo-/hetero-alpha-crystallin is indispensable for understanding its underlying function: chaperone activity. We then performed deuteration-assisted small-angle neutron scattering (DA-SANS) method on alpha-crystallin. The existence of subunit exchange, which is one of subunit dynamics was clearly confirmed with DA-SANS. It is also found that the subunit exchange in hetero-alpha-crystallin is different from homo-alpha-crystallin. It is supposed that such difference of subunit exchange might be deeply related to its chaperone activity.

3Pos034 Refinement of Cryo-EM Structures Using Scattering Factors of Charged Atoms

Koji Yonekura, Saori Maki-Yonekura, Rei Matsuoka, Yoshiki Yamashita, Fumie Iwabuki, Maiko Tanaka (*RIKEN SPring-8 Center*)

Electron scattering relates directly to the distribution of Coulomb potential, which allow us to analyze charged states of amino acids and metals in biological macromolecules. We explored a suitable treatment of electron scattering factors of charged atoms for refinement of the atomic models against electron diffraction data from three-dimensional protein crystals. This approach found the proper electrostatic setting to produce atomic models with improved statistics and to better reflect experimental data. Structure refinement for single particle analysis also benefits from this analysis. We are applying the analysis to cryo-EM structures of an energizer membrane protein for studying charged states in the functional site and how the protein works upon ion conducting.

3Pos035 Visualization of 11- and 34-fold rotational symmetries in the MS ring of the bacterial flagellum by electron cryomicroscopy

Akihiro Kawamoto[1], Ayana Kaido[2], Miki Kinoshita[1], Tomoko Miyata[1], Tohru Minamino[1], Takayuki Kato[1], Keiichi Namba[1,3] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Dept. Food Science and Nutrition., Doshisha Women's College of Liberal Arts*, [3]*QBiC, RIKEN*)

The bacterial flagellum is a motility nanomachine with a rotary motor and a helical propeller. The MS ring is a rotor composed of a single membrane protein, FliF, and acts as the base for flagellar assembly. The structure has been visualized electron cryomicroscopy (cryoEM) and image analysis to have the 26-fold rotational symmetry. However, the resolution is still limited to see the structural detail. We report a high-resolution cryoEM structure of the Salmonella MS ring, which clearly shows 11- and 34-fold rotational symmetries in the central core and S ring region, respectively. These symmetries suggest that the former acts as the template for the helical assembly of rod proteins into a tubular structure with 11 protofilaments and the latter for the C ring assembly.

3Pos036 単粒子コヒーレント X 線回折像の類似積判定のためのマルチステップアルゴリズム

Multistep similarity detection algorithm for single particle X-ray coherent diffractions

Atsushi Tokuhisa[1], Osamu Miyashita[1], Florence Tama[1,2] ([1]*AICS, RIKEN*, [2]*Department of Physics, Nagoya University*)

Coherent diffraction patterns observed by X-ray free electron laser (XFEL) provide information on biomolecular conformations and dynamics. We are exploring new algorithms which enable structure modeling from limited experimental data by solving inverse problems. We need to evaluate agreements between the noisy and sparse experimental data and a large number of candidate models. Both speed and accuracy of the similarity detection is important for realizing this strategy. Previously, we have shown that the similarity detection algorithm between 2D diffraction patterns is effective in this approach. In this meeting, we will present new multistep algorithms to quickly evaluate the agreement between diffraction patterns using 1D intensity profile.

3Pos037 Molecular determinants of the ATP binding properties of the ε subunit from bacterial ATP synthases

Alexander Krah[1,2], Yasuyuki Kato-Yamada[3], Changbong Hyeon[2], Shoji Takada[1] ([1]*Dept. Biophys., Kyoto Uni.*, [2]*KIAS*, [3]*Dept. Life Sci., Rikkyo Uni.*)

The main producer of ATP, the universal energy source in all living cells, F-type ATP synthases are driven by an electrochemical gradient across the membrane. Vice versa, this enzyme can hydrolyze ATP to maintain this electrochemical gradient. To prevent a shortcut of ATP, in most bacteria subunit ε of the ATP synthase undergoes a large conformational change from the non-inhibitory down-state (ATP bound to ε) to the ATPase inhibitory up-state (ATP released from ε) when the ATP concentration passes a certain threshold. The ATP binding strength and thus ATP release from subunit ε in various organisms varies from the μM to the mM range. Here we discuss the structural basis for the divergent ATP binding properties obtained by MD simulations.

3Pos038 Microtubule stability and the tubulin molecule interactions within the microtubule lattice

Kenta Hirasada, Daisuke Yamamoto, **Miho Katsuki** (*Fukuoka Univ., Faculty of Science*)

Tubulin molecules assemble into longitudinal protofilaments (pf) within microtubules (MT) with adjacent tubulins interacting laterally and longitudinally. We previously demonstrated that Mal3 protein (homologue of EB1) induces a more seam-like lattice packing.
In the presence of Mal3 MTs sliding on a kinesin surface were more bent during sliding suggesting that pfs may have freedom to shift longitudinally after their incorporation into the lattice. Atomic force microscopy showed that MTs split into individual protofilaments, which then shrink independently. These observations suggest that the tubulin lateral interactions between pfs are weaker than the longitudinal interactions within pfs and that MT stability might be controlled by at least two layers of mechanism.

3Pos039 隣り合うチューブリン存在下と非存在下における α/β チューブリン C 末端の異なる三次元空間分布
Three-dimensional distributions of α/β-tubulin C-terminal tails and the influence of neighboring tubulins

Koji Umezawa[1,2], Yukinobu Mizuhara[3], Jun Ohnuki[3], Mitsunori Takano[3] ([1]*Grad. Sch. of Sci. & Tech., Shinshu Univ.*, [2]*IBS, Shinshu Univ.*, [3]*Grad. Scl. of Adv. Sci. & Eng., Waseda Univ.*)

Microtubule is a tubular assembly of α and β tubulin heterodimer. The C-terminal tail (CTT) of α and β tubulin is related to the assembly. However, the role of CTT in the assembly formation is elusive because CTT is disordered. Then, we have investigated the three-dimensional distribution of CTT by molecular dynamics simulation for the heterodimer in the absence or the presence of neighboring heterodimers. The result of a single heterodimer showed that CTT of α-tubulin (αCTT) distributed differently than that of β tubulin (βCTT). The αCTT lay on the longitudinal surface of microtubule while the βCTT covered the lateral surface. In presence of neighboring heterodimers, αCTT and βCTT shifted its location and interacted with an adjacent tubulin, stabilizing the assembly.

3Pos040 F アクチンの水和状態は Mg^{2+}/Ca^{2+} イオンに強く依存する
Strong Mg/Ca Ion Dependence of Hydration State of F-actin

Makoto Suzuki[1], Asato Imao[1], George Mogami[1], Ryotaro Chishima[1], Takahiro Watanabe[1], Takaya Yamaguchi[1], Nobuyuki Morimoto[1], Tetsuichi Wazawa[2] ([1]*Grad. School of Eng. Tohoku Univ.*, [2]*The Institute of Scientific and Industrial Research, Osaka University*)

Hydration-state difference of F-actin by the bound divalent cations has been revealed through precision microwave dielectric relaxation (DR) spectroscopy. G- and F-actin in Ca- and Mg-containing buffer solutions exhibit dual hydration components comprising restrained water and hypermobile water (HMW). The hydration state of F-actin is strongly dependent on the ionic composition. In every buffer tested, the HMW signal Dhyme of F-actin is stronger than that of G-actin. Dhyme value of F-actin in Ca2mM-buffer is markedly higher than in Mg2mM-buffer. Moreover, in the presence of 2 mM Mg^{2+} ion, the hydration state of F-actin is changed by adding a small fraction of Ca^{2+} ion (0.1 mM) and becomes closer to that of the Ca-bound form in Ca2.0-buffer.

3Pos041 アクチンフィラメントに結合したローダミンファロイジン蛍光のゆらぎ
Fluctuation of rhodamine-phalloidin fluorescence along actin filaments

Taro Ueda[1,3], Saku Kijima[2,3], **Takahiro Suzuki**[1] ([1]*Dept. of Physics, Waseda Univ.*, [2]*Biomed. Res. Inst., AIST*, [3]*Grad. Sch. Life Environ. Sci., Univ. of Tsukuba*)

Fluorescence intensity (F.I.) of rhodamine (Rh) is affected by its local environment, and the F.I. of Rh-phalloidin (RhPh) is enhanced by binding to actin filaments. During the initial phase of the binding process of RhPh to actin filaments observed by 30 s-interval time lapse imaging, F.I. was non-uniform and punctate along the filaments, and moreover, the fluorescent puncta apparently moved along the filaments. When filaments fully bound with RhPh were partially bleached, the fluorescence was again punctate, and the puncta moved along the filaments. The puncta movements were suppressed when filaments were fixed with glutaraldehyde, suggesting that the puncta movements reflect conformational dynamics of actin filaments that affect the local environment of Rh.

3Pos042 OH 伸縮振動のラマン分光によるミオシン S1 および他のいくつかのタンパク質の水和状態の測定
Hydration study on myosin subfragment-1 (S1) and some other proteins by Raman OH-atretching spectroscopy

Yuki Ochiai[1], George Mogami[1], Tetsuo Taniuchi[2], Makoto Suzuki[1] ([1]*Grad. Sch. Eng., Univ. Tohoku*, [2]*IMRAM, Univ. Tohoku*)

In this study, Raman OH-stretching spectroscopy was used to analyze the hydration state of myosin S1 and some other proteins, lysozyme, ovalbumin, BSA at 293K. Firstly, each protein dissolved in DMSO was measured to assign the non-water Raman bands such as CH-stretching (2900 - 3100 cm-1) and fluorescence bands emitted by the protein in the frequency range from 2700 to 4000 cm-1. After removing the non-water bands from the raw Raman spectrum, the spectrum between 2700 and 4000 cm-1 was decomposed into the bulk water component and the hydration layer component. The hydration numbers of these proteins were in good agreement with the results obtained by the previous dielectric relaxation studies.

3Pos043 ウルトラファインバブル水中のタンパク質構造の研究

Study of protein structure in ultra-fine bubble water

Mitsuhiro Hirai[1], Satoshi Ajito[1], Kosuke Takahashi[1], Noboru Ohta[2], Tatsuo Iwasa[3] ([1]*Grad. Sch. Sci. Tech., Gunma Univ.*, [2]*Jpn. Syn. Rad. Res. Inst.*, [3]*Muroran Inst. Tech.*)

Ultra-fine bubble (UFB) is usually defined as small bubbles with the diameter less than 1 micro-m, which is different from microbubbles generally having approximately 1 ~ 100 micro-m in diameter. UFB is colorless and transparent to the naked eye, and is stable for several weeks or so in solutions. Recently, the industrial applications of UFB have been focused and highlighted in the various fields such as agricultural and fishery industries, and medical therapy. However, the detailed mechanism underlying the performance of UFB, especially the effect of UFB on biological materials, is not known. Therefore, we investigated the effect of UFB on protein structure by using small-and-wide angle X-ray scattering.

3Pos044 3D−RISM 理論を応用した溶液中における Met−enkephalin の構造揺らぎの解析

Analysis of structural fluctuations of Met-encephalin in the solution phase by means of 3D-RISM theory

Masatake Sugita[1], Fumio Hirata[2] ([1]*Dept. of Bioinfo., Col. of Life Sci., Ritsumeikan Univ.*, [2]*Toyota Phys. & Chem. Res. Inst.*)

Recently, B. Kim and F. Hirata derived a new statistical mechanics formulation of characterizing the structural fluctuation of a complicated solute correlated with the molecular solvents based on the Generalized Langevin Equation and 3D-RISM theory. This formulation suggests that the Hessian matrix of the free energy corresponds to the inverse of the variance covariance matrix of the solute molecule in the solution phase.
In this study, we analyze structural fluctuation of Met-enkephalin immersed in water by calculating the second order derivative of the solvation free energy in addition with the potential energy, and diagonalizing the hessian matrix. After that comparing the results with those from the Normal Mode analysis and MD simulation.

3Pos045 分子動力学シミュレーションとエネルギー表示理論を用いた共溶媒変性効果の自由エネルギー解析

Free energy analysis of cosolvent effect through molecular dynamics simulation and energy-representation method

Yu Yamamori, Nobuyuki Matsubayasi (*Grad. Sch. of Eng. Sci., Osaka Univ.*)

We tackled the long-time problem how urea works to destabilize the native structure of protein when it is added to pure-water solvent. We propose the clear answers: the urea-induced denaturation is governed by the direct mechanism through the van der Waals interaction without the decisive role of the backbone or side chain. What should be emphasized is that this conclusion is based on the fine-grained (all-atom) molecular dynamics simulation and the high-precision calculation of the transfer free energy of a protein from pure-water solvent to urea-water mixed solvent with energy-representation method.

3Pos046 タンパク分子内情報伝達を実現する構造基盤の探索—既知の構造から

Search for Common Structural Basis of Mechanical Communication in Proteins: from Known Structures

Yuichi Togashi (*RcMcD, Hiroshima Univ.*)

Communication within protein machines is ubiquitous, and a representative case is allosteric regulation in enzymes. Roles of mechanics in such communication are particularly interesting. We have developed a simple scheme to detect mechanical communication by using steered molecular dynamics simulations of coarse-grained elastic network models of proteins (Y. Togashi, NOLTA IEICE, 2016). To explore real allosteric enzymes, first, we apply this method to a class of known ligand binding sites of proteins, to extract their common mechanical communication patterns. In this presentation, the methodology and current results will be presented.

3Pos047 CAPAXIS と PyMOL を用いたウイルス粒子脱殻の描画・操作

Modeling of uncoating of virus capsid by using CAPAXIS and PyMOL

Shunsuke Sato[1], Aya Kosugi[1], Go Wabanabe[2], Shigetaka Yoneda[2] ([1]*Grad. Sch. Sci., Kitasato Univ.*, [2]*Sch. Sci., Kitasato Univ.*)

We have developed software library, CAPAXIS, for analysis on icosahedral symmetric virus capsid structures. CAPAXIS has functions such as classification of rotation axes, calculation of cell numbers, generation of the entire structures and superposition of capsid structures with different coordinate axes. CAPAXIS has been implemented in the molecular visualization system, PyMOL. We added new developments on CAPAXIS to model deformation of virus capsid structure in the uncoating process. Moreover, rotation axes and positions in the crystal cells for all the 512 capsid structures in the ViperDB were analyzed with CAPAXIS and inter-capsid interaction was clarified.

3Pos048 生体分子の分子動力学計算を取り扱う高速な QM/MM 理論の開発

Development of rapid QM/MM approach for biomolecular simulations

Hiroaki Nishizawa[1], Hisashi Okumura[1,2] ([1]*IMS*, [2]*Sokendai*)

A quantum mechanical/molecular mechanical (QM/MM) approach based on the density-functional tight-binding (DFTB) theory is a useful tool for analyzing chemical reaction systems in detail. In this study, an efficient QM/MM method is developed by the combination of the DFTB/MM and particle mesh Ewald (PME) methods. Because the Fock matrix, which is required in the DFTB calculation, is analytically obtained by the present method, the Coulomb energy is accurately and rapidly computed. For assessing the performance of this method, DFTB/MM calculations are conducted for a system consisting of two amyloid-β peptides and a zinc ion in explicit water under periodic boundary conditions. Furthermore, the behavior of the zinc ion is compared between the MM and DFTB/MM calculations.

3Pos049 タンパク質の基準振動モードのネットワーク解析：中心性指標の計算

Network analysis of normal modes of proteins: calculations of various centrality measures

Hiroshi Wako[1], Shigeru Endo[2] ([1]*Sch. of Soc. Sci., Waseda Univ.*, [2]*Sch. of Sci., Kitasato Univ.*)

One of the challenging problems in protein dynamics is how to characterize internal motions of a protein. For example, a conformational change from apo to holo states in enzyme observed in their PDB data is known to be associated with some of the lowest-frequency normal mode motions. It implies that characterization of the lowest-frequency normal modes of a protein is useful to find possible motions to be related to its function. We defined a network of residues in a protein for each normal mode based on the correlative motions among residues, and then applied network analysis methods. In particular, we calculated various centrality measures such as degree and betweenness to characterize it, expecting to reveal the relationships between structural dynamics and function.

3Pos050 タンパク質構造変化における経路の多様性：分子動力学シミュレーションによる解析

A variety of pathways for a conformational change of a protein investigated by molecular dynamics simulation

Sotaro Fuchigami (*Grad. Sch. of Medical Life Science, Yokohama City Univ.*)

Conformational changes in proteins are often crucial for their function. When a large conformational change such as a domain motion occurs, its pathway is not identical but diverse because of the intrinsic flexibility of a protein. However, the molecular details remain unclear. In the present study, we selected lysine-, arginine-, ornithine-binding protein as a target protein which undergoes large domain motions, and performed all-atom molecular dynamics simulations from the closed conformation without ligands in explicit water. Almost all trajectories showed conformational changes from the closed form to the open form, some of which occurred immediately and others slowly. We will discuss the similarity and differences in the pathways of conformational change.

3Pos051 カメレオンモデルによる NtrC の構造転移機構の研究

Mechanism of conformational transition of NtrC studied by using chameleon model

Shinya Abe[1], Atsushi Mizuno[2], Masaki Sasai[1], Tomoki P. Terada[1] ([1]*Dept. Comput. Sci. Eng., Grad. Sch. Eng., Nagoya Univ.*, [2]*Dept. Appl. Phys., Sch. Eng., Nagoya Univ.*)

Nitrogen regulatory protein C (NtrC) is a single-domain protein which exhibits two-state allosteric behavior involving the rearrangement of an α-helix. It has been argued that formation of nonnative hydrogen bonds is required for the conformational transition of NtrC, which is in contrast to the cracking mechanism with local unfolding, proposed for a multi-domain protein adenylate kinase. We have applied the chameleon model, which is a coarse-grained model to describe free energy landscape of conformational transition (Terada et al., J. Phys. Chem. B (2013)), to characterize the allosteric transition of NtrC. We will discuss the contribution of nonnative contacts and local unfolding in the transition state ensemble between inactive and active states of NtrC.

3Pos052 Oct4 の 2 つの DNA 結合サブドメインを結ぶ柔軟な linker 領域の自由エネルギー地形

Free-energy landscape of the flexible linker connecting two DNA-binding subdomains of Oct4

Tomonori Hayami[1,2], Shoji Takada[3], Haruki Nakamura[1], Junichi Higo[1] ([1]*Inst. Protein Res., Osaka Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Dept. Biophys., Grad. Sch. Sci., Kyoto Univ.*)

Oct4 is known as a transcription factor required for making induced pluripotent stem cells (iPSCs). An Oct4 DNA-binding domain consists of two DNA-binding subdomains connected by a linker, which is an intrinsically disorder region.
A crystal structure shows that the N-terminal part of the linker adopts helix when the Oct4 DNA-binding domain binds to DNA. It was proposed that the linker has a crucial role for reprogramming to pluripotency, although the linker does not interact with DNA directly.
We obtained a conformational ensemble of the linker by performing enhanced conformational sampling, virtual-system coupled adaptive umbrella sampling (V-AUS), and computed the free-energy landscape at 300 K. Conformational changes occurring in the linker will be discussed.

3Pos053 タンパク質の協同的な折れたたみとループのつながり方の関係

Relation between cooperative protein folding and loop connections

Nobu C. Shirai[1], Shintaro Minami[2] ([1]*Center for Information Technologies and Networks, Mie University*, [2]*Graduate School of Information Science, Nagoya University*)

When we talk about protein folding, we tend to focus on hydrophobic cores of a protein structure. How about loops? Minami et al. suggested that loop connections between secondary structure elements (SSEs) in hydrophobic cores are important for robustness against mutations in their database analysis. In order to investigate the effect of loops on protein folding, we compared thermodynamic stability of proteins with different loop connections by coarse-grained simulations. We constructed Go-like models of a hydrophobic core composed of a α-helix layer and a β-sheet layer, and then added five loops connecting the SSEs in several ways. As a result of simulations, it was shown that number and position of layer-crossing loops change the cooperativity of protein folding.

3Pos054 酸曝露後中和による抗体のフォールディングと凝集: 二種の光子相関分光法による追跡

Folding versus aggregation of an antibody initiated by pH-shift stress: Double tracking by photon correlation spectroscopies

Hiroshi Imamura, Akira Sasaki, **Shinya Honda** (*Natl Inst Adv Indust Sci. Tech.*)

Acid treatment in the purification process of an antibody drug induces its unfolding and aggregation, the mechanism of which is yet to be elucidated. By using static and dynamic light scattering, we found that evolution of the aggregates proceeded via two phases: a rapid formation of oligomers initiated by the neutralization of pH was followed by a slow growth of the aggregates. The aggregates were characterized as a fractal with the dimension of 1.98. Tracking the dye-labeled monomer by using fluorescence correlation spectroscopy revealed that the folded monomer was not involved in the aggregates, while the unfolded monomer was. We proposed a model of the antibody aggregation, incorporating folding versus aggregation into Smoluchowski aggregation kinetics.

3Pos055 NMRを用いた血清環境での相互作用解析

NMR approach for understanding protein interactions in serum environments

Saeko Yanaka[1,2], Rina Yogo[1,2], Hirokazu Yagi[1,2], Koichi Kato[1,2] ([1]*Department of Bioorganization Research, Okazaki Institute for Integrative Bioscience and Institute for Molecular Science, National Institutes of Natural Sciences*, [2]*Department of Structural Biology and Biomolecular Engineering, Graduate School of Pharmaceutical Sciences, Nagoya City University*)

In the blood stream, secretory proteins interact with various components to activate the immune system. Methods to directly observe interactions in the serum are scarce, hampering the understanding of the immune system. Using NMR, we observed HSQC spectra of 15N-labeled IgG domains in the serum environments. Our data indicated selective perturbations of HSQC peaks, enabling identification of the sites involved in interactions with serum components. Interestingly, human polyclonal IgG caused similar spectral perturbations. This suggests that the IgG domains carry antigenic determinants recognized by a subset of endogenous antibodies, demonstrating that the NMR method enables identification of their epitopes dealing with heterogeneous systems.

3Pos056 酸化と酵素切断がLDLの物性に与える影響

Physical properties of low-density lipoprotein after oxidation or proteolytic enzyme treatment

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Oxidized low-density lipoproteins (LDLs) is a potent risk factor for the development of cardiovascular disease. Recently, using AFM, we reported that elastic modulus of LDL is decreased by its metal oxidation. However, the mechanism of this change is not well investigated. We postulated that the fragmentations of apolipoprotein B-100 might be a reason for this change. In this study, we measured the elastic modulus of LDL particles before and after incubation with enzymes that induce the fragmentation of LDL. We also measured DSC to observe the lipid phase transition of LDL before and after the enzyme treatment. We would like to discuss the effect of fragmentation of the apolipoprotein on the elastic modulus change of LDL and phase transition of the lipid in LDL.

3Pos057 イムノグロブリンGのマルチドメイン構造形成におけるエントロピー効果

Entropic stabilization of the multi-domain architecture in immunoglobulin G

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Immunoglobulin G (IgG) is a multi-domain protein, where several homo- and hetero-dimeric interactions are involved. We showed the multi-domain architecture of the Fc region entropically enhanced the conformational stability of CH3 domain. We conducted a guanidine-induced unfolding reaction against the Fc region, isolated CH2 monomer and CH3 homo-dimer proteins. After reaction equilibrium, the unfolded fractions of CH3 domain in the Fc region at each guanidine concentration were apparently smaller than those of isolated CH3 homo-dimer protein at the same protein concentration. We concluded that this stabilization would stem from the proximity effect of two CH3 domains in the Fc region, which increased the effective concentration of the dimerization reaction.

3Pos058 統計熱力学に基づいたサーモフィリックロドプシンの熱安定化変異体の作製

Identification of thermostabilizing mutations for thermophilic rhodopsin based on statistical thermodynamics

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Thermophilic rhodopsin (TR), which is a 7-transmembrane protein derived from the extreme thermophilic bacterium thermus thermophilus JL-18 (Optimum growth temperature is about 75 degrees Celsius), absorbs visible light and functions as a proton pump. It features exceptionally high thermostability. On the basis of the high-resolution structure of TR recently determined by us (T. Tsukamoto et al., JBC (2016)), we predicted even more stabilizing mutations of TR using our physics-based free-energy function (S. Yasuda et al., J. Phys. Chem. B (2016)). The thermostability of the mutations was then measured in experiments: Some of them actually exhibited higher thermostability than wild type. Its implication will be discussed.

3Pos059 Amorphous aggregation of cytochrome c with inherently low amyloidogenicity is characterized by the phase diagram

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Despite extensive investigations on cytochrome c (Cytc), the mechanisms of its aggregation remain largely unknown. We herein examined the aggregation of three physiologically-relevant types of Cytc and its amyloidogenic fragment.
Although the aggregation propensity of holo-Cytc was low due to high solubility, markedly unfolded apo-Cytc, lacking heme, strongly promoted amorphous aggregation with increases in hydrophobicity. Ag-bound Cytc increased fibrillar aggregation. However, mature amyloid fibrils were not detected for any of the Cytc variants or its fragment. These revealed the intrinsically low amyloidogenicity of Cytc. Moreover, the phase diagram constructed using solubility and aggregate types is useful for a comprehensive understanding of protein aggregation.

3Pos060 The virial coefficients based on the rotaitional diffusion as a criterion of the protein crystallizaton

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The virial coefficients are useful parameters for describing protein-protein interactions. We formulated a virial coefficient based on the rotational diffusion of lysozyme (HEWL) and examined closely its validity when used as a criterion for the protein crystallization. The rotational diffusion coefficients were determined using the steady state and time-resolved fluorescence anisotropy of HEWL stained by a fluorescence probe. The virial coefficients were evaluated from the linear and/or quadratic dependences of the relative rotational diffusion rates (D_{rot}/D^0_{rot}) on the HEWL concentration. The resulting virial coefficients showed the attractive interactions were induced according to the species of monovalent cations closely correlating with the HEWL crystallization.

3Pos061 The hydration state near the binding site of human Serum Albumin revealed by the time-resolved fluorescence spectrum of Trp214

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Protein stability and interactions with small molecules would be influenced by the hydration of proteins surface. However, the detail has not been cleared because analyzing methods is limited. Trp residue is a powerful probe for studying the protein hydration because it shifts the fluorescence maximum according to the interaction with surrounding water molecule(s). The time required for the spectral shift can reveal well the hydration state around Trp. Here, we estimated the hydration of HSA near the Trp214 by measuring the time-resolved fluorescence spectrum by the methods using streak scope and global analysis of time correlated single photon counting through the full spectral region. Furthermore, the correlations with the ligand binding were investigated.

3Pos062 アクチンフィラメントの圧電特性 III
Piezoelectric property of an actin filament III

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Actin filaments are shown to alter the affinity with actin-binding proteins (ABPs) such as myosin and cofilin in response to external forces, implying that actin plays a key role in mechanobiology. However the physical mechanism remains unclear. In our previous molecular dynamics (MD) study, we presented a novel concept of “piezoelectric allostery” where a locally applied force causes an electrostatic response of myosin allosterically (Ohnuki et al., Phys. Rev. E, 2016), which was also observed in an actin filament (Annu. Meeting, 2015). We here examine by MD simulation whether the actin-myosin interaction is affected by the piezoelectric effect of the actin filament.

3Pos063 テトラヒメナ外腕ダイニン重鎖（Dyh3p)における運動系の開発と運動特性
Motor domain-based motility system and motile properties of alpha heavy chain in Tetrahymena outer arm dynein

Masaki Edamatsu (*Department of Life Sciences, Graduate School of Arts and Sciences, The University of Tokyo*)

Axonemal dyneins power ciliary motility and their motor functions are regulated by accessary proteins bound to the tail regions. To understand the essential properties of dynein motility, the functional motor domain of the alpha heavy chain (Dyh3p) in Tetrahymena outer arm dynein was purified, and the motile properties were examined using an in vitro motility system. The purified protein caused microtubules to glide at a velocity of 5.0 µm/s with their minus-end trailing. In addition, the motility was inhibited in an ATP concentration-dependent manner, which is in contrast with kinesin-1. This method will enable further molecular studies on diverse axonemal dyneins and ciliary motility.

3Pos064 タンパク質の NMR 解析が困難な系にも有効なシグナル帰属法
Signal assignment strategy for protein NMR under challenging conditions

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NMR analyses of proteins are interfered with enhanced relaxation of large proteins, proteins in molecular crowding environments, and so on. Therefore it is important to develop analysis strategies only with relaxation-tolerant experiments. It is also desirable to shorten measurement time for unstable targets, or for maximal signal gain per time. We have developed an amino-acid selective isotope labeling strategy, Stable Isotope Encoding (SiCode). This enables amino-acid typing only with relaxation-tolerant two-dimensional spectra of small number of labeled samples, which helps to reduce total measurement time, even from low signal-to-noise-ratio spectra by model fitting analysis. We will demonstrate this strategy under challenging conditions such as in-cell NMR.

3Pos065 二量子遷移 EPR 距離測定における短距離成分の影響
Effects of Short Distance Components on Double Quantum Coherence EPR Distance Measurements

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In EPR distance measurements method, two aminoacid residues are labeled by nitroxide spin labels by using the site directed spin labeling method (SDSL), and the distance between the labeled residues is obtained from an analysis of the dipolar interaction between the electron spins on the two labels. Using the double quantum coherence (DQC) or the electron-electron double resonance (DEER) method, we can detect a distance up to ~8 nm, because these methods can measure very small dipolar interactions. On the other hand, for a short distance less than ca. 2 nm, a large splitting causes difficulties in the analysis of distances. In this paper, we discuss these problems and effects on the resulting distance distribution data.

3Pos066 天然変性タンパク質の SAXS プロフィル評価法を開発するための新たな枠組み
A novel framework for developing the evaluation method of SAXS profile of IDP

Yasutaka Seki[1], Shigeyoshi Nakamura[2] ([1]*Kochi Med. Sch.*, [2]*Kitakushu Nat. Coll. of Tech.*)

Small-angle X-ray scattering (SAXS) has long been used as an important means for determining global-structure parameters of biopolymers in solution. It can be applied not only to natively globular proteins but also to intrinsically disordered proteins (IDP). In the latter case, however, we need to evaluate a large number of SAXS profiles for averaging them, because the IDP consists of an ensemble with diverse conformations. It is possible to evaluate high accuracy SAXS profiles by using MD simulation, but the computational cost is impractically high for IDPs. We propose a new framework for developing the evaluation method of SAXS profiles with high accuracy and low computational cost. In the framework, the SAXS profiles by using MD is employed for reference data.

3Pos067 SAXSとアミノ酸残基レベル二次構造情報からのタンパク質立体構造の構築
Protein structure constructed with SAXS and secondary structures at amino acid residue level

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Previously we developed a novel algorithm for constructing protein structures with small-angle X-ray scattering (SAXS) restraints, and implemented as SAXS_MD. We applied this program to nine proteins with various fold, and performed the molecular dynamics calculation with SAXS and NMR-derived secondary structures as restraints to successfully construct appropriate structures as accurate as 3 - 4 Å of RMSD. In this study we used the secondary structural information at amino acid residue level obtained from such as VUV-CD, instead of NMR, and calculated restrained-MD along with SAXS information. For all-alpha proteins, the resultant structure was as accurate as 5 Å of RMSD.

3Pos068 タンパク質複合体の解離過程の分子動力学
A Steered Molecular Dynamics to Understand the Dissociation Process of Protein Complex

Yutaka Ueno, Yuki Mochizuki (*AIST Kansai, Biomedical Research*)

On evaluating the binding affinity of the protein to its target molecule based on atomic models, we studied a dissociation process by molecular dynamics simulations. Since a slow dissociation rate directly contributes its strong binding affinity, the simulation helps us to design molecular interactions. Using a conventional molecular dynamics program, constant force pulling for a molecular fragment that bind to the protein was applied. The dissociation event was smoothly accelerated by a temperature control, yielding distortions of the interacting molecular surface. Calculated dissociation times for several proteins were compared with the dissociation rate constant reported by surface plasmon resonance experiments.

3Pos069 脂質ナノディスクとZMW法を用いた高濃度リガンドでの計測が可能な膜タンパク1分子計測系の構築
Nanodiscs platform on ZMWs for single-molecule imaging of membrane proteins at high ligand concentration

Keisuke Tsukada, Kazushi Isomura, Tomotaka Komori, Sotaro Uemura (*Dep. Bio. Sci., Grad. Sch. Sci., Univ. Tokyo*)

The single molecule measurements of membrane proteins have been widely studied. However, the background fluorescent noise has hindered single-molecule observation at physiological concentrations of fluorescent ligands. We have combined Zero-mode waveguides (ZMWs) and the Nanodisc technology to overcome this limitation. ZMWs are nanoscale structures that give sufficient signal/noise ratios for imaging even in the presence of several micromolar fluorochromes. Using the techniques, we succeeded in visualizing the interaction of single cytochrome P450 3A4 molecules and fluorescent ligands. We will discuss the properties of the second binding site on the protein, which is expected to be bound only at physiological concentration of the ligand and exhibit cooperativity.

3Pos070 X線1分子追跡法によるTRPV1チャネルの分子運動解析
3D Motion Maps of TRPV1 cation channel depicted by Diffracted X-Ray Tracking Method

Kazuhiro Mio[1], Keigo Ikezaki[2], Hiroshi Sekiguchi[3], Yuhuku Matsushita[2], Tai Kubo[1], Yuji C. Sasaki[2] ([1]*Molprof, AIST*, [2]*Frontier Science, Adv. Material Sci., Univ. of Tokyo*, [3]*JASRI*)

The TRPV1 is a nonselective cation channel that responds to various signals. Structure of TRPV1 was recently revealed in atomic resolution by the cryo-EM, however gating mechanisms are largely unknown. To understand the dynamics in channel function, we adopted the Diffracted X-ray Tracking (DXT) technique. Individual protein was labeled with gold nanocrystals, and the motion of X-ray diffraction spots from the crystal were investigated as intramolecular movement. TRPV1, which was introduced “Met tag” for labeling nanocrystal and “His tag” for substrate absorption, was expressed in HEK293, purified, and immobilized on the Ni-NTA coated polyimide substrates. Intramolecular motions against capsaicin, acidic conditions, and activating temperature were analyzed.

3Pos071 細胞性粘菌の生きた細胞での膜タンパク質の拡散の網羅的解析
Comprehensive Diffusion Analysis of Membrane Proteins in Living Dictyostelium Cells

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Protein diffusion in cell membranes plays a key role in many signaling processes. To clarify the relationship between diffusion and membrane protein’s structure in living cell membrane, I measured diffusion coefficients of 24 kinds of membrane proteins in cell membrane by using single molecule imaging and I estimated the number of diffusion state which each membrane protein has. In comparison with theoretical models in artificial membrane, fastest diffusion state of each membrane protein obeyed the Saffman-Delbruck model which predicts a logarithmic dependence of a protein’s diffusion coefficient on its inverse hydrodynamic radius. This result suggested membrane proteins in fastest state diffuse in cell membrane’s region which is similar to artificial membrane.

3Pos072 細菌Ⅲ型分泌装置の回転運動によるエフェクター輸送の制御
Rotation of needle-like type III secretion apparatus directly regulates its effector transport

Takashi Ohgita, Kohei Fukuda, Kyoko Momiyama, Naoki Hayashi, Naomasa Gotoh, Hiroyuki Saito (*Kyoto Pharm. Univ.*)

Bacteria inject effectors to induce infection into host cells via type III secretion apparatus (T3SA). Previously, we found out that T3SA rotates like flagellum (FASEB. J. 27, 2013). Based on the simulation suggesting that hydrophobic helix in T3SA is required for effector transport, it is hypothesized that effector transport would be regulated by T3SA rotation. In this study, we quantified the transport speed to examine this. Our results demonstrated that effectors are transported at ≈6 molecules/min, leading to the estimation of rotation speed of T3SA bearing microbead to be ≈10 s/rotation. Since this estimation roughly agrees with the rotation speed (≈20 s /rotation) observed previously, it is suggested that effector transport is directly regulated by T3SA rotation.

3Pos073 抗体修飾ナノニードルと AFM を用いた引っ張り試験による細胞骨格の機械的特性の解析

Analysis of mechanical property of cytoskeleton by tensile test for intermediate filament using antibody-modified nanoneedle and AFM

Moe Susaki[1], Itaru Takeda[2], Ayana Yamagishi[3], Yuta Takano[2], Tomoko Okada[3], Chikashi Nakamura[1,2,3] ([1]*Fac. Eng., Tokyo Univ. Agric. Technol.*, [2]*Grad. Sch. Eng., Tokyo Univ. Agric. Technol.*, [3]*Biomed. Res. Inst., AIST*)

Nestin, an intermediate filament protein, is suspected to be involved in metastasis of cancer cells. We found that knockout of nestin gene in highly metastatic breast cancer cell SC2 caused an increase in elastic modulus of the cell body, resulting in lowered infiltrative property. In order to elucidate nestin's contribution to mechanical property of cytoskeletal structure, we investigated vimentin which co-polymerizes with nestin. By using AFM and anti-vimentin-antibody-modified nanoneedle, we performed tensile test for vimentin filament in living cells and evaluated mechanical property of cytoskeletal network containing vimentin. Between wild type SC2 and nestin-knockout cell, obviously different shapes of force curves were obtained in the test.

3Pos074 テンダム遺伝子を用いた無細胞タンパク質発現ノイズの解析

Stochastic gene expression in cell-free system by tandem genes

Shiori Fujimoto, Yi Zhang, Kazuhito Tabata, Hiroyuki Noji (*Grad. Sch. Eng., Univ. Tokyo*)

Gene expression noise can be categorized into extrinsic noise and intrinsic noise. The fluctuations in the amount of components involved in gene expression are the primary factor of extrinsic noise, resulting in a positively correlated fluctuation. The intrinsic noise results from the inherent stochastic biochemical process, resulting in an uncorrelated fluctuation.
We prepared a DNA sequence composed of tandemly arranged two genes to investigate the cause of gene expression noise in cell-free system. The tandem structure ensures that the same copy numbers of different genes can always be encapsulated into each compartment. We confirmed a strong correlation in expression and this preliminary result suggests that extrinsic noise is predominant in the cell-free system.

3Pos075 凍結トラップ結晶構造解析と時間分解分光を用いた P450nor の反応中間体の解析

Reaction Intermediate Analysis of P450nor Using Freeze-Trap X-ray Crystallography and Time-Resolved Spectroscopy

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P450nor is a heme enzyme that catalyzes the reduction of NO to N_2O in the denitrification. In the reaction, NO is reduced by two electrons directly transferred from NADH. The resulting highly-activated species (intermediate-I) is unique and has been of high interest in coordination chemistry, but its electronic and geometric structures are not yet known. Here, we succeeded in solving the crystal structure of intermediate-I. We performed single-shot time-resolved visible microspectroscopy for a crystal using caged-NO at 20°C and found that intermediate-I appears in the crystal very slowly (on the second timescale), which allowed for its freeze-trapping. To further analyze the electronic and protonation states of intermediate-I, time-resolved IR spectroscopy is on-going.

3Pos076 結晶状態ヘモグロビンの大規模四次構造変化の直接観測

Direct observation of large-scale quaternary motions of hemoglobin in a crystalline state

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Many proteins undergo large conformational changes that are relevant to their functions. The most famous example is the T (tense) to R (relaxed) transition in human hemoglobin. However, the transition path between these two end states is still unknown, which limits our understanding of the allosteric mechanism. To monitor directly this process, we introduce a novel crystal form that allows quaternary conformational changes occurring in the internal hemoglobin molecules. A combination of such crystals and X-ray crystallography enables the observation of large-scale quaternary motions induced by changing external conditions. Our findings give a comprehensive picture of the equilibrium conformers and transition pathway for hemoglobin.

3Pos077 構造状態と関係したヘモグロビンのピコ秒ダイナミクスの変化

Changes in the picosecond dynamics of hemoglobin related to the structural states

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Hemoglobin (Hb) is an allosteric protein, which adopts two different quaternary states, the tense (T-) and relaxed (R-) states. The structures and functions of these two states are well characterized, but much less is known about their dynamics, particularly on the pico- to nano-second time scales. To investigate how the dynamics is related to these structural states, we carried out quasielastic neutron scattering experiments on human deoxy-Hb and CO-Hb, which correspond to the T- and R-states, respectively, using the backscattering spectrometer BL02 (DNA) at MLF/J-PARC. It was found that the dynamics are different between deoxy-Hb and CO-Hb such that the dynamics of CO-Hb is enhanced. This implies that the dynamics change is involved with the mechanism of allostery.

3Pos078 四量体ヒトヘモグロビンにおける 2 つの α 鎖に特有の Fe-His 結合と四次構造との関連

Distinct Fe-His bond of two α subunits in human $\alpha_2\beta_2$ tetramer hemoglobins and their quaternary structures

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We investigated properties of α chain by using β-mutant half-met hemoglobin M (Hb M), in which heme iron of β chain is occupied by Fe^{3+} instead of Fe^{2+}. Hb Ms are excellent specimens to elucidate a regulation mechanism of oxygen affinity of α chain discriminatively, since Hb Ms are valence-hybrid ($Fe^{2+}Fe^{3+}$) binding O_2 only to Fe^{2+}. Three half-met Hb Ms examined include Hb M Hyde Park (βH92Y), Hb M Saskatoon (βH63Y) and Hb M Milwaukee (βV67E). In the present study, we point out appreciable heterogeneity of Fe^{2+}-His frequency in contrast to undetectable one of normal β chain in $Fe^{3+}Fe^{2+}$ and discuss a relation between the heterogeneity and O_2 binding properties of Hbs M and other Hbs which can bind O_2 to Fe^{2+} of α chain only.

3Pos079 神経保護作用を持つヒトニューログロビンとヘテロ三量体 G_i 蛋白質 α サブユニットとの相互作用に重要なアミノ酸残基の特定

Identification of residues crucial for the interaction between human neuroprotective protein “neuroglobin” and $G\alpha_i$

Nozomu Takahashi, Keisuke Wakasugi (*Dep. of Life Sci., Grad. Sch. of Arts and Sci., Univ. of Tokyo*)

Neuroglobin (Ngb) is a globin widely expressed in the brain. Mammalian Ngb protects neuronal cells under conditions of oxidative stress. We previously showed that human Ngb acts as a guanine nucleotide dissociation inhibitor (GDI) for the α-subunits of heterotrimeric G_i proteins and inhibits the decrease in cAMP concentration, leading to protection against cell death. In the present study, we show that Glu53, Glu60, and Glu118 of human Ngb are crucial for both the neuroprotective activity and interaction with $G\alpha_{i1}$. Moreover, we demonstrate that Lys46, Lys70, Arg208, Lys209, and Lys210 residues of $G\alpha_{i1}$ are important for its binding to human Ngb. We propose a molecular docking model of the complex between human Ngb and $G\alpha_{i1}$.

3Pos080 ドメインスワッピングによるミオグロビン二量体の形成

Formation of myoglobin dimer by domain swapping

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Myoglobin (Mb) is a monomeric hemoprotein possessing eight α-helices, and functions as an oxygen storage protein *in vivo*. Mb can form a domain-swapped dimer by treatment with ethanol. Based on the structural change of the hinge region from a loop structure to an α-helical structure upon Mb dimerization, we constructed Mb dimers by designing the amino acid sequence of the hinge region. We investigated the amount of the dimers obtained from *E. coli* cell extractions compared to the monomers among WT and mutant proteins. We also investigated the structures and thermodynamic properties of the dimers.

3Pos082 共鳴ラマン分光法による 2 価コバラミンの軸配位子に依存した構造変化の検出

Resonance Raman Study of Cobalamin (II): Axial Ligands-Dependent Structural Change

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Cobalamin (Cbl) and its derivatives consist of a corrin ring containing a Co ion and two axial ligands. They have physiologically important roles in metabolism. Previous investigation revealed formation of a six-coordinated Cbl(II) [1]. It was reported to undergo pH-dependent structural changes between six- and five-coordinated Co with a $pK_a = 4.8$ as a result of on-off reaction of one of the axial ligands. We measured resonance Raman spectra of SO_2^-Cbl(II) (base-on and off) and Cbl(II) (base-on) generated by reduction of OHCbl(III) with sodium dithionite at pH 8 and pH 3 or with DTT at pH 8. We detected changes in Raman bands due to the axial ligand changes.

1. Salnikov et al., Dalton Trans. 40, 9831-9834 (2011)

3Pos083 タンパク質中のヘムの歪みの統計的解析

Statistical analysis of heme distortion in protein

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Heme proteins have a wide variety of functions that can be attributed to the modulability of the molecular and electronic structures of hemes. In order to access the structural variety of hemes in proteins, we extracted the skeletal structures of hemes from 3748 entries in Protein Data Bank. According to the principal component analysis, the most significant variation for the entire samples was along the saddling mode. In contrast, the most significant one for hemes with cysteine axial ligands (heme-Cys) was along the ruffling mode. Heme-Cys showed a bimodal distribution along the mode axis, which was reasonably corresponded to two types of Cys coordination. Structural characteristics for other axial ligands and the significance of them will also be discussed.

3Pos084 アンサンブルドッキングを用いた CYP1A2 化合物の代謝部位予測

Prediction of site of metabolism of compounds for CYP1A2 by ensemble docking simulation

Hiroaki Saito[1], Taku Mizukami[2], Yoshinori Hirano[1], Takao Otsuka[1], Noriaki Okimoto[1], Makoto Taiji[1] ([1]*RIKEN Quantitative Biology Center (QBiC)*, [2]*Japan Institute of Science and Technology (JAIST)*)

We investigate the computational method, which predicts the site of metabolism (SOM) of compounds for CYP1A2 by ensemble docking simulation. The docked compound poses obtained from the ensemble docking simulation are used for the analysis of accessibility of atoms of the compounds, which coordinate to the heme iron in CYP1A2. The top 3 ranked atoms by this analysis can be possible SOM for the compounds, and we found the predicted SOMs are correspond to those which are experimentally known, in 78.9 %. The obtained accessibility data for the compounds can be used for the more accurate SOM prediction by combining with the reactivity data obtained from the SMARTCyp. We found that the new score function with the reactivity data can find the correct SOM of compounds in 89.5%.

3Pos085 本来の構造と機能を保持したウシミトコンドリア呼吸鎖複合体の精製

Purification of native mitochondrial respiratory complexes from bovine heart

Satoru Shimada, Shigefumi Uene, Marika Oosaki, Ryoko Takahashi, Harunobu Shimomura, Kaoru Mieda, Shintaro Maeda, Masahide Hikita, **Kyoko Shinzawa-Itoh** (*Grad. Sch. Life Sci., Univ. Hyogo*)

Mitochondrial respiratory chain complexes, complex I (NADH dehydrogenase), complex III (cytochrome bc_1 complex) and complex IV (cytochrome c oxidase), via Q_{10} and cytochrome c (cyt c), sequentially transport electrons and reduce O_2 to H_2O. During this process, protons are actively transported across the mitochondrial inner membrane, and the generated proton gradient is utilized for ATP synthesis by complex V (F_1F_O ATP synthase). To investigate the mechanisms of mitochondrial respiration, not only the structure of each complex but also the interactions between complexes must be examined in detail using the native purified sample. We have established a highly repeatable method of preparing native each complexes and supercomplex from bovine heart mitochondria.

3Pos086 部分フッ素化リン脂質二分子膜中膜タンパク質バクテリオロドプシンの構造と安定性に対するフッ化アルキル鎖長依存性

Structural stability of bacteriorhodopsin in partially fluorinated analogs of DMPC with different perfluoroalkyl chain lengths

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Previously we reported that a membrane protein bacteriorhodopsin (bR) in vesicles of a novel partially fluorinated analog of DMPC with the perfluorobutyl group in the myristoyl chain (F4-DMPC) adopts the native-like higher order structure and photocycle at physiological temperature. In this study, a novel partially fluorinated phosphocholine with the perfluorooctyl group (F8-DMPC) was used for reconstitution experiments. At 30 oC, bR in F8-DMPC vesicles has also trimeric structure and a photocycle, which are very similar to native purple membrane. Thermal stability of the reconstituted bR in F4-DMPC and F8-DMPC vesicles were compared. The reconstituted bR in F8-DMPC vesicle showed much higher stability against heat, which is comparable to native purple membrane.

3Pos087 高圧下で界面活性剤を用いて昆虫細胞膜から可溶化したPBANR（クラス-A GPCR）はリガンド結合能を保持する

PBANR, a class-A GPCR, solubilized under high hydrostatic pressure retains its ligand binding ability

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The effect of high hydrostatic pressure (HHP) on the solubilization of a class-A G protein-coupled receptor, the silkmoth pheromone biosynthesis-activating neuropeptide receptor (PBANR), was investigated. The membrane fraction of insect cells expressing PBANR was subjected to HHP treatment (200 MPa) at room temperature for 1-16 h in the presence of 0-2.0% (w/v) *n*-dodecyl-β-D-maltopyranoside (DDM). The solubilization efficiency of PBANR was largest following 6-h HHP treatment with 0.4-1.0% DDM. Fluorescence-detection size-exclusion chromatography demonstrated that the ligand binding ability of PBANR was retained and lost after 6-h solubilization with 1.0% DDM under HHP and ambient pressure, respectively, demonstrating the stabilization effect of HHP on solubilized PBANR.

3Pos088 GraDeR：単粒子解析等の膜タンパク質資料調整

GraDeR: membrane protein preparation for single particle cryoEM & more

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We developed a method, named GraDeR, which substantially improves the preparation of membrane protein complexes for structure determination by single-particle cryo-electron microscopy (cryo-EM). In GraDeR, glycerol gradient centrifugation is used for the mild removal of free detergent monomers and micelles from lauryl maltose-neopentyl glycol detergent stabilized membrane complexes, resulting in mono- disperse and stable complexes to which standard processes for water-soluble complexes can be applied.

3Pos089 エーテル型部分フッ素化リン脂質膜中の膜タンパク質バクテリオロドプシンのサーモクロミズム

Thermochromism of bacteriorhodopsin in partially fluorinated di-o-tetradecylphosphocholine vesicles

Masaya Miyazaki[1], Naoyuki Tuchida[1], Toshinori Motegi[1], Takashi Kikukawa[2], Toshiyuki Takagi[3], Hiroshi Takahashi[1], Hideki Amii[1], Toshiyuki Kanamori[3], Masashi Sonoyama[1] ([1]*Fac. Sci. Tech., Gunma. Univ.*, [2]*Grad. Sch. Sci., Hokkaido. Univ.*, [3]*AIST*)

It has been demonstrated that a novel partially fluorinated ester phospholipid (F4-DMPC) is promising as a possible material for structural and functional analyses of membrane proteins. In this study, a novel partially fluorinated ether phospholipid (F4-DTPC) has been employed for biophysical studies of a membrane protein bacteriorhodopsin (bR). The wavelength of absorption maximum of bR in F4-DTPC vesicles showed significant red-shift with decreasing temperature, indicating that bR is a temperature sensor in F4-DTPC vesicles. What is responsible for the thermochromism of bR was explored by spectroscopic measurements at various pH values. It was revealed that the temperature-dependent changes in pKa of Asp85 in a plausible candidate for the thermochromism of bR.

3Pos090 GXXXG モチーフによる膜貫通ヘリックスの二量体形成：会合トポロジー制御下での一分子 FRET 研究

GXXXG-mediated dimerization of transmembrane helices: single-molecule FRET detection with controlled association topology

Yoshiaki Yano, Yuta Watanabe, Katsumi Matsuzaki (*Grad. Sch. Pharm Sci., Kyoto Univ.*)

Small-residue-mediated interhelical packings are ubiquitously found in helical membrane proteins, although their interaction dynamics and lipid dependence remain mostly uncharacterized. Here a recently developed liposomal single-molecule FRET technique was used to examine the effect of a GXXXG motif on dimerization of the de-novo designed (AALALAA)3 helix. The association topology was controlled by reduction of a disulfide-linked helix dimer in liposomes. The motif evoked transient dimerization in both parallel and antiparallel topologies with decreases of association free energy of 20 and 15 kJ mol-1, respectively. Unexpectedly, membrane cholesterol completely abolished these associations, indicating a critical role of the lipid on the Gly-mediated interaction.

3Pos091 脂質膜分子による上皮成長因子受容体の膜近傍ドメイン構造制御機構

Conformational regulation of the juxtamembrane domain of epidermal growth factor receptor by membrane lipid molecules

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The transmembrane (TM) and juxtamembrane (JM) regions bridge the extracellular and intracellular domains of single-pass membrane proteins including EGFR, and play an important role for activation. While interaction of JM with lipid molecules regulates EGFR dimerization, the precise mechanism remains unclear. By using synthesized fluorescent peptides consist of TM and JM regions, we have shown that negatively charged lipids stabilize JM dimer conformation. Here, we further investigate how the JM conformation is regulated by membrane lipids, especially the effects of PIP2 molecules and phosphorylation of JM. These peptides and lipids were reconstituted into nanodisc, and applied to single-molecule FRET analysis to monitor the conformational dynamics of JM regions.

3Pos092 ESR による銅ポンプ P 型 ATPase における 金属イオン配位子の動的構造：ATP 効果の研究

Structural dynamics in metal ion coordination of copper pump P-type ATPase as studied by EPR spectroscopy: Effect of ATP

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All cells have a Na/K pump P-type ATPase. We have studied bacterial Cu^{2+}-selective pump CopB to explore the coordination chemistry of the metal binding sites in P-type ATPases. Our ESR studies previously showed that paramagnetic Cu^{2+}- ions are bound to an ATPase in Type-2-like coordination. Here, we studied ESR spectroscopy on Cu^{2+}- bound during ATP hydrolysis or in the presence of ATP analog. No or slight spectral change was found in the frozen state after ATP addition. At room temperature, the line-shape was broader than in frozen state in the absence of ATP. In the presence of nonhydrolyzable ATP analog (AMPPCP) the line-shape became sharper like frozen state. This suggested that Cu^{2+}-coodination was dynamic or had multiple states and changed during ionic transport.

3Pos093 高速 AFM による ABC タンパク質の動態観察

High-speed atomic force microscopy shows conformational changes of nucleic binding domains of ABC protein

Shohei Takigaura[1], Mikihiro Shibata[1,2], Kazuaki Yamahara[3], Yasuhisa Kimura[3], Kazumitsu Ueda[3,4], Takayuki Uchihashi[1,2], Toshio Ando[2] ([1]*Dep. Phys. Kanazawa Univ.*, [2]*Bio-AFM FRC*, [3]*Div. App. Lif. Sci. Kyoto Univ.*, [4]*iCeMs*)

ABC proteins are one of the membrane protein and actively transport substrates across the cell membrane. ABC proteins consist of two NBDs which are membrane-associated ATPases and two TMDs. Since conformational changes of NBD domains related to ATP hydrolysis are important for the active transport of substrates, to better understand the molecular mechanism of ABC proteins, direct visualization of conformational changes of NBD domains at work is required. In this study, we have used HS-AFM to visualize conformational changes of NBD domains of CmABCB1. As a result, we have succeeded in capturing the dynamic structural change of NBD domains of CmABCB1 in the solubilized state. And also, we attempt to observe the dynamics of CmABCB1 in lipid bilayer with nanodiscs.

3Pos094 高速原子間力顕微鏡による電位依存性プロトンチャネルの直接観察

Direct observation of voltage-gated proton channels by high speed AFM

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Voltage-gated proton channel (Hv1/VSOP) mediates proton conductance across the plasma membrane in response to changes in membrane potential. Recent study in X-ray crystallography provides atomic structure in resting state. However, the information about the molecular dynamic behavior of Hv1/VSOP is lacking.

In this study, we applied the high speed atomic force microscopy (HS-AFM) to image dynamic structural changes and interactions of individual Hv1/VSOP. Purified Hv1/VSOP was reconstituted in artificial lipid and observed by HS-AFM. We succeeded to observe the molecules showing globular structure and characteristic ring-like structure. These direct observations will be useful for studying the structural dynamics of voltage-gated proton channel at single molecule level.

3Pos095 多剤輸送担体 EmrE の基質結合エントロピー利得に対する水分子の寄与

Contribution of water molecules for the gain in the substrate binding entropy to multidrug resistance transporter, EmrE

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We reported previously the pH dependent entropic contribution to binding energy between multidrug resistance transporter, EmrE and its substrate. Here, we had examined a heat capacity change (ΔC_p) upon tetraphenylphosphonium ion (TPP^+) binding to EmrE at pH 6.0 and 7.4 with isothermal titration calorimetry. Both exhibited a similar large negative ΔC_p (~-0.4 kcal/mol/K). This might suggest that the TPP^+ induced structural changes and protein flexibility are similar under the both conditions. Hence, large entropic contribution in alkaline solution is primarily due to solvent entropy from release of water molecules upon binding rather than conformational entropy. Based on the thermodynamics properties, the entropic effects of substrate binding to EmrE will be discussed.

3Pos096 cd1NiR:cNOR 複合体構造を安定化する相互作用の理論解析

Theoretical analysis of interaction that stabilizes cd1NiR:cNOR complex structure

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Microbial denitrification has been explored in various fields, such as global warming and water treatment. In this process, one of the problems is how nitric oxide, toxic to cells, moves from nitrite reductase (NiR) to nitric oxide reductase (NOR). Recently, Tosha et al. determined the crystal structure of cNOR in complex with cd1NiR using Pseudomonas aeruginosa. However, it is difficult to dispel doubts about the complex formation due to the experimental conditions like a crystal packing effect. Thus, we aim to show great potential for the complex formation in vivo with molecular dynamics simulation. In this study, a plausible explanation is built by focusing on two types of interaction, which contribute to the stability of the complex structure.

3Pos097 3 次元立体構造が不明の膜タンパク質に対する耐熱化置換体の特定

Identification of Thermostabilizing Mutations for a Membrane Protein Whose Three-Dimensional Structure is Unknown

Yuta Kajiwara[1], Satoshi Yasuda[2,3], Yuuki Takamuku[2], Takeshi Murata[2,3,5], Masahiro Kinoshita[4] ([1]*Graduate School of Energy Science, Kyoto University*, [2]*Graduate School of Science, Chiba University*, [3]*Molecular Chirality Research Center, Chiba University*, [4]*Institute of Advanced Energy, Kyoto University*, [5]*JST, PRESTO*)

We recently developed a physics-based method for identifying thermostabilizing mutations of a membrane protein. The method employs a free-energy function F focused on the translational entropy of hydrocarbon groups within the lipid bilayer. The method was illustrated for the adenosine A2a receptor by utilizing its experimentally determined three-dimensional (3D) structure. The success rate for the identification was remarkable high. In this work, we postulate that the 3D structure is unknown. We construct candidate models for the 3D structure using the homology modeling and select the model giving the lowest value to the change in F upon protein folding. The performance achieved in this work is only slightly lower than that in the recent work.

3Pos098 In silico screening of novel stress response factors regulated by mitochondrial inner membrane proteases

Kenichiro Imai, Yoshinori Fukasawa, Kentaro Tomii, Paul Horton (*AIRC, AIST*)

Importance of proteolytic regulations by mitochondrial proteases recently is emerged in mitochondrial and cellular stress responses. However the proteolytic regulations are poorly understood due to limited numbers of substrates. In this study, we focus on mitochondrial rhomboid protease PARL, which cleaves a transmembrane domains of substrate depending on mitochondrial membrane potential. To find novel substrates, we then developed prediction pipeline combining mitochondrial targeting sequence prediction, transmembrane domain prediction optimized for mitochondrial inner membrane and PARL cleavage site prediction. We will report the detail of the pipeline and discuss relevance between predicted substrates and mitochondrial and cellular stress responses.

3Pos099 MD シミュレーションを用いた RNA 二重らせん構造の熱安定性予測
Predicting RNA Duplex Dimerization Free-Energy Changes upon Mutations Using Molecular Dynamics Simulations

Shun Sakuraba[1], Kiyoshi Asai[1,2], **Tomoshi Kameda**[2] ([1]*Grad. Sch. Frontier Sci., Tokyo Univ.*, [2]*AI center, AIST*)

The binding free energy of RNA-RNA pairs is a fundamental value that represent the structural stability of RNA complex. We report the comparison of RNA-RNA binding free-energies from the molecular dynamics simulation and from the experiments. Linear regression from 9 double-stranded RNA sequences of 6 base pairs yielded the mean absolute deviation of 0.55 kcal / mol and the R2 value of 0.97, showing a quantitative agreement with experimental data. The achieved accuracy shows that the MD simulation with the current molecular force field is capable of estimating the thermodynamic properties of RNA molecules.

3Pos100 単層および多層カーボンナノチューブ上における蛍光 DNA の蛍光強度の塩基配列依存性
Base sequence dependence for fluorescence intensity of fluorescent dye-labeled DNA on single- and multi-walled carbon nanotubes

Shusuke Oura, Kazuo Umemura (*Tokyo Univ. of Sci.*)

It is known that fluorescence from fluorescent dyes directly adsorbed onto single- or multi-walled carbon nanotubes (SWNTs and MWNTs, respectively) are quenched by carbon nanotubes due to electron transfer. We examined the quenching ratios of fluorescence when fluorescein-labeled 30-mers of thymine or adenine (Fluor-T30 and Fluor-A30, respectively) were reacted with the T30-wrapped SWNTs or MWNTs. Quenching ratios of Fluor-T30 in SWNT and MWNT samples were 28 ± 3.1% and 36 ± 2.0%, and those of Fluor-A30 were 11 ± 1.9% and 32 ± 1.9%, respectively. The data show that SWNT conjugates have better resolution for the base-sequences of Fluor-ssDNA, which would be helpful information for the single-base mismatch level DNA detection using quenching phenomenon.

3Pos101 メタダイナミクスとアルケミカル変換法を用いた定量的結合活性予測
Approach to the quantitative prediction of the binding affinity using metadynamics and alchemical transformation

Yoshiaki Tanida, Azuma Matsuura (*FUJITSU LABORATORIES LTD.*)

Metadynamics simulation has been performed to explore the (meta)stable conformation of theophylline with an RNA aptamer. Subsequently, we have applied the alchemical free energy calculation for each binding pose. The binding affinity predicted is in good agreement with the experimental value.

3Pos102 紫外線損傷 DNA における Flippling 機構
On the Flipping-out mechanism of the UV-induced DNA damage

Ryuma Sato, Ryuhei Harada, Yasuteru Shigeta (*Center of comp. Sci., Univ. Tsukuba*)

Although DNAs are often damaged by UV light irradiation, which causes skin cancers, DNA photolyases (PHRs) in a photolyase/cryptochrome family repair the UV-induced DNA damages. On the other hand, the cryptochromes cannot repair DNA lesions in vivo. In the repair mechanisms, PHRs recognize DNA lesions and bind to them. Upon the binding to the DNA lesions, the damaged parts have been flipped out. However, it is unclear that whether the flipping-out occurs as the DNA alone or that in a protein complex.
In this study, we performed MD and QM simulations to address how the flipping-out mechanism occurs. Through free energy and interaction energy analyses, it is found that the flipping-out cannot be induced in the DNA alone.

3Pos103 Comparison of Multi-Dyes Quenching by Single-Walled Carbon Nanotube Dispersion with Single Stranded DNA

Ying Tan, Katsuki Izumi, Kazuo Umemura (*Tokyo University of Science*)

Single-walled carbon nanotube (SWCNT), could cause fluorescence quenching of dyes. In this study, three kinds of dyes, uranine (Ur), acridine orange (AO) and Rhodamin B (RB), were added into SWCNT dispersion with thymine 30 mers (T30-SWCNT). Three groups of quenching assay due to T30-SWCNT, Ur/AO, Ur/RB, and AO/RB, were carried out respectively, quenching intensity variations were compared and competitive adsorptions were investigated. It was found that AO adsorbed onto T30-SWCNT surface prior to Ur and RB, while Ur showed a best quenching-restrain performance. The needed dye concentration was extremely low and the dispersion of dye was prepared simply without sonication process, which were important for applying the fundamental quenching studies to multiplexed sensing.

3Pos104 サイトキニン脱水素酵素における独特なフラビン-基質配置に関する量子化学的研究

Quantum chemical study on unusual flavin-substrate alignment in cytokinin dehydrogenase

Kyosuke Sato (*Dept. Mol. Physiol., Facult. Life Sci., Kumamoto Univ.*)

In most flavin-dependent amine oxidases and dehydrogenases, including D- and L-amino acid oxidase, and sarcosine oxidase, the substrate binds to the active site such that the substrate C and N atoms involved in double bond formation are located near the flavin N5 and C4a atoms, respectively. Quantum chemical calculation revealed that the transition state energy is minimized in this configuration. Although the substrate of cytokinin dehydrogenase binds to the active site using an alignment different from many other flavin-dependent oxidases and dehydrogenases, the quantum chemical calculations confirmed that the transition state energy is minimized in this alignment.

3Pos105 生体分子の電子状態解析のための大規模第一原理 DFT 計算手法の開発

Large-scale DFT calculation method for electronic-structure analysis of biomolecules

Ayako Nakata[1], Takao Ostuka[2], David R. Bowler[3], Tsuyoshi Miyazaki[1] ([1]*NIMS*, [2]*RIKEN*, [3]*UCL*)

First-principles DFT calculations have been powerful tools to investigate electronic structures of biomolecules. However, conventional DFT calculation methods can treat up to about a thousand atoms because of the high computational cost.

We have recently developed an efficient method, "multi-site method", to reduce the computational cost of DFT calculations significantly while keeping accuracy, and introduced it to our own large-scale DFT code CONQUEST. We have also introduced an eigenstate-calculation method called "Sakurai-Sugiura method" to obtain molecular orbitals (MOs) in specific energy regions efficiently. The density of states and MOs of DNA systems in water consisting of thousands of atoms are investigated by first-principles DFT with the present methods.

3Pos106 生体分子系における定温オーダーN法第一原理分子動力学計算

Constant temperature order-N first-principles molecular dynamics calculations of biomolecular system and short-time behavior

Takao Otsuka[1], Makoto Taiji[1], David R. Bowler[2], Tsuyoshi Miyazaki[3] ([1]*RIKEN QBiC*, [2]*UCL*, [3]*NIMS*)

Molecular simulation methods are now commonly used to explore biological phenomena of proteins and/or biomolecules. Such molecular simulation methods are expected to help us understand the mechanism of biological phenomena. Nowadays, quantum mechanics/molecular mechanics (QM/MM) hybrid methods or its molecular dynamics (QM/MM-MD) method are also used well in this field. Recently, we succeeded to introduce stable MD method to the framework of our order-N first-principles density functional theory methodology. In this study, we demonstrate our order-N first-principles molecular dynamics calculations with temperature controlled method and investigate the short-time behavior for hydrated DNA system.

3Pos107 両親媒性抽出剤を用いた水相から有機相へのリン酸化合物の抽出

Extraction of phosphoric compounds from aqueous phase into an organic phase with an amphiphilic extractant

Hideyuki Komatsu (*Bioinfo. & Biosci., Kyushu Inst. Tech.*)

Extraction of metal ions from an aqueous solution into an organic solvent with an amphiphilic extractant is useful in applications of environmental and industrial fields. Such an organic solvent/amphiphilic extractant system has been developed to extract phosphoric compounds such as nucleotides from aqueous solutions by use of alkyl amine as an amphiphilic extractant. Because phosphate exists as four ionic forms depending on pH, pH of solution should be controlled for the accurate evaluation of the extraction. In this study, the pH-controlled condition of extractant/solvent system has been established by tuning of alkyl amine and buffer concentrations, and the extraction of phosphoric compounds by using this system will be characterized and thermodynamically evaluated.

3Pos108 リン酸イオンの水和エネルギー空間分割解析

Spatial-Decomposition Analysis of Hydration Energy of Phosphate Ions

George Mogami[1], Nobuyuki Matubayasi[2], Makoto Suzuki[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Eng. Sci., Osaka Univ.*)

Ions such as alkali halide, ATP, and phosphate, and muscle protein actin form hypermobile water which have higher dielectric relaxation frequency than bulk water (G. Mogami et al., 2013, JPCA; 2011, Biophys. Chem.; S. R. Kabir et al., 2003, BJ). Recently, we developed a framework to visualize the distribution of hydration energy of those ions (G. Mogami et al., 2016, JPCB). We carried out spatial-decomposition analyses of hydration energy for monoatomic ions and demonstrated a favorable ion-water interaction at the expense of water-water interaction beyond second hydration layer. In this study, we have performed the spatial-decomposition analysis of hydration energy of mono-, di-, and tri-phosphate ions and have revealed the hydration structure around polyatomic ions.

3Pos109 実効相互作用を用いた電解質中のマクロアニオンの分子シミュレーション

Molecular simulation of macroanions in an electrolyte solution based on the effective potential

Ayumi Suematsu, **Ryo Akiyama** (*Dept. Chem., Kyushu Univ.*)

In a dilute electrolyte solution like-charged macromolecules, such as DNAs and acidic proteins, are repel with each other via the electric bilayers. The thickness depends on the electrolyte concentration. Then, the "liquid"-"crystal" transition of the macroanion system is observed in the dilute electrolyte condition. The mechanism is almost same as the Alder transition. However, the width of coexisting region obtained by experiments is much wider than the prediction of hard spheres model. We carried out molecular simulations for macroanions in an electrolyte solution based on the effective one-component model. The effective potentials were calculated by using the integral equation theory. We will discuss the calculated results.

3Pos110 MM/3D-RISM 法を用いたシクロデキストリン誘導体とロクロニウム臭化物の結合自由エネルギーの予測

Estimation of binding free energies for inclusion processes of Rocuronium bromide by cyclodextrin derivatives using MM/3D-RISM method

Yuji Hayashino[1], Masatake Sugita[1], Fumio Hirata[2], Takeshi Kikuchi[1] ([1]*Dept. of Bioinfo., Col. Life Sci., Ritsumeikan Univ.*, [2]*Toyota Phys. & Chem. Res. Inst.*)

To seek the drug-candidate compounds using computational methods, it's necessary to evaluate binding free energy between bound and unbound states of target molecules immersed in the solution phase. We have suggested that the MM/3D-RISM method is effective to evaluate the binding free energy since the method enables calculations of the solvent distribution and fluctuation around a host-guest system with reasonable accuracy.
As the target molecules, we used some cyclodextrin derivatives and rocuronium bromide. At first, we applied the umbrella sampling method to seek the most stable binding mode. After that we performed MD simulation for resampling the fluctuated structures around the most stable binding mode. Then we estimated the binding free energy.

3Pos111 ボルンエネルギーとクーロンエネルギー間のバランスの物理的理解と計算指針

Physical understanding and computational guideline for the balance between Born and Coulomb energies

Dan Parkin, Yukinobu Mizuhara, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

The stability of ionic bond in aqueous solution is marginal and therefore difficult to accurately calculate due to the counterbalance between Born energy increase and Coulomb energy decrease upon bond formation. The generalized Born (GB) model, where water is treated as a dielectric, has been widely used to study the folding and binding in biomolecules. However, failures to accurately reproduce electrostatic interactions in the GB model have been pointed out. The key lies in evaluating the "effective Born radii", which involves critical parameters that largely affect association-dissociation thermodynamics. By showing the parameter dependence of Born and Coulomb energies, we clarify the underlying physics and present a guideline for accurate simulation.

3Pos112 何故無細胞タンパク質合成系においてはそんなに多くのリボソームが必要なのか？

Why we need so many ribosomes in cell-free protein synthesis?

Yue Xu, Yi Zhang, Hiroyuki Noji (*Grad. Sch. Eng., Univ. Tokyo*)

Central dogma defines the flow of genetic information, which was proved to be general in both living cells and cell-free system, but does not give the details of the flow. Our previous work succeeded in cell-free protein synthesis from single DNA molecule in cell-sized chambers, and revealed a great difference (>10^3-fold) between the number of ribosome and synthesized protein. Here we decreased the number of ribosome in chamber, and found that the synthesis of fully active protein suddenly stops when it falls below a certain number around 10^3. Interestingly, a similar threshold concentration was also confirmed in bulk synthesis with excess DNA addition. Our preliminary result suggests that non-specific adsorption on the chamber wall seems not to be the dominant factor.

3Pos113 一細胞トランスクリプトーム解析へ向けた、PCR を含まないライブラリー調製

Amplification-free library construction for single-cell transcriptome analysis

Tetsuo Fujinami, Yusuke Oguchi, Mai Yamagishi, Yoshitaka Shirasaki, Sotaro Uemura (*Grad. Sch. Sci. Univ. Tokyo*)

The recent progress in sequencing technologies enables comprehensive transcriptome analysis even at the single-cell level. To analyze an extremely small quantity of RNA particularly extracted from a single-cell, however, the inescapable PCR amplification during library preparation interferes with accurate quantification.
To overcome this, we developed a new library construction method without PCR. RNA extracted from a cell, were added 3' poly(A) tail and captured onto the poly(T)-coated glass surface, followed by fluorescent dNTPs incorporation. By counting directly each molecule with a single-molecule imaging system, we succeeded to quantify nucleic acids at sub-pM concentrations.
Here, we also discuss the potential of this method for single-cell transcriptome analysis.

3Pos114 無細胞タンパク質合成系に向けた最良 T7 プロモーター配列の探索

Improvement of T7 promoter sequence for cell-free protein synthesis

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T7 RNA polymerase (RNAP) is commonly used in cell-free transcription/translation system. It recognizes T7 promoter sequence and initiates mRNA synthesis. We attempt to achieve further improvement of T7 promoter that is more suitable for the in vitro cell-free system. In the present study, we developed a chamber array system to explore the improved T7 promoter sequences that more adapt to the cell-free system. We prepared mutation libraries of T7 promoter sequence by saturation mutagenesis followed with a downstream reporter gene, Venus. Because of the completely random sequences, quite a few chambers showed fluorescent signals. We recovered DNAs from such chambers, and the following investigation is ongoing.

3Pos115 細胞集積度によるパラクラインシグナリングの制御により遺伝子発現の安定性が変化した

Paracrine signaling modulated by the accumulation of cells altered the stability of gene expression

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Macrophages are widely known to accumulate and function at a site of infection though the accumulation effects on the gene expression are not deeply investigated yet. In the previous meeting, we demonstrated that the expression of some genes in individual LPS-stimulated macrophages became high and homogeneous by increasing the cellular density. Here, we conclude this cellular density-dependent gene regulation was based on the modulation of paracrine signaling effect, based on both the result of an ELISpot/RNA-FISH assay and simulations of paracrine signaling under different cellular densities. The accumulation of macrophages to the inflammatory site is therefore indicated as one of the key processes to fulfill a stable response even within highly heterogeneous cells.

3Pos116 Causal role of DNA methylation ?: A computational model

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Gene regulation in eukaryotes involves complex interactions between underlying gene regulatory network and the local chromatin structure in vicinity of the gene giving rise to epigenetic information processing. DNA methylation plays a central role in the heritable epigenetic information transfer. However, based on recent global genome methylation pattern data, Bestor et al. [PNAS (2015)] questioned the causal role of DNA methylation in gene regulation dynamics. We present a computational model of eukaryotic gene expression which captures the interplay of the above mentioned hierarchy and resolves the ambiguity risen from global DNA methylation patterns and dynamic role of DNA methylation in gene regulation with quantitative consistency.

3Pos117 An in silico Approach to Investigating Gene Variants of Unknown Significance in a Clinical Context

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Next-generation sequencing has improved diagnoses of inheritable disease, but also increased detection of variants of unknown significance (VUS), posing diagnostic challenges. In silico analyses can help predict the pathogenicity of VUSs. Three probands with Marfan Syndrome (MFS) were each identified to have a novel VUS of potential pathogenicity, in addition to their MFS-causing FBN1 mutation. Mutations occurred in MYH11, COL5A2 and NOTCH1 genes. In silico analyses showed the mutated residues are evolutionary conserved, and located at sites of potential structural and functional importance. The findings highlight the value of in silico techniques in interpreting VUSs and may enrich clinical investigations when establishing the biological relevance of novel mutations.

3Pos118 遺伝子発現におけるポリアミンの DNA 高次構造との関係性 Relationship between DNA higher order structure and Gene-Expression with Polyamines

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We have studied the effect of polyamines on gene expression activity by adapting an in vitro luciferase assay. It was found that polyamines accelerate the gene expression at low concentrations, whereas, at high concentrations, a significant inhibition is marked. We also examined DNA higher order structure at different concentration of polyamines by use of AFM. It became clear that DNA takes compact conformation at the polyamine concentrations to cause the inhibition. At lower concentrations, loosely condensed state of DNA is observed. Such conformational behavior is discussed in relation to the gene expression activity.

3Pos119 *Guillardia theta* におけるロドプシン様遺伝子群の発現解析 Expression analysis of microbial rhodopsin-like genes in *Guillardia theta*

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The Cryptomonad *Guillardia theta* has 44 genes encoding microbial rhodopsin-like proteins in their genomes. Light-driven ion-pump activity was reported for some rhodopsins based on heterologous *E. coli* expression system. However, neither conversion of light into metabolic energy nor expression in native cells remains known. To find their physiological roles, we investigated the expression pattern of these genes in various growth conditions. The nitrogen (N) deficiency induced color change to green from brown in growing cells. The 11 and 25 rhodopsin-like genes were expressed in N-sufficient and N-deficient cells, respectively. These results suggested that some of expressed genes contribute to make the energy for N assimilation.

3Pos121 細胞性粘菌の Tyr143 変異アクチンのカルボキシ末端領域にある Phe352、Met355 と Trp356 の側鎖の二形性 Dimorphism of the side-chains of Phe352, Met355, and Trp356 in the carboxyl-terminal region of Dictyostelium actin mutants

Yuki Gomibuchi[1], Taro Q.P. Uyeda[2], Takeyuki Wakabayashi[1] ([1]*Teikyo Univ.*, [2]*Waseda Univ.*)

The frontal region of actin around Tyr143 has been proposed to be involved in polymerization and the interaction with myosin. Mutations of Tyr143 (Y143F, Y143I, Y143W) resulted in the changes in polymerizability and the activation of myosin ATPase. Surprisingly, the crystal structures at 1.93-2.03 Å resolution showed that the side chains of the residues 352-356 (FQQMW) changed their configurations in a concerted manner, with the main chain being unchanged. Whereas Trp356 of the wild-type actin is buried, its side chains of mutants were also exposed to the medium (dimorphism). The side chains of other residues (352-355) changed their configurations in accordance with the changed Trp356. Thus, the carboxyl-terminal region of the mutant actins showed overall dimorphism.

3Pos122 筋収縮制御メカニズムの解明を目指した細いフィラメントの立体構造解析 CryoEM structural analysis of muscle thin filament composed of actin filament, tropomyosin and troponin

Yurika Yamada[1], Keiichi Namba[1,2], Takashi Fujii[1] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*RIKEN QBiC*)

Muscle contraction is driven by cyclic interactions of myosin in the thick filament with the thin filament composed of actin, tropomyosin (Tm) and troponin (TnC, TnI, TnT). It is thought that the binding of Ca2+ released from sarcoplasmic reticulum to TnC causes a conformational change of Tm on the actin filament to allow actin-myosin interaction. To understand this regulatory mechanism, the structure of thin filament at high resolution is necessary. We obtained the structure of skeletal muscle thin filament at 20 Å resolution but crosslinking was necessary to stabilize it. To prevent the dissociation of Tm and Tn, we expressed human cardiac Tn complex and slightly modified Tm in E. coli and reconstituted the thin filament. The structural analysis is under way.

3Pos123 中性子準弾性散乱により明らかとなった心筋症原因変異がもたらすトロポニンの動力学異常

Effects of a cardiomyopathy-causing mutation on the internal dynamics of troponin revealed by quasielastic neutron scattering

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Dynamics of the wild-type troponin (Tn) and a cardiomyopathy-causing mutant of Tn (TnT_{K247R}) was studied by quasielastic neutron scattering in both the $\pm Ca^{2+}$ states. It was found that in the $-Ca^{2+}$ state, both the residence time and the amplitudes of atomic motions are smaller for the mutant. On the other hand, in the $+Ca^{2+}$ state, the amplitudes of atomic motions are larger for the mutant with no difference in the residence time. This modified dynamics of the mutant in the $-Ca^{2+}$ state should perturb the Ca^{2+}-signal transmission pathway in Tn, resulting in the larger fluctuations upon Ca^{2+}-binding. This should weaken the interaction with tropomyosin, which directly controls the force production process, leading to the reported functional aberration of the mutant.

3Pos124 ウニのコネクチン様タンパク質の構造解析

Sequential analysis of connectin-like protein in sea urchin

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Connectin is a giant elastic protein that extends from Z- to M-line, keeping the integrity of sarcomere structure. Connectin-like proteins have been found also in invertebrate muscles, but still not in sea urchin muscle. Using genomic information, we have found a connectin-like protein in sea urchin, and determined its sequence by RT-PCR. This connectin-like protein that is estimated to have 80 kbp consists of tandem Ig domains, long PEVK regions and a tandem fibronectin type 3 domain. As is the case of vertebrate connectin, this connectin-like protein seemed to sustain longitudinal integrity of sea urchin muscle bearing its passive tension, for mild trypsin treatment accelerated the relaxation of passive tension at particular condition in our study.

3Pos125 Mg ポリマー再考

Revisiting "Mg-Polymer"

Mahito Kikumoto, Shuuichi Takeda, Yuichiro Maeda (*Structural Biology Research Center, Nagoya-Univ.*)

In order to study the dynamic property of polymerized actin to clarify the relationship between polymorphism and actin state/structure, we have been searching for another state of the polymerized actin. When Physarum actin was polymerized with Mg2+, the polymerized actin showed a low viscosity and a high ATPase activity. This state was designated "Mg-polymer" in 1967. Independently, it was found that rabbit muscle actin polymerized in the presence of β-actinin (=CP) and Mg2+ has similar properties. Because of its high ATPase activity, this state may be a good model for analyzing the ATPase mechanism and/or polymorphism of the actin filament. We set out revisiting "Mg-polymer" to re-identify mechanistic, structural and biochemical characters of this state.

3Pos126 細いフィラメント上のトロポミオシンのモデル：スピンラベル ESR 距離マップ

Modeling for tropomyosin position in the thin filament by distance measurements using spin-labeling dipolar EPR spectroscopy

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We have determined the interspin distance (within 2.5 nm) between 15N-based spin labeled tropomyosin (Tm) and 14N-based spin labeled Cys374 of actin and between spin labeled Tm and actin (Mn^{2+}). We constructed the model of Tm position on actin filament by 2.5-nm search. A model showed a small Ca^{2+} dependent movement (<0.5 nm) of Tm along the thin filament and that Tm is flexible and flared at the NC junction without Ca^{2+}. Flexibility of Tm at NC junction is also suggested by the fact that the distance between cardiac troponin(cTn)-T and Tm at NC junction included the population with the distance unexpectedly longer than that in a crystal structure. The flexibility and dynamics of Tm and cTnT at the NC junction may play an important role as muscle switch and regulator.

3Pos127 筋原線維懸濁液の ATP 分解素過程中のプロトン NMR 緩和経過

Spin-spin relaxation of 1H NMR signals from myofibril suspension during cross-bridge cycling

Tetsuo Ohno, Hitomi Sano (*Dept. Physiol., The Jikei Univ. Sch. Med.*)

The dynamic changes of water molecules structure surrounding contractile proteins might play an important role in cross-bridge cycling during contraction. The spin-spin relaxation process of 1H-NMR signals from suspension of myofibril prepared from rabbit could be well represented by the summation of several exponentials indicating that water molecules in the suspension could be conveniently grouped into several components based on the relaxation time constant (T2). The slowest two components (T2 around 0.4s and 0.15s) dominated over faster relaxation components. This may suggest that the potential of the water molecules existing around myofibril is high.

3Pos128 骨格筋タンパク質と水の相互作用を融点から探る

Interaction between water and myoproteins revealed by melting points

Naoya Nakahara[1], Tetsuo Ohno[1], Masako Kimura[2], Sumiko Kimura[1], Shigeru Takemori[1] ([1]*Jikei Univ. Sch. Med.*, [2]*Kagawa Nutri. Univ.*)

MRI reflects content and states of tissue water. Interaction between water and macromolecules is considered to restrict molecular motional freedom similarly to freezing of water. With differential scanning calorimetry (DSC) on skinned fibers, we observed extra latent heat absorption with temperature rise at sub-zero temperatures of -25, & -22°C. Heat-denaturation of myosin and actin increased these extra latent heat decreasing basal specific heat capacity at -80~+20°C. The extra latent heat and the specific heat capacity depended on the presence of thin and/or thick filaments in sarcomere. These strongly suggest the extra latent heat and specific heat capacity to be tightly coupled with inter myofibrillar interaction that realizes muscle contraction.

3Pos129 T-plastin の 2 つのアクチン結合ドメインとアクチンフィラメントとの結合性の比較

Comparison of binding affinities of two actin-binding domains of T-plastin to actin filament

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T-plastin is an actin-binding protein and a member of plastin family. This family, including I-plastin(fimbrin), L-plastin, and T-plastin, cross-links actin filaments. T-plastin distributes to various non-muscle cells, but its function remains unknown. T-plastin includes two EF-hand domains, actin binding domain 1 and 2(ABD1 and ABD2) from the N-terminus. In this study, we expressed ABD1 and AD2 in E. coli, and examined the binding of each domain of T-plastin to actin filament. We show that the binding of ABD1 was reduced by Ca2+, and as a result, cross-linking of actin filaments by ABD1 and ABD2 was inhibited by Ca2+.

3Pos130 心筋細胞集団の伝搬のゆらぎの局所・全体相関の解明のためのオンチップ心筋細胞ネットワーク解析技術の開発

Development of On-chip Cardiomyocyte Network Analysis Assay for Understanding of Fluctuation Correlation in Cell-to-cell Conduction

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To understand the community effect of cell network, we have developed a constructive on-chip cardiomyocyte network assay using processed agarose microstructures on a 64-channel, 10-μm multielectrode array chip. Comparing the fluctuation of propagations and waveforms of external field potentials of cells on the neighboring electrode, this system enables us to analyze the fluctuation of cell-to-cell conductions of cardiomyocyte in various network patterns, e.g. small cluster, 1-D line, and 2-D sheet. After administration of proarrhythmic compounds, we examined the influence of local fluctuation for propagation in the whole networks in various patterns, and found that the spatial pattern dependence in local-whole network fluctuation correlation as their community effect.

3Pos131 肥大型心筋症特異的なトロポミオシン変異体(V95A,D175N)のアクトミオシン収縮速度・収縮力への異なる影響

Tropomyosin's HCM mutants (V95A, D175N) differently affect the actomyosin sliding velocity and force

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HCM is a disease of cardiac insufficiency, and its major causes are mutations of sarcomeric proteins. We studied the effect of human α-tropomyosin (Tpm) mutants (V95A, D175N) in contraction. Thin filament reconstituted with recombinant Tpm was examined with *in vitro* motility assays using optical tweezers, and the sliding velocity and force were measured. At saturating [Ca^{2+}] force was significantly less when V95A Tpm was used, and significantly more when D175N was used, compared to WT. These results are different from those on muscle fibers, presumably because different experimental conditions were used. It demonstrates the importance of investigating the same mutations with different experimental approaches to understand the molecular mechanisms of a cardiac disease.

3Pos132 温めた心筋細胞に備わった収縮振動は遅い Ca^{2+}変動に対して周期を一定に保つ

Contractive oscillations intrinsic to heating cardiomyocytes maintain the period against late Ca^{2+} variations

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The heart beat regulate Ca^{2+} variations. But 38~42 °C cardiomyocyte could express contractive oscillations independent of Ca^{2+} variations. The Ca^{2+} variation frequency of isolated cardiomyocyte is slower than heart because pacemaker cells are removed. And the period of contractive oscillations are similar to heart beat, so we could separate Ca^{2+} variations and Contractive oscillations. Interestingly, amplitude of the contractive oscillations are regulated by Ca^{2+} variations, on the other hand, period of the contractive oscillations are independent of Ca^{2+} variations. Using the cardiomyocyte simulation based on chemical and mechanical state of myosins, we create the model of the period maintain mechanism. This oscillation stability may essential to heart beat.

3Pos133 周期性伸展刺激における伸展周期と心筋細胞の応答の関係

Relation between stretch cycles and response of cardiomyocytes in cyclic stretch stimulation

Chiho Nihei, Tomoyuki Kaneko (*LaRC, Grad. Sci. Eng., Hosei Univ.*)

Mechanical stress plays important roles in metabolism and development. Especially, the stress is involved in structural and functional maintenance of hearts. In this experiment, we applied mechanical stress to cardiomyocytes isolated from chicken embryo by using the cultured cell stretch system, and researched changes of shape and orientation in the cells. The result showed cardiomyocytes oriented vertical to the stretch direction on only conditions that stretch cycles was high frequency (60 cycles / min). However, cardiomyocytes contract in the direction of cellular long axis in vivo, and cellular long axis and direction of mechanical stress are parallel. These evidence suggested that cardiomyocytes might have other mechanism in response to cyclic stretch stimulation.

3Pos134 Mg^{2+}/Ca^{2+} 交換及び温度変化による F-アクチンの三次構造変化

Tertiary structure of F-actin affected by Mg^{2+}/Ca^{2+} and temperature

Takaya Yamaguchi, George Mogami, Makoto Suzuki (*Grad. Sch. Eng., Univ. Tohoku*)

The tertiary structural change of F-actin (FA) by exchanging Mg^{2+}/Ca^{2+} ions have been revealed by circular dichroism (CD) spectroscopy in the short and long wave length ranges from 200 to 240 nm and from 280 to 320 nm. FA in Mg(2mM)-containing buffer (Mg-FA) exhibited a Cotton effect around 290 nm which was attributed to the Trp arrangement. This Cotton effect around 290 nm was found to decrease sensitively by addition of Ca^{2+} ion in the range from 0.03 to 0.1 mM. This tertiary structural change of FA was reversibly controlled by addition and reduction of Ca^{2+} ion concentration using EGTA. Moreover, Mg-FA exhibited a thermosensitive tertiary structural change in the range from 10 to 25 ℃, while Ca-FA exhibited less temperature dependence.

3Pos135 Does homo hexamer function as a stator of rotary motor?

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V_1 of bacterial V-ATPase is an ATP driven rotary molecular motor, consisting of central rotor of DF and stator hexamer A_3B_3 which is a member of AAA+ family. Although most of AAA+ proteins form homo hexamer, stators of both V_1 and F_1 are composed of hetero hexamer (A_3B_3 or $\alpha_3\beta_3$). In this study, we inquire whether homo hexamer functions as a stator of rotary molecular motor. We constructed expression system of FliI derived from bacteria flagellar. The ATPase active $FliI_6$ was obtained by gel filtration column chromatography. Then we constructed the expression system of FliJ which is a coiled coil protein like rotor proteins. We are trying to reconstitute $FliI_6$ - FliJ complex, and to observe rotation of the inserted FliJ relative to $FliI_6$ by single molecular observation.

3Pos136 全原子 MD と粗視化 MD を組み合わせたマルチスケール MD 解析による V1-ATPase の回転機構の解明
Rotation mechanism of V1-ATPase elucidated by multi-scale MD analysis

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V_1-ATPase is a molecular motor constructed by the A_3B_3 hexamer ring and the central stalk. Coupling with an ATP hydrolysis, the A_3B_3 ring change own conformations and their packing manner, and then the conformational change induces a 120-degree rotation of the stalk. To understand the relationship between the conformational change and the rotation, we performed a multiscale MD simulation in which coarse-grained and all-atom MD simulations were combined using a fluctuation matching. In the simulation, we found two key movements for 120-degree stalk rotation: spontaneously taking the intermediate state `bindable-like' structure and creation of the space to avoid steric hindrances.

3Pos137 Probing the biophysical properties of a Thermoalkaliphilic F1 ATPase gives insight into adaptation and regulation

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Alkaliphilic F1Fo ATP synthases are highly adapted, performing oxidative phosphorylation at high pH against an inverted pH gradient (acidin/ alkalineout) and have a tightly regulated ATP hydrolysis activity that can be relieved in the presence of LDAO or mechanical activation. Here we explore the rotary dynamics of TA2F1, a thermoalkaliphilic ATP synthase, with single-molecule analysis. Reported are the enzymatic and biophysical properties of TA2F1, including kinetics, regulatory features, and the stepping characteristics of this molecular motor. Torque measurements revealed the highest torque for an F1 molecule described using fluctuation theorem. The mechanism of LDAO activation and implications of a high torque in terms of extreme environment adaptation are presented.

3Pos138 高速 AFM で明らかにする回転軸の無い腸内連鎖球菌由来 V_1-ATPase の一方向的協同性度合
The Extent of Unidirectional Cooperativity in Rotorless *Enterococcus hirae* V_1-ATPase Revealed by High-speed AFM

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V_1-ATPase is the soluble part of V-ATPase and known to be a rotary motor similar to F_1-ATPase. We previously revealed that the $\alpha_3\beta_3$ ring of F_1-ATPase has intrinsic cooperativity in adopting conformational states, even without the central γ shaft. Here we used high-speed AFM to investigate if such cooperativity also exists in the A_3B_3 ring of V_1-ATPase. Under the microscope, the A_3B_3 ring at ~μM ATPγS showed conformational changes of the A subunits propagating in the anticlockwise direction, but its transition to the clockwise direction was also observed, suggesting weaker cooperativity in V_1-ATPase. We would like to discuss the cooperativity of the rotorless A_3B_3 complex, while presenting more data obtained using ATP and a low ATP hydrolysis mutant (B(R350K)).

3Pos139 リン酸結合蛋白を封入したフェムトリットル体積のドロップレットアレイによる無機リン酸検出
Detection of inorganic phosphate by phosphate binding protein encapsulated in femtoliter droplet arrays

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Detection of inorganic phosphate (Pi) is an important factor in elucidating mechanisms of ATPase-driven motor proteins. Low temporal resolution of previous Pi quantification methods such as an end-point assay followed by a color reaction had been overcome by real-time measurements of Pi-dependent increase in fluorescence intensity of rhodamine-labeled phosphate binding protein (Rho-PBP). Here we further applied this method to femtoliter chamber arrays to develop a system for detection of small numbers of Pi dissociated from single-molecule enzymes. The rotary motor protein F_1-ATPase was encapsulated in droplet chambers together with Rho-PBP, and the increase in Pi concentration due to ATP hydrolysis has been monitored by fluorescence response of Rho-PBP.

3Pos140 Unveiling the chemomechanical coupling of F1 ATPase of Paracoccus denitrificans

Mariel Zarco - Zavala[1], Duncan G G McMillan[1], Toshiharu Suzuki[1], Hiroshi Ueno[1], Francisco Mendoza-Hoffmann[2], Jose J. Garcia-Trejo[2], Hiroyuki Noji[1] ([1]*Department of Applied Chemistry, Graduate School of Engineering, The University of Tokyo*, [2]*Departament of Biology, Chemistry Faculty, National Autonomous University of Mexico*)

P. denitrificans (Pd) is a bacterium related to the protomitochondria that harbors a respiratory chain extremely similar to that of mitochondria. The Pd-ATPase has a tightly regulated hydrolytic activity controlled by a novel alpha-proteobacteria exclusive inhibitor the "ζ subunit", that appears to be functionally related to the IF1. Here, we establish an E. coli expression system for PdF1 suitable for single molecule analysis, and have started its biochemical and biophysical characterization. Recent studies have shown differences in the chemomechanical coupling of bacterial and mitochondrial F1, thus, characterization of the rotatory catalysis of PdF1 will provide an insight into the adaptations of ATPase machinery upon the endosymbiosis process.

3Pos141 腸球菌由来 ADP 結合型 V_1-ATPase の X 線結晶構造解析

Crystal structures of the ADP-bound V_1-ATPase from *Enterococcus hirae*

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V_1-ATPases are ATP-driven rotary motors in various membrane systems. We recently reported the crystal structures for the Enterococcus hirae V_1-ATPase, corresponding to the catalytic dwell state waiting for ATP hydrolysis. Here, we present the crystal structures obtained by soaking nucleotide-free V_1 crystals in ADP. In the presence of 20 μM ADP, two ADP molecules bound to two of three binding sites and cooperatively induced conformational changes of the third site to an ATP-binding mode, corresponding to the ATP-binding dwell. In the presence of 2 mM ADP, all nucleotide-binding sites were occupied by ADP to induce conformational changes corresponding to the ADP-release dwell. Based on these and previous findings, we propose a novel V_1-ATPase rotational mechanism model.

3Pos142 *Enterococcus hirae* 由来 V_1-ATPase アルギニンフィンガー変異体が示す特異な回転特性

Arginine finger mutant of *Enterococcus hirae* V_1-ATPase shows unusual rotational behaviors

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V_1-ATPase (V_1) is a rotary molecular motor driven by ATP hydrolysis. To understand the chemo-mechanical energy conversion mechanism of V_1, we conducted single-molecule analysis of an arginine finger mutant (B(R350K)) of *Enterococcus hirae* V_1. The B(R350K) mutant showed 300-times lower maximum velocity and 400-times lower Michaelis constant than those of wild-type, indicating that not ATP biding, but ATP hydrolysis and/or product release become very slow as compared with the wild-type. Furthermore, interestingly, B(R350K) mutant frequently showed very short pauses at high [ATP] and backward steps in the presence of high [ADP]. In this presentation, we will discuss chemo-mechanical coupling mechanisms which cause these unusual rotational behaviors of the B(R350K) mutant.

3Pos143 Structural analysis by NMR on C-terminal region of FliG, an essential motor component of Vibrio Na^+-driven flagella

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The flagellar motor protein complex plays a crucial role in bacterial movement. We have been focusing on the sodium-driven flagellar motor protein complex in Vibrio alginolyticus, and demonstrated that the C-terminal domain of FliG (FliGC) which is believed to interact with the stator protein PomA. To unravel the molecular mechanism of rotational motion, we characterize the structural and dynamic property of wild type and mutant FliGC defective in motility. From NMR analysis, changes in structure and dynamics were observed in several FliGC mutants. Moreover, molecular dynamics simulation showed conformational changes in FliGC. From these results, we conclude that the structural re-arrangement in FliGC is correlated to the change in rotational direction of flagellum.

3Pos144 サルモネラ属菌 FliFG 融合変異型べん毛モーターの動態機能計測

Functional analysis of a FliFG deletion-fusion mutant flagellar motor

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The bacterial flagellar motor is a bidirectional rotary motor. The rotator of the motor is composed of FliF, FliG, FliM and FliN proteins. Initially, 26 copies of FliF form the core ring called the MS ring with 26-fold rotation symmetry in the inner membrane. FliG then binds to FliF at the 1:1 ratio. Finally, the FliN-FliM complexes bind to FliG and form the C ring with 34-fold rotational symmetry. To examine the effect of this symmetry mismatch between the MS ring and C ring on the motor function, we studied a FliF-FliG deletion fusion mutant that has the C ring with a smaller diameter than the wild-type and an altered rotation symmetry. The rotation direction of the mutant motor was biased to the clockwise direction. Detailed functional analysis is under way.

3Pos145 光ピンセットを用いたバクテリアべん毛モーターの最大トルクの計測

Maximum torque generated by the bacterial flagellar motor measured by optical tweezers

Taishi Kasai[1], Yoshiyuki Sowa[1,2] ([1]*Reserch center for Micro-Nano Tech. Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*)

The bacterial flagellar motor is a rotary molecular machine which is powered by the ion flux across the cell membrane. The motor consists of a rotor of 50 nm in diameter surrounded by up to ~11 stator units. Multiple stator units interact with the rotor to generate torque independently in a working motor. The stator units dynamically exchange between the motor and a pool in the cell membrane during motor rotation, and their stability in the motor depends on the external mechanical load to the motor. Therefore, the measurement of motor torque under controlled load conditions is important for understanding energy conversion mechanism of the motor. In this study, we measured maximum torque of the motor by combining optical tweezers and nanometry with high-speed camera.

3Pos146 べん毛モーターのトルク特性とその個体差の精密測定

Precise measurement of torque characteristics and individual variability of bacterial flagellar motor

Kento Sato, Shuichi Nakamura, Seishi Kudo, Shoichi Toyabe (*Grad. Sch. Eng., Tohoku Univ.*)

The bacterial flagellar motor rotates driven by ion flows. The measurement of torque characteristic is critical for revealing the motor design principle. However, fundamental torque characteristics including the maximum torque which indicates the motor performance has not been accurately measured. In this study, we precisely measured the torque characteristics of flagellar motor of a tethered salmonella by using electrorotation. We applied an external torque of controlled magnitude and precisely measured both the external torque and the viscous torque loaded on the cell body. We report our measurement of the torque-speed relationship under a broad range of load for an individual motor of an individual cell.

3Pos147 The mechanism of Vibrio alginolyticus polar flagellum growth

Chien-Jung Lo, Meiting Chen (*Department of Physics, National Central University*)

Bacterial flagella are self-assembled external tubular filaments. Flagellins, flagellum monomers, are pumped out from the basal body associated with six integral membrane proteins (FlhA, FlhB, FliO, FliP, FliQ and FliR) and three soluble proteins (FliH, FliI, and FliJ) through the nanometer transportation system. As flagellin reaches the end of flagellar filament, it becomes the new extending part of the filament. In order to study this nanometer size self-assembled system, we have developed a fast flagellar protein binding assay to directly monitor the length of the polar flagella of Vibrio alginolyticus. The flagellar growth rates of wild-type, long, and short flagella strains were measured. We will discuss the mechanism of the flagellar growth.

3Pos148 Functional Analysis of Slow-Motile Mutations in Flagellar Stator MotA/B of Salmonella

Seyedehnoorolhoda Shajaripourjaberi, Naoya Terhara, Tohru Minamino (*frontier biosciences*)

Stator of proton driven bacterial flagellar motor composed of two cytoplasmic membrane proteins, MotA and MotB. The torque that is required for rotating the flagellar filament is generated by the electrostatic interaction between FliGc of the rotor and highly conserved charged residues of cytoplasmic loop of MotA, which located between TM2 and TM3. Mutants that have single-mutation in these residues show slow-motility phenotype. We characterized these mutants by measurement of their motility and found that these mutants decreased motility compare to wild type cells. We measured the rotational speed of motors by bead assays and obtained elaborated understanding of torque generation mechanism by analyzing the torque-speed relationship of motors of the slow-motile mutants.

3Pos149 Rng2によるF-アクチンとミオシン間の協同的相互作用の制御
Regulation of cooperative interaction between myosin and F-actin by Rng2

Taiga Imai[1], Masak Takaine[2], Kentaro Nakano[2], Osamu Numata[2], Taro Uyeda[3], Kiyotaka Tokuraku[1] ([1]*Muroran institute of technology*, [2]*University of Tsukuba*, [3]*Waseda University*)

Rng2 is a protein that localizes in the contractile ring (CR) of fission yeast and relates to the formation and control of the CR. However, the regulatory mechanism of CR by Rng2 has remained unknown. Recently, we found that Rng2CHD, which is a F-actin binding domain of Rng2, strongly inhibited sliding of F-actin on HMM in in vitro motility assay. In this study, we examined the effect of Rng2CHD for cooperative interaction between HMM-GFP and F-actin by fluorescence microscopy. The results revealed that substoichiometric amount of Rng2CHD in the presence of ATP inhibited the cooperative binding of the HMM-GFP to F-actin, suggesting that Rng2 regulates of the cooperative interaction between myosin and F-actin.

3Pos150 SH1ヘリックス内に変異をもつミオシンIIはアクチンフィラメントの滑りの活性化エネルギーを減少させる
Myosin II SH1 helix mutant lowers the activation energy for sliding of Factin

Shigeru Chaen[1], Kotomi Shibata[1], Tsubasa Koyama[1], Atsushi Suenaga[1], Sosuke Iwai[2] ([1]*Dept. Biosci. Nihon Univ.*, [2]*Dept. Biol. Hirosaki Univ.*)

The SH1 helix region of myosin II is a joint that links the converter subdomain to the rest of the myosin motor domain and possibly plays a key role in the arrangement of the converter/lever arm. Mutations within the SH1 helix in human myosin IIs have been shown to cause diseases. To reveal whether these SH1 helix mutations affect thermal properties of myosin II, here we introduced the E683K or R686C point mutation into the SH1 helix in Dictyostelium myosin II. We found that these mutations decreased both the thermal stability of myosin II and the activation energy of a rate-limiting process involved in actin movement. These results suggest that SH1 helix is a key structural element that determines the flexilbility and thermal properties of myosin motor.

3Pos151 プログラム可能なミオシンアセンブリの設計と高解像1分子イメージング
Design of a programmable myosin motor assembly and nanometer-precision single-molecule imaging

Masashi Ohmachi[1], Keisuke Fujita[1], Keigo Ikezaki[2], Toshio Yanagida[1,3], **Mitsuhiro Iwaki**[1,3] ([1]*QBiC, RIKEN*, [2]*Univ. of Tokyo*, [3]*Osaka Univ.*)

Molecular machines typically coordinate together and emerge physiological functions in cells. Muscle is such an assembly system mainly composed of actomyosin. While mechanical motion of isolated single myosins and ensemble motion of myosin filament are well characterized, how single myosins in an assembly behave or individual myosins in an assembly are coordinated together is unclear. To resolve the internal dynamics in muscle at single molecule resolution, we have developed a programmable myosin filament composed of skeletal myosin II constructs and 3D DNAorigami, where we can freely control number, spacing, orientation and species of myosins. We'll discuss nanometer-precision single-molecule imaging of our programmable myosin filaments.

3Pos152 単純化された筋繊維の計算モデルを用いた筋収縮におけるバイアスブラウン運動の寄与の研究
Contribution of biased Brownian motion in muscle contraction studied by a simplified computational model of muscle fiber

Daisuke Watanabe, Masaki Sasai, Tomoki P. Terada (*Dept. Comput. Sci. Eng., Grad. Sch. Eng., Nagoya Univ.*)

Using coarse-grained molecular dynamics, we have obtained free energy landscapes for different structural/chemical states of a single myosin head moving along an actin filament (Nie et al., PLoS Comput. Biol. (2014)). In the present study, in order to elucidate the roles of biased Brownian motion and structural/chemical changes of myosin in muscle contraction, we have constructed a simplified computational model of muscle fiber consisting of a single actin filament and three myosin filaments in a half sarcomere, in which each myosin head experiences dynamic change of free energy landscape. We discuss the contribution of biased Brownian motion in the macroscopic characteristics of muscle fibers, such as relationships among displacement, velocity and generated force.

3Pos153 Rng2 によるアクチンフィラメントの構造変化と、HMM で駆動されるアクチン運動の協同的阻害

Structural changes of actin filaments induced by Rng2, and the resultant inhibition of actin movement on HMM

Yuki Hayakawa[1], Keiko Hirose[2], Masafumi Yamada[2], Kien X. Ngo[1], Noriyuki Kodera[4], Masak Takaine[3], Kentaro Nakano[3], Osamu Numata[3], Taro Uyeda[1,2] ([1]*dep physics, Waseda Univ.*, [2]*Biomed Res Inst, AIST*, [3]*Grad. School of Life and Environ Sci., Univ. Tsukuba*, [4]*Bio-AFM Res Ctr, Kanazawa Univ.*)

Rng2 is an actin-binding IQGAP protein essential for the formation of contractile rings in fission yeast, but how this is done is unknown. In *in vitro* motility assays, calponin-homology actin binding domain of Rng2 (Rng2CHD) strongly inhibited sliding of actin filaments on HMM surfaces. IC_{50} (~0.01 μM) was much lower than K_d (~0.2 μM), suggesting that each bound Rng2CHD molecule inhibits productive interaction of many neighboring actin protomers with myosin II motors. Intriguingly, Rng2CHD did not inhibit movement by myosin V in vitro. In negatively-stained electron micrographs, the filaments interacting with Rng2CHD appeared rough and fragile. We are currently performing high-speed AFM observations to further gain insight into the structural changes induced by Rng2CHD.

3Pos154 Effect of external mechanical stress on collective motion of microtubules

Tamanna Ishrat Farhana[1], Arif Md. Rashedul Kabir[2], Daisuke Inoue[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. Chem. Sci. & Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*)

The collective motion of microtubules (MTs) driven by motor proteins in vitro is known to produce fascinating patterns including local streams, aster, vortices pattern and so on. These collectively moving MTs can reorient to preferential direction on application of controlled external mechanical perturbation. Previously, our group has demonstrated that, the application of uniaxial mechanical stretching in single step and cyclic manner can reorient the spatial arrangement of the motile MTs. In this work, we present the formation of an ultra large vortex pattern upon application of mechanical perturbation through indentation stress. The effect of different parameters related to formation vortex has also been investigated and will be discussed in details.

3Pos155 微小管内 GDP-チューブリンの精密な周期決定

Determination of accurate axial tubulin repeat in GDP-microtubules

Shinji Kamimura[1], Hiroshi Imai[1], Toshiki Yagi[2], Tomohiro Shima[3,4], Yasushi Okada[4,5], Hiroyuki Iwamoto[6] ([1]*Dept. Biol. Sci., Chuo University*, [2]*Dept. Life Sci., Pref. Univ. Hiroshima*, [3]*Dept. Biol. Sci., Univ. Tokyo*, [4]*Lab. Cell Polarity Regulation., Qbic, RIKEN*, [5]*Dept. Phys., Univ. Tokyo*, [6]*Life & Environmental Div., SPring-8*)

The axial repeat of tubulin molecules in microtubules is roughly 4 nm and has conventionally been used as one of the most convenient standard scales to estimate molecular or organelle size for electron microscopy. However, in our previous report, the axial repeat varied depending on experimental conditions, e.g., temperature, with microtubule stabilizer and GTP-hydrolysis states. It was also shown that the tubulin axial repeat in porcine brain GTP-microtubules, which are mainly composed of GDP-tubulin, was almost constant with a low coefficient of thermal expansion of 3.7×10^{-5}/degree. Here, we determined the accurate repeat of GDP-tubulin molecules after careful revision of camera length, X-ray wavelength as well as the angles of specimen, beam and detector tilting.

3Pos156 高精度な微小管分離に向けた微小管の持続長設計

Design of Microtubule Persistence Length Toward High-precision Microtubule Sorting

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We propose a microtubule (MT) sorting system by varying MT persistence length. Six MT groups were polymerized under different conditions, and their persistence length were investigated. Result shows nucleotide in a polymerization buffer and/or tubulin concentration (growth rate) had significant effects; GMPCPP or slowly polymerized MTs were stiffer than GTP or fast polymerized ones. We also found MTs polymerized below the growth rate of ~1.0 micrometer/min showed high and constant persistence lengths, while persistence length decreased above the growth rate. Furthermore, additive effects of persistence length and our previously reported electrical charge enabled to demonstrate sorting of two MT groups with ~85% precision in a microfluidic device with 3 kV/m applied.

3Pos157 Regulated swarming of molecular robots prepared from a DNA programmed biomolecular motor system

Jakia Jannat Keya[1], Ryuhei Suzuki[1], Arif Md. Rashedul Kabir[2], Daisuke Inoue[2], Kazuki Sada[1,2], Akinori Kuzuya[3], Akira Kakugo[1,2] ([1]*Graduate School of Chemical Sciences and Engineering, Hokkaido University*, [2]*Faculty of Science, Hokkaido University*, [3]*Faculty of Chemistry and Bioengineering, Kansai University*)

Swarming is a fascinating display of coordinated behavior of living organisms which most often give rise to emergent functions. Researchers have been motivated to construct molecular robots which may exhibit artificial swarm intelligence similar to observed in nature. Here by integrating molecular devices through a bottom up approach, we developed molecular robots and demonstrated their swarming. We used biomolecular motor protein kinesin as actuator which propelled microtubules (MTs) in an in vitro motility assay. As a processor highly selective and programmable material DNA was used that allowed logical operations of swarming of the MTs. The swarming pattern and motion of MTs was regulated through DNA input signals which led to designing for a more complex system.

3Pos158 構成論的手法を用いた鞭毛の機能的再構築

Functional reconstitution of flagellar axonemes by self-organization of microtubules, dynein docking complex and outer arm dyneins

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The coordination among dynein arms makes flagella beat in an organized manner. To reveal this mechanism, we reconstituted axonemal structures in vitro in a bottom-up manner. Biotinylated tubulins were polymerized into microtubules and attached on avidin-coated glass surface. High-salt extract of dynein arms from Chlamydomonas axonemes were added to the microtubules. Dynein formed regular arrays on the microtubules in a self-organized manner. Microtubules freshly added to these dynein-microtubule complexes formed bundles with them. Adding ATP made a pair of microtubules display occasionally repetitive associations and dissociations cycles. These results suggest that repetitive bending of the axoneme is simply derived from dynein-microtubule interactions.

3Pos159 電子顕微鏡トモグラフィーによって明らかになった細胞質ダイニンのモータードメインの配置

Orientation of two motor domains of cytoplasmic dynein characterized using electron computed tomography

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Cytoplasmic dynein has a complex and massive structure containing two identical heavy chains. The heterogeneous architecture of the dynein gives us difficulty with crystallization or averaging a large number of molecules to achieve high resolution. In this study, each three-dimensional structure of cytoplasmic dynein bound to microtubule was constructed by electron tomography. This approach succeeded in visualizing AAA+ domain of the dynein head and its stalk associating microtubule. The two motor domains of dynein AAA+ rings faced each other with the flat side on the inside. This conformational feature of dynein was reproduced using cryo-EM. Structural characterization by electron tomography analysis has uncovered native cytoplasmic dynein structure.

3Pos160 ポリグルタミン酸化酵素欠損マウスの気管繊毛が示す異常な跳ね上がり運動

Abnormal "hopping" of mouse tracheal cilia deficient in tubulin polyglutamylation

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Motility of cilia is the fundamental mechanism for exclusion of harmful materials from mammalian trachea. It has been revealed that posttranslational modification of microtubules constituting the axoneme affects structure and motility of cilia. Indeed, 2D trajectories of cilia deficient in tubulin polyglutamylation lost asymmetry and caused accumulation of mucus (Ikegami, 2010). Here we reinvestigated motility of cilia from the knockout mice under a 3D tracking microscope. Beads attached to demembranated single cilia from knockout mice displayed a transient large upward movement at the beginning of a recovery stroke more frequently than the wild type mice. Therefore, constant low trajectory of the recovery stroke may contribute to stable fluid flow.

3Pos161 クラミドモナス鞭毛の波形変化に関わるタンパク質の探索

Searching a putative protein responsible for switching waveform of Chlamydomonas flagella

Junya Kirima[1], Misaki Shiraga[1], Hiroaki Kojima[2], Kazuhiro Oiwa[1,2] ([1]*Grad. Sch. Life Sci., Univ. Hyogo*, [2]*Adv. ICT Res. Inst., NICT*)

Chlamydomonas flagella show a photo-shock response against strong light exposure, in which the waveform temporally changes from a ciliary type to a flagellar type. Since the response is elicited by increase in the intracellular Ca concentrations, the calcium binding proteins in an axoneme would play some roles of the waveform changes. Among uncharacterized several proteins with the EF-hand motif, we focused on FAP85. The western blotting with anti-FAP85 antibody confirmed that FAB85 exists even in the mutant axonemes lacking major axonemal component (oda1, oda2, ida1, ida5, pf2, pf3, pf14, pf18 and mbo1). Immunofluorescence microscopy on wild type axonemes showed FAP85 exists along a whole flagellum. These results suggest FAP85 is localized on doublet microtubules.

3Pos162 バクテリア・カビ由来セロビオヒドロラーゼの結合、解離、プロセッシブ運動の一分子蛍光観察

Single-molecule fluorescence analysis of binding, dissociation, and processive movement of bacterial and fungal cellobiohydrolases

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Cellobiohydrolase is a linear motor protein that hydrolyzes crystalline cellulose processively. It consists of catalytic domain, cellulose binding module (CBM) and linker. Here we visualized binding, dissociation, and processive movement of intact and domains of *Cellulomonas fimi* Cel6B (CfCel6B) and *Trichoderma reesei* Cel6A (TrCel6A) with single-molecule fluorescence imaging. Binding rate constants of CBM from CfCel6B and TrCel6A were 35 % and 7.5% of intact, respectively. These results indicate that structural differences in CBM from CfCel6B and TrCel6A affect initial binding to cellulose surface. From the translational rate and duration time of movement of intact, the values of processivity for CfCel6B and TrCel6A were estimated as 52 and 68, respectively.

3Pos163 分子モーター RecBCD による混雑環境下でのタンパク質-DNA 複合体の除去

Sequential eviction of crowded nucleoprotein complexes by the RecBCD molecular motor

Tsuyoshi Terakawa[1], Redding Sy[1,2], Silverstein Timothy[1], Greene Eric[1] ([1]*Columbia Univ.*, [2]*Univ. of California*)

In physiological settings, all nucleic acids motor proteins must travel along substrates that are crowded with other proteins. However, the physical basis for how motor proteins behave in these highly crowded environments remains unknown. Here, we use real-time single molecule imaging, kinetic Monte Carlo simulations and Molecular dynamics simulations to determine how the ATP-dependent translocase RecBCD travels along DNA occupied by tandem arrays of DNA-binding obstacle proteins. We demonstrate that RecBCD forces each protein into its nearest adjacent neighbor, causing rapid disruption of the underlying protein-nucleic acid interface. This result suggests that molecular crowding itself completely alters the mechanism by which RecBCD removes protein obstacles from DNA.

3Pos164 RHAU ヘリカーゼがグアニン４重鎖構造の安定性を調節するメカニズムの一分子研究

RHAU helicases regulate G4 stability during the ATPase cycle revealed from single-molecule analysis

Huijuan You[1], Jie Yan[1,2] ([1]*MBI, National University of Singapore*, [2]*Dep. of Physics, National University of Singapore*)

RHAU helicase specifically resolves thermodynamically stable four-stranded G-quadruplex (G4) structures that are implicated in a wide array of biological processes. Importantly, RHAU has been suggested to be a major source of G4 resolving activity in cells. Through direct probing the G4 stability at single-molecule level, we show that binding of RHAU stabilizes G4 in the absence of nucleotide, while it destabilizes G4 when coupled to ATP hydrolysis. We also show that the unfolding kinetics of a G4 structure can be modulated by different ATPase states of RHAU. These findings provide important insights into the ATP-dependent RHAU-mediated G4 destabilization mechanism.

3Pos165 転写バーストは DNA 上の RNA ポリメラーゼの相互作用によって内因的に引き起こされる

Transcriptional bursting is intrinsically caused by interplay between RNA polymerases on DNA

Keisuke Fujita[1,2], Mitsuhiro Iwaki[1,2], Toshio Yanagida[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. of Front. Biosci., Osaka Univ.*)

Transcriptional bursting has been broadly studied as the potential source of cell-to-cell variability. Although molecular mechanisms of transcriptional bursting have been proposed, it is still unclear whether transcriptional bursting is intertwined with many transcriptional regulatory factors or is an intrinsic characteristic of RNA polymerase on DNA. Here, we designed an in vitro single-molecule measurement system to analyze the kinetics of transcriptional bursting. The results indicated that transcriptional bursting is caused by interplay between RNA polymerases on DNA. In the meeting, we plan to introduce a novel experimental design to confirm our model in vivo and modulate the kinetics of transcriptional bursting in vivo.

3Pos166 金ナノプローブで明らかにされたリニア分子モーター霊菌 *Serratia marcescens* 由来キチナーゼ A の 1 nm ステップ運動

One nanometer steps in the motion of a linear molecular motor *Serratia marcescens* chitinase A resolved by gold nanoprobe

Akihiko Nakamura[1,2], Ryota Iino[1,2,3] ([1]*Okazaki inst. for Integrative Bioscience*, [2]*The Graduate University for Advanced Studies (SOKENDAI)*, [3]*Institute for Molecular Science*)

Serratia marcescens chitinase A (*Sm*ChiA) is a linear molecular motor moving on and hydrolyzing crystalline chitin. Here we directly visualized steps and pauses of *Sm*ChiA with gold nanoparticle as a probe. Step sizes were 1.1 nm and -1.2 nm for forward and backward steps, respectively, consistent with the length of product chitobiose (~1 nm). The ratio of forward to backward steps was 18, corresponding to the energy difference of 2.9 kBT. Interestingly, 2.4 nm forward steps were also observed, and chitotetraose without one acetyl group was detected with MALDI-TOF Mass. These results indiciate *Sm*ChiA skips one chitobiose unit if deacetylated. Distribution of pause was fitted by single exponential decay with time constant of 28.6 ms, indicating single rate-limiting step.

3Pos167 海洋性ビブリオ菌のべん毛形成に関わる DnaJ ファミリータンパク質 SflA のペリプラズム領域の構造特性

Structural property of the periplasmic TPR domain of SflA, a DnaJ family protein involved in flagellation of *Vibrio alginolyticus*

Mayuko Sakuma[1,2], Satoshi Inaba[2], Shoji Nishikawa[3], Takehiko Nishigaki[2], Seiji Kojima[2], Katsumi Imada[3], Michio Homma[1,2] ([1]*Radioisotpe Res. Cent., Nagoya Univ.*, [2]*Div. Biol. Sci. Grad. Sch. Sci., Nagoya Univ.*, [3]*Dept. of Macromol. Sci., Grad. Sch. Sci., Osaka Univ.*)

SflA is a 35kDa single transmembrane protein involved in negative regulation of flagellation in marine bacterium *Vibrio alginolyticus*. SflA consists of the N-terminal periplasmic region ($SflA_N$), the C-terminal cytoplasmic J-domain, which is conserved in DnaJ family proteins, and a transmembrane helix connecting the two regions. We have solved the crystal structure of $SflA_N$ at 2.0 Å resolution. The core structure of $SflA_N$ adopts a Tetratricopeptide repeat (TPR) motif, suggesting that it binds to some other protein molecules. $SflA_N$ forms a dimer and the dimer structure can be superimposed on SycD, a type III secretion chaperon containing TPR. By comparing these structures, we predict the amino acids involved in protein-protein interaction in $SflA_N$.

3Pos168 FlhF がもつ GTPase モチーフへの変異によるビブリオ菌極べん毛数と位置への影響

Effect of mutations in the GTPase motif of FlhF on the number and location of the polar flagellum of Vibrio alginolyticus

Shota Kondo, Michio Homma, Seiji Kojima (*Division of Biological Science, Graduate School of Science, Nagoya University*)

Vibrio alginolyticus has a single polar flagellum whose number and placement are regulated positively by FlhF. FlhF is a GTPase and homolog of signal recognition particle (SRP) protein Ffh and SRP receptor FtsY. FlhF is localized at cell pole and direct the formation of flagellum. To investigate the mechanism of FlhF localization, we introduced random mutations to *flhF* by hydroxylamine and isolated mutants that cannot generate flagellum at the cell pole. Mutations were mapped on the GTPase motif of FlhF. The mutant FlhF proteins showed reduced polar localization than wild type and still could associate with membrane. These results indicate that the GTPase motif of FlhF has a critical role for the polar localization.

3Pos169 細菌 Rhomboid プロテアーゼ GlpG の生理的基質の探索：GlpG のべん毛 III 型分泌装置機能への関与の可能性

Screening of physiological substrates of *E. coli* rhomboid protease GlpG: possible involvement of GlpG in the flagellar function

Yohei Hizukuri, Kosuke Terushima, Yoshinori Akiyama (*Inst. Virus Res., Kyoto Univ.*)

Rhomboid family intramembrane proteases are found in all kingdoms of life. In eukaryotes, they are involved in various important cellular events such as proteolytic activation of membrane-bound EGFR ligands. In contrast, their substrates and physiological roles in prokaryotes remain largely unknown. To explore the function of an *Escherichia coli* rhomboid homologue, GlpG, we screened a total of 204 predicted single-spanning transmembrane proteins encoded in the *E. coli* genome for their cleavage by GlpG, and found that a component of the flagellar type III secretion apparatus received a GlpG-dependent cleavage *in vivo*. We are now investigating the physiological significance of this finding.

3Pos170 集合に共役したべん毛モーター固定子 MotB のペリプラズム領域における構造変化の変異体解析

Mutational studies of the assembly-coupled conformational change in the periplasmic region of a flagellar stator protein MotB

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Bacterial flagellar motor generates torque by the rotor-stator interaction that couples with the ion flow through the channel in the stator. Stator units become active only when they are anchored around a rotor via the periplasmic region of the stator B subunit. We determined the crystal structure of this region from MotB (MotBC) and proposed a large conformational change during the stator incorporation into the motor. A mutation L119P in helix α1 of MotBC causes increased proton conduction and incorporation into the motor, suggesting that this mutant stator mimics the active conformation. Here we report the crystal structure of MotBC(L119P) and further mutational analyses to characterize and test the conformational change in the MotBC during the stator incorporation.

3Pos171 細菌べん毛モーター固定子蛋白質 MotA および回転子蛋白質 FliF の構造解析のための条件検討

Screening for the structural analysis of the stator and the rotor proteins MotA and FliF in the bacterial flagellar motor

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The bacterial flagellar motor is driven by the interaction between the stator and the rotor. The stator is composed of MotA and MotB, and the rotor rings are composed of FliF, FliG, FliM and FliN. The detailed molecular structures of those proteins except for MotA and FliF have already been reported. In this study, we performed the purification and biochemical analysis of MotA and FliF from thermophilic bacteria. The purified MotA formed tetrameric complex and crystallized, though more improvement was needed for X-ray analysis. The full-length and some periplasmic fragments of FliF were well purified and one of them formed stable dimer. We are now trying to grow better crystals of those proteins for structure analysis. We will discuss the progress in the poster.

3Pos172 FlhA の構造変換がべん毛蛋白質輸送順序の決定に重要である

Conformational rearrangements of FlhA is critical for ordered protein export during flagellar assembly

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A transmembrane export gate protein FlhA forms a nonameric ring structure in the bacterial flagellar type III export apparatus. FlhA not only acts as a sorting platform for flagellar chaperone-export substrate complexes accompanied by the cytoplasmic ATPase complex but also plays an important role in the energy coupling mechanism with the ATPase complex. The C-terminal cytoplasmic domain of FlhA (FlhAc) adopts two distinct, open and closed conformations. The conformational switch between these two forms is critical for the export activity. Here, we carried out structure-based functional analysis of FlhA. We show that the conformational switch of FlhAc affects the FlhAc-FlhAc interaction, thereby coordinating flagellar protein export with assembly.

3Pos173 サルモネラべん毛モーターにおける MotA Met-206 の役割

Roles of MotA Met-206 in rotation and assembly of the Salmonella flagellar motor

Yuya Suzuki[1], Kodai Oono[1], Shuichi Nakamura[1], Fumio Hayashi[2], Kenji Oosawa[3], Yusuke V. Morimoto[4], Seishi Kudo[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Center for Inst. Anal.*, [3]*Div. Mol. Sci., Fac. Sci. and Tech., Gunma Univ.*, [4]*QBiC, Riken*)

The bacterial flagellar motor is composed of a rotor and a stator, which converts proton influx through stator units into rotation. The stator consists of MotA and MotB. We previously performed rotation assays of the Salmonella mutant motor with a point mutation M206I in MotA and found that MotA-Met206 plays a key role for protonation of the stator. Here we examined the effect of MotA-Met206 on stator assembly into the motor by labeling the stator unit with fluorescent protein. Fluorescent observation showed that subcellular stator localization was somewhat reduced by the M206I mutation. This result suggests that MotA-Met206 could contribute not only to protonation of stator but also to stator assembly into the motor.

3Pos174 細菌べん毛 III 型タンパク質輸送の *in vitro* 再構築

***In vitro* reconstitution of the bacterial flagellar type III protein export**

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The flagellar export apparatus is used to construct the bacterial flagellum. It is composed of six membrane components and three soluble components. Proton-motive force is primarily required for protein export and the hydrolysis energy of ATP is important for the efficient export. However, the molecular mechanism is still unclear. Here we present an inverted membrane vesicle (IMV)-based *In vitro* transport assay system that enables the measurement under well-controlled conditions. We succeeded in transporting flagellar proteins into the IMV and constructing the axial structure in the IMV. Addition of the soluble components in solution dramatically enhanced protein transport into the IMV, suggesting that they play an important role for efficient protein export.

3Pos175 反転膜を用いたべん毛 III 型蛋白質輸送の蛍光による検出

Fluorescence detection of the flagellar type III protein export using the inverted membrane vesicles

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The type III secretion system (T3SS) is bacterial protein export machinery used to deliver virulence proteins into their host cells. The flagellar protein export apparatus is a member of the T3SS family used for construction of the bacterial flagellum. To elucidate the molecular mechanism of the type III protein transport, we recently have constructed an in-vitro transport assay system of the flagellar export apparatus using inverted membrane vesicles (IMV). However, immunoblotting detection of translocated proteins into IMV limits the accurate measurement of protein export. Here we show the detection of proteins translocated into the IMV by fluorescence. This technique enables the accurate and quantitative analysis of the type III protein export.

3Pos176 バクテリアべん毛輸送ゲート複合体の発現系の構築と精製

Expression and purification of the bacterial flagellar type III export gate complex

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The bacterial flagellum is a supramolecular motility machine. The flagellar type III export apparatus, which is composed of a transmembrane export gate complex and a cytoplasmic ATPase complex, transports flagellar component proteins from the cytoplasm to the distal end of the growing structure for construction of the bacterial flagellum. The export gate complex is composed of FlhA, FlhB, FliO, FliP, FliQ and FliR and is localized in the basal body MS ring formed by FliF. Here, we constructed a co-expression plasmid encoding FlhA, FlhB, FliF, FliG, FliO, FliP, FliQ and FliR. We over-expressed these proteins in Salmonella and purified the MS ring together with the export gate complex. We will discuss the assembly mechanism and stoichiometry of the export gate components.

3Pos177 三次元的形状解析を用いたらせん形細菌の構造的理解

3D microscopic observation of the cell shape of spiral shaped bacteria

Hajime Tahara, Shuichi Nakamura (*Grad. Sch. Eng. Univ. Tohoku*)

Leptospira are spiral shaped bacteria. When swimming in a fluid, Leptospira transform the shapes of the cell ends by rotating the internal flagella. In this study, we de-energized Leptospira by addition of an uncoupler and examined diversity of the cell shapes by three-dimensional microscopy. We found that the de-energized cells are classified into at least three groups of morphology. We also showed that the morphology observed at the de-energized state influences motility form when the cell being resurrected by removing uncoupler. We propose a plausible model that the cell morphology somehow determined by physical interaction between the internal flagellar and cell membrane affects motility form of Leptospira.

3Pos178 Coordinated cell-body rotation in spirochete motion

Kyosuke Takabe, Seishi Kudo, Shuichi Nakamura (*Grad. Sch. Eng., Univ. Tohoku*)

Leptospira are spirochetes bacteria characterized by a right-handed helical cell body (protoplasmic cylinder, PC) and two intracellular flagella. Leptospira bend the anterior and posterior ends to spiral-shaped (S) and hook-shaped (H), respectively, for translation. Coordination of the rotation of both ends is crucial for unidirectional swimming. Previously, we showed a correlation between the PC and H rotations, and the S rotation could correlate with unidentified rotating parts of cell body for rotational balance. Here, we showed the possibility that not only rotations of the three parts (PC, S and H) but also "gyration" of an entire cell-body are involved in swimming of Leptospira, which will be valuable to understand the spirochete motion in further detail.

3Pos179 局所的な照明によって誘起されたスピロプラズマの遊泳方向の反転

Reversal motion of *Spiroplasma* induced by partial illumination

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Spiroplasma are helically shaped bacteria. They developed a distinct swimming strategy without the use of flagella. They simultaneously exhibit different helicities in bodies, and swim by the continuous propagation of kinks locating between two helices. To investigate the dynamics of the cytoskeletal structure inside cells, we illuminated strong light in the presence of FM 4-64 and specifically damaged the front tip of the cell body. Notably, the cells changed their swimming direction. In contrast to this reversal of the direction, the illumination to the rear end showed no significant effect on the swimming. These observations suggest that free rotation of the cytoskeletal structure around the front tip is requirement to control of the generation of kinks.

3Pos180 ロッド様に直線状で固く，野生型より長い変異型フックの金ナノ粒子標識によるべん毛モーターの高分解能回転計測

High resolution measurements of flagellar motor rotation by nanogold attached to a straight, solid hook mutant longer than the wild-type

Shuichi Nakamura[1,2], Yusuke V. Morimoto[2,3], Tohru Minamino[2], Keiichi Namba[2] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Frontier BioSci., Osaka Univ.*, [3]*QBiC, Riken*)

The bacterial flagellar motor is a rotary nanomachine fueled by proton motive force. We previously showed stepwise rotations of the Salmonella motor spinning under physiological conditions and proposed a model that the dwell time corresponds to the period of strong stator-rotor binding and that the rotation between dwells is driven by a biased Brownian motion. However, the response dynamics of the probe attached to the filament was problematic because of the elasticity of the hook and filament. Here we performed rotation assays using a mutant having a solid, straight and somewhat longer hook (120 nm) but no filament and observed very stable rotations by labelling nanogold directly to the hook. This assay will allow us to understand the motor mechanism more precisely.

3Pos181 シュードモナス属べん毛モーターの回転計測

Rotation assay of the Pseudomonas flagellar motor

Taro Hariu, Takuto Tensaka, Seisi Kudo, Shuichi Nakamura (*Grad. Sch. Eng., Tohoku Univ.*)

Pseudomonas syringae is a bacterium having multiple polar flagella fueled by proton motive force. Two sets of stator units, MotA/B and MotC/D, associate with the flagellar motor either simultaneously or independently and their output characteristics are likely to be different from each other; however qualitative measurements have not been performed. In this study, we measured torque of the P. syringae flagellar motor by tethered cell assay. We found that the stall torque of the P. syringae motor is larger than that of well characterized proton-driven E. coli motors, suggesting highly efficient energy conversion or high proton conductivity in P. syringae flagellar motor. We will also show the results of rotation assays of mutant motors lacking MotA/B or MotC/D.

3Pos182 ガラスに付着したビブリオ菌を用いたべん毛フリッキングの解析

Analysis of flagellar flicks of Vibrio cells stuck to a glass slide

Taichi Ohnuki, Shuichi Nakamura, Shoichi Toyabe, Seisi Kudo (*Grad. Sch. Eng., Univ. Tohoku*)

A phenomenon called "flicking" is observed for single-polar flagellated bacteria. The flicking occurs when a cell changes its swimming direction from backward to forward. During the flicking, the angle between the cell axis and the flagellar helical axis changes largely, resulting in up to 90° changes in the locomotion direction. The direction change is important for efficient chemotaxis. The flicking has been considered to occur as a result of "buckling" of the hook. However, detailed mechanism of the flagellar flicking is still unknown. In this study, we investigate the dynamic motion of the polar flagellum of V. alginolyticus stuck to a glass slide with a dark-field microscope, which enables us a long-term observation and detailed analysis of the flicking.

3Pos183 大腸菌の忌避刺激で見られた時間遅れは FliM の共同的な振る舞いにより説明される

Simulation of delays in repellent responses of *Escherichia coli* using a conformational spread model

Takashi Sagawa[1], Yoshiyuki Sowa[2], Ikuro Kawagishi[2], Kazuhiro Oiwa[1], Hiroaki Kojima[1] ([1]*Adv. ICT Res. Inst., NICT*, [2]*Dept. of Front. Biosci., Hosei Univ.*)

We measured flagellar rotational senses of *Escherichia coli* as chemotactic output to find significant delays in attractant and repellent responses. By contrast, no such delay was detected for repellent responses in the FRET analyses on the interaction between the motor component FliM and the signaling protein CheY (Sourjik, PNAS, 2002). To reconcile this apparent discrepancy, we applied a conformational spread model to simulate stochastic transitions in conformational states and ligand-binding states of 34 FliM monomers in a C-ring of the motor. The simulation reproduced the difference in the delay time between the flagellar rotation and the FliM-CheY interaction, suggesting that the delay is derived from cooperative conformational changes among FliM subunits.

3Pos184 大腸菌二成分制御系 AtoS, AtoC の相互依存的細胞内局在

Interdependent co-localization of the histidine kinase AtoS and the response regulator AtoC of *Escherichia coli*

Takahide Endo[1], Yukiko Miyao[1], Kentaro Yamamoto[1], Masatoshi Nishikawa[1], Yoshiyuki Sowa[1,2], Ikuro Kawagishi[1,2] ([1]*Dept. Frontier Biosci., Hosei Univ.*, [2]*Research Center for Micro-Nano Technology, Hosei Univ.*)

Two-component regulatory systems (TCS) mediate responses to various environmental stimuli. The *Escherichia coli* TCS consisting of the histidine kinase AtoS and the response regulator AtoC is involved in the gene regulation in response to short-chain fatty acids. Here we examined their cellular localization by constructing fusions to fluorescent proteins. Expression of AtoS-GFP and AtoC-TagRFP were co-localized at cell poles. However, when expressed alone, the former is distributed throughout the cytoplasmic membrane, and the latter showed dotted localization in the cytosol. His_6-tagged AtoC also induced polar localization of AtoS-GFP. These results suggest that AtoS and AtoC interdependently localize at cell poles, possibly facilitating signal amplification.

3Pos185 大腸菌は、アミノ酸種を識別する：データ駆動的アプローチにより明らかにする単細胞生物の化学知覚

Escherichia coli identify amino-acid species : unicellular organism's chemical perception revealed by using a data-driven approach

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Detection and recognition of chemical substances are primitive but vital for living organisms to survive. Organisms can detect and recognize various chemicals by using a relatively small number of receptors, integrate the information, and take actions. We focus on the organisms' ability of flexible recognition of environmental chemicals (information), and characterize the ability in a data-driven manner. We observe chemotactic behavior of E. coli against several chemical attractants, and analyze relationship between chemical stimuli and chemotactic response with a statistical framework. Our study reveals that E. coli recognize chemical species during chemotaxis, and show a novel sensory perception of unicellular organisms for processing environmental information.

3Pos186 コレラ菌走化性受容体 Mlp24 のアミノ酸受容能はカルシウムイオンで増強される

Ca^{2+} potentiates attractant responses to amino acids mediated by the chemoreceptor Mlp24 of *Vibrio cholerae*

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Amino acid chemotaxis of *Vibrio cholerae* is mediated mainly by two major chemoreceptors, Mlp24 and Mlp37. We solved structures of the periplasmic domain of Mlp24 with or without attractant amino acids and found that the ligand-binding pocket contains a Ca^{2+} ion even in the *apo* form. The Ca^{2+} ion appears to restrict the dynamics of the loop which is proximal to the ligand-binding site. In this study, we examined roles of the Ca^{2+} ion in its ligand recognition. ITC analyses confirmed that the periplasmic fragment of Mlp24 actually binds Ca^{2+}, although the affinity is relatively low (K_D is in the sub-millimolar range). Moreover, Ca^{2+} decreased the K_D values of the fragment for certain amino acids. These effects were supported by *in vivo* assays for amino acid responses.

3Pos187 バクテリアのマグネットコンパスの人為的反転

Artificial polarity-reversal of bacterial magnetic compass

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Magnetotactic bacteria synthesize nano-sized magnetic crystals, magnetosome, which function as tiny compasses allowing the microbes to navigate using the earth's geomagnetic field. Here, we applied the external magnetic fields to *Magnetospirillum magneticum* AMB-1 under an optical microscope, and quantitatively controlled their swimming direction at the single-cell level with magnetic tweezers. Additionally,the polarity of their swimming was artificially reversed by applying the quick reversal of the large magnetic field, indicating the polarity-reversal of their nano-compasses. We establish the novel technique to manipulate bacterial "taxis" independent from swimming motility, which can be applied to other bacterial motility with the labelling of isolated magnetosome.

3Pos188 Importance of receptor cooperativity on the switching coordination of flagellar motors on a single *Escherichia coli* cell

Yong-Suk Che[1], Hiroto Takahashi[2], Akihiko Ishijima[1], Hajime Fukuoka[1] ([1]*Dept. Frontier Biosci., Osaka Univ.*, [2]*IMRAM, Tohoku Univ.*)

Our previous work shows the rotational switching of two different motors on a same cell is highly coordinated, and the dynamic change in CheY-P concentration derived from cooperative work of chemoreceptor cluster would regulate its coordination. Here, we measured the switching coordination in a cell co-expressing wild-type and mutant receptors (Tsr), which defects the cooperativity for the attractant response. The coordination was abolished depending on the expression level of the Tsr mutant, suggesting that Tsr mutant collapsed cooperativity of receptor array to affect the CheY-P production. To estimate the strength of cooperativity, we are trying to quantify the expression levels of wild-type and mutant receptors. Details of these results will also be discussed.

3Pos189 変性蛋白質センサーとしての大腸菌ヒスチジンキナーゼ BaeS

The histidine kinase BaeS of Escherichia coli may sense denatured proteins

Tohru Umemura[1], Yoshiyuki Sowa[1,2], Ikuro Kawagishi[1,2] ([1]*Dept. Frontier Bio., Hosei Univ.*, [2]*Research Center for Micro-Nano Technology, Hosei Univ.*)

The histidine kinase BaeS of *Escherichia coli*, together with the response regulator BaeR, has been implicated in responses to indole and divalent cations but the direct stimulation it senses has been eluded. We recently found that membrane stress or intracellular harmful compound(s) induces a BaeSR-mediated response. Here we report that heat shock, cold shock, reduction, acidification and overexpression of proteins led to the aggregation of GFP-tagged BaeS. Without such stress, deletion of a chaperon protein gene caused the aggregation of BaeS-GFP. We hypothesize that BaeS is a sensor for unfolded proteins just as the eukaryotic sensor IRE1 for ER stress. To test this hypothesis, imaging analyses on interaction between BaeS and the chaperon protein are under way.

3Pos190 G タンパク質共役型受容体の動的なホモ・ヘテロダイマー形成：二色同時蛍光１分子観察による解析

Dynamic homo- and hetero-dimerizations of G-protein coupled receptors: An analysis by dual-channel single fluorescent molecule observation

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G-protein coupled receptor, GPCR, is one of the largest receptor families. Unlike class-C GPCRs, it has been controversial whether class-A GPCRs, consisting of over 80 % of all GPCRs, work as monomers or dimers. In particular, since hetero-dimers are thought to work for signal sorting and integration, its nature has been widely studied for decades. However, it is still unclear even for dynamics. By employing dual-color single-molecule observation technique, we found that two dopamine receptors, D1R and D2R form their temporal homo-dimers as well as temporal hetero-dimers with the similar lifetimes while they can exist as monomers. These findings might reveal how and why homo-/hetero-dimers and monomers are important in signal transduction processes.

3Pos191 共焦点画像解析と 1 分子計測を用いた ERK シグナル伝達系のボトルネックの解明

Unraveling origins of bottleneck effects for ERK signal transduction using confocal image analyses and single molecule imaging

Kazunari Mouri, Yasushi Okada (*RIKEN QBiC*)

PC12 cells select fates optically depending on nutrient and stimulation conditions (Mouri K., et al., PLOS Comput. Biol., 2013). The translocation of ERK is a key step to convert analog stimuli into digital fates (Shindo Y., et al., Nat. Commun., 2016). Integrating FRAP and a mathematical model, we enabled simultaneous estimation of the influx and efflux of ERK. We combined this method with RICS (raster image correlation spectroscopy) using high-resolution confocal images, and estimated the absolute number of transport molecules per nuclear pore. Also, we succeeded to measure single molecules of ERK on nuclear pores using TIRF. These suggest that import flux is much slower than that of importinβ, and ERK translocation works as a bottleneck of signal transduction processes.

3Pos192 三量体 G タンパク質の制御を介した走化性レンジの拡張機構

Dynamic range extension of eukaryotic chemotaxis via regulation of heterotrimeric G protein dynamics

Yukihiro Miyanaga[1,2], Yoichiro Kamimura[1,2], Masahiro Ueda[1,2] ([1]*Frontier Biosciences, Osaka Univ.*, [2]*QBiC, Riken*)

Chemotactic eukaryote cells can sense chemical gradients over a wide range of concentrations via trimeric G protein signaling. Recently we identified a regulator of G protein dynamics named Gip1, which extends the chemotactic range of *Dictyostelium* cells. However the whole rage of eukaryotic chemotaxis is not fully explained by the regulator, suggesting some additional mechanisms. Here we report three G protein-related events working in different concentration ranges for chemotaxis, that are the activation, Gip1-dependent translocation and complex formation with receptor of G protein, with EC50s of 1 nM, 10 nM and 300 nM, respectively. Chemotactic cells may switch or combine three different G protein regulation mechanisms to achieve wide-range chemotaxis.

3Pos193 走化性タンパク質の細胞内動態と細胞応答の同時計測

Simultaneous observation of the intracellular chemotactic proteins and the cellular behavior

Hajime Fukuoka[1], Hiroto Takahashi[2], Akihiko Ishijima[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*IMRAM, Tohoku Univ.*)

Chemotaxis system is performed by many kinds of proteins. To understand this system as the protein's behavior in a cell, the activity and the dynamics of chemotaxis proteins should be correlated with the cellular response, which is monitored through the rotation of flagellar motor. In this study, to monitor the activity of receptor array, we developed functional fusion of CheA and fluorescent protein (FP), and we are trying to simultaneously measure the motor rotation and FRET derived from the conformational change in receptor array. To investigate the dynamics of signaling molecule CheY during cellular response, the change of polar localization CheY-FP and the motor rotation was simultaneously measured. We would like to discuss about our results at the annual meeting.

3Pos194 ライブセルイメージングと薬剤実験に基づく動的な誘引物質勾配場における HL60 細胞の走化性運動の解析

Pharmacological and live-cell imaging analysis of chemotactic HL60 cells under dynamically changing chemoattractant gradient

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The direction of neutrophil chemotaxis is thought to be determined by a mechanism that senses the difference in the chemoattractant concentration across the cell body. However, several lines of evidence suggest that the cells may also be utilizing temporal information. Here, we show that neutrophil-like HL60 cells exhibit forward movement and concomitant PH-Akt translocation only in the rising phase of a propagating fMLP gradient. Furthermore, cells pharmacologically impaired of the front-rear polarity exhibited more marked asymmetry in the migratory response. Our data suggest that there is a distinct mode of chemotaxis driven by transient changes in the chemoattractant concentration and that this could be partially overridden by the pre-existing polarity.

3Pos195 マクロファージの走熱性メカニズムの解明

Thermotaxis mechanism of mouse macrophage

Hideo Saitou, Seine Shintani, Hideo Higuchi (*Grad. Sch. Sci., The university of Tokyo*)

To understand the transient processes of macrophage at temperature jump, we observed the motility of mouse macrophage. The temperature jump was initiated by local illumination of medium by the infrared laser (1550nm). The macrophage moved toward the heat source while stretching filopodia and lamellipodia with time delay after the illumination. To understand the mechanism of the thermotaxis, we observed the motility of macrophage in the presence of inhibitors for Rho family such as RhoA, Rac1, cdc42, for myosin and depolymerizing agents for actin filament and microtubule. The macrophages in the presence of ML-141 were able to migrate but did not polarize in the direction of the temperature gradient and thermotaxis towards heat source was abolished.

3Pos196 免疫受容体シグナルを担う足場分子LATは細胞膜に繋留された小胞で機能する：1分子イメジングによる解明

The immune signal adaptor molecule LAT works on cytoplasmic vesicles tethered to the plasma membrane: a single-molecule imaging study

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A transmembrane adaptor protein, Linker for Activation of T cells (LAT), is a key molecule for mediating the activated-receptor signal to IP3-calcium responses, which eventually lead to histamine release in immune mast cells. Using single-molecule tracking and LAT-knock-out mast cells prepared by the CRISPR/Cas9 system, we found the following. Approximately 10% LAT molecules existed in cytoplasmic vesicles, which we termed LAT vesicles. Each LAT vesicle contained one or two LAT molecules, is tethered to the plasma membrane (PM) by exocyst complexes for lifetimes of 6 s or 73 s, contained several types of phosphoinositides and their kinases, and served as the key signaling platform, which entirely differed from LAT monomers or clusters in the PM.

3Pos197 PIP3とRasの自己組織的な局在形成過程とその制御メカニズムの解析

Analysis of Self-organized Domain Formation and Regulation Mechanism of PIP3 and Ras

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Spontaneous cell migration is induced by self-organization of PIP3-enriched domain on cell membrane that acts as a signal of actin polymerization. In previous study, we found PI3K, PIP3 and Ras compose positive feedback loop and it can amplify local PIP3 fluctuations. To examine details of the regulation, we analyzed responses to several inhibitors of the signaling pathway, and found active Ras shows well consistent localization with PIP3 in many cases. On the other hand, in the PI3K null strain and PIP3 overproducing strains, activated Ras shows self-organization in spite of a defect on PIP3 signaling pathway. These results suggest Ras is dominant in self-organization of signal molecules including PIP3.

3Pos198 Controlling contractile instabilities in the actomyosin cortex

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The actomyosin cell cortex is an active contractile material for driving cell- and tissue morphogenesis. The cortex has a tendency to form a pattern of myosin foci, which is a signature of potentially unstable behavior. Here we report that feedback between active Rho and myosin induces a contractile instability in the cortex. We discover that an independent RhoA pacemaking oscillator controls this instability, generating a pulsatory pattern of myosin foci and preventing the collapse of cortical material into a few dynamic contracting regions. Our work reveals how contractile instabilities that are natural to occur in mechanically active media can be biochemically controlled in order to robustly drive morphogenetic events.

3Pos199 増殖中の培養細胞における遊離コレステロールの分布

Distribution of free cholesterol in MDCK cells during the migration on culture dish

Yoshikatsu Ogawa (*AIST BMD*)

Cholesterol is one of the components for mammalian cells. The distribution of Cholesterol in cells is not homogeneous. Our group developed a fluorescent chemical probe (R-Chol) for studying the distribution of cholesterol in cells by microscopy. On the other hand, one of the roles of cholesterol in cells is supposed to maintain the flexibility and stability of cell membranes. Therefore, we hypothesized that cholesterol might be transported to the local area of the membrane which undergoes a dynamic rearrangement (e.g., Leading edge of epithelial cell). We stained MDCK cells with R-Chol to investigate its distribution in the cells by fluorescent microscopy during wound healing assays. Some spots of fluorescent signals were observed around the leading edges of cells.

3Pos200 獲得免疫応答におけるTリンパ球の単一細胞測定系の開発

Development of a single cell assay system for T lymphocytes in adaptive immunity

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Adaptive immunity is involved in the specific antigen recognition between T lymphocytes (T cells) and antigen presenting cells (APCs). It has been believed that T cells show antigen-specific response even at single cell level. In this study, using single cell manipulation and live cell imaging, we developed a single cell assay system that can monitor T cell activation by T cell-APC interaction. We found that reactivity and specificity of single T cell in the first contact with specific antigen on single APC was very poor, whereas reactivity and antigen-specificity was drastically increased when contacted with APC interacting with activated T cells. The findings suggest that the surrounding condition of APC affects T cell response in antigen recognition.

3Pos201 Drug response of lymphocytic leukemia cells to anticancer drug is affected by experience of cell division prior to treatment

Akihisa Seita, Takahiro Yamada, Yuichi Wakamoto (*Department of Basic Science, Graduate School of Arts and Science, University of Tokyo*)

Even in clonal population, a small fraction of cancer cells survive anti-cancer drug treatment. Obtaining information that distinguishes survivors from the other requires tracking single-cell lineages before and after environment changes. Here, using a new microfluidic device for long-term and high-throughput tracking of mammalian cell lineages, we observed that lymphocytic leukemia cells (L1210) exhibit different levels of responsiveness to Mitomysin-C. Furthermore, it is suggested cells dividing before treatment have higher viability than non-dividing cells. We are now investigating a quantitative relationship between drug sensitivity and cellular growth history.

3Pos202 ErbB レセプターの相互リン酸化ネットワーク
Cell and signal specific phosphorylation networks of the ErbB receptor family

Hiraku Miyagi, Michio Hiroshima, Atsushi Mochizuki, Yasushi Sako (*RIKEN*)

ErbB family proteins are receptor tyrosine kinases on the animal cell surface. Abnormal expression and/or mutation of ErbBs correlate with human cancers. The four members of ErbB family form ligand-induced homo/heterodimers for mutual phosphorylation. However, cell type and ligand specificity of the ErbB dimerization network remains to be elucidated. Here, we detected phosphorylation of ErbB proteins after two types of ligands treatment in the three types of human carcinoma cell lines using immunoblotting method. As a result, we identified distinctive responses of ErbB network depending on the cell lines and ligand species. We suggest phosphorylation networks among subtypes of ErbBs, which are different between cell types and signals.

3Pos203 膜活性ポリマーによる脂質二分子膜ナノディスクの形成
Lipid bilayer nanodiscs formed by designed membrane-active polymers

Kazuma Yasuhara, Jin Arakida, Masaya Inoue, Jun-ichi Kikuchi (*Grad. Sch. Mat. Sci., Nara Inst. Sci. Tech.*)

Membrane proteins play important roles in various biological processes including signal transduction, material transport, and energy production. There is a great need for the development of a molecular platform, which mimics the local environment of biomembranes, for the analysis of membrane proteins. We have designed and synthesized amphiphilic copolymers to form lipid bilayer nanodiscs through the direct fragmentation of a membrane. Screening of the polymer library revealed that nanodisc formation activity of the polymer depends on the structural characteristics of the polymers such as amphiphilicity and molecular weight. Additionally, it was confirmed that a lipid bilayer structure was maintained in the nanodiscs formed by the polymers.

3Pos204 可溶性高分子で生成された水性相分離のマイクロ液滴内への生細胞の含有
Entrapment of Living Cells inside Micro-Droplet under Aqueous/Aqueous Segregation with Solvable Polymers

Tadashi Fujimoto[1], Naoki Nakatani[1], Kanta Tsumoto[2], Chwen-Yang Shew[3], Kenichi Yoshikawa[1] ([1]*Fac. Life Medical Sciences, Univ. Doshisha*, [2]*Grad. Sch. Eng, Univ. Mie*, [3]*Chemistry, City. Univ. New York*)

It is getting clearer that aqueous environment around living eukaryotic cells play an essential role to decide the fate-decision of stem cells. Here, we will report specific localization of living cells in a micro segregated aqueous/aqueous two phase system, composed with two different water-solvable polymers. We have adapted a pair of polymers, polyethylene glycol (PEG) and dextran, and obtained the solution composed with droplets rich in dextran in the environment rich in PEG by taking suitable composition near critical state of the phase separation. It was found that red blood cells are selectively localized inner side of the droplet rich in dextran.

[Ref] K. Tsumoto, et al., Life, 5, 459 (2015).

3Pos205 粗視化モデルによる脂質分子の集合体の形成過程に関する理論的研究
Theoretical study on the process of the formation of lipid molecule cluster by coarse-grained model

Shogo Kinoshita, Satoshi Nakagawa, Makoto Wada, Seiichiro Ito, Kazutomo Kawaguchi, Hidemi Nagao (*Nat. Sci. Kanazawa Univ.*)

Amphiphilic molecules can spontaneously form various structures in water solution such as spherical micelle, rod-like micelle, spherical vesicle, and so on. The purpose of this study is to present an implicit model describing the process of the association and the dissociation of lipid molecule clusters. We use a coarse-grained model which consists of two spherical particles. We consider the tilt and the bending potential to form a bilayer structure. We can find that the molecules form a disk-like micelle in the case of only the tilt potential, and that the molecules spontaneously assemble into some spherical micelles and rod-like micelles in the case of the tilt potential and the bending potential.

3Pos206 ベシクルに大小 2 種のコロイド粒子を内包させた系の相分離ー朝倉・大沢理論の拡張—
Phase Separation of Two Kinds of Colloidal Particles in Giant Vesicles -Extension of Asakura-Osawa Theory-

Yuno Natsume[1], Kazumi Itoh[1], **Yuhei Natsume**[2], Kensuke Kurihara[3,4,5] ([1]*Japan Women's Univ.*, [2]*Chiba Univ.*, [3]*Okazaki Institute for Integrative Bioscience*, [4]*Institute for Molecular Science*, [5]*Research Center for Complex Systems Biology, The Univ. of Tokyo*)

The excluded volume effect plays an essential role to the biomolecular conformation made for a restricted extent. Here, we investigated the phase separation of two kind of colloidal particles (hard spheres) encapsulated into phospholipid giant vesicles with diameter of 10~20 μm. We concentrated our attention on cases where the volume fractions (10 vol%) of large spheres (whose diameters were 1.0 μm) were ten times as large as those of small ones (0.1μm). Under the influence of this situation, we obtained the following remarkable results of experiments; Large spheres were condensed into the inner part, while the small ones were localized in outer shell. This characteristic phase separation was discussed from the viewpoint of the extension of Asakura-Oosawa theory.

3Pos207 種々の脂質組成によるタンパク質内包リポソームの構造学的研究

Structural study of liposomes encapsulating proteins depending on lipid composition and species

Kosuke Takahashi, Mitsuhiro Hirai (*Grad. Sch. Eng., Gunma Univ.*)

Liposomes are expected to be effective drug-delivery systems (DDS) due to their high adaptability to a living body and superior applicability to various types of drugs. We reported on the encapsulation proteins within the lipid-raft model liposomes and the characterization of the structure by using X-ray scattering and shell-modeling analysis. In addition, recently, we have found that the encapsulation of proteins in liposomes improved the heat-resistant property of those structures [1]. In the present study, we have clarified the thermal structural stability of liposomes encapsulating proteins by changing lipid composition and species under molecular crowding environment.

[1] M. Hirai et al., J. Phys. Chem. B, 119, 3398 (2015) ; J. Synchrotron Rad., 20, 869 (2013).

3Pos208 アシル鎖長の異なるホスファチジルエタノールアミン二重膜の熱および圧力相転移

Thermotropic and barotropic phase transitions in bilayer membranes of phosphatidylethanolamines with varying acyl chain lengths

Masaki Goto[1], Shigeru Endo[2], Nobutake Tamai[1], Hitoshi Matsuki[1] ([1]*Grad. Sch. of Biosci. and Bioindus., Tokushima Univ.*, [2]*Fac. of Engin., Tokushima Univ.*)

We have investigated thermotropic and barotropic phase transitions in bilayer membranes of PEs with two linear saturated acyl chains (Cn = 12 (DLPE), 14 (DMPE), 16 (DPPE) and 18 (DSPE)) in terms of the temperature-pressure phase diagrams and the thermodynamic quantities of the phase transitions. The stability of the gel phase for the PE bilayers changed from metastable to stable depending on pressure and the chain length. The hydrated crystal phase of the PE bilayers was much more stable than that of PC bilayers although the chain-melting behavior of both bilayers was almost comparable. These findings are attributable to the strong interaction between polar head groups of PE molecules with smaller numbers of interlamellar water molecules.

3Pos209 親水性高分子により基板との距離を制御した人工膜への膜タンパク質再構成

Patterned model membrane with hydrophilic polymer brushes for the functional incorporation of membrane proteins

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Substrate-supported planar lipid bilayers (SPBs) are a versatile model of the biological membrane. However, functional studies of membrane proteins using SPBs is challenging due to the proximity between the membrane and the substrate. To overcome this limitation, we fabricated a micropattterned SPB with a hydrophilic polymer layer (polymer brush) between the membrane and the substrate. The membrane was composed of polymeric and fluid bilayers. The polymer brush was formed from the surface of the polymeric bilayer. We incorporated Bacteriorhodopsin (BR) as a model of the membrane-embedded proteins into the fluid bilayer. We discuss the effect of polymer brush-supported SPBs on the mobility and the proton-pumping function of BR.

3Pos210 バクテリオロドプシン球殻構造体の形成機構

A mechanistic insight into the formation of bacteriorhodopsin vesicle

Daisuke Yamamoto, Risa Mutoh (*Fac. Sci. Fukuoka Univ.*)

Bacteriorhodopsin (bR) is a membrane protein that works as a light-driven proton pump. While bR forms a two-dimensional crystal in the physiological conditions, its membrane structure is known to be converted into the uniformly-sized vesicles in the presence of octyl-thioglucoside (OTG), of which the mechanism is unknown. Here, we directly observed the bR-vesicles at molecular resolution by atomic force microscopy. The observed inter-molecular distance of bR in the membranes well fitted to a model that is constructed based on the thermodynamic equilibrium of OTG. Our results strongly suggest that the formation of bR-vesicle is caused by the difference of bR-OTG interaction between at the cytoplasmic side and at periplasmic side of bR.

3Pos211 ホスホリパーゼ Cβ 1 の C 末端には脂質膜のチューブ形成能がある

Membrane tubulation ability of phospholipase Cβ1 C terminal domains

Takehiko Inaba[1], Takuma Kishimoto[2], Motoshide Murate[1], Takuya Tajima[1,3], Mitsuhiro Abe[1], Asami Makino[1], Nario Tomishige[1], Reiko Ishitsuka[1], Yasuo Ikeda[3], Shinji Takeoka[3], Toshihide Kobayashi[1,4] ([1]*RIKEN*, [2]*Kyorin Univ. Sch. Medicine*, [3]*Waseda Univ.*, [4]*UMR 7213 CNRS, University of Strasbourg*)

Liposome based microscopic screening reveals that phospholipase Cβ1 (PLCβ1) induces lipid membrane tubulation. This activity is derived from the C terminal domains and did not require the N terminal lipase X-Y domain and lipid binding PH domain. The overexpression of PLCβ1 in cell also induced the plasma membrane tubulation. The deletion of C terminal domains abolished the membrane localization and tubulation activity, whereas the mutation of lipase activity did not affect them. The reduction of PLCβ1 by knockdown caused the significant reduction of caveolae formation and the induction of evagination in plasma membrane. These results indicate that PLCβ1 play the role in plasma membrane morphogenesis, especially in the caveolae maintenance.

3Pos212 巨大分子系シミュレーションに向けた粗視化脂質モデルの開発

Developing a coarse-grained model of lipid for large molecular simulations

Suguru Kato, Shoji Takada (*Kyoto University*)

The lipid molecules are essential for the biological membranes, which include the cell membrane, nuclear membrane, Golgi apparatus, endoplasmic reticulum, and so forth. With the standard atomistic molecular dynamics (MD) methods, it is difficult to simulate large molecular systems such as a whole organelle because of the high computational costs. In contrast, coarse-grained MD simulations have an advantage of less computational cost and can deal with longer time scape of large systems. Here, we develop a coarse-grained model for lipid based on simulation data by the short atomistic MD simulations of the lipid membrane. It allows us to calculate the large system with low computational costs.

3Pos213 アルギニンペプチドの膜透過を促進する両親媒性ペプチドによる脂質パッキングの変化

Alteration of Lipid Packing State by Amphipathic Peptides Promoting Membrane Penetration of Octaarginine

Tomo Murayama, Shiroh Futaki (*ICR, Kyoto Univ.*)

An amphipathic helix is a common motif for membrane-active peptides/proteins, including antimicrobial peptides and curvature inducing proteins. We previously reported that an amphipathic N-terminal segment of epsin-1 induced positive membrane curvature and promoted membrane translocation of R8, a representative cell-penetrating peptide (Pujals et al. ACS Chem. Biol., 8, 1894 (2013)). To investigate the relationships between these activities, we studied the effects of amphipathic peptides derived from membrane-remodeling proteins on membrane translocation of R8. In addition to the assessment of their curvature inducibility, we monitored the lipid packing state of plasma membranes under the treatment with these peptides.

3Pos214 FTIR-ATR のプリズム上に作製した皮膚角層モデル膜への物質透過解析

Permeation of substances into stratum corneum model membranes prepared directly on FTIR-ATR prism

Kohei Oka, Satoru Kato (*Grad. Sch. Sci. & Tech., Univ. Kwansei Gakuin*)

We improved the method that we proposed previously for permeation analysis of thin lipid film. In this study, we prepared stratum corneum model membrane composed of only one kind of ceramide, cholesterol and free fatty acid with hydrocarbon chain length of C20 by directly spraying the lipid solution (chloroform/methanol) onto on the FTIR-ATR prism. The permeation process was analyzed by detecting the molecules penetrating into the region where evanescent field exists. We succeeded to obtain stable data for a fairly long period by eliminating the effect of heavy water vapor in the air with FTIR. Using the improved method we analyzed semi quantitatively the permeation process of some substances through the model membrane.

3Pos227 神経振動活動のシナプス入力による位相依存的な調節

Phase-dependent modulation of neural oscillations by synaptic inputs

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Oscillatory neural activities have been implicated in various types of information processing in the CNS. We examined effects of synaptic stimuli on periodic spiking in cerebellar Purkinje cells and local field potential oscillation in a land mollusk. We found that in both systems, the response depends on the stimulus phase. We also showed that common input to multiple oscillatory elements modulates the synchrony in a phase-dependent manner, and that regulation of the input phase is important for the generation of appropriate response in synchrony. These results suggest that neural responses are in general phase-dependent, and that neural networks are designed to utilize the oscillatory dynamics.

3Pos228 線虫の whole-brain イメージングデータに関する位相同期解析

Phase synchronization analysis of whole-brain imaging data of C. elegans

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There are 302 neurons in the nervous system of C. elegans. About 170 neurons of them densely locate in the head region. We developed a 4D imaging system to measure the neural activities in the head region as a worm lives. To understand how the central nervous system works as a whole, therefore, phase synchronization analysis is carried out for the neural imaging data of the wild-type and some mutants which have defect in neural connectivity. The degree of phase synchronization is quantified by two indices; phase coherence index (the Kuramoto order parameter) and phase locking index. We find that anti-phase synchronization plays an important role to organize collective dynamics of the neurons. We also carried out correlation clustering for the neural imaging data.

3Pos229 神経細胞から伸長する神経突起の特性のオンチップ１細胞解析

Minimum Requirements of Microchannel Patterns for Building of Stable Neuronal Circuits in On-chip Cell Network Assay

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For patterning of constructive stable neuronal circuits, we have examined minimum requirements of structures of micropatterns for guiding the directions of neurite elongation as desired. We used the photothermal spot heating system for flexible micropatterning. The system consists of four major parts; a cultivation dish coated with an agarose thin layer, a 1064/1480-nm dual infrared laser, a phase-contrast microscope with a CCD camera, and a computer enabling semi-automated micropatterning. Using the system, we found the minimum requirements of micropattern shapes for neuronal network formation, e.g., the minimum microchannel width for neurite extension was 8 μm. By using the acquired condition, we successfully formed artificial stable neuronal circuits on a chip.

3Pos230 フェムト秒レーザー光刺激による神経回路網の誘発応答特性

Spatio-temporal activity pattern in neuronal network evaluated by femtosecond laser-induced stimulation

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In order to identify functional connections in living neuronal networks, it is indispensable to study the spatio-temporal activity pattern in neuronal networks introduced by precise stimulation at single cell level. Here, we applied femtosecond laser-induced stimulation to evaluate neuronal activity in neuronal networks. After loading a fluorescent Ca^{2+} indicator into neurons, spontaneous Ca^{2+} spikes in neuronal networks were confirmed. When femtosecond laser pulses irradiated into target neuron, intracellular Ca^{2+} immediately increased at the laser spot. Subsequently, synchronous Ca^{2+} spikes in other neurons close to the target neuron was frequently observed, suggesting that propagation of neuronal activity was evoked by laser-induced stimulation.

3Pos231 培養神経回路網におけるネットワークグラフ構造の培養日数依存的変化

Developmental changes of graph structures in cultured neurons -the analysis with functional-connection map

Nanami Hirata, Wataru Minoshima, Hidekatsu Ito, Suguru Kudoh (*Department of Human System Interaction, School of Science and Technology, Kwansei Gakuin University*)

Recently, cultured neuronal networks are focused on as a model of the fundamental neuronal circuit in the brain. We calculated conditional probability of evoked action potential at a focused neuron on a specific electrode just after the evoked action potential at a certain neuron, and extracted the graph structure based on the conditional probabilities of co-occurrence of events. Then we quantified the features of the graph structure of the neuronal circuit. As a results, the local clustering coefficients and the assortativity coefficient of the network suggested that the functional-connectivity-map was moderately disassortative and sparse. In addition, the local clustering coefficients slowly decreased during culture days, suggesting that he network structure changed to be more segmented.

3Pos232 エピカテキンは、ヨーロッパモノアラガイの呼吸行動の中枢リズム発生器である RPeD1 ニューロンの興奮性を変化させる

Epicatechin alters the electrophysiological activity of RPeD1 in the pond snail, Lymnaea

Yoshimasa Komatsuzaki[1], Minoru Saito[2], Ken Lukowiak[3] ([1]*Dept. Phys., CST, Nihon Univ.*, [2]*Grad. Sch. Integ. Basic Sci., Nihon Univ.*, [3]*Hotchkiss Brain Inst., Cumming Sch. of Med., Univ. of Calgary*)

We investigated how epicatechin(epi) brings about its enhancing effects on RPeD1, which is a necessary site for long-term memory(LTM) formation. RPeD1 is 1 of 3 neurons that comprise the central pattern generator(CPG) that drives aerial respiratory behaviour. We recorded from RPeD1 in semi-intact preparations 24h after a single 0.5h training session in epi and found that the firing and bursting rate significantly decreased. We next performed in vitro operant training. After the training, naïve snails exposed to epi prior to recording exhibited significantly increased RPeD1 excitability. These results suggest that epi alters RPeD1 excitability in such a way as to make it more difficult for the CPG to drive aerial respiratory behaviour and leads to enhanced LTM formation.

3Pos233 Increasing reproducibility of activity patterns in nenural network during culture days

Takumi Okada, Wataru Minoshima, Hidekatsu Ito, Suguru Kudoh (*Dept. of Human System Interaction, School of Sci. and Tech., Kwansei Gakuin University,*)

The representations of outer world in the brain are considered to be undertaken by spatiotemporal activity patterns of the circuits. In this study, we analyzed the transition of the internal states of the rat hippocampal neurons cultured on a multi-electrodes-array-dish. We analyzed the feature of center of gravities (CG) of the activity locations. As a result, we confirmed that the reproducibility of the neuronal-network-activity increased during culture days. CG-analysis also reduces detailed activity pattern. Thus now we are applying similarity analysis to 64-dimensional feature vectors of neuronal activity. Electrical-activity-pattern at a certain 5-ms-width-window was represented as a 64-dimensional-“0-1”-feature-vector, and its stability was also analyzed.

3Pos234 超高感度な匂い識別は、受容体 - G 蛋白質の初期一過性相互作用と前梨状皮質振動性応答に部分的に支配される

Supersensitive odor discrimination is controlled in part by initial interactions of receptor-G-protein and cortical oscillatory responses

Takaaki Sato[1], Reiko Kobayakawa[2], Ko Kobayakawa[2], Makoto Emura[3], Shigeyoshi Itohara[4], Takashi Kawasaki[1], Riichi Kajiwara[5], Ichiro Takashima[1], Toshio Iijima[6], Akio Tsuboi[7], Hiroyoshi Matsumura[8] ([1]*AIST*, [2]*Kansai Med. Univ.*, [3]*Takasago Internat'l. Corp.*, [4]*BSI., RIKEN*, [5]*Sch. Sci. & Technol., Meiji Univ.*, [6]*Grad. Sch. Life Sci., Tohoku Univ.*, [7]*Nara Med. Univ.*, [8]*Ritsumeikan Univ.*)

Compared to WT mice, ΔD mice w/o all dorsal ORs showed selective major loss of odor detection sensitivity to (+)-enantiomers with sub-ppq-level sensitivity to (-)-enantiomers in Y-maze behavioral tasks. Moreover, ΔD mice showed enantiomer odor discrimination paradox with $>10^{10}$-fold sensitivity reductions, indicating that the most sensitive dorsal ORs play a critical role in odor coding. Using mutated OR and a homology model, we found that both Glu at the 2nd residue of helix 8 and hydrophobic cores of helix 8/TM1-2 are important for rapid Ga activation via initial transient interactions. A wavelet correlation analysis suggests that the olfactory system extracts sensory information by summating multiple OR signals in the third-order neurons via feedforward inhibition.

3Pos235 定量的マンガン造影 MRI による全脳神経活動計測

Whole brain activity mapping using quantitative activation-induced manganese-enhanced MRI

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To reveal brain functions and responsible regions for neurological disorders, the changes of the neuronal activities in whole brain should be analyzed. For this aim, we developed a new MRI technique, quantitative activation-induced manganese-enhanced MRI (qAIM-MRI). qAIM-MRI is based on the use of manganese ion (Mn^{2+}) as a surrogate marker of Ca^{2+} influx. Mn^{2+} can pass through the voltage-dependent Ca^{2+} channels and is therefore known to be taken up by neurons in activity-dependent manner. Mn^{2+} shorten the longitudinal relaxation time (T_1) of proton (H^+), which can be quantified by MRI scanner. We succeeded in non-invasively measurement of the history of the neuronal activities in whole brain, and in revelation of the region with activity changes in various conditions.

3Pos236 Foraging behavior of Caenorhabditis elegans

Chien Jung Lo, **Mao Ting Cheng** (*Dept. of Physics, NCU, Taiwan*)

Caenorhabditis elegans is a free-living nematode about 1 mm in length and 100 μm in width. C.elegans is the ideal model organism in many field of research because of their well-studied fate of its 1031 cells. We aim to study foraging behavior of C.elegans. Understanding the foraging behavior of this model oraganism C.elegans represents the epitome of a searching mode of this simple living creature. We design spatial food patterns for C. elegans to search. During the foraging process, we record images of their locomotions to define the foraging states and reveal the pattern changing of foraging behavior. We found the C. elegans has different searching patterns in absence of food and approching the food. We will discuss the details of their searching states of foraging.

3Pos237 2次元系および3次元系における細胞の自発運動動態
Spontaneous cell migration dynamics in 2D and 3D environment

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Cell migration is essential in many physiological functions. Recently, plasticity in migration modes depending on in vivo environments attracts considerable attention in researches on development and disease. Here, single-cell migration dynamics in 2D and 3D environment were studied by statistical analysis of cellular trajectories, mathematical modeling and simulation. In 2D migration of mouse T-cell, distinct modes and transitions between them were identified, while in 3D ex vivo migration, only smooth migration was detected. Characteristic statistical features found in Dictyostelium cell migration were also found in mouse T-cell migration, and the same class of model was tested in both cell types. Functional significance of fluctuation in cell migration was discussed.

3Pos238 活動量と睡眠との関連解析
Association between physical activity and sleep efficiency

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Recently several wearable sensors are used in the context of individual's healthcare. One of important feature of life is sleep quality. However, relationship between daily physical activity and night sleep quality has not been examined in general. Almost 200 healthy adults were recruited in this study. Over two weeks, participants wore a activity sensors to monitor their activity in whole day. Also participants involved completing questionnaire related sleep twice in a day, at the awake and sleep. The statistical properties of activity data, quantified by digital integration of acceleration, were examined. We will report the association between statistical features of duration of active/resting activity state and sleep features.

3Pos239 光誘起チャネルロドプシンに関する理論研究
Theoretical study on molecular mechanism of photo-induced gate opening of channelrhodopsin

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Channelrhodopsins (ChR) are light-sensitive cation channels formed by seven-transmembrane helices, which have shown experimental utilities in optogenetics. However, although the crystal structure of the closed-state of ChR was determined in 2012, the ion conducting open-state structure and the molecular mechanism of the ion conduction remain unknown. In this study, the gate-opening conformational changes of ChR induced by photo-isomerization of the retinal chromophore were studied in atomic detail with molecular dynamics (MD) simulations and QM/MM RWFE-SCF free energy geometry optimization. Water/ion distributions of the channel in the photo-activation process are then examined by 3D-RISM theory with conformational samples obtained by MD simulations.

3Pos240 古細菌型TSAモチーフ配列をもつ真正細菌由来ハライドイオンポンプロドプシンの光反応解析
Cl^--pumping Photoreaction of a Bacterial Halide-ion Pumping Rhodopsin with an Archaeal-type TSA motif

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Light-driven halide-ion pumping rhodopsins have been discovered from halophilic archaea, marine bacteria, and cyanobacteria. These proteins have functionally important motifs in the 3rd-helix, TSA, NTQ, and TSD, respectively. Here, we focused on a halide-ion pump discovered from a marine bacterium *Rubricoccus marines* SG-29^T. This protein has the archaeal TSA motif with close phylogenetic relationship to cyanobacterial halide-ion pumps. To characterize the *R. marines* halide-ion pump, we firstly expressed it in *E. coli* cells. The functional expression and inward Cl^--pump activity were confirmed by the light-induced pH change measurement. To elucidate the Cl^--pumping photoreaction, we performed spectroscopic and kinetic analyses. Details will be discussed in this meeting.

3Pos241 *Mastigocladopsis repens* halorhodopsinのフォトサイクルの解析とTSDモチーフの役割の解明
Photocycle of *Mastigocladopsis repens* halorhodopsin and the role of its TSD motif

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Mastigocladopsis repens halorhodopsin (MrHR) is a novel light-driven Cl^--pumping rhodopsin. MrHR and its homologs completely conserve the Thr, Ser and Asp residues (TSD motif) on the C helix. Here, we show the analysis results of MrHR photocycle and roles of the TSD motif. Different from other Cl^- pumps, MrHR binds I^- but cannot transport it. The I^--bound form showed the photocycle lacking the intermediates after L, suggesting that Cl^- moves to cytoplasmic side during L decay. Mutations revealed that Thr74 and Ser78 are important to the Cl^- binding. Replacements of Asp85 did not change Cl^--binding affinity, but greatly prolonged the decay of N and O mixture, which appears after L. Thus, Asp85 may play a role in Cl^--releasing reaction.

3Pos242 ハロロドプシンの陰イオン輸送サイクルにおけるレチナール色素の異性化
Isomerization of the retinal chromophore during the anion pumping cycle of halorhodopsin

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A recent crystallographic study of reaction states of the light-driven chloride ion pump halorhodopsin from Natronomonas pharaonis (pHR) has shown that an N-like reaction state (N') in which the retinal chromophore takes on the 13-cis/15-syn configuration occurs after the N state; that is, isomerization around the retinal Schiff base linkage occurs in the N-to-N' transition. In this study, we analyzed flash-induced absorption changes in pHR-rich claret membrane to clarify the reaction kinetics of the anion uptake process. The result suggests that at high halide ion concentrations, a large fraction of the N' state decays into the 13-cis/15-syn isomer (HR').

3Pos243 X 線小角散乱法を用いたシロイヌナズナ phototropin1 と変異体の構造及び機能研究

Structural and functional study of Arabidopsis phototropin1 and its mutants by using small-angle X-ray scattering

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Phototropin (phot), a blue-light (BL) receptor protein of plants, works to maximize the efficiency of photosynthesis. Phot comprises two light-oxygen-voltage sensing domains (LOV1 and LOV2) that absorb blue light, and a serine/threonine kinase domain (STK) responsible for BL-dependent phosphorylation leading to cellular signaling cascades. BL-excited LOV2 is primarily responsible to activate STK, but the detail how LOV2 activates STK is still unclear. Here we investigated LOV2-STK fragment of phot1 from Arabidopsis and its mutants by using small-angle X-ray Scattering at SPring-8. BL-dependent SAXS changes, the rearrangement of functional domains are analyzed. In addition, the influences of mutations to phosphorylation activity will be discussed based on the SAXS data.

3Pos244 Chromophore conformation in active site of orange carotenoid protein studied by Raman optical activity spectroscopy

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Orange carotenoid protein (OCP) is a photoreceptor protein which has a carotenoid pigment to sense light and works for photoprotection of the light-harvesting complex of cyanobacteria. Since the X-ray crystal structure was unveiled in 2003, OCP has been actively studied over this decade in terms of its structure and function. However, the molecular mechanism of the light-activated photoprotection has yet to be clarified. In this study, we use Raman optical activity spectroscopy and examine the conformational change of the active site of OCP. The detailed conformational analysis of OCP will be presented on the basis of the ROA spectra.

3Pos245 Functional characterization of a microbial rhodopsin from the marine eubacterium *Rubricoccus marinus* SG-29

Saki Inoue[1], Susumu Yoshizawa[2], Takashi Tsukamoto[1,3], Yuki Sudo[1,3] ([1]*Fac. Pharm. Sci., Okayama Univ.*, [2]*AORI, Univ. of Tokyo*, [3]*Grad. Sch. Med. Dent. & Pharm. Sci., Okayama Univ.*)

Recent advances in genomics has enabled to discover new types of retinal proteins. Here we characterized the function of a microbial rhodopsin from the marine eubacterium *Rubricoccus marinus* SG-29. It was successfully expressed in *Escherichia coli* cells as a recombinant protein showing pink color in the presence of retinal. Notably, the cell suspension was alkalized upon illumination and the alkalization was ceased by addition of the protonophore CCCP, indicating the "inward'' proton transport activity of the protein. Further spectroscopic analysis of the purified protein also revealed functional characteristics including light-dark adaptation and spectral blue-shift upon acidification. Together with other findings, their implications will be discussed.

3Pos246 Signaling kinetics of Cyanabacterial phytochrome (Cph1) studied by the transient grating method

Kimitoshi Takeda, Masahide Terazima (*Department of chemistry, Kyoto University*)

Cph1 is a member of phytochromes which are red and far-red light sensor proteins, and consists of a photosensory module (PSM) and an output module (OPM). Although light absorption triggers conformational changes via photoisomerization of the chromophore, the reaction dynamics has not been clarified yet. Here, we investigated the photoreaction of full length Cph1 by using the transient grating method. So far, we have revealed the following dynamics. Cph1 forms a dimer in the ground state. Upon red light excitation, the secondary structure changes in the vicinity of chromophore, which is followed by an association of the PSM part in the dimer. This may change the quaternary structure of Cph1 including the OPM, which should be a key step in the signaling process of Cph1.

3Pos247 In situ 光照射固体 NMR によるセンサリーロドプシン II の光中間体の解析

Characterization of photo intermediates in sensory rhodopsin II as revealed by in-situ photo-irradiation solid-state NMR

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Sensory rhodopsin II (NpSRII) transmits a negative phototaxis signal by photoisomarization of a retinal chromophore. To reveal the detailed photoreaction pathway, we measured 13C and 15N NMR signals of [14, 20-13C]Ret and [ε-15N]Lys -labelled NpSRII using in-situ photo-irradiation CP-MAS NMR. Under the green light (520 nm) illumination, three discrete M-intermediates and O-intermediate were successfully detected. And also, using the UV light at 365 nm to excite the intermediates, we trapped bR-like N-intermediate (N'-intermediate). We also calculated chemical shift values of retinal by means of quantum chemical calculation to reveal the difference of interaction of retinal with protein. In the result, we revealed structural changes of retinal in photocycle.

3Pos248 異なる生物種におけるフォトトロピン光反応の多様性

Diversity of photoreaction of phototropins among different organisms

Yusuke Nakasone[1], Koji Okajima[4], Kenichi Hitomi[3], John Christie[3], Satoru Tokutomi[2], Masahide Terazima[1] ([1]*Kyoto University*, [2]*Osaka Prefecture University*, [3]*Scripps research institute*, [4]*Keio University*)

Phototropins are blue-light dependent kinases in plants and green algae. They contain two LOV domains as light sensing modules and their photoreactions have been studied. We have found that the reactions of the LOV domains from Chlamydomonas reinharditii (Cr) are different from those of the LOV domains from Arabidopsis thaliana (At) and Ostreococcus tauri (Ot). In a case of Cr-phot, the LOV1 domain showed a drastic diffusion coefficient change upon photoexcitation, whereas the LOV2 did not undergo global reaction. This observation was contrary to the case of At- and Ot-phot, in which the excitation of the LOV2 domains resulted in the unfolding of their C-terminal helices. These findings suggest that phots have diversity in their signaling mechanism.

3Pos249 EPR 法による Photozipper 反応過程の解析
Reaction mechanism in Photozipper monitored by Electron Paramagntic Resonance

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Photozipper protein (PZ) is a synthetic gene encoding N-terminally truncated monomeric Aureochrome-1 containing a basic region/leucine zipper (bZIP) domain and a light-oxygen-voltage (LOV) domain. PZ acts as a transcriptional control element, regulated by blue light (BL). In order to clarify the reactions, the radical formation was investigated by electron paramagnetic resonance (EPR). In the mutants, C254S and C254A, blue light induced the neutral flavin radicals in the presence of DTT. It takes 2-3 min to form the radicals after illuminating BL with sigmoid function. The dependences of the light intensities and the sample concentration show that the sigmoidal kinetics reflects the formation of dimerization of the PZ.

3Pos250 Substrate recognition of the (6-4)photolyase

Yuma Terai[1], Takahiro Yumiba[1], Tomoko Ishikawa[2], Takeshi Todo[2], Junpei Yamamoto[1], Shigenori Iwai[1] ([1]*Grad. Sch. Eng. Sci., Osaka Univ.*, [2]*Grad. Sch. Med., Osaka Univ.*)

Exposure of DNA to ultraviolet light induces DNA damage, which inhibits proliferation of cells. The (6-4) photolyase is a unique enzyme that can specifically repair the (6-4) photoproducts, one of the UV-induced DNA damage, in a light-dependent manner, and its reaction mechanism has been investigated thus far. However, the substrate recognition mechanism remains unclear.
In this study, we synthesized various types of the oligonucleotides containing the (6-4) photoproduct, and binding of the purified (6-4) photolyase to the substrates was comprehensively analyzed. In combination with the mutational analysis of the amino acid side chains, we found an important interaction between DNA and enzyme that governs the binding of the (6-4) photolyase.

3Pos251 (6-4)光回復酵素による逐次的 2 光子 DNA 修復における逆電子移動の観測
Monitoring of the back electron transfer in the successive two-photons DNA repair by the (6-4) photolyase

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The (6-4) photolyase (6-4PL) is a flavoprotein that utilizes blue light to repair the (6-4) photoproduct (6-4PP), one of the UV-induced DNA damage. We have reported that the photorepair of the 6-4PP by the 6-4PL consists of two successive and distinctive photoreactions. The first photoreaction contributes to the formation of an intermediate and the second does to the completion of the repair. Because both photoreactions require photo-induced electron transfer (ET) from the excited state of fully-reduced FADH-, the electron should be transferred back to the transiently-formed FADH radical in each reaction.
In this study, we monitored the back ET with a single or repetitive laser flash(es), and successfully discriminated the back ETs of the first and second photoreactions.

3Pos252 イチョウの葉の微量色素分析
Precise pigment analysis of ginkgo leaves

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In photosystem (PS) I one molecule of Chl a' is present as a heterodimer of Chl a'/a, and in PS II two pheophytin (Phe)a molecules are present. There is no information about Chl a' and Phe a in primitive plants, e.g. gymnosperm. In this study, we performed precise pigment analysis of leaves of Cycas revoluta, Pinus thunbergii, Ginkgo biloba and Ephedra minima. The Chl a/Chl b ratio of angiosperm leaves is ca. 3.5, and the Chl a'/ Phe a is 1.1. Cycas revoluta showed almost the same stoichiometry. Ephedra minima showed higher a/b ratio, 4.2, but almost the same Chl a'/ Phe a ratio, 1.0. However, Pinus thunbergii showed lower a/b ratio of 2.7, and a'/ Phe a, 0.7. Ginkgo biloba showed similar lower a/b ratio of 2.7, and drastically lower Chl a'/ Phe a ratio of 0.4.

3Pos253 新奇クロロフィルを持つシアノバクテリアより光化学系 II 標品の単離精製とエネルギー移動機構の解析
Analysis of energy transfer system of photosystem II complexes isolated from new chlorophyll containing cyanobacterium

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Chlorophylls (Chls) play important roles in light harvesting, energy transfer, charge separation and electron transfer during photosynthetic reaction. Recently, more red-shifted Chl *f* was found in novel cyanobacterium. The Chl content of this cyanobacterium depends on the cultivation light. Chl *f* is present only in cells grown under far-red light (>700 nm). The photochemical and photophysical functions of Chl *f* are not known in photosystem II complexes (PS II). Therefore, we analyzed steady and time-resolved fluorescence spectroscopy for an investigation of PS II isolated from cells grown under far-red light. We discuss characteristics of PS II isolated from Chl *f* containing cyanobacterium.

3Pos254 Aggregation of chlorophylls d and f in n-hexane

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Chlorophylls (Chls) a and b are familiar in higher plants, Chl d is the major pigment in Acaryochloris marina, and Chl f is a unique chlorophyll in some cyanobacteria cultured under only far-red light. Recently, we came across the aggregation of Chls d or f in n-hexane. The absorption maxima were 755nm for Chl d and 831nm for Chl f, while the corresponding monomer maxima in diethyl ether were 686nm and 695nm. The QY band of Chl f aggregate is broader than that of Chl d aggregate. The QY/Soret raito of Chl f aggregate in hexane is as same as that of monomer Chl f in diethyl ether. Aggregation was not seen in Chls a, b, pheophytins a, b, d and f in hexane. The results suggest that Mg is indispensable for aggregation and position of formyl group is also important.

3Pos255 緑藻ミル糸状体における培養時光強度に依存したカロテノイドの蓄積

Extra accumulation of carotenoids upon intense irradiation during culture of a siphonous green algae, *Codium fragile*

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In the oxygenic photosynthetic organisms, carotenoids bound to the light-harvesting complexes are highly conserved: lutein, 9'-cis neoxanthin and violaxanthin. *Codium* species have siphonaxanthin and its ester instead of lutein, and are called as a siphonous green alga. Recently we have found that it accumulate extra all-trans neoxanthin (tNx) when cultivated under high irradiation conditions. In this study, we precisely compared the pigment profiles of the unialgal culture of *Codium fragile* (KU-654, KU-MACC) under four strengths of irradiances with three different kinds of light-sources. As a result, the accumulation of tNx and all-trans violaxanthin was promoted in accordance with the photon density of cultivation. Plausible function of extra tNx was discussed.

3Pos256 チラコイド膜での光還元に対する共溶媒の効果

Effect of co-solvents on photo-reduction in thylakoid membranes

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We examined photo-redox activity in thylakoid membrane containing PS1 and PS2 for photosynthesis in the presence of co-solvents, namely sucrose, glucose, betain, and saponin using a colorimetry of reduction of 2,6-dichloroindophenol (DCIP). In addition, 2,5-dibromo-6-isopropyl-3-methl-1,4-benzoquinone (DBMIB) was used to inhibit electron transport from PS2 to PS1, distinguishing PS2 activity only. In the combination of PS1 and PS2, the photo-reduction of DCIP was increased by 50% with the increase in concentrations of betain above 0.1 M, whereas sugars up to 1.5 M decreased the reduction. In the PS2 only, both betain and sugars increased the reduction. Interestingly, saponin, which is a detergent containing in plants, also increased the reduction.

3Pos257 Identity of chlorophyll e

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We serendipitously came across the formation of Chl d from Chl a with papain in aqueous acetone. Papain is a proteolytic and thiol protease. Bromelain is the same group protease present in pineapple. In this paper, we incubated Chl a with pineapple in aqueous acetone. The expected conversion of Chl a to d was not observed, but a new peak was detected. Absorption property of the new pigment is the same as that of 151-OH-lactone Chl a. We found that 151-OH-lactone Chll a shows the absorption spectrum similar to that of Chl e discovered in yellow-green algae Tribonema bombycinum and Vaucheria hamata. Chlorophyll e was reported to resemble Chl c with respect to its absorbality in a sugar column. Chl e might be 151-OH-lactone Chl a or 151-OH-lactone chlorophyllide a.

3Pos258 Solubility and stability of chlorophylls in algal oil

Terumitsu Kanjoh[1], Mikihide Demura[2], Masaki Yoshida[2], Makoto Watanabe[2], Masataka Nakazato[3], Masami Kobayashi[1] ([1]*Div. Materials Sci., Fac. Pure Appl. Sci., Univ. Tsukuba*, [2]*Fac. Life Environ. Sci., Univ. Tsukuba*, [3]*Chl. Tes. Inst.*)

There is scarce information about the solubility and stability of chlorophylls (Chls) in algal oils, *e.g.*, squalene and squalane. In this work, we examined the solubility and stability of Chls *a, b, d* and pheophorbides (Phdes) *a, b* in squalene and squalane. Chlorophylls *a, b* and *d* were found to be dissolved in squalene easily, but slightly in squalane. Pheophorbides *a* and *b* without long chain were insoluble in both oils. Note that squalene has six C=C double bonds, and squalane has no C=C double bond. The results suggest that chlorophylls have higher solubility in squalene possessing a lot of double bonds. It is of interest to note that Chls *a, b* and *d* are stable in squalene rather than in squalane, which runs counter to our expectations.

3Pos259 Complex formation between carbon nanomaterials and photosystem complexes

Shota Tanaka, Tatsuya Tomo (*Grad. Sch. Sci., Tokyo Univ. of Sci.*)

The hybrid device using carbon nanomaterial and photosystem complexes has been promoted in recent years, since the quantum yield of photosynthetic energy and electron transfer is nearly 100%. To obtain reduction power from water, "Z-scheme" system is effective reaction which involved in two photosystem complexes. In this study, we isolated photosystem complex from Thermosynechochoccus elongatus and applied graphen oxide as carbon nanomaterial. Because graphen is an excellent electron carrier. The reduced graphen oxide and photosystem complex was successfully obtained by visible light irradiation. This was confirmed by oxygen evolving measurements, photoluminescence and X-ray photoelectron spectroscopy. The implications of these results will be discussed.

3Pos260 緑化途上トウモロコシ生葉の極低温顕微分光による光合成タンパク質前駆体の蛍光スペクトル同定

Spectral identification of late precursors to photosynthetic proteins by cryogenic microscopy of greening etiolated Zea mays leaves

Hirotomo Nagasawa, Tomofumi Chiba, Yutaka Shibata (*Graduate School of Science, Tohoku University*)

When angiosperms are germinated under dark conditions, protochlorophyllide, a late precursor to chlorophyll (Chl), is accumulated. Once irradiated with light, Chl is synthesized by a light-dependent enzyme and the assemblies of photosystems (PSs) are started. The purpose of the present study is to reveal the in vivo assembly of PSs during this greening process.

In a C4 plant, the distributions of the two PSs are different depending on the distance from the vascular bundle. We conducted the fluorescence microscopy on leaf sections of greening Zea mays by using the home-build cryogenic confocal microscope. We identified the fluorescence spectra of late precursors to PSs by the singular-value decomposition analysis. We will discuss the assembly intermediates to PSs.

3Pos261 北海道で採取した紅色非硫黄細菌による酢酸塩からの光水素生成

Phototrophic hydrogen production from acetate by purple non-sulfur bacteria from rivers in Hokkaido

Mayoka Kanoh[1], Kazuma Tazawa[1], Seigo Kumakura[2], **Masahiro Hibino**[1,2] ([1]*Div. Sustain. Enviro. Eng., Muroran Inst. Tech.*, [2]*Dept. Appl. Sci., Muroran Inst. Tech.*)

Hydrogen has been considered a potential fuel for the future, mainly due to its recyclability and nonpolutiong nature. Hydrogen has to be produced renewably and in large scale, with environmentally benign processes. Biological hydrogen production stands out as an environmentally harmless process carried out under mild operating conditions with renewable resources. Herein we have been studying photoproduction of hydrogen by purple non-sulfur bacteria from rivers in Hokkaido. The cell growth, substrate consumption kinetics and the substrate conversion efficiency of acetate into hydrogen have been investigated in batch-type photobioreactor experiments for hydrogen production by growing cells of the purple non-sulfur bacteria. The details of the results will be discussed.

3Pos262 低線量或いは高線量 X 線被ばく後のマウス肝臓におけるメタボローム解析

Metabolome analyses in livers of mice exposed to low or high-dose X-ray-irradiation

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Radiation induces many molecular alterations in organisms. Biological markers, which indicate radiation effects and are used for predicting late effects in the future, are expected to be identified. Metabolomics has attracted much attention. Low molecular weight metabolites, which are more directly related to biological activities in organisms than DNA or proteins, are evaluated in metabolomics. Here, metabolic analyses were performed in livers of mice irradiated at 0.1, 0.5 and 2 Gy with X-rays. Dose-dependent or dose-specific alterations were evaluated. Metabolites in the liver were analyzed by CE-TOFMS at one month after irradiation. Altered metabolites will be discussed in the viewpoint of characters and usage as biomarkers for evaluating radiation effects.

3Pos263 ビタミンDによって誘発される単球の分化と酸化ストレスの関係

Relations of oxidative stress and monocytic differentiation induced by vitamin D_3

Naoya Matsunaga, Kiyotaka Murakami, Wakako Hiraoka (*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*)

To elucidate the mechanism of immune enhancement, we focused on the signal transduction related to oxidative stress in 1α ,25-dihydroxyvitamin D_3 (vitamin D_3)-induced differentiation. PLB-985, human myeloid cells, were stimulated with vitamin D_3 (200 nM) and Phorbol 12-myristate 13-acetate (PMA) (100 nM) for monocytic differentiation. Microscopy and electron spin resonance (ESR) spin-trapping analysis showed that reactive oxygen species (ROS) were generated in vitamin D_3 stimulation and vitaminD_3 enhanced Nox2 activation. We intend to report a more detailed process using Nox inhibitors and ROS scavengers.

3Pos264 プリオンオクタペプチドと二価金属錯体結合によってひきおこされるレドックス不均衡

Redox imbalance induced by coordination of divalent metals in octarepeat region of human PrP

Shinnosuke Kondo, Wakako Hiraoka (*Dept. Phys., Grad. Sch. Sci. & tech., Meiji Univ.*)

Prion protein (PrP) has metal-binding sites, and its misfolding leads to neurodegenerative diseases. While the physiological function of PrP is still unknown, its redox and metal-reservoir potentials are considered to be significant. We focused on the octapeptide (PHGGGWGQ) acting as the Cu^{2+}-binding site located in the octarepeat region of human PrP. In our last presentation at the 53th BSJ, we revealed that the binding process of octapeptide-Cu^{2+} was reversible and that the other divalent ions were partially substituted for Cu^{2+}. In this study, we investigated the reactivity of metal-binding octapeptide with ROS via ESR spin-trapping analysis and found that the metal-binding octapeptide accelerated the conversion from superoxide into hydroxyl radical.

3Pos265 超音波に誘発される CMNB-ケージ基と脂肪酸の分解

Ultrasound-induced scission of CMNB-caged moiety and fatty acids

Kengo Takei[1], Takuya Wada[1], Asuka Kato[1], Masato Mutoh[2], Wakako Hiraoka[1] ([1]*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Mater. & Human Env. Sci., Shonan Inst. of Tech.*)

We have determined the efficiency of molecular scission in caged compounds and fatty acids irradiated with ultrasound at 28 kHz. CMNB-caged fluorescein was used as a caging model, from which fluorescein can be released after the scission of the caged moiety. Caged compound, oleic acid, and stearic acid were solvated in PBS or methanol in order to estimate the effect of hydroxyl radicals generated via the sonolysis of water. DART mass spectroscopy showed that the fluorescein was liberated from the caging after irradiation. In the case of fatty acid, no product attributed to selective scission was observed after irradiation. Hydroxyl radical was considered to be the most effective factor in the ultrasound-induced molecular scission of both caged compounds and fatty acids.

3Pos266 一分子観察における DNA 二重鎖の光切断に対する PEG 保護作用

Protective effect of PEG against DNA double-strand breaks caused by photo irradiation through single molecule observation

Moe Usui, Yuko Yoshikawa, Kenichi Yoshikawa (*Facul. Life Med. Sci., Doshisha Univ.*)

We have performed quantitative evaluation on the double-strand breaks of genome-sized giant DNA (T4 DNA; 166 kbp) caused by photo irradiation at difference concentrations of PEG, polyethylene glycol. Under the irradiation of visible light with 491 nm, real-time observation by use of fluorescence microscopy was carried out for the DNA molecules stained by a cyanine dye, YOYO-1, where YOYO-1 also plays the role to generate reactive oxygen species under photo irradiation. We have measured the breaking time of DNA from the start of irradiation. It has become clear that PEG decreases the probability of double-strand breaks. We will discuss the mechanism in relation to the consumption of reactive oxygen species with PEG.

3Pos267 **機械学習を用いた機能未知スプライシングアイソフォームの機能性推定**
Evaluation of functionality of uncharacterized splicing isoforms using machine learning techniques

Pramote Teerasetmanakul, Masafumi Shionyu (*Grad. Sch. Bio-Sci., Nagahama Inst. Bio-Sci. Tech.*)

Although many isoforms produced by alternative splicing have been discovered, isoform-specific functions are not well studied experimentally. It is expected uncharacterized splicing isoforms from the same gene have distinctive functions to diversify its gene function. However, structure-based method estimated many splicing isoforms were nonfunctional in previous studies. We developed a method based on machine learning techniques that estimates isoform functionality starting from some biological properties, such as expression level of protein or mRNA. Our method revealed many uncharacterized splicing isoforms, which were estimated to be products of splicing noise by structure-based method in previous studies, had some biological functions.

3Pos268 **Predicting protein-protein interactions using sequence homology and machine learning methods**

Yifan Tang, Wei Cao, Tohru Terada, Kazuya Sumikoshi, Shugo Nakamura, Kentaro Shimizu (*Grad. Sch. Agr., Univ. Tokyo*)

Protein-protein interactions plays an important role in cellular processes. These processes are regulated and performed by the interaction between proteins. Therefore, identification of PPIs could help us have a deep understanding of protein functions and various biological processes. We developed a method for predicting PPIs using sequence homology and machine learning methods. As for homology-based method, we predicted that a given pair of proteins interact with each other if the homologous proteins interact. Various sequence features are used for the machine learning methods including support vector machine, random forest and so on. We compared results of prediction by homology-based method and machine learning methods and designed the integration of these methods.

3Pos269 **クロマチン構造形成における単純反復配列の機能的役割**
Functional Roles of Simple Repeat Sequences in Chromatin Conformations

Takeru Kameda[1], Atsushi Ikegaya[1], Takeshi Sugawara[2], Naoaki Sakamoto[1,2], Akinori Awazu[1,2] ([1]*Dept. of Mathematical and Life Sciences, Hiroshima University*, [2]*Research Center for the Mathematics on Chromatin Live Dynamics*)

Non-coding DNA regions in eukaryote genomes have been investigated extensively. These regions often include a variety of simple repeat sequences (Micro-satellite). Such repeat sequences are known to induce several diseases by their extensions in human genome. On the other hand, they also have been expected to play crucial roles to determine the chromatin conformations. Thus, in this study, we analyzed the characteristics of simple repeat sequences like di- nucleotides and tri-nucleotides repeats to infer possible functional roles of such repeat sequences to regulate the local chromatin conformations. We also focused on the distributions of repeat sequences in human chromosomes to infer their roles by the comparison with those of CTCF binding sites.

3Pos270 **翻訳伸長因子 1A の配列情報に基づいた機能分岐に関わる重要な残基の予測**
Prediction of key residues involving functional divergence based on sequence information of translation elongation factor 1A

Yosuke Kondo, Satoru Miyazaki (*Fac. Pharm., Tokyo Univ. Sci.*)

Eukaryotic elongation factor 1A (eEF1A) is a GTP-binding protein and elongates new polypeptide chains. While there are two types of eEF1As, eEF1A1 and eEF1A2, whose sequence identity is 92%, the binding affinities for GTP and GDP are different. In addition, eEF1A1 is involved in apoptosis, whereas overexpression of eEF1A2 causes cancer. However, it is unknown how the functional divergences are triggered. In this study, we aligned sequences homologous to human eEF1A and then obtained alignment sites including residues not only mutated different properties from human's but also conserved within each group of some organisms. Because of consistency with post-translational modification sites of eEF1A, we concluded that the obtained residues should be analyzed experimentally.

3Pos271 **天然変性領域の機能部位：Protean Segments が効果的に相互作用できる理由**
Interface property of protean segments: intrinsically disordered regions that undergo disorder-to-order transitions upon binding

Divya Shaji, Takayuki Amemiya, Ryotaro Koike, **Motonori Ota** (*Grad. Sch. Inf. Sci., Nagoya U.*)

Protean segments (ProSs) are the short regions in intrinsically disordered proteins that can undergo disorder-to-order transitions upon binding. It has been indicated that interactions of ProSs are effective: the number of contacts per residue of ProS interface is large. To reveal the properties of ProS interface that are responsible for the interaction efficiency, we classified the interface into core, rim and support, and analyzed them based on the relative accessible surface area (rASA). We found that the balance between a small core and a large rim, and the large solvent exposure of the rim in the monomeric state, are the key to the disorder-to-order transition and the effective interactions of ProSs.

3Pos272 **Immuno-Navigator: a co-expression database for cell type-specific network inference in the immune system**

Alexis Vandenbon (*IFReC, Osaka University*)

Correlation of expression can offer useful hints regarding gene function or underlying regulatory mechanisms. Large amounts of expression data are now publicly available, allowing researchers to estimate expression correlation over thousands of samples. However, extracting information from correlation data is not straightforward, because the raw data was generated by different laboratories under different conditions. Here we present Immuno-Navigator, a coexpression database for cells of the immune system, which addresses this issue. We show how our database can be used for generating hypotheses for further experimental analysis. We demonstrate how it confirms known facts in immunology and successfully predicts key regulators in naturally occurring regulatory T cells.

3Pos273 光回復酵素／クリプトクロムファミリーの機能発現に重要なアミノ酸残基部位の探索

In search for amino acid positions that determine the molecular function of photolyase/cryptochrome family

Daichi Yamada[1], Kei Yura[1,2] ([1]*Cent. Info. Biol., Ochanimizu Univ.*, [2]*NIG*)

Photolyase (PHR) is one of the enzymes that repairs UV-damaged DNA. Cryptochrome (CRY) is homologous to PHR, but no longer repairs UV-damaged DNA and functions as a photoreceptor or as a component of the biological clock. Both of them assume a strikingly similar protein architecture, though the cause of the functional difference remains unknown. In this study, we scrutinized their amino acid sequences and narrowed down the amino acid positions important for distinguishing the functions of PHR/CRY family. Specifically, we built an alignment and a phylogenetic tree, and compared the highly conserved positions. The differences in the conservation pattern of amino acid residues can be an explanatory cue for the difference in the function of PHR/CRY family.

3Pos274 Refining the performance of k-mer count similarity prediction and examining use of different scoring matrix for better pairwise alignment

Kazunori Yamada, Kengo Kinoshita (*Tohoku University*)

The method counting the number of sharing k-mers between pairwise sequences and calculating its inverse number is one of a simplified prediction method of similarity between two biological sequences. The performance of the method largely depends on the k-mer length to be counted. In this study, we examined the effect of various length of k-mers on the performance of the similarity prediction and developed a more refined similarity predictor using machine learning methods with various k-mers as input vector. In addition, we examined the effect of changing substitution matrix based on the predicted similarity of pairwise sequences on the accuracy of pairwise alignment.

3Pos275 緑藻の走光性と多細胞性の関係

Relation between phototaxis and multicellularity of green algae

Keisuke Yamada, Yoshihiro Murayama (*Tokyo Univ. of Agri. and Tech.*)

We have investigated phototactic behavior of two species of spherical green alga, Volvox consisting of 2000 somatic cells and Pleodorina consisting of 128 cells, to reveal the relation between the phototaxis and the multicellularity. Each somatic cell has an eyespot and two flagella, and both algae can swim toward light by coordinating flagellar beating. Algae in a chamber were stimulated sinusoidally by varing light intensity from both sides of the chamber, and the motion of algae were observed. The response to the light stimulus was characterized by a mobility and a relaxation (response) time. We have found that both algae display approximately same mobility but the relaxation time of Pleodorina is shorter than that of Volvox.

3Pos276 染色体の凝縮が染色体の構築や分離に与える影響

Effects of chromatin condensation on chromosome construction and segregation

Yuji Sakai[1,2], Masashi Tachikawa[1], Atsushi Machizuki[1], Kazuhisa Kinoshita[3], Tatsuya Hirano[3] ([1]*RIKEN, Theoretical Biology Laboratory*, [2]*RIKEN, iTHES*, [3]*RIKEN, Chromosome Dynamics Laboratory*)

The spatial organization of chromosomes in mitosis is a fundamental question of biology. Upon entry into mitosis, a mass of chromatin distributed in the interphase nucleus is converted into a discrete set of chromosomes with high condensed rod-shape structures. The condensen protein complex is response for the chromosome condensation. It is considered that the condensin has two functions, making twisted loops in chromosome and getting gather in chromosome axis.

In this study, we investigate effects of these condensin functions on the chromosome costruction and the segregation by using coarse-grained molecular dynamics simulations.

3Pos277 クロマチン動態とコンタクトマップの関係を深く理解するための数理的研究

A mathematical study for deep understanding of relationship between chromatin dynamics and contact map

Masaki Nakagawa (*RcMcD, Hiroshima Univ.*)

In recent years, chromosome conformation capture (3C) method is well developed. The contact map of chromosomes is expected to give rich information about the chromosome conformation. However, the chromatin fiber constructing chromosomes is flexible unlike proteins. Therefore, it is generally difficult to understand complete relationship between chromatin dynamics and the contact map.

In this study, we focus on topological associated domain (TAD) structures appearing in contact maps, and will discuss theoretically about relationship with loop structures in chromatin fibers by using abstract polymer models.

3Pos278 Enhancement of sampling space in multivariate analysis of experimental big data in various biological sciences

Jiyoung Kang[1], Kazuhiko Yamasaki[2], Masaru Tateno[1] ([1]*Univ. of Hyogo*, [2]*Biomed. Res. Ins. AIST*)

Recently, mathematical analyses of experimental big data have become crucial in various biological disciplinary. The multivariate curve resolution alternating least-squares (MCR-ALS) method is widely used to decompose multidimensional complicated data, such as spectral data, to several pure spectra. In this study, we analyzed our circular dichroism (CD) spectra data of double-stranded DNA (228 bp) in the complex with a peptide under various concentrations, but the MCR-ALS failed. So, we combined our simulated annealing (SA) protocol (i.e., the SA-MCR-ALS), and thereby the analysis succeeded to obtain reasonable pure spectra. Thus, our method should be a standard tool for the extended sampling of various multivariate data, such as microarray and image data.

3Pos279 血糖値調節におけるインスリン・Cペプチドの血中動態の数理モデルを用いた解析

Mathematical model analysis of blood glucose regulation with insulin and C-peptide

Kaoru Ohashi[1], Masashi Fujii[1], Shinsuke Uda[1], Hisako Komada[2], Kazuhiko Sakaguchi[2], Wataru Ogawa[2], Shinya Kuroda[1] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Grad. Sch. Med., Univ. Kobe*)

Homeostatic control of blood glucose is regulated by a complex feedback between glucose and insulin. Assessment of insulin secretion function in humans has been challenging because insulin concentration is inferred from hepatic insulin extraction. However, C-peptide, equimolar secreted with insulin, is negligibly degraded by the liver, and enables simultaneous assessment of insulin secretion and hepatic insulin extraction. We constructed and selected mathematical models representing the measurements of blood insulin and C-peptide of 121 human subjects with healthy and type 2 diabetes, and discuss relationships among estimated parameters about insulin secretion and degradation by the liver and peripheral tissues.

3Pos280 生命科学研究用の特化型シミュレータ群の具現化

Implementation of a group of special-purpose simulators for life science researches

Hideto Katsuma[1,2], Jun Takayama[2], Yukako Tohsato[2], Koji Kyoda[2], Shuichi Onami[1,2] ([1]*Grad. Sch. System Inform., Kobe Univ.*, [2]*Lab. Dev Dyn., RIKEN QBiC*)

Simulators are important tools in life science researches. Most cell simulators developed so far are multifunctional general-purpose simulators. A strong point of a general-purpose simulator is the capacity to deal with various life phenomena by using the multifunctionality. However, the multifunctionality often complicates the structure of simulator. To circumvent this problem, we are developing special-purpose simulators, each of which deals with a specific life phenomenon and implements one geometry model and one mathematical model. To deal with various life phenomena, we employ the concept of 'a group of special-purpose simulators'. As a first step, we developed a group of special-purpose simulators for intracellular calcium waves.

3Pos281 遺伝子発現量制御メカニズムのタイプ分類と遺伝子機能の関係

Relationship between regulatory pattern of gene expression level and gene function

Masayo Inoue, Katsuhisa Horimoto (*molprof, AIST*)

The regulation of gene expression is essential for the normal functioning of all living organisms. We studied the mechanisms of regulation of gene expression levels by using a simple mathematical model and available DNA microarray expression data from the GEO data repository. Based on our model, the gene expression relationships between a TF and its regulating gene were classified into four types that correspond to different gene regulatory mechanisms. Pathway analysis revealed that each relation type corresponded to distinct gene functions. This finding indicates that gene expression is regulated structurally (not arbitrarily) according to the gene function; thus, systematic regulatory control may exist at the whole-cell level.

3Pos282 T細胞の胸腺における細胞数のダイナミクスのモデリングと推定

Modeling and inferring dynamics of T cell population in thymus

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A development of T cell, a type of immune cells, in the thymus is a dynamic process that generates the repertoire of T cell population. Thymic epithelial cells (TECs) are responsible for shaping the appropriate repertoire by selecting functional and nonself-reactive T cells. We still have much less knowledge on the dynamic aspects of the selection and differentiation in thymus especially from the viewpoint of regulations by TECs.
In this study, we construct a mathematical model on the recovery dynamics of T cells after X-ray irradiation by employing experimental data on joint dynamics of T cell and TEC populations. The inferred model suggests positive/negative correlation between medullary/cortical TECs and the stable population size of the T cells.

3Pos283 触媒反応系から成る細胞のトレードオフ

Trade-off in a protocell model with catalytic reactions

Atsushi Kamimura, Kunihiko Kaneko (*The University of Tokyo*)

Cells integrate diverse molecules to keep their reproduction taking essential resources from environment. It is commonly observed that cells with higher fitness in one environment can lead to reduced fitness in another. Such negative correlations are generally called trade-offs, and, together with varying environments, are likely considered as one of the major sources to produce the great diversity of cells. Here, by considering a protocell model consisting of catalytic molecules and resources, we will discuss how such trade-offs of cell growth can be integrated into theoretical treatment. In particular, we will emphasize an importance of reaction steps of complex formation to show the trade-offs.

3Pos284 SpineにおけるSmall-volume effect: Robust, Sensitive, Efficientな情報伝達のメカニズム

Small-volume effect enables robust, sensitive and efficient information transfer in the spine

Masashi Fujii[1], Kaoru Ohashi[1], Yasuaki Karasawa[2], Minori Hikichi[1], Shinya Kuroda[1] ([1]*Dept. Biol. Sci., Grad. Sch. of Sci., Univ. of Tokyo*, [2]*Dept. Neurol., Grad. Sch. of Med., Univ. of Tokyo*)

The spine is an extremely small platform (0.1 μm^3, 10^4-fold smaller than a cell) for information processing in a neuron, and contains merely tens to hundreds molecules. Why is the spine so small that reactions inevitably become stochastic? We previously showed that, despite such noisy conditions, the spine exhibits robust, sensitive and efficient features of information transfer using the probability of Ca^{2+} increase; however, the mechanisms are unknown. Here, we show that the small-volume effect enables robust, sensitive and efficient information transfer in the spine volume, but not in the cell volume. We propose that the small-volume effect is the functional reason why the spine has to be so small.

3Pos285 Curvature-driven splitting of a planar traveling wave

Kazuya Horibe[1], Ken-ichi Hironaka[2,3,4], Katsuyoshi Matsushita[2], Koichi Fujimoto[2] ([1]*Grad. Sch. Info, Univ. Osaka*, [2]*Grad. Sch. Sci., Univ. Osaka*, [3]*CDB, Inst., Riken*, [4]*JSPS Research Fellow(PD)*)

Although reaction-diffusion equations provide the variety of travelling waves dynamics such as collision and annihilation mainly on a flat surface, little is known about the role of curved surface. Here we numerically show the sequence of dynamics, i.e., bending, collision, annihilation and splitting of a travelling wave, can be driven by a curved surface represented by the two dimensional Gaussian function. The sequence to wave splitting occurred above a threshold ratio of the height and width of the bell shape. The wave split utilizing the curvature information could function as a signal processing on convoluted surface such as amoeboid cells and brain cortex.

3Pos286 Quantification of dynamic mechano-response of myoblast using stimulus responsive matrix

Marcel Hoerning[1], Masaki Nakahata[2], Akihisa Yamamoto[1], Mariam Veschgini[3], Stefan Kaufmann[3], Yoshinori Takashima[2], Akira Harada[2,4], Motomu Tanaka[1,3] ([1]*iCeMS, Kyoto University*, [2]*Osaka University*, [3]*Heidelberg University*, [4]*ImPACT*)

Stimulus-responsive materials can reversibly alter the elasticity and thus enable the detailed study of biologically relevant cell dynamics [Yoshikawa et al., JACS, 133, 2011]. In this study, we use hydrogels that are cross-linked by supramolecular host-guest interactions [Nakahata et al, Nature Commun. 2, 2011]. We show that the morphological dynamics of C2C12 myoblasts can be reversibly controlled by real-time induced changes in the mechanical microenvironment. We track the stress-sensitive responses of the cells by quantifying the actin (de-)polymerization dynamics and other morphological parameter.

3Pos287 再生ヒドラにおける形状ダイナミクスと対称性の破れの定量化

Quantification of Morphological Dynamics and Symmetry Break in Regenerating Hydra Tissues

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The freshwater polyp Hydra is a paradigm for an unlimited regenerative capacity, where a whole organism can be regenerated from a broad range of initial conditions. To understand the symmetry break (body axis formation) of a regenerating Hydra, the morphological dynamics is quantitatively studied using mode analysis. We introduce a non-invasive experimental system that prevents the loss of information concerning the active fluctuation during regeneration. Using both regenerates (cut tissue segment) and reaggregates (reaggregation of dissociated single cells), we show that the dynamics of the modes and translational motion is strongly related to the symmetry break. The results suggest the importance of cytoskeletal structure formation and emergence of hydroskeletons.

3Pos288 2D swarming bacteria

Chien Jung Lo, **Ching Yuan Lin** (*Dept. of Physics, NCU, Taiwan*)

Motility is one of the charming feature of living organism. From physics point of view, active-matter is composed of large number of individual active agents with simple rules of movement leading to complex collective behaviors. Due to the experimental difficulties of manufacturing identical active agents, theoretical work leads the research work in this field. In our research, we use surface moving bacteria as the active-material to study the emerging behavior of active nematics in 2-dimension system. We especially focus on a jamming state of active nematics where all of the local force are balanced. We also investigate the jamming process and define the specific topological structure in this unique jamming state.

3Pos289 聴覚刺激によって引き起こされる脳波の引き込み現象と確率共鳴

Auditory entrainment and stochastic resonance in EEGs

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We here report auditory entrainment and stochastic resonance in electroencephalograms (EEGs). EEGs were recorded with an 8-channel electroencephalograph where each channel was assigned to one of the 8 electrode positions of the international 10-20 electrode method. When a periodic pulse-type auditory stimulus (24-36 Hz) was applied to the left ear of the subject, a sharp peak was observed at the same frequency in the power spectrum for EEGs from two electrodes (CP5, CP6). Next, the stimulus intensity was decreased until the sharp peak in the power spectrum disappeared. When a noisy stimulus was applied to the right ear with the periodic stimulus to the left ear, the sharp peak appeared again. In addition, auditory signal detection was improved in a similar way.

3Pos290 Observation of conformational dynamics of FliI by HS-AFM

Kei Adachi[1], Jun-ichi Kishikawa[3], Hiroyuki Terashima[2], Takayuki Uchihashi[1], Katsumi Imada[2], Ken Yokoyama[3], Toshio Ando[1] ([1]*Coll. Sci. & Eng., Kanazawa Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Facul. Biosci., Kyoto Sangyo Univ.*)

FliI is a component protein of the bacterial flagellar export apparatus that transports the flagellar specific proteins across the cell membrane to the distal end of the growing flagellum. It is known that FliI is a homohexameric ATPase and forms a complex structure with FliJ similar to the $\alpha_3\beta_3$ hexamer with the γ subunit of F_1-ATPase. However, the functional mechanism of FliI, such as conformational dynamics in ATP hydrolysis, is still unknown. In this study, we directly observed the conformational changes of FliI induced by ATP hydrolysis using a high-speed AFM. In the presentation, we will show the oligomerization process of FliI in the presence of ATP visualized by the high-speed AFM and discuss the dynamics relevant to the physiological function of FliI.

3Pos291 高速原子間力顕微鏡と光ピンセットの複合システム

Combined system of high speed atomic force microscopy(HS-AFM) and optical tweezers

Shin'nosuke Yamanaka[1], Akane Goto[1], Hiroki Watanabe[2], Takayuki Uchihashi[1,3], Toshio Ando[1,3] ([1]*Grad. Sch. Sci., Kanazawa Univ.*, [2]*RIBM Co., Ltd.*, [3]*Bio-AFM FRC., Kanazawa Univ.*)

High-speed AFM (HS-AFM) enables us to directly visualize dynamic events on biological molecules. On the other hand, optical tweezers have been widely used to manipulate individual molecules through a bead binding to the target molecules. If we could combine the HS-AFM and the optical tweezers, the HS-AFM would be a more versatile tool because the combined system will allow us to observe dynamic behaviors of molecules under loads. For this purpose we incorporated the dual beam optical tweezers into the HS-AFM combined with fluorescent microscopy. In the presentation we will discuss the instrumentation of the combined system and sample preparation for HS-AFM imaging of single molecule under the external forces.

3Pos292 脂質膜の曲率に依存したタンパク質-脂質膜の相互作用の直接観察のための高速 AFM 用基板の開発

Development of HS-AFM substrate for observation between proteins and lipid membrane depending on the physical shape of lipid membrane

Takahiro Toyoda[1], Shin'nosuke Yamanaka[1], Akane Goto[1], Hiroki Watanabe[2], Shunsuke Shozui[1], Mikihiro Shibata[1,3], Takayuki Uchihashi[1,3] ([1]*Dept. of phys., Kanazawa Univ.*, [2]*RIBM Co., Ltd.*,, [3]*Bio-AFM FRC., Kanazawa Univ.*)

It is recently illustrated that some proteins associate with and/or dissociate from lipid membrane by recognizing the physical shape or curvature of lipid membrane. Direct observations of these phenomena at sub-molecular level by HS-AFM would promote a better understanding of these mechanisms. To realize this observation, we are developing a substrate that can change the physical shape of lipid membrane. Using a nano-sphere imprinting, we succeeded in the fabrication of a substrate with submicrometer-sized holes over a wide area, on which lipid membrane or lipid membrane with membrane proteins were covered. Thereby, flat area and area with a curvature were created on those membrane surfaces. We will analyze behaviors of proteins and lipid molecules on both areas.

3Pos293 長時間１蛍光分子追跡法による接着班分子の動的リクルートの解明

Transient recruitment of focal adhesion molecules revealed by super-long single molecule tracking

Taka-aki Tsunoyama[1], Kenichi G.N. Suzuki[2,3], Takahiro K. Fujiwara[2], Akihiro Kusumi[1,2,4] ([1]*OIST*, [2]*Inst. Integrated Cell-Material Sciences (WPI-iCeMS), Kyoto Univ.*, [3]*NCBS/inStem, India*, [4]*Inst. Frontier Medical Sciences, Kyoto Univ.*)

Super-long single-molecule imaging (>100 s) developed by us by greatly reducing photobleaching and photoblinking was employed for studying focal adhesion (FA)-protein dynamics in living cells. Integrin molecules entered the FA region quite readily and diffused there, with intermittent myosin-dependent stationary periods lasting for 40 s, during which integrin linked actin filaments (AFs) and extracellular matrix (ECM). This result suggests that the AF-ECM linkage was mediated by many integrin molecules recruited to the FAs one after another. Other cytoplasmic FA proteins were also dynamically and temporarily recruited to the FAs (1~10 s), and interestingly, ~20% of them were recruited by way of diffusion on the cytoplasmic surface of the plasma membrane.

3Pos294 個々の酵母細胞内 ATP 濃度の可視化により明らかになった、酸化ストレス下における細胞内 ATP 濃度の低下

In vivo imaging of cytoplasmic ATP in living yeast cells reveals a profound effect of oxidative stress on ATP level

Masak Takaine[1,2], Hiromi Imamura[3], Satoshi Yoshida[1,2] ([1]*Gunma Univ. Initiative for Adv. Res.*, [2]*Gunma Univ. Inst. for Mol. and Cell. Regulation*, [3]*Lab. of Funct. Biol., Grad. Sch. of Biostudies, Kyoto Univ.*)

Cells use ATP as a source for chemical energy. Therefore, it is important to understand the dynamic changes in intracellular ATP level (ATP_i) in individual cells for deciphering cellular energy metabolism. However, it has long been technically challenging.
Here, we used a recently developed ATP indicator QUEEN and visualized the ATP_i in living yeast cells. We verified that QUEEN is a reliable indicator of ATP_i in yeast and found that ATP_i rapidly drops in response to the exposure to oxidative stress as well as glucose depletion.
Our study demonstrates a powerful use of the biosensor for visualizing ATP dynamics and reveals that yeast cells have a mechanism to adjust ATP_i upon environmental stress.

3Pos295 PDLIM2 の相互作用タンパク質 MKRN2 は、NF-κB の p65 サブユニットに対する新規ユビキチン E3 リガーゼとして機能する

PDLIM2-interacting protein MKRN2 functions as a novel E3 ligase for p65 subunit of NF-κB

Chanyoung Shin[1,2], Yuma Ito[1], Makio Tokunaga[1], Takashi Tanaka[2], **Kumiko Sakata-Sogawa**[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*IMS, RIKEN*)

PDLIM2 functions as a nuclear ubiquitin E3 ligase for p65 subunit of NF-κB, negatively regulating NF-κB -mediated inflammatory responses. However, it remains unclear how PDLIM2 activity can be controlled. Here we show that MKRN2, a RING finger domain-containing ubiquitin E3 ligase, is associated with PDLIM2 and critical for PDLIM2-mediated termination of NF-κB activation. Forced expression of MKRN2 promoted polyubiquitination and degradation of p65. Notably, knockdown of MKRN2 by siRNA impaired PDLIM2-mediated polyubiquitination and degradation of p65, indicating requirement of MKRN2 for the activity of PDLIM2. These results delineate a novel role of MKRN2 in negatively regulating NF-κB -mediated inflammatory responses, cooperatively with PDLIM2.

3Pos296 リポ多糖刺激における炎症抑制タンパク質 PDLIM2 活性化の生細胞イメージング定量解析

Live cell imaging and quantitative analysis of anti-inflammatory protein PDLIM2 activation upon LPS stimulation

Shota Ichikawa[1], Yuma Ito[1], Takashi Tanaka[2], Makio Tokunaga[1], Kumiko Sakata-Sogawa[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*IMS-RCAI, RIKEN*)

NF-κB plays an important role in the initiation of immune responses. PDLIM2 is an nuclear E3 ubiquitin ligase, which terminates NF-κB activation in the nucleus to prevent excessive inflammatory responses. However, detailed mechanism of PDLIM2 activation is still elusive. Aiming to clarify the activation mechanism of PDLIM2 by LPS stimulation, we established an observation system using 3T3 cells, which expresses TLR4 constitutively. We confirmed the nuclear translocation of PDLIM2 together with NF-κB upon LPS stimulation. We also analyzed the effect of mutations in PDLIM2 on the localization and the function.

3Pos297 mRNA の一分子追跡によるストレス顆粒形成初期機構の調査

Investigating initiation mechanism of stress granule formation by tracking single mRNA particles

Masamichi Imaseki[1], Ko Sugawara[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm Sci., Univ. of Tokyo*, [2]*JST, PRESTO*)

mRNA is an essential gene product in cells and undergoes various regulations such as stress granule (SG) formation. It is suggested that during SG formation, polysomal mRNAs first form small granules, then aggregate to be large granules. However, the molecular mechanism of initiating granule formation is unknown. In this research, we directly observed GAPDH mRNA in cos7 cell by using single particle tracking (SPT). Analysis of SPT results showed that the fraction ratio of confined diffusing particles decreased and the immobile fraction increased in 10 minutes after arsenite-stress induction. The change in diffusion constants of particles were also observed. These results indicate that the molecular dynamics of mRNA is highly variable during initiation of SG formation.

3Pos298 高速近接場光学顕微鏡の開発

Development of high-speed near-field optical microscopy

Takayuki Umakoshi[1], Shingo Fukuda[2], Takayuki Uchihashi[1,2,3], Toshio Ando[1,2,3] ([1]*Bio-AFM FRC, Kanazawa Univ.*, [2]*Coll. Sci. & Eng., Kanazawa Univ.*, [3]*CREST-JST*)

High-speed near-field optical microscopy is promising for high-rate capture of single molecule fluorescence images at super-resolution. Its combination with high-speed AFM will expand the usefulness of both microscopy techniques in biological studies. To establish such a system, we developed near-field optical microscopy and combined it with our tip-scan high-speed AFM system. First, near-field light was effectively generated at an AFM tip by tightly focused laser, and near-field fluorescence signal was efficiently detected through confocal configuration. Second, we fabricated a metallic AFM tip to generate stronger near-field light, allowing faster imaging. We will describe details of these techniques and show near-field fluorescence imaging achieved.

3Pos299 転写伸長制御に関わるタンパク質動態のイメージング定量解析

Quantitative image analysis of dynamics of promoter-proximal pausing related proteins

Shinnosuke Kunimi, Yuma Ito, Yuki Yamaguchi, Kumiko Sakata-Sogawa, Makio Tokunaga (*Sch. Life Sci. Tech., Tokyo Inst. Tech.*)

Promoter-proximal pausing of RNA polymerase II is one of the important rate-limiting steps in transcription. In the establishment of pausing, elongation factors DSIF and NELF are recruited to Pol II soon after transcription initiation, where initiation factor TFIIE is thought to play a regulatory role in the recruitment of DSIF. The function of these factors are regulated by phosphorylation with cyclin-dependent kinases. Aiming to elucidate the dynamic propaties of the factors under cyclin-dependent kinase control, we performed fluorescence recovery after photobleaching (FRAP) analysis. The results indicated that dynamics of DSIF was notably affected with the inhibitors for Cdk7 or P-TEFb. We will discuss the effects of the inhibitors on NELF and TFIIE dynamics.

3Pos300 大気圧電子顕微鏡を用いた分泌腺組織の水中観察

Secretory glands imaged in aqueous solution by atmospheric scanning electron microscopy (ASEM)

Toshiko Yamazawa[1], Naotoshi Nakamura[2], Mari Sato[3], Chikara Sato[3] ([1]*Dept. Mol. Physiol., Jikei Univ. Sch. Med.*, [2]*Dept. Statistical Genetics, Kyoto Univ.*, [3]*Biomed. Res. Inst., AIST*)

Mammals have 3 pairs of major salivary glands, the parotid glans, submandibular glands and sublingual glands. The disfunction of these secretory organs immediately leads to various diseases, such as Mikulicz disease or Sjögren's syndrome, by poorly understood mechanisms. We report the direct observation of tissue in aqueous solution by atmospheric scanning electron microscopy (ASEM). The secretion granules in salivary glands were fixed, stained with phosphotungstic acid and visualized in aqueous liquid using the ASEM. The results indicate that the ASEM has a potential that can be a new method of detecting an abnormality of the secretory granules.

3Pos301 ベイズ推定を用いた透過型電子顕微鏡画像の CTF 補正の自動化

Development of automated CTF correction of transmission electron microscopic images using the Bayesian estimation

Koji Hisanaga, Takuo Yasunaga (*Kyushu Institute of Technology Graduate School of Computer Science and System Engineering*)

Cryo-electron microscopy has been used for observation of protein in a cell or in vitro at the molecular level. However, it is necessary to take defocused electron micrographs for the purpose of improvement of contrast, which results in blurring of the images. The blurring function is represented as CTF (contrast transfer function) in the Fourier space. The correction of CTF is important to obtain 3D reconstructed maps at the higher resolutions. Various programs for automated correction of CTF have been developed, but the decisive method is absent in every condition, especially at low S/N images. Here, we proposed a new tool to estimate defocusing values by using Bayesian estimation and report the evaluation of the tool.

3Pos302 様々な生物種の温度測定に利用でき且つ速い温度変化を測定可能な蛍光性温度プローブタンパク質

Genetically encoded ratiometric fluorescent thermometer with wide temperature range and rapid response

Masahiro Nakano[1], Yoshiyuki Arai[1], Ippei Kotera[2], Kohki Okabe[3,4], Yasuhiro Kamei[5], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ.*, [2]*RIES, Hokkaido Univ.*, [3]*Grad. Sch. Pharma., Univ. Tokyo*, [4]*JST, PRESTO*, [5]*NIBB*)

Temperature is a fundamental physical parameter responsible for biological events. Although conventional thermometers have found several important phenomena such as heat generation in mitochondria, development of a thermometer with a sensitivity to wide temperature range and rapid response is still desired to quantify temperature change in not only homeotherms but also poikilotherms from cellular level to *in vivo* level. Here, we report development of a genetically encoded fluorescent thermometer with the above desired property and demonstrate quantitative monitoring fast temperature changes in mitochondria as well as the thermometry in a living fish embryo.

3Pos303 新規誘電率顕微鏡(SE-ADM)による生きたそのままの細胞の液中ナノスケール観察
Nanoscale observation of intact living cells in a medium with low radiation damage using scanning electron-assisted dielectric microscopy

Tomoko Okada, **Toshihiko Ogura** (*Advanced Industrial Science and Technology (AIST), Biomedical Research Institute*)

High-resolution imaging of biological specimens by scanning electron microscopy (SEM) provides valuable insight to biological structures. However, SEM observations of biological specimens under high vacuum conditions require specific sample preparations involving glutaraldehyde fixation, negative staining and cryo-techniques. We recently developed a new technology of SE-ADM system based on FE-SEM(1). Here, we demonstrate the first nanoscale observation of living cultured mammalian cells using SE-ADM system with a newly developed culture dish holder(2). As a result, intact cells and organelles are clearly visible in high-contrast and high-resolution images in medium.
(1) T. Ogura, BBRC, 459, pp. 521-528 (2015), (2) T. Okada & T. Ogura, Sci. Rep., 6, 29169(2016).

3Pos304 **Improvement of photostability of fluorescent dyes by using lanthanide ions**

Takuma Imoto[1], Shin Mizukami[2], Kazuya Kikuchi[1,3] ([1]*Grad. Sch. Eng, Osaka Univ.*, [2]*IMRAM, Tohoku Univ.*, [3]*IFReC*)

Fluorescent dyes are widely used in advanced optical microscopic applications such as single-molecule imaging and super-resolution imaging with high-intensity laser illumination. In these experiments, photobleaching of fluorophores is one of the critical problems to be overcome. Generally, chemical reactions that start from the long-lifetime T1 state are thought to be the main photobleaching processes. Therefore, to shorten lifetime of the T1 state can be a rational strategy to enhance the photostability of various fluorescent dyes, and a compound to shorten the T1 state lifetime is called TSQ (Triplet State Quencher). We hypothesized that lanthanide ions work as TSQs, and found some lanthanide ions in solution enhanced the photostability of fluorophores.

3Pos305 **Optical measurement of diffusion and pH in nanopores of protein crystals**

Kazuo Mori, Bernd Kuhn (*OIST*)

Protein crystals are nanoporous materials due to gaps between the stacked proteins. We use molecular probes and imaging techniques to study key characteristics of the water/salt/buffer-filled nanopores. Lysozyme and thaumatin crystals were labeled with the fluorescent dye Rhodamine or the pH-sensitive dye SNARF-1 by diffusion. We use Fluorescence Recovery After Photobleaching (FRAP) to study dye diffusion in the pores. For measuring pH changes in the nanopores we used ratiometric fluorescence imaging. We can continuously observe pH changes in the nanopores after a pH change of the bath solution. In steady state we find a lower pH in the crystal nanopores compared to the bath which might be caused by an accumulation of protons in the pores due to hindered diffusion.

3Pos306 アポフェリチンを用いた複合(MRI 造影・発光)希土類ナノ粒子の作製
Synthesis of rare earth hybrid nanoparticles in the apoferritin cavity

Keita Kontani, Hideyuki Yoshimura (*Meiji University*)

To get homogeneous nanoparticles (NPs), protein (apoferritin) cavity has been utilized as a reaction chamber. Protein shells served as a template to restrain particle growth and as a coating to prevent coagulation between NPs. Apoferritin is known to mineralize several metal ions in the cavity. Here we report synthesis of hybrid rare earth NPs, Eu and Gd, in the apoferritin cavity. Using this hybrid NPs, Gd is expected to work as a contrast enhancement reagent for MRI and Eu is expected to work as a photoluminescent marker. The Eu/Gd NPs exhibit emission peak at 614nm and the longitudinal relaxation time (T1) of 1H (10MHz, 12uM Gd, room temperature) is 1.7s which is shorter than pure H2O solution 3.1s.

3Pos307 標的遺伝子高感度検出に向けた自己組織化単分子膜修飾金ナノ粒子の分散安定化
Optimized modification of SAMs for suppression of non-specific binding gold nanoparticles for High-sensitivity Target Genetic Assay

Keiko Esashika, Takaha Mizuguchi, Toshiharu Saiki (*Sci. Rech., Keio Univ.*)

Gold nanoparticles (AuNPs) are the most promising because they exhibit a large scattering cross-section in the visible region, as well as chemical stability, nontoxicity, and biocompatibility. Homogeneous DNA assays using AuNPs require the reduction of non-specific binding between AuNPs to improve sensitivity in detecting the target molecule. We employed alkanethiol self-assembled monolayers (SAMs) for modification of the AuNP surface to attain both good dispersability and high hybridization efficiency. The alkanethiol SAM increases the repulsive interaction between AuNPs, reducing non-specific binding and promoting the extension of surface-immobilized ssDNA into the solvent, thus enhancing the hybridization process.

3Pos308 血中循環腫瘍細胞を測定するためのサイズ分画機能を備えた画像認識型セルソーターの開発
Development of Size Classifying Imaging Cell Sorter for Identifying of Circulating Tumor Cells

Moe Iwamura[1], Masao Odaka[2], Kenji Matsuura[2], Akihiro Hattori[2], Hideyuki Terazono[2], Kenji Yasuda[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*)

Imaging cell sorter has been developed and improved to acquire circulating tumor cells (CTCs) in blood with recognizing their morphometric characteristics such as shapes and sizes of cells and cell clusters. The imaging cell sorter is equipped with a high speed CCD camera, a real-time image analyzer and a microfluidic chip having a feature of size classification exploiting microfabrication technology. By using this system, we effectively identified the difference of sizes of particles and clusters combining microfluidic size separation and image recognition. According to those size classification features, we found that the analysis time was improved significantly and practically applicable to identify CTCs in blood.

3Pos309 血中循環腫瘍細胞を無染色で識別するためのオン・チップ高機能画像認識型細胞分取装置の開発

Development of Functional On-Chip Imaging Cell Sorter for Identification of Non-Labeled Circulating Tumor Cells

Masao Odaka[1], Akihiro Hattori[1], Kenji Matsuura[1], Hideyuki Terazono[1], Moe Iwamura[2], Kenji Yasuda[2] ([1]*WASEDA Biosci. Res. Inst. Singapore (WABIOS), Waseda Univ.*, [2]*Dept. Physics, Waseda Univ.*)

We have developed an on-chip imaging cell sorter to identify target cells based on their morphological characteristics, such as size, shape of cells and nucleus, to overcome the limitation of conventional antibody-based cell recognition. As a practical application, we applied our imaging cell sorter for recognition of circulating tumor cells (CTCs). However, we needed to improve the efficiency of sorting process because the population of CTCs in blood is extremely small in number. By the microfabricated functional flow channel and sample preparation, we improved more than a hundred-fold efficiency to identify and separate clustered cells. The results indicate that this functional imaging cell sorter can be utilized enough for practical non-labeled CTC detections.

3Pos310 ゲル媒質中で反応拡散系によるパターン形成を行う DNA 論理ゲート

DNA logic gate performs Reaction-Diffusion Pattern formation in gel medium

Keita Abe[1], Ibuki Kawamata[2], Shin-ichiro M. Nomura[2], Satoshi Murata[2] ([1]*Dpt. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Eng., Tohoku Univ.*)

Pattern formation plays a key role in the process of development in biology. The process is totally bottom-up self-organization where all the chemical reactions take place spontaneously. Mimicking such pattern formation by an artificial system enables us to fabricate a dynamic molecular assembly capable of creating a defined structure automatically. In our research, we aim to form patterns with reaction-diffusion system implemented as an designed chemical reaction network of DNA. Here, we succeeded in generating bisector lines from two or more sources which are given as an initial pattern. The lines appear using DNA AND gates anchored in a field of hydrogel, in which short DNA strands diffuse from the sources to the AND gates.

3Pos311 電極埋め込み型ナノポアの AC ゲート電位による DNA の挙動制御

DNA motion and translocation controlled by nanopore with embedded gate electrode

Naoto Sakashita, Yuta Kato, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

Silicon based nanometer scale pore, nanopore has paid attention since its ability to detect single molecules and their structures mainly by measuring ionic current profiles. Various groups have demonstrate the sensing capability by nanopore toward the next generation of DNA sequencing. One of the crucial issues of nanopore analysis is the extremely fast speed of the DNA passing through a pore. Here, we present a novel method to slow the DNA speed by various alternating waveforms on an embedded gate electrode in nanopores without conventional bias voltages across the pore membrane. By taking advantage of the asymmetric geometry of a SiN membrane supported by a Si substrate, ratchet-like DNA motions can be demonstrated by optimized sawtooth waves.

3Pos312 *In vitro* selection of novel peptide agonists for human somatostatin receptor subtype-2 using water-in-oil microdroplets

Takashi Sakurai[1], Ryo Iizuka[1], Yasuyuki Nakamura[2], Jun Ishii[3], Akihiko Kondo[2], Ayaka Iguchi[4], Dong H. Yoon[4], Tetsushi Sekiguchi[5], Shuichi Shoji[4], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Grad. Sch. of Eng., Kobe Univ.*, [3]*Org. of Adv. Sci. and Technol., Kobe Univ.*, [4]*Dept. of Nanosci. and Nanoeng., Waseda Univ.*, [5]*Res. Org. for Nano & Life Innov., Waseda Univ.*)

G protein-coupled receptors (GPCRs), including somatostatin receptors (SSTRs), are one of the most important drug-targets in the pharmaceutical industry. Thus, intense efforts have been devoted to screening new GPCR ligands for potential drug candidates from chemical libraries. However, for many GPCRs, such attempts have failed. Then, we have developed an *in vitro* selection method to obtain peptide agonists for GPCRs using water-in-oil microdroplets. Using this method, we successfully identified novel peptide agonists for human SSTR subtype-2 (SSTR2) from a library of partially randomized SST sequences. This result demonstrates that our method will be a powerful platform for identifying novel peptide agonists for human GPCRs.

3Pos313 ナノポア計測の周波数解析による複数種類の microRNA のパターン認識

Pattern Recognition for MicroRNA Expressions by using Fourier Analysis on Nanopore Sensing

Akihiro Tamotsu[1], Moe Hiratani[2], Masayuki Ohara[2], Ryuji Kawano[3] ([1]*Tokyo Univ. of Agri. and Tech. Dept. Biotech. and Life Sci.*, [2]*Tokyo Univ. of Agri. and Tech. Dept. Biotech. and Life Sci.*, [3]*Tokyo Univ. of Agri. and Tech. Dept. Biotech. and Life Sci.*)

MicroRNA (miRNA) has been attracted as an ultra-early diagnostic marker for every cancers. In bladder cancer, it has been known that two types of miRNA were overexpressed. Although it is possible to detect miRNA with the current data of nanopore measurement, the discrimination of miRNA sequence is still in challenge because the each current signal obtained by nanopore measurement is similar. In this study, we applied Fourier analysis to the current data to recognize the miRNA pattern in bladder cancer. As the result, the frequency ingredients of signals were different between one type of miRNA and two types of miRNA. This difference suggests that the nanopore measurement can be applied to the diagnosis of bladder cancer by integrating with Fourier analysis.

3Pos314 DNA を用いたナノ粒子 3D プリンタの実現

Nanoparticle 3D Printing by DNA Bonding

Yuki Sakamoto, Shoichi Toyabe (*Grad. Sch. Eng. Applied Physics, Univ. Tohoku*)

Assembly of dynamic and chemically-functional nanostructures is demanded for realizing nanomachines. However, conventional lithographic processes have a difficulty in constructing such structures. Here, we demonstrate a novel method to build a functional nanomachine by assembling particles of various properties. Our method is based on DNA strands attached to the particle surface. These DNA strands are designed to form hairpin at room temperature and unfold when heated. We simultaneously trapped and heated particles by optical tweezers with IR laser. When particles are brought close, complementary DNA strands on different particles are hybridized to connect particles. We demonstrate assembling a micro robot arm moving by magnet and a bacterium-driven swimming structure.

3Pos315 DNA ナノ構造の DNA ハイドロゲルへの繰り返し電子解離／会合

Repeatable electronic dissociation/association of DNA nanostructures on DNA hydrogels

Keitel Cervantes, Ibuki Kawamata, Shin-Ichiro Nomura, Satoshi Murata (*Tohoku University*)

Repeatable association/dissociation of double-stranded DNA is key for realizing DNA nanomachines. Fast electronic melting on surfaces is a promising method; however, factors such as pH and Joule heating may contribute to the dissociation. To establish pure electronic control, we need to exclude those factors. Here, we developed an experimental setup to electronically control the dissociation/association of DNA nanostructures to complementary-DNA hydrogels. The DNA gel inside a capillary was placed between Pt electrodes. A strong voltage drove the dissociation and diffusion of the nanostructure from the gel. Association was also possible by applying an inverse milder voltage. This repeatable mechanism may be used for reusable molecular devices and molecular computers.

3Pos316 作製済みリポソームへのマイクロピペットの穿刺

Microinjection into already made liposome

Shota Sato, Shin Yoshida, Tomoyuki Kaneko (*LaRC, FB, Hosei Univ.*)

Liposomes were impossible to add any substance to after liposomes were prepared.
We tried to put substance into liposome by microinjection with micropipette into liposome prepared by spontaneous transfer with inner solution mixed with gelatin.
We succeeded in putting fluorescent substance by microinjection into liposome held by another micropipette. We could observe fluorescent dye was in aqueous core of the liposome.
It was suggested that gelatin made possible micro injection by forming cortex supporting membrane of liposome.

3Pos317 Quantification of intersample differences in T cell populations

Ryo Yokota[1], Yuki Kaminaga[2], Tetsuya Kobayashi J.[1,2] ([1]*Institute of Industrial Science, the University of Tokyo*, [2]*School of Engineering, The University of Tokyo*)

Diversity of T cell receptors (TCR) determines the way how the T cell population responds to pathogens. Inferring and categorizing the diversities for T cell populations (repertoire) are crucial for evaluating immunological states of our body. Previous methods mainly used the count statistics of TCR sequences but discarded the direct information of the amino acid sequences of TCRs. In this study, we propose a new method that is based not on the count statistics but on the similarity in TCR sequences. As the results, we show almost the same sample-clustering structure as the one estimated by previous methods. Moreover, our method can also identify the specific sequences causing the sample difference, which is impossible with the previous approach.

3Pos318 多チャンネル局所化学刺激システムの開発

Development of the multi-channel local chemical stimulation system

Masaru Kojima[1], Tatsuoya Furusawa[1], Hajime Fukuoka[2], Yasushi Mae[1], Tatsuo Arai[1] ([1]*Grad. Sch. Eng. Sci., Osaka Univ.*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*)

For analyzing detailed and localized biological phenomenon, a local environmental control technique is important. For example, when analyzing the detailed and localized properties of single cells, this technique is desired. In here, we developed a multi-channel local environmental chemical stimulation system. By using micro pipettes, this system could control the local reagent concentration dynamically, freely and automatically. In this system, to reduce the diffusion of chemical solution, spout pipette and suction pipette were used. Multi-channel pipette was used for realizing spout of multi-solution. Furthermore, as an evaluation of this system, peeling off of certain area of confluent cells was demonstrated.

3Pos319 多電極電位計測システムを用いた心筋細胞に対するテルフェナジンの影響

Effect of Terfenadine to cardiomyocytes on multi electrode array system

Mitsuki Maruyama, Tomoyuki Kaneko (*LaCR, Hosei Univ.*)

Cardiotoxicity testing at present in vitro could not accurately detect the toxicity in clinical, e.g. anti-allergic drugs of Terfenadine.
Cardiotoxicity of Terfenadine was investigated by multi electrode array (MEA) system which is a simple and noninvasive technique to measure the extracellular field potential of excitable cells. We measured the extracellular potential of cardiomyocytes isolated from chick embryo (E13) by MEA system and analyzed inter spike interval (ISI), field potential duration (FPD) and Short Term Variability (STV).
Although FPD did not show the prolongation, STV showed 5 times higher than before applying drugs.
It was suggested that MEA system could be used for a new indication of the cardiotoxicity because STV was the strong tool of the arrhythmia.

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名字（Family Name）のアルファベット順にソートしています。すべて、オンラインで入力されたデータのまま、表示しています。

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Araki, Mitsugu (荒木 望嗣) 1Pos019
2Pos084
Arakida, Jin (荒木田 臣) 3Pos203
Aramaki, Shinji (荒牧 信二) 2SGA-04
Aramaki, Shinji (荒牧 慎二) 1Pos153
1Pos156
Arata, Toshiaki (荒田 敏明) 3Pos065
Arata, Toshiaki (荒田 敏昭) **3Pos092**
3Pos126
Arata, Yukinobu (荒田 幸信) **2Pos130**
Arif Md. Rashedul, Kabir (Arif Md. Rashedul Kabir) 1Pos289*
Ariga, Katsuhiko (有賀 克彦) 3SGA-04
Ariga, Takayuki (有賀 隆行) 1Pos159
2Pos159
Arikawa, Keisuke (有川 敬輔) **1Pos017**
Arisaka, Fumio (有坂 文雄) **1SGA-08**
2Pos061*
Arisaka, Katsushi (有坂 勝史) 1Pos228*
Ariyama, Hirotaka (有山 弘高) **2Pos291**
Ariyoshi, Mariko (有吉 眞理子) **3SCA-02**
Arizono, Misa (有薗 美沙) 2SBP-06
Asahina, Yuya (朝比奈 雄也) 2Pos054
Asai, Kiyoshi (浅井 潔) 3Pos099
Asano, Teizo (浅野 禎三) 2Pos231
Asano, Yuki (浅野 友紀) **2Pos057**
Asanuma, Hiroyuki (浅沼 浩之) 2Pos158
Ashida, Keita (芦田 慶太) 1Pos227*
1Pos230*
Ashida, Naoko (芦田 名伯子) **2Pos199**
Ashida, Yuto (蘆田 祐人) 2Pos142*
Ashikawa, Hiroki (芦川 裕樹) 1SGP-01
2Pos285
Atsuji, Shogo (厚地 省吾) **2Pos058***
Aurangazeb, Kabir (アウロンゴゼブ コビル) 2Pos026
Awazu, Akinori (粟津 暁紀) 1Pos173
2Pos110
2Pos265
3Pos269
Ayukawa, Shotaro (鮎川 翔太郎) **3SEA-01**
Azai, Chihiro (浅井 智広) 2Pos246
Azuma, Yusuke (東 裕介) **1Pos316**
Baba, Junpei (馬場 淳平) 1Pos085
Baba, Kentarou (馬場 健太郎) 3Pos032
Baba, Mihori (馬場 みほ里) 1SDP-02
2Pos145*
Baba, Tomoyuki (馬場 知之) **1Pos231***
Babe, Natsuki (馬部 菜月) 2Pos017
Baker, Bradley (Baker Bradley) **1SAA-02**
Bannai, Hiroko (坂内 博子) **2SBP-06**
Bausch, Andreas (Bausch Andreas) **1SDP-07**
Bavi, Navid (Bavi Navid) 1Pos215
Bekker, Gert-Jan (Bekker Gert-Jan) **1Pos019**
3Pos019
Bellve, Karl (Bellve Karl) 1SEA-04
Bhattacharyya, Bhaswati (Bhattacharyya Bhaswati) **1Pos279**
Bito, Haruhiko (尾藤 晴彦) 2SGP-03
Blocker, Ariel J (Blocker Ariel J) 1Pos006
Bowler, David R. (Bowler David R.) 3Pos105
3Pos106
Brandani, Giovanni (Brandani Giovanni) **2Pos107**
Brettel, Klaus (Brettel Klaus) 3Pos251
Brown, Jason (Brown Jason) 1SEA-04
Brown, Leonid S. (Brown Leonid S.) 2Pos255
Brzezinski, Peter (Brzezinski Peter) 1Pos089*
Bustamante, Gilbert (ブスタマンテ ギルバート) 2Pos118
Candelli, Andrea (Candelli Andrea) 2Pos112
Cao, Wei (曹 巍) 3Pos268
Carl H., Johnson (Carl H. Johnson) 1Pos287*
Cervantes, Keitel (Cervantes Keitel) **3Pos315**
Cervantes-Salguero, Keitel (セルバンテスーサルゲロ ケイテル) 3SGA-04
Chadani, Yuhei (茶谷 悠平) **2SDP-03**
Chaen, Shigeru (茶園 茂) **3Pos150**
Chan, Siu Kit (Chan Siu Kit) **1Pos001**
Chan, Siu Kit (陳 兆傑) 3Pos242
Chang, Jae-won (張 宰源) 1Pos303
2Pos050*
2Pos070*
Chang, Kaichun (張 凱鈞) 2Pos108
Chang, Mari (張 マリ) **1Pos038***
1Pos078*
Chang, Wen-Hsuan (Chang Wen-Hsuan) 1Pos114
Chantal, Barberot (シャンタル バルベロ) 2Pos031
Charoenwattanasatien, Ratana (Charoenwattanasatien Ratana) **1Pos023**
Chatake, Toshiyuki (茶竹 俊行) 1Pos119
2Pos078
3Pos077

Chatani, Eri (茶谷 絵理) **1SCP-04**, 1Pos010, 1Pos013*, 2Pos122
Che, Yong-Suk (蔡 栄淑) **3Pos188**
Chen, Eric H.-L. (Chen Eric H.-L.) 2Pos052*
Chen, Meiting (Chen Meiting) 3Pos147
Chen, Po-Ting (Chen Po-Ting) 2Pos052*
Chen, Rita P.-Y. (Chen Rita P.-Y.) 2Pos052*
Cheng, Cheng (成 せい) **3Pos239**
Cheng, Mao Ting (Cheng Mao Ting) **3Pos236**
Chi, Hung-Yuan (Chi Hung-Yuan) 1Pos114
Chi, Peter (冀 宏源) 1Pos117*
Chiam, Keng-Hwee (Keng-Hwee Chiam) 1Pos170
Chiba, Hitoshi (千葉 仁志) 3Pos056
Chiba, Mayuka (千葉 真優香) 1Pos305
Chiba, Shinobu (千葉 志信) 2SDP-03
Chiba, Tomofumi (千葉 智史) 3Pos260
Chikenji, George (千見寺 浄慈) 1Pos047, 1Pos273, **2Pos266**, 3Pos015, 3Pos018
Chirifu, Mami (池鯉鮒 麻美) 2Pos027
Chishima, Ryotaro (千島 亮太郎) 3Pos040
Cho, Kyunghak (Cho Kyunghak) **2Pos200**
Choi, Hyonjin (チェ ヒョンジン) 2Pos199
Choi, Hyunjin (Choi Hyunjin) **1Pos192**
Chowdhury, Srikanta (Chowdhury Srikanta) 2Pos253
Christie, John (Christie John) 3Pos248
Cook, Gregory M. (Cook Gregory M.) 3Pos137
Craft, Julie (Craft Julie) 1SEA-04
Craige, Branch (Craige Branch) 1SEA-04
Cross, Robert (Cross Robert) 2Pos073
Cryershinozuka, Kazuho (クライヤー篠塚 一帆) **1Pos271***
Danno, Keisuke (檀野 圭右) **1Pos169**
Das, Sumita (Das Sumita) **1Pos276***
Das, Sumita (ダス スミタ) 1Pos275
Date, Kohei (伊達 公平) 1Pos242
Davis, Tim (Davis Tim) 2Pos157
Dellarole, Mariano (デラロール マリアノ) 2Pos061*
Demura, Makoto (出村 誠) 1Pos195, 1Pos241, 1Pos242, 2Pos068, 3Pos240, 3Pos241
Demura, Mariko (出村 茉莉子) 1Pos024
Demura, Mikihide (出村 幹英) 3Pos258
Dezawa, Takuma (出澤 拓磨) **1Pos075**
Diez, Stefan (Diez Stefan) **2SBA-06**
Ding, Da-Qiao (丁 大橋) 2Pos264
Dohra, Hideo (道羅 英夫) 2Pos205
Dohra, Hideo (道羅、英夫、) 1Pos194, 1Pos198
Doi, Katumi (土居 克実) 1Pos066
Doi, Nobuhide (土居 信英) 1Pos082, 2Pos093, 2Pos204*
Doi, Satoko (土井 聡子) **2Pos253**
Doi, Tomoko (土井 知子) 1Pos002
Dokainish, Hisham (Dokainish Hisham) **3Pos025**
Doung Hyun, Yoon (Doung Hyun Yoon) 2Pos315*
Doura, Tomohiro (堂浦 智裕) 2Pos257*
Driessen, Rosalie (Driessen Rosalie) **2Pos112**
Drummond, Douglas (Drummond Douglas) **2Pos073**
Ebihara, Mika (蛯原 三華) **1Pos200**
Ebina, Teppei (蝦名 鉄平) 3Pos020
Ebisawa, Shinichi (蛯澤 伸一) 1Pos049
Ebisawa, Tatsuki (海老沢 樹) 3Pos087
Ebisuya, Miki (戎家 美紀) **3SEA-02**
Edamatsu, Masaki (枝松 正樹) **3Pos063**
Eiraku, Mototsugu (永楽 元次) 1Pos281
Ekimoto, Toru (浴本 亨) 3Pos136
Elstner, Marcus (Elstner Marcus) 2Pos042
Emoto, Hikaru (江本 光) 1Pos164
Emura, Makoto (江村 誠) 3Pos234
Endo, Aiko (遠藤 愛子) **3Pos135**
Endo, Masayuki (遠藤 政幸) **3SGA-01**, 1Pos203, 1Pos213, 2Pos315*
Endo, Satoshi (遠藤 智史) 2Pos026
Endo, Shigeru (猿渡 茂) 2Pos008, 3Pos049
Endo, Shigeru (遠藤 茂) 3Pos208
Endo, Takahide (遠藤 貴秀) **3Pos184**
Endo, Toshiya (遠藤 斗志也) **3SFA-01**, 3SFA-99
Enoki, Sawako (榎 佐和子) **2SCA-03**, 2SCA-99, 1Pos135
Enomoto, Akiko (榎本 暁子) **2Pos236**
Enomoto, Atsushi (榎本 篤) **1SAP-06**
Eric, Greene (Eric Greene) 3Pos163
Ernst, Oliver P (エルンスト オリバー) 2Pos227
Esashika, Keiko (江刺家 恵子) 2Pos314*, **3Pos307**
Etchuya, Kenji (越中谷 賢治) 2Pos024, 3Pos010
Ezaki, Soichiro (江崎 宗一郎) **1Pos039***
Ezaki, Yuji (江崎 裕児) 2Pos123*
Fangjia, Luo (Fangjia Luo) **2Pos077**
Farhana, Tamanna Ishrat (Farhana Tamanna Ishrat) **3Pos154**
Feig, Michael (Feig Michael) 2Pos060
Fischer, Niels (Fischer Niels) 3Pos088
Flechsig, Holger (Flechsig Holger) **1Pos072**
Forgarty, Kevin (Forgarty Kevin) 1SEA-04
Fuchigami, Sotaro (渕上 壮太郎) **3Pos050**
Fuda, Hirotoshi (布田 博敏) 3Pos056
Fuji, Ritsuko (藤井 律子) 3Pos255
Fuji, Shuji (藤井 修治) 2Pos134
Fujii, Hajime (藤井 哉) **2SGP-03**
Fujii, Masashi (藤井 雅史) 3Pos279, **3Pos284**
Fujii, Nao (藤井 奈緒) 2Pos313
Fujii, Noriko (藤井 紀子) 3Pos033
Fujii, Ritsuko (藤井 律子) 1Pos252*
Fujii, Soichiro (藤井 聡一郎) **2Pos294**
Fujii, Takashi (藤井 高志) 2SDP-05, 3Pos122
Fujii, Yuki (藤井 裕紀) **2Pos277***
Fujimori, Akira (藤森 亮) 3Pos262
Fujimori, Fumihiro (藤森 文啓) 2Pos206
Fujimori, Taihei (藤森 大平) 1SAP-04, **2Pos186**
Fujimoto, Akira (藤本 陽) 3Pos004
Fujimoto, Kazuya (藤本 和也) **2Pos156**
Fujimoto, Koichi (藤本 仰一) 1SGP-03, 3Pos285
Fujimoto, Shiori (藤本 栞理) **3Pos074**
Fujimoto, Tadashi (藤本 直) **3Pos204**
Fujinami, Tetsuo (藤波 哲郎) **3Pos113**
Fujioka, Yoichiro (藤岡 容一朗) 3SAA-03
Fujioka, Yuji (藤岡 祐次) **3Pos230**
Fujisawa, Takahiro (藤澤 貴宏) **1Pos168**
Fujisawa, Tetsuro (藤澤 哲郎) 1Pos012, **1Pos015**
Fujisawa, Tomotsumi (藤澤 知績) **3Pos244**
Fujishiro, Shin (藤城 新) **2Pos113**
Fujishiro, Shunsuke (藤代 峻輔) **1Pos268**
Fujita, Hideaki (藤田 英明) 3Pos200
Fujita, Katsumasa (藤田 克昌) **1SBA-04**
Fujita, Keisuke (藤田 恵介) 3Pos151, **3Pos165**
Fujiwara, Ikuko (藤原 郁子) **2Pos139**
Fujiwara, Kazuo (藤原 和夫) 1Pos049, 2Pos062
Fujiwara, Kei (藤原 慶) 1Pos082, 2Pos093, 2Pos204*
Fujiwara, Kentaro (藤原 健太郎) **3Pos255**
Fujiwara, Satoru (藤原 悟) 1Pos059, **3Pos077**, 3Pos123
Fujiwara, Takahiro K. (藤原 敬宏) 3Pos196, 3Pos293
Fujiwara, Toshimichi (藤原 敏道) 1Pos008*
Fujiyoshi, Ayako (藤吉 彩子) 2Pos120
Fujiyoshi, Satoru (藤芳 暁) **2SEP-03**
Fujiyoshi, Takafumi (藤吉 貴史) **2Pos123***
Fujiyoshi, Yoshinori (藤吉 好則) 1Pos002, 1Pos097, 3Pos088
Fukada, Harumi (深田 はるみ) 2SDP-05
Fukasawa, Yoshinori (深沢 嘉紀) 3Pos098
Fukuda, Asahi (福田 朝陽) **2Pos082**
Fukuda, Ikuo (福田 育夫) **2Pos086**, 2Pos270
Fukuda, Kohei (福田 昂平) 3Pos072
Fukuda, Shingo (福田 真悟) 3Pos298
Fukui, Kiyoshi (福井 清) 3Pos016
Fukumori, Yoshihiro (福森 義宏) 3SDA-04, 1Pos022, 1Pos306*, 2Pos289
Fukumura, Hiroshi (福村 裕史) 1Pos075
Fukumura, Takuma (福村 拓真) 3Pos176
Fukunaga, Koichi (福永 興壱) 1Pos318*
Fukunaga, Yuko (福永 優子) 2Pos175
Fukuoka, Hajime (福岡 創) 3Pos188, **3Pos193**, 3Pos318
Fukuoka, Mayuko (福岡 万佑子) 2Pos026
Fukushima, Ryosuke (福島 綾介) 1Pos295
Fukushima, Seiya (福島 誠也) 1Pos191, **3Pos197**
Funatsu, Takashi (船津 高志) 1SBP-04, 2Pos069, 2Pos116, 2Pos195*, 2Pos196, 2Pos273, 2Pos279, 2Pos284, 2Pos292, 2Pos294, 2Pos315*, 3Pos297, 3Pos312
Furuike, Yoshihiko (古池 美彦) 1Pos286, **2Pos003**, 3Pos013, 3Pos031
Furukawa, Arata (古川 新) **3SCA-06**
Furukawa, Shinya (古川 信也) 1Pos093
Furuno, Tadahide (古野 忠秀) **1Pos176**
Furusawa, Chikara (古澤 力) **1SGP-04**
Furusawa, Tatsuoya (古澤 達也) 3Pos318
Furuta, Akane (古田 茜) **1Pos141**
Furuta, Ken'ya (古田 健也) 2SBA-00, **2SBA-02**, 1Pos141, 1Pos143, 1Pos290*
Furuta, Tadaomi (古田 忠臣) 1Pos271*, 2Pos045
Furutani, Yuji (古谷 祐詞) **2SAA-01**, 2Pos227
Furuya, Akane (古谷 茜) 2Pos244
Fushiki, Shinji (伏木 信次) 2Pos175
Futaki, Shiroh (二木 史朗) 3Pos213
Futamata, Hiroyuki (二又 裕之) 1Pos197
Fuzino, Yasuhiro (藤野 泰寛) 1Pos066
Gao, Yi Qin (Gao Yi Qin) 2Pos105
Garcia-Trejo, Jose J. (García-Trejo José J.) 3Pos140
Genda, Makoto (源田 真) **1Pos130***
Gengyo-Ando, Keiko (安藤 恵子) 1SAA-05
Gerle, Christoph (げーれ くりすとふ) **3Pos088**
Getzoff, Elizabeth D (Getzoff Elizabeth D) 2Pos101
Ghatak, Chiranjib (ガターク チランジブ) **1Pos196**
Gi, Senka (魏 川華) **1Pos132***
Gohara, Mizuki (郷原 瑞樹) 3Pos143
Gomibuchi, Yuki (五味渕 由貴) **3Pos121**
Gonda, Kohsuke (権田 幸祐) 1Pos105
Gong, Jian Ping (龔 剣萍) 2Pos282*
Goto, Akane (後藤 朱音) **1Pos308**, 3Pos291, 3Pos292
Goto, Hitoshi (後藤 仁志) 1Pos205
Goto, Masaki (後藤 優樹) 1Pos209

3Pos208
Goto, Takashi (後藤 孝) 2Pos286
Goto, Yuji (後藤 佑児) 2Pos050*
Goto, Yuji (後藤 祐児) 1Pos063*
2Pos058*
2Pos064*
2Pos065
3Pos026
3Pos059
Gotoh, Mizuho (後藤 みずほ) **1Pos229**
Gotoh, Naomasa (後藤 直正) 3Pos072
Greimel, Peter (Greimel Peter) 2Pos206
Grill, Stephan (Grill Stephan) 3Pos198
Gu, Chan (Gu Chan) 2Pos105
Guentert, Peter (Guentert Peter) 2SFP-05
Haga, Hisashi (Haga Hisashi) 2Pos187
Haga, Hisashi (芳賀 永) **1SAP-03**
Haga, Kei (芳賀 慧) 3Pos004
Haga, Yukari (芳賀 ゆかり) 1Pos097
Hakuno, Fumihiko (伯野 史彦) 1Pos166*
Hamada, Daizo (濱田 大三) 2SDP-00
2Pos050*
Hamada, Hiroshi (濱田 博司) 2Pos176
2Pos177
2Pos179
Hamada, Tsutomu (濱田 勉) 1Pos190*
Hamaguchi, Takashi (濱口 貴史) 2Pos076*
Hamaguchi, Tasuku (浜口 祐) 2Pos164
2Pos166*
Hamao, Kozue (濱生 こずえ) 1Pos183
Hamasaki, Yuuta (濱崎 雄太) 3Pos289
Hambly, Brett D. (Hambly Brett D.) 3Pos117
Han, Sung-Sik (Han Sung-Sik) 2SCP-05
Han, Yong-Woon (韓 龍雲) **2Pos099**
Hanashima, Akira (花島 章) 3Pos124
Hanawa, Yutaka (塙 優) 3Pos254
Handa, Yusuke (反田 祐介) **2Pos002**
Hanson, Benjamin (Hanson Benjamin) 1Pos142*
Happo, Naohisa (八方 直久) 1SEP-04
Hara, Kanako (原 伽奈子) 2Pos237
Harada, Akira (原田 明) 3Pos286
Harada, Ryuhei (原田 隆平) 1Pos027
2Pos036
2Pos060
2Pos270
3Pos102
Harada, Yoshie (原田 慶恵) 1Pos317
2Pos099
2Pos165
2Pos315*
Harata, Masahiko (原田 昌彦) 1Pos310
2Pos300
Hariu, Taro (針生 太郎) **3Pos181**
Harris, Sarah (Harris Sarah) 1Pos142*
Haruyama, Takamitsu (春山 隆充) **2Pos040**
Hasan, Moynul (ハーサン モイヌル) **2Pos209**
Hasegawa, Kazuya (長谷川 和也) 1SEP-02
Hasegawa, Masashi (長谷川 将司) 1Pos256
Hasegawa, Sena (長谷川 世納) 1Pos296
Hasegawa, Shin (長谷川 慎) **1Pos137***
Hasegawa, Taisuke (長谷川 太祐) **3SBA-02**
2Pos254
Hasegawa, Takeshi (長谷川 毅) **2Pos129***
Hasegawa, Yoshitaka (長谷川 賢卓) 1Pos235
Hasegawa, Yusuke (長谷川 雄将) 1Pos197
Hasemi, Takatoshi (長谷見 崇俊) **3Pos241**
Hashiguchi, Nobuto (橋口 惟斗) 1Pos042
Hashimoto, Eri (橋本 江里) **1Pos195**
Hashimoto, Hideki (橋本 秀樹) 1Pos252*
Hashimoto, Kanako (橋本 佳奈子) **2Pos242**
Hashimoto, Kazuhito (橋本 和仁) 1Pos102*
Hashimoto, Ken-ichi (橋本 賢一) 1Pos215
Hashimoto, Mami (橋本 麻美) **3Pos086**
Hashimoto, Masaki (橋本 真宜) 1Pos224
Hashimoto, Naoki (橋本 直記) 2Pos254
Hashimoto, Shota (Hashimoto Shota) 1Pos276*
Hashimoto, Shota (橋本 翔多) **1Pos275**
Hashimoto, Takashi (橋本 貴志) **2Pos305**
Hashimoto, Yu (橋本 優) 1Pos106
2Pos147
Hashimoto, Yuichi (橋本 優一) **2Pos240***
Hata, Hiroaki (畑 宏明) **2Pos151**
Hatada, Yuji (秦田 勇二) 2Pos273
Hatakeyama, Tetsuhiro S. (畠山 哲央) **2SGP-05**
1Pos288
Hatanaka, Yusuke (畑中 悠佑) 1Pos235
Hatori, Kuniyuki (羽鳥 晋由) 1Pos149
2Pos225*
3Pos256
Hattori, Akihiro (服部 明弘) 2Pos287
2Pos318
3Pos130
3Pos229
3Pos308
3Pos309
Hattori, Motoyuki (服部 素之) 1Pos095*
Hauer, Florian (Hauer Florian) 3Pos088
Hawkins, Taviare L. (Hawkins Taviare L.) 3Pos156
Haya, Kazumi (羽矢 和未) **1Pos057***
Hayakawa, Masayuki (早川 雅之) **1Pos292***
Hayakawa, Satoshi (早川 智士) **2Pos055***
Hayakawa, Yuki (早川 悠貴) **3Pos153**
Hayami, Tomonori (速水 智教) **3Pos052**
Hayashi, Fumio (林 史夫) 3Pos173
Hayashi, Fumio (林 文夫) **1SFP-06**
2Pos201*
2Pos218
3Pos209
Hayashi, Hideyuki (林 秀行) 1Pos027
Hayashi, Humio (林 文夫) 2Pos214*
Hayashi, Kouichi (林 好一) 1SEP-04
Hayashi, Kumiko (林 久美子) 1Pos137*
Hayashi, Masahito (林 真人) **1Pos182**
1Pos188*
2Pos226*
Hayashi, Naoki (林 直樹) 3Pos072
Hayashi, Shigehiko (林 重彦) 3SBA-02
1Pos099
1Pos245*
2Pos022*
2Pos254
3Pos239
Hayashi, Tatsuya (林 達也) **1Pos277**
Hayashi, Tetsuya (林 哲也) 2Pos135
Hayashi, Tomohiko (林 智彦) **2Pos125**
Hayashi, Yugo (林 有吾) **3Pos028**
Hayashi, Yuki (林 勇樹) 2Pos053
Hayashi, Yuuki (林 勇樹) 1Pos032*
1Pos035
1Pos038*
1Pos054*
1Pos058
1Pos078*
2Pos025
2Pos028*
2Pos089
2Pos092*
2Pos164
Hayashino, Yuji (林野 裕至) **3Pos110**
Hayashizaki, Takahiro (林崎 貴大) 2Pos141
Hazawa, Masaharu (羽澤 勝治) 1Pos306*
Heyning, Olivier (Heyning Olivier) 2Pos112
Hiata, Fumio (平田 文男) 2Pos129*
Hibino, Kayo (日比野 佳代) **2SGP-02**
1Pos315*
2Pos111
2Pos278
Hibino, Masahiro (日比野 政裕) 1Pos206
3Pos261
Hieu Chi, Dam (Hieu Chi Dam) 2Pos128
Higashi, Masahiro (東 雅大) 2Pos251*
Higo, Akiyoshi (肥後 明佳) 1Pos079
Higo, Junichi (肥後 順一) 3Pos052
Higo, Tomboy (肥後 智也) **1Pos153**
Higo, Tomoya (肥後 智也) 2SGA-04
Higuchi, Hideo (樋口 秀男) 1Pos105
1Pos134
1Pos145*
2Pos142*
2Pos155
2Pos192*
3Pos132
3Pos195
Higuchi, Kae (樋口 佳恵) 3Pos064
Higuchi, Masayuki (樋口 真之) **3Pos139**
Higuchi, Yoshiki (樋口 芳樹) 1Pos067
3Pos080
Hijikata, Atsushi (土方 敦史) 1Pos267
Hijikata, Atsushi (土方 敦司) **1Pos269**
2Pos152
3Pos143
Hikichi, Minori (引地 穣) 3Pos284
Hikima, Takaaki (引間 孝明) 1Pos012
3Pos031
3Pos243
Hikiri, Simon (肥喜里 志門) **2Pos085***
Hikita, Masahide (引田 理英) 3Pos085
Hinde, Elizabeth (Hinde Elizabeth) **2SCP-04**
Hioki, Mayu (日置 茉優) **1Pos234**
Hippler, Michael (Hippler Michael) 1Pos023
Hippler, Michael (Michael Hippler) 2Pos244
Hirai, Anna (平位 杏奈) **1Pos018**
Hirai, Mitsuhiro (平井 光博) 1Pos014
1Pos053
3Pos043
3Pos207
Hiraiwa, Tetsuya (平岩 徹也) **1Pos283**
Hirakawa, Hirokazu (平川 博一) 3Pos262
Hirakawa, Rika (平川 利佳) **1Pos151**
Hirakawa, Takeshi (平川 健) 1Pos320
Hiramatsu, Hirotsugu (平松 弘嗣) 2Pos193*
Hiramoto-Yamaki, Nao (平本-山木 菜央) 3Pos196
Hirano, Minako (平野 美奈子) **1Pos218**
1Pos223
Hirano, Tatsuya (平野 達也) 3Pos276
Hirano, Yoshinori (平野 秀典) 3Pos084
Hiraoka, Wakako (平岡 和佳子) 3Pos263
3Pos264
3Pos265
Hiraoka, Yasushi (平岡 泰) 2Pos264
Hirasada, Kenta (平佐田 健太) 3Pos038
Hirashima, Naohide (平嶋 尚英) 1Pos176
Hirata, Fumio (平田 文男) 3Pos044
3Pos110
Hirata, Hiroaki (平田 宏聡) **1Pos170**
Hirata, Keisuke (平田 啓介) 2Pos027
Hirata, Kunio (Hirata Kunio) 2Pos077
Hirata, Kunio (平田 邦生) 1SEP-02
Hirata, Nanami (平田 菜々美) **3Pos231**
Hiratani, Moe (平谷 萌恵) **2Pos311***
3Pos313
Hirate, Taiki (平手 太樹) **3Pos129**
Hiratsuka, Yuichi (平塚 祐一) 2Pos316*
Hirokawa, Nobutaka (廣川 信隆) 2SGP-06
1Pos133
Hirokawa, Takatsugu (広川 貴次) 2Pos084
Hiromasa, Yasuaki (廣政 恭明) 1Pos066
Hironaka, Ken-ichi (廣中 謙一) 3Pos285
Hirono, Masafumi (廣野 雅文) **2Pos172**
Hirono, Moritoshi (廣野 守俊) 3Pos227
Hirosawa, Koichiro M. (廣澤 幸一朗) **3Pos196**
Hirose, Keiko (広瀬 恵子) 1Pos155
2Pos181
3Pos153
Hirose, Osamu (広瀬 修) 2Pos133
3Pos228
Hirose, Takuji (Hirose Takuji) 1Pos081
Hiroshima, Akinori (広島 明宣) 1Pos026
Hiroshima, Michio (廣島 通夫) 1Pos104
1Pos107
2Pos299
3Pos202
Hirota, Shun (廣田 俊) 1Pos067
1Pos094*
2Pos051
3Pos080
Hirotsune, Shinji (広常 真治) 2Pos175
3Pos159
Hirusaki, Kotoe (蛭崎 琴絵) 2Pos200
Hisabori, Toru (久堀 徹) 1Pos079
Hisanaga, Koji (久永 浩司) **3Pos301**
Hisatomi, Osamu (久冨 修) 1Pos260
1Pos261

1Pos262
2Pos230
3Pos249
Hitomi, Kenichi (人見 研一) 3Pos248
Hiyama, Miyabi (樋山 みやび) **1Pos249**
Hizukuri, Yohei (檜作 洋平) **3Pos169**
Hoerning, Marcel (ホールニング マルセル) **3Pos286**
Hojo, Hironobu (北條 裕信) 2Pos054
Hojo, Masaru (北條 賢) 1Pos014
Hojo, Yasushi (北條 泰嗣) 1Pos235
Holliger, Philipp (Holliger Philipp) 2SBA-03
Holme, M N (HOLME M N) 2Pos219
Holstein, Thomas W. (Holstein Thomas W.) 3Pos287
Homma, Michio (本間 道夫) 1Pos004
1Pos008*
2Pos152
3Pos143
3Pos167
3Pos168
3Pos170
3Pos171
Homma, Noriyasu (本間 経康) 3Pos235
Honda, Gen (本田 玄) **1Pos171**
2Pos186
Honda, Hajime (本多 元) 1Pos184
2Pos141
Honda, Naoya (本田 尚也) 2Pos254
Honda, Natsuki (本田 菜月) **1Pos159**
Honda, Satoshi (本田 諭志) 2Pos198
Honda, Shinya (本田 真也) 2Pos275
2Pos306
3Pos054
3Pos057
Honda, Takaaki (本多 孝明) **2Pos284**
Hongo, Kenta (本郷 研太) 2Pos090
Honma, Mareki (本間 希樹) **2SFP-03**
Hori, Yuichiro (堀 雄一郎) **1SCP-02**
Hori, Yuki (堀 裕基) 2Pos090
Hori, Yutaka (堀 豊) 3Pos115
Horibe, Kazuya (堀部 和也) **3Pos285**
Horigane, Makoto (堀金 慎) 2Pos286
Horii, Keitaro (堀井 啓太郎) **1Pos074**
Horii, Yusuke (堀井 湧介) **2Pos193***
Horikoshi, Miki (堀越 未希) **1Pos210**
Horimoto, Katsuhisa (堀本 勝久) 3Pos281
Horiuchi, Kaoru (堀内 かほる) **2Pos179**
Horton, Paul (ホートン ポール) 3Pos098
Hoshida, Masayuki (星田 政行) 1Pos184
Hoshikawa, Miho (星川 美穂) 2Pos039*
Hoshina, Hitoshi (星名 仁志) 1Pos131
Hoshino, Daisuke (星野 太佑) 2Pos167*
2Pos189*
Hoshino, Masaru (星野 大) 2SDP-05
Hoshino, Yumi (星野 由美) **2SBP-04**
Hosogi, Naoki (細木 直樹) 3Pos007
Hosokawa, Chie (細川 千絵) 3Pos230
Hosokawa, Tomohisa (細川 智永) **2SGP-04**
Hosoya, Hiroshi (細谷 浩史) 1Pos154
Hotta, Kohji (堀田 耕司) 1Pos227*
1Pos230*
2Pos194*
2Pos277*
Hou, Feng (侯 峰) 2Pos001*
Hsu, WeiLin (許 維麟) **2Pos045**
Hugues, Chate (Hugues Chaté) 1Pos285
Hui, Shu-Ping (惠 淑萍) 3Pos056
Hukuda, Wakao (福田 青郎) 3Pos118
Hukushima, Koji (福島 孝治) **2SFP-02**
Hull, J.Joe (Hull J. Joe) 3Pos087
Hummer, Gerhard (Hummer Gerhard) 1Pos127
Husimi, Yuzuru (伏見 譲) **1SDA-04**
1SDA-99
Hussmann, Frauke (Hussmann Frauke) 2Pos073
Hyeon, Changbong (Hyeon Changbong) 3Pos037
Hyodo-Otsu, Tomoko (兵土(大津) 知子) 2Pos147
I M. Mahaputra, Wijaya (I M. Mahaputra Wijaya)
1Pos125
Ichihashi, Norikazu (市橋 伯一) 2Pos258*
Ichikawa, Shota (市川 翔太) 2Pos300
3Pos296
Ichimura, Taro (市村 垂生) 1Pos133
Ichinose, Junnya (一ノ瀬 純也) 1Pos223
Ichinose, Takako (一ノ瀬 孝子) 1Pos180
Ichinose, Yu (市瀬 悠) 1Pos184
Ichioka, Takayuki (市岡 隆幸) 1Pos024
3Pos067
Ide, Soichiro (井出 宗一郎) **3Pos020**
Ide, Toru (井出 徹) 1Pos191
1Pos218
1Pos223
Ifuku, Kentaro (伊福 健太郎) 1Pos252*
Iguchi, Ayaka (井口 彩香) 2Pos315*
3Pos312
Iguchi, Kota (井口 公太) 1Pos169
Ihara, Kunio (井原 邦夫) 1Pos001
Ihara, Sigeo (井原 茂男) 1SFA-05
Iida, Mamiko (飯田 真美子) **2Pos067***
Iida, Naoya (飯田 直也) **2Pos149**
Iida, Shinji (飯田 慎仁) **1Pos142***
Iida, Takahiro (飯田 高広) 3Pos030
Iida, Tatsuya (飯田 龍也) **3Pos142**
Iida, Yoshihiro (飯田 禎弘) **2Pos091**
Iijima, Toshio (飯島 敏夫) 3Pos234
Iino, Ryota (飯野 亮太) 2SCA-03
2Pos148
2Pos156
3Pos001
3Pos138
3Pos142
3Pos162
3Pos166
Iino, Yuichi (飯野 雄一) 3SAA-02
3Pos228
Iinuma, Ryosuke (飯沼 良介) 2Pos315*
Iitsuka, Yoshiaki (飯塚 芳朗) 2Pos307
Iizuka, Ryo (飯塚 怜) 2Pos069
2Pos116
2Pos273
2Pos294
2Pos315*
3Pos312
Ikawa, Keisuke (井川 敬介) 1SAP-02
Ikebe, Jinzen (池部 仁善) **2Pos115**
2Pos270
Ikeda, Kazuho (池田 一穂) 1Pos137*
Ikeda, Masami (池田 修己) 2Pos272
Ikeda, Shiro (池田 思朗) 2SFP-05
Ikeda, Takanari (池田 隆成) 1Pos176
Ikeda, Yasuo (池田 康夫) 3Pos211
Ikegami, Koji (池上 浩司) 2Pos174*
3Pos160
Ikegami, Takahisa (池上 貴久) 2Pos244
Ikegawa, Masaya (池川 雅哉) 1Pos169
Ikegaya, Atsushi (池谷 淳) 3Pos269
Ikeguchi, Masamichi (池口 雅道) 1SGA-05
1Pos049
2Pos062
Ikeguchi, Mitsunori (池口 満徳) 1Pos009
1Pos319
2Pos085*
2Pos146*
Ikemizu, Shinji (池水 信二) 2Pos027
Ikemoto, Mitsushi J. (池本 光志) 1Pos186
Iketaki, Kentaro (池滝 憲太郎) 1Pos166*
Ikeya, Teppei (池谷 鉄兵) **2SFP-05**
Ikezaki, Keigo (池崎 圭吾) 1Pos231*
1Pos303
2Pos050*
2Pos070*
3Pos026
3Pos070
3Pos151
Ikura, Teikichi (伊倉 貞吉) 1Pos040
2Pos074
Imada, Katsumi (今田 勝己) 3Pos186
Imada, Katsumi (今田 勝巳) 1Pos004
1Pos008*
2Pos002
2Pos005
2Pos072
2Pos177
3Pos167
3Pos170
3Pos171
3Pos172
3Pos174
3Pos175
3Pos290
Imada, Yasuhiro (今田 康博) 1SEP-01
3Pos083
Imai, Hiroo (今井 啓雄) 1Pos234
2Pos232*
2Pos236
Imai, Hiroshi (今井 洋) 2Pos165
3Pos155
Imai, Kenichiro (今井 賢一郎) **3Pos098**
Imai, Mizue (今井 瑞依) 1Pos093
Imai, Ryosuke (今井 亮輔) **1Pos315***
2Pos263
Imai, Taichi (今井 太一) 2Pos277*
Imai, Taiga (今井 大河) **3Pos149**
Imaizumi, Miku (今泉 美玖) 1Pos298
Imamoto, Yasushi (今元 泰) 2Pos230
2Pos235
Imamura, Hiromi (今村 博臣) **3SAA-04**
2Pos285
3Pos294
Imamura, Hiroshi (今村 比呂志) 1Pos013*
2Pos029
2Pos275
3Pos054
3Pos057
Imamura, Motonori (今村 元紀) 2Pos143*
3Pos138
Imanishi, Michie (今西 三千絵) **2Pos248**
Imao, Asato (今尾 麻人) 3Pos040
Imaseki, Masamichi (今関 眞倫) **3Pos297**
Imashimizu, Masahiko (今清水 正彦) **2Pos106**
Imoto, Daisuke (井元 大輔) 1SAP-04
1Pos284
2Pos183
Imoto, Takuma (井元 琢真) **3Pos304**
Inaba, Kazuo (稲葉 一男) 2Pos001*
2Pos169
2Pos181
Inaba, Satomi (稲葉 理美) 1Pos033*
1Pos036*
1Pos040
Inaba, Satoshi (稲葉 敏) 3Pos167
Inaba, Takehiko (稲葉 岳彦) 2Pos206
3Pos211
Inagaki, Naoyuki (稲垣 直之) 3Pos032
Inagaki, Shigenori (稲垣 成矩) **2Pos298***
Inagawa, Marina (稲川 麻織奈) 2Pos289
Inariyama, Keiichi (稲荷山 慶一) **3Pos029**
Inobe, Tomonao (伊野部 智由) **2SDP-06**
2SDP-99
Inoh, Yoshikazu (伊納 義和) 1Pos176
Inomata, Kohsuke (猪股 晃介) 3Pos064
Inomata, Naoki (猪股 直生) 2SDA-01
Inoshima, Yasuo (猪島 康雄) 1Pos037
Inoue, Daisuke (Inoue Daisuke) 3Pos154
3Pos157
Inoue, Daisuke (井上 大介) 1Pos289*
Inoue, Haruki (井上 晴幾) 2Pos167*
2Pos189*
Inoue, Keiichi (井上 圭一) **2SAA-03**
1Pos238*
1Pos239
1Pos240*
1Pos244
1Pos263*
1Pos264
1Pos265
2Pos239
2Pos240*
2Pos252
2Pos256
3Pos119
Inoue, Keiya (井上 慶也) **3Pos010**
Inoue, Kodai (井上 広大) 1Pos024
Inoue, Masanao (井上 真尚) **1Pos093**
Inoue, Masatoshi (井上 昌俊) 2SGP-03

Inoue, Masaya (井上 雅也) 3Pos203
Inoue, Masayo (井上 雅世) **3Pos281**
Inoue, Rintaro (井上 倫太郎) **3Pos033**
Inoue, Saki (井上 紗希) **3Pos245**
Inoue, Seiya (井上 誠也) 1Pos067
Inoue, Takanari (井上 尊生) **1SBP-03**
Inoue, Yosuke (井上 耀介) **2Pos004**
Inoue, Yumi (井上 由美) 3Pos144
3Pos172
Inui, Takashi (乾 隆) 2Pos058*
Inui, Yayoi (乾 弥生) 1Pos320
Irie, Katsumasa (入江 克雅) **1Pos097**
Irle, Stephan (イレ ステファン) 1Pos030
Isaka, Yuta (井阪 悠太) **3Pos136**
Isei, Yuhta (伊世井 湧太) **3Pos252**
3Pos254
Iseki, Mineo (伊関 峰生) 1Pos248*
Ishibashi, Ken-ichi (石橋 健一) 1Pos024
Ishida, Atsuhiko (石田 敦彦) 1Pos065
Ishida, Hisashi (石田 恒) **1Pos070**
Ishida, Kentaro (石田 研太郎) 1Pos158
1Pos162
2Pos307
3Pos311
Ishida, Motohiko (石田 元彦) 2SFA-04
2Pos183
3Pos194
Ishida, Norihito (石田 規人) 1Pos039*
Ishiguro, Naotaka (石黒 直隆) 1Pos037
Ishiguro, Ryo (石黒 亮) 1Pos012
1Pos015
Ishihara, Ayumi (石原 あゆみ) **2SGA-04**
1Pos153
Ishihara, Shuji (石原 秀至) 1SAP-04
1Pos284
2Pos183
Ishihara, Takeshi (石原 健) 3SAA-02
3Pos228
Ishii, Jun (石井 純) 3Pos312
Ishii, Kentaro (石井 健太郎) 3Pos030
Ishii, Kunihiko (Ishii Kunihiko) 1Pos126
Ishii, Kunihiko (石井 邦彦) **3SBA-01**
2Pos051
Ishii, Noriyuki (石井 則行) **1Pos186**
Ishii, Shoko (石井 頌子) 1Pos091
Ishii, Shuya (石井 秀弥) **3Pos131**
Ishijima, Akihiko (石島 秋彦) 3Pos188
3Pos193
Ishikawa, Daisuke (石川 大輔) **3SGA-05**
1Pos203
1Pos213
Ishikawa, Haruto (石川 春人) **3SBA-04**
Ishikawa, Isamu (石川 勇) 3Pos007
Ishikawa, Tomoko (石川 智子) 3Pos250
Ishikita, Hiroshi (石北 央) 2Pos042
2Pos245*
Ishikuro, Daiki (石黒 大輝) **2Pos049**
Ishimori, Koichiro (石森 浩一郎) 1Pos086
1Pos089*
1Pos093
1Pos195
3Pos059
Ishioka, Daisuke (石岡 大輔) 2Pos220
Ishitani, Ryuichiro (石谷 隆一郎) 1Pos095*
Ishitsuka, Reiko (石塚 玲子) 3Pos211
Ishiura, Masahiro (石浦 正寛) 3Pos030
Ishiwata, Daiki (石渡 大貴) **3Pos162**
Ishiwata, Shin'ichi (石渡 信一) 2SBA-05
1Pos147
1Pos183
1Pos229
2Pos170
3Pos131
Ishizuka, Toru (石塚 徹) 2Pos041
Ishizuka-Katsura, Yoshiko (石塚 芳子) 3Pos141
Ishrat Fahana, Tamanna (Ishrat Fahana Tamanna)
1Pos152*
Islam, MM. (Islam MM.) **3Pos027**
Islam, Md Zahidul (イスラム、 エムディ ザヒドゥル、)
1Pos194
Islam, Md. Sirajul (Islam Md. Sirajul) 1Pos289*
Islam, Md. Zahidul (イスラム エムディ ザヒドゥル)
2Pos207
2Pos209
Islam, Mohammad M. (イスラム モハマド･M) 2Pos061*
Islam, Mohammad Monirul (Islam Mohammmad Monirul)
1Pos062
Isman, Kurniawan (Isman Kurniawan) 2Pos021
Isogai, Yasuhiro (磯貝 泰弘) **2Pos029**
Isogaki, Tsubasa (磯垣 翼) **2Pos300**
Isojima, Hiroshi (磯島 広) 1Pos135
Isomura, Kazushi (磯村 和志) **1Pos116**
3Pos069
Isozaki, Naoto (磯崎 直人) **3Pos156**
Isumi, Miyu (伊角 実優) **1Pos004**
Itabashi, Takeshi (板橋 岳志) 2Pos170
Ito, Akira (井藤 彰) **2SDA-04**
Ito, Hidekatsu (伊東 嗣功) 3Pos231
3Pos233
Ito, Hideki (伊藤 秀城) 2SCP-06
Ito, Koreaki (伊藤 維昭) 2SDP-03
Ito, Manami (伊藤 真奈美) **1Pos291***
Ito, Masaharu (伊藤 正晴) 2Pos238
Ito, Masahiro (伊藤 政博) 3SDA-02
2Pos153*
Ito, Masaki (伊藤 正樹) **1Pos289***
Ito, Nobutoshi (伊藤 暢聡) 1Pos022
1Pos040
2Pos074
Ito, Seiichiro (伊藤 誠一朗) 3Pos205
Ito, Shingo (伊東 真吾) **1Pos030**
Ito, Shota (伊藤 奨太) 1Pos238*
1Pos239
1Pos240*
1Pos244
1Pos248*
1Pos263*
1Pos264
2Pos122
2Pos256
Ito, Shuichi (伊東 修一) 1Pos299
Ito, Takuya (伊藤 拓哉) 1Pos296
Ito, Yoshihiro (Ito Yoshihiro) 1Pos081
Ito, Yuji (伊藤 優志) 1Pos113*
Ito, Yuko (伊藤 裕子) 2Pos146*
Ito, Yuma (伊藤 由馬) **1Pos310**
1Pos312
1Pos313
2Pos295
2Pos300
3Pos295
3Pos296
3Pos299
Ito, Yutaka (伊藤 隆) 2SFP-05
1Pos011
Itoh, Hideki (伊藤 秀城) 1Pos229
Itoh, Hiroyasu (伊藤 博康) 1Pos106
2Pos147
Itoh, Kazumi (伊藤 一実) 3Pos206
Itoh, Kyoko (伊東 恭子) 2Pos175
Itoh, Satoru (伊藤 暁) **1Pos025**
Itoh, Shigeru (伊藤 繁) 2Pos246
Itoh, Yuji (伊藤 優志) **1Pos110***
Itoh, Yukihiro (伊藤 幸裕) 1Pos069
Itoh-Shinzawa, Kyoko (Itoh-Shinzawa Kyoko) 2Pos077
Itoh-Shinzawa, Kyoko (伊藤-新澤 恭子) 1Pos088
Itohara, Shigeyoshi (糸原 重美) 3Pos234
Itoh－Shinzawa, Kyoko (伊藤－新澤 恭子) 1Pos086
Itou, Tatsuro (伊藤 竜朗) **3Pos179**
Itou, Yoshiko (伊藤 喜子) 1Pos153
Iwabe, Naoyuki (岩部 直之) 2Pos233
Iwabuki, Fumie (岩蕗 文恵) 1Pos005
3Pos034
Iwadate, Yoshiaki (岩楯 好昭) 1Pos160
1Pos163
1Pos164
Iwai, Ryosuke (岩井 良祐) **1Pos056**
1Pos061
2Pos268
Iwai, Shigenori (岩井 成憲) 1Pos068
2Pos101
3Pos250
3Pos251
Iwai, Sosuke (岩井 草介) 3Pos150
Iwaki, Masayo (岩城 雅代) **1Pos216**
1Pos234
2Pos044
2Pos250*
Iwaki, Mitsuhiro (岩城 光宏) 1Pos138
3Pos151
3Pos165
Iwaki, Shigehiro (岩木 薫大) 3SCA-06
Iwamoto, Hiroyuki (岩本 裕之) 1Pos133
2Pos138
2Pos171
2Pos173
3Pos155
Iwamoto, Masayuki (岩本 真幸) 1Pos098
1Pos225
Iwamoto, Shigeto (岩本 成人) 2Pos076*
Iwamura, Moe (岩村 萌絵) **3Pos308**
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Kojima, Masaru (小嶋 勝) **3Pos318**
Kojima, Risa (小島 理沙) 2Pos246
Kojima, Sakiko (小嶋 佐妃子) 2Pos201*
2Pos218
Kojima, Seiji (小嶋 誠司) **3SDA-01**
1Pos004
1Pos008*
3Pos143
3Pos167
3Pos168
3Pos170
3Pos171
Kojima, Toshinori (小島 紀徳) 1Pos296
Kokabu, Yuichi (小甲 裕一) 3Pos136
Kokaji, Yuko (小鍛治 優子) 3Pos256
Komada, Hisako (駒田 久子) 3Pos279
Komatsu, Hideyuki (小松 英幸) **3Pos107**
Komatsu, Masaaki (小松 雅明) 2SDP-05
Komatsuzaki, Naoya (小松崎 直也) **2Pos078**
Komatsuzaki, Tamiki (Komatsuzaki Tamiki) 2Pos187
Komatsuzaki, Tamiki (小松崎 民樹) **1SBA-08**
Komatsuzaki, Yoshimasa (小松崎 良将) **3Pos232**
Komazawa, Kosuke (駒澤 光佑) 1Pos215
Komori, Tomotaka (小森 智貴) 1Pos116
3Pos069
Kon, Takahide (昆 隆英) 1Pos142*
Konagaya, Akihiko (小長谷 明彦) 1Pos289*
Kondo, Akihiko (近藤 明彦) 3Pos312
Kondo, Hiroko (近藤 寛子) 2Pos015
Kondo, Hiroko X. (近藤 寛子) **2Pos044**
3Pos083
Kondo, Kazunori (近藤 和典) 1Pos136
Kondo, Shingo (近藤 慎吾) 2Pos229*
Kondo, Shinnosuke (近藤 真之介) **3Pos264**
Kondo, Shota (近藤 翔太) **3Pos168**
Kondo, Takao (近藤 孝男) 1Pos286
2Pos003
Kondo, Yosuke (近藤 洋介) **3Pos270**
Kondo, Yuichi (近藤 雄一) **2Pos155**
Konno, Hiroki (紺野 宏記) 2Pos040
Konno, Masae (今野 雅恵) 2Pos239
2Pos252
3Pos119
Konno, Takashi (今野 卓) **3Pos021**
Kono, Fumiaki (河野 史明) **1Pos059**
3Pos077
Kono, Hidetoshi (河野 秀俊) 1Pos119
2Pos115
Kontani, Keita (紺谷 啓太) **3Pos306**
Koshiba, Takumi (小柴 琢己) **3SAA-01**
Koshiyama, Kenichiro (越山 顕一朗) 2Pos210
Kosodo, Yoichi (小曽戸 陽一) 2Pos131
Kosuda, Satoshi (Kosuda Satoshi) 2Pos016
Kosugi, Aya (小杉 綾) 3Pos047
Kosugi, Takahiro (小杉 貴洋) **2SBA-01**
1Pos077
2Pos088
2Pos096
Kotani, Norito (小谷 則遠) 1Pos307
Kotera, Hidetoshi (小寺 秀俊) 2Pos156
3Pos156
Kotera, Ippei (小寺 一平) 2SDA-03
3Pos302
Kouyama, Tsutomu (神山 勉) 1Pos001
3Pos242
Koyama, Tsubasa (小山 翼) 3Pos150
Koyanagi, Mitsumasa (小柳 光正) **2SAA-06**
2Pos234
Koyasako, Kotaro (小屋迫 光太郎) **3Pos159**
Kozai, Toshiya (小財 稔矢) 2Pos049
3Pos014
Kozaki, Yuko (小崎 裕子) **1Pos265**
Kozawa, Yuichi (小澤 祐市) 2SAP-04
Kozlowski, Pawel M. (Kozlowski Pawel M.) 3Pos082
Kozuka, Jun (小塚 淳) 2Pos299
Krah, Alexander (Krah Alexander) **3Pos037**
Kubo, Hiroki (久保 宏樹) 1Pos001
3Pos242
Kubo, Mikako (久保 美香子) 1Pos024
Kubo, Minoru (久保 稔) 1SEP-02
1SEP-06
1Pos091
3Pos075
Kubo, Shintaroh (久保 進太郎) **1Pos144**
Kubo, Shoutaro (久保 翔太郎) **3Pos061**
Kubo, Tai (久保 泰) 1Pos231*
3Pos070
Kubo, Tomohiro (久保 智広) **1SEA-04**
Kubota, Hiroaki (久保田 寛顕) **1Pos147**
Kubota, Rinko (窪田 倫子) 2Pos310*
Kuboyama, Masahiro (久保山 正浩) 2Pos304
2Pos308
Kuddus, Md. Ruhul (Kuddus Md. Ruhul) **2Pos068**
Kudo, Hisashi (工藤 恒) 1Pos054*
1Pos078*
2Pos025
2Pos028*
Kudo, Seishi (工藤 成史) 3Pos146
3Pos173
3Pos178
3Pos182
Kudo, Seisi (工藤 成史) 3Pos181
Kudoh, Suguru (工藤 卓) 3Pos231
3Pos233
Kudoh, Suguru N. (工藤 卓) 3Pos230
Kugimiya, Akimitsu (釘宮 章光) 2Pos015
Kuhara, Atsushi (久原 篤) 1Pos228*
Kuhn, Bernd (Kuhn Bernd) **1SAA-06**
3Pos305
Kumagai, Mai (熊谷 真衣) **2Pos101**
Kumagai, Yusuke (熊谷 祐介) 2Pos049
Kumagai, Yutaro (熊谷 雄太郎) 2SFA-03
Kumakura, Seigo (熊倉 聖悟) 3Pos261
Kunida, Katsuyuki (国田 勝行) 2Pos167*
2Pos189*
Kunihara, Tomoko (椢原 朋子) **1Pos054***
1Pos058
Kunimi, Shinnosuke (國見 慎之介) **3Pos299**
Kunitomo, Hirofumi (国友 博文) 3SAA-02
Kurahashi, Yuhi (倉橋 雄飛) **1Pos125**
Kuraoka, Yusho (倉岡 遊正) **1Pos201**
Kurihara, Hiroki (栗原 裕基) 1Pos277
2Pos133
Kurihara, Kensuke (栗原 顕輔) 1Pos189
2Pos224
3Pos206
Kurihara, Yukiko (栗原 由紀子) 2Pos133
Kurisaki, Ikuo (栗崎 以久男) **2Pos031**
Kurisu, Genji (栗栖 源嗣) 1Pos003*
1Pos023
1Pos142*
2Pos246
2Pos249
Kurita, Jun-ichi (栗田 順一) 1Pos009
Kurobe, Atsushi (黒部 淳史) 2Pos062
Kuroda, Daisuke (黒田 大祐) 2Pos015
Kuroda, Kenichi (黒田 賢一) 1Pos204
Kuroda, Shinya (黒田 真也) 2Pos167*
2Pos189*
3Pos279
3Pos284
Kuroda, Y (Kuroda Y.) 3Pos027
Kuroda, Yutaka (Kuroda Yutaka) 2Pos016
Kuroda, Yutaka (黒田 裕) **1SGA-06**
1Pos062
2Pos061*
3Pos020
Kuroi, Kunisato (黒井 邦巧) **2Pos227**
Kurokawa, Chikako (黒川 知加子) 1Pos213
Kurokawa, Kazuo (黒川 量雄) 1Pos314
Kuruma, Yutetsu (車 兪澈) 2Pos222
Kurumizaka, Hitoshi (胡桃坂 仁志) **1SFA-01**
Kusaka, Katsuhiro (日下 勝弘) 2Pos078
Kushiro, Keiichiro (久代 京一郎) 2Pos052*
Kusumi, Akihiro (楠見 明弘) 1SFP-01
3Pos190
3Pos196
3Pos293
Kuwabara, Naoyuki (桑原 直之) 3Pos092
Kuwajima, Kunihiro (桑島 邦博) **1Pos050**
Kuwata, Kazuo (桑田 一夫) 2Pos026
Kuwayama, Hidekazu (桑山 秀一) 2Pos135
Kuzoo, Shouhei (葛生 祥平) 3Pos187
Kuzuya, Akinori (Kuzuya Akinori) 3Pos157
Kuzuya, Akinori (葛谷 明紀) 2Pos158
Kyoda, Koji (京田 耕司) 3Pos280
Lane, E. Birgitte (Lane E. Birgitte) 2SCP-06
Le, Minh N. T. (Le Minh N. T.) 2Pos134
Lechtreck, Karl (Lechtreck Karl) 1SEA-04
Lee, Jong-Bong (Lee Jong-Bong) **2SCA-04**
Lee, Jooyoung (李 柱榮) 1Pos050
Lee, Nam Ki (Lee Nam Ki) **2SCA-06**
Lee, Seohyun (Lee Seohyun) **1Pos105**
Lee, Young-Ho (Lee Young-Ho) 3Pos059
Lee, Young-Ho (李 映昊) 2Pos058*
Levadnyy, Victor (Levadnyy Victor) 2Pos208
Levadnyy, Victor (レバツニー ビクター) 2Pos209
Leverenz, Ryan (Leverenz Ryan) 3Pos244
Li, Chen (李 辰) **2Pos079**
Li, Chun-Biu (Li Chun-Biu) 2Pos187
Li, Hung-Wen (Li Hung-Wen) 1Pos114
Li, Hung-Wen (李 弘文) 1Pos117*
Li, Xing (李 興) **1Pos014**
Lian, Tengfei (Lian Tengfei) 2Pos105
Lim, Chwee Teck (Chwee Teck Lim) 1Pos170
Lim, Wei Ming (林 偉銘) **1Pos312**
2Pos295
Limviphuvadh, Vachiranee (Limviphuvadh Vachiranee) 1Pos268
Lin, Ching Yuan (Lin Ching Yuan) **3Pos288**
Lin, Sheng-Yao (Lin Sheng-Yao) **1Pos114**
Lin, Yuxi (Lin Yuxi) **3Pos059**
Liu, Cuihua (劉 翠華) 3Pos262
Lo, Chien Jung (Lo Chien Jung) 3Pos236
3Pos288
Lo, Chien-Jung (Lo Chien-Jung) **3Pos147**
Lo, Yu-Hua (Lo Yu-Hua) 3Pos087
Lu, Chih-Hao (盧 致豪) **1Pos117***
Lu, S.-W. (呂 淑文) 2Pos241
Lu, Yaxin (Lu Yaxin) 3Pos117
Lucas, Robert (Lucas Robert) 2Pos234
Lukatsky, David (David Lukatsky) 2Pos106
Lukowiak, Ken (Lukowiak Ken) 3Pos232
Lwin Aye, Seaim (Seaim Lwin Aye) **2Pos093**
Ma, Yue (馬 越) **1Pos122**
Mabuchi, Issei (馬渕 一誠) 1Pos154
Machiyama, Hiroaki (町山 裕亮) **3Pos200**
Machizuki, Atsushi (望月 敦史) 3Pos276
Madigan, M. T. (Madigan M. T.) 2Pos241
Mae, Yasushi (前 泰志) 3Pos318
Maeda, Kayo (前田 佳代) 2Pos140
Maeda, Ryo (前田 亮) **3Pos091**
Maeda, Shintaro (前田 晋太朗) 3Pos085
Maeda, Yuichiro (前田 雄一郎) 3Pos125
Maekawa, Shohei (前川 昌平) 2Pos218
Maeno, Akihiro (前野 覚大) **2Pos056**
Maeshima, Kazuhiro (前島 一博) 1Pos315*
2Pos111
2Pos263
Mahmud, Md. Nuruddin (Mahmud Md. Nuruddin) **1Pos037**
Makabe, Koki (真壁 幸樹) **2Pos090**
Maki, Koichiro (牧 功一郎) **2Pos047**
Maki, Kosuke (槇 亙介) 1Pos001
1Pos052
3Pos030
Maki, Kousuke (槇 亙介) 3Pos242
Maki-Yonekura, Saori (眞木 さおり) **1Pos005**
3Pos034
Makino, Asami (牧野 麻美) 2Pos206
3Pos211
Makino, Yoshiteru (槇野 義輝) **3Pos247**
Makino, Yoshiteru (槇野 義輝) 1Pos237

Mamun, Abdullah Al (Mamun Abdullah Al) 3Pos082
Manavalan, Balachandran (マナバラン バラチャンドラン) 1Pos050
Martin, Douglas (Martin Douglas) 2Pos073
Martinac, Boris (Martinac Boris) 1Pos215
Maruta, Shinsaku (丸田 晋策) 1Pos136
1Pos139
2Pos304
2Pos305
2Pos308
Maruyama, Kouichi (丸山 耕一) 3Pos262
Maruyama, Mitsuki (丸山 充貴) **3Pos319**
Maruyama, Shintaro (丸山 慎太郎) **2Pos143***
3Pos138
3Pos141
Maruyama, Tatsuro (丸山 達朗) 1Pos095*
Maruyama, Yohei (丸山 洋平) **2Pos157**
Masahide, Terazima (寺嶋 正秀) 3Pos012
Masaike, Tomoko (政池 知子) 2Pos144
2Pos174*
3Pos139
3Pos160
Masaki, Noritaka (正木 紀隆) **1Pos304**
Mase, Yoko (間瀬 瑤子) 3SCA-04
Mashima, Tsukasa (真嶋 司) 2Pos067*
Masubuchi, Takeya (増渕 岳也) **2Pos315***
Masuda, Ryosuke (益田 稜介) **1Pos138**
Masuda, Shinji (増田 真二) **2SAA-05**
Masuda, Yuji (Masuda Yuji) 3Pos009
Masuhara, Kaori (増原 香織) **2Pos304**
2Pos308
Masumoto, Hiroshi (増本 博) 2Pos286
Masuyama, Yota (増山 陽太) **3Pos015**
3Pos018
Matsubara, Hitomi (松原 瞳) **1Pos193**
Matsubara, Ken (松原 賢) 1Pos303
Matsubayasi, Nobuyuki (松林 伸幸) 3Pos045
Matsuda, Hiroki (松田 洋樹) 2Pos078
Matsuda, Keisuke (松田 佳祐) 3Pos095
Matsuda, Kyohei (松田 恭平) **1Pos148**
Matsuda, Mariko (松田 麻理子) 3SCA-02
Matsuda, Naoki (松田 直樹) **2Pos167***
Matsuda, Noriyuki (松田 憲之) **3SFA-06**
Matsuda, Takahiro (松田 昂大) 2Pos282*
Matsuda, Tomoki (松田 知己) 1Pos309
2Pos298*
3Pos175
Matsukawa, Tadashi (松川 忠司) 3Pos185
Matsuki, Hitoshi (松木 均) 1Pos209
3Pos208
Matsumoto, Atsushi (松本 淳) 1Pos070
3Pos005
Matsumoto, Daiki (松本 大輝) **1Pos313**
Matsumoto, Shogo (松本 正吾) 3Pos087
Matsumoto, Takashi (松本 崇) 1Pos023
Matsumoto, Tomoharu (松本 友治) 2Pos140
Matsumoto, Yudai (松本 雄大) **2Pos072**
Matsumura, Hiroyoshi (松村 浩由) 3Pos234
Matsumura, Yoshitaka (松村 義隆) **1Pos024**
3Pos067
Matsunaga, Naoya (松永 直也) **3Pos263**
Matsunaga, Sachihiro (松永 幸大) 1Pos320
Matsuno, Asuka (松野 明日香) **2Pos007***
Matsuo, Atsushi (松尾 篤史) **1Pos305**
Matsuo, Tatsuhito (松尾 龍人) 1Pos059
3Pos077
3Pos123
Matsuo, Yuta (松尾 雄大) **2Pos213***
Matsuoka, Koji (松岡 浩司) 1Pos034
Matsuoka, Rei (松岡 礼) 1Pos005
3Pos034
Matsuoka, Satomi (松岡 里実) 1Pos191
3Pos197
Matsushima, Kouji (松島 綱治) 1Pos039*
Matsushita, Katsuyoshi (松下 勝義) **2Pos184**
3Pos285
Matsushita, Masaki (松下 雅季) **1Pos108**
Matsushita, Takashi (松下 卓史) 2Pos269
Matsushita, Yufuku (松下 祐福) **2Pos050***
3Pos026
Matsushita, Yuhuku (松下 祐福) 1Pos303
3Pos070
Matsutani, Yuki (松谷 優樹) 2Pos230
Matsuura, Azuma (松浦 東) 2Pos083
3Pos101
Matsuura, Kenji (松浦 賢志) 2Pos287
2Pos318
3Pos130
3Pos229
3Pos308
3Pos309
Matsuzaki, Katsumi (松崎 勝巳) 3Pos090
Matsuzaki, Kohei (松崎 興平) **1Pos135**
Matsuzaki, Mari (松崎 茉莉) 2Pos248
Matsuzaki, Mizuki (松崎 瑞季) **2Pos140**
Matsuzaki, Yuri (松崎 由理) **2SEA-04**
Matubayasi, Nobuyuki (松林 伸幸) 3Pos108
Matushita, Yuhuku (松下 祐福) 2Pos070*
Maurer-Stroh, Sebastian (Maurer-Stroh Sebastian) 1Pos268
Mayanagi, Kouta (眞柳 浩太) 3SCA-01
Mayanagi, Kouta (真柳 浩太) 1Pos156
McMillan, Duncan G G (McMillan Duncan G G) 3Pos140
McMillan, Duncan G. G. (McMillan Duncan G. G.) **3Pos137**
Mehta, Shalin (Mehta Shalin) 1SAA-03
Memtily, Nassirhadjy (Memtily Nassirhadjy) 3Pos002
Mendoza-Hoffmann, Francisco (Mendoza-Hoffmann Francisco) 3Pos140
Mieda, Kaoru (三枝 馨) **3Pos082**
3Pos085
Mikami, Nagisa (三上 渚) 2Pos144
2Pos146*
Miki, Ayako (三木 彩子) **1Pos033***
Miki, Kunio (三木 邦夫) 1Pos022
Miki, Masao (三木 正雄) 3Pos126
Miki, Motohiro (三木 元博) 3Pos004
Mikoshiba, Katsuhiko (御子柴 克彦) 2SBP-06
Mikoshiba, Naoki (御子柴 直紀) 1Pos083*
2Pos095*
Mikuni, Shintaro (三國 新太郎) 2Pos100
2Pos114*
Minagawa, Noriko (Minagawa Noriko) 1Pos081
Minagawa, Yoshihiro (皆川 慶嘉) 2SCA-03
1Pos084
2Pos148
3Pos142
Minami, Kosuke (南 皓輔) 3SGA-04
Minami, Shintaro (南 慎太朗) **1Pos273**
2Pos266
3Pos053
Minamino, Tohru (Minamino Tohru) 3Pos148
Minamino, Tohru (南野 徹) **3SDA-03**
2Pos072
3Pos035
3Pos144
3Pos172
3Pos174
3Pos176
3Pos180
Minamino, Tooru (南野 徹) 3SCA-06
Minato, Seiya (港 聖也) 2Pos307
Minato, Shotaro (湊 翔太郎) 2Pos286
Mino, Hiroyuki (三野 広幸) 1Pos254
1Pos256
2Pos249
3Pos249
Minobe, Reina (見延 玲奈) **1Pos256**
Minoshima, Wataru (箕嶋 渉) 3Pos231
3Pos233
Mio, Kazuhiro (三尾 和弘) **3Pos070**
Misawa, Norihiko (三沢 典彦) 1Pos252*
Mishima, Daisuke (三島 大輔) 2Pos220
Mishima, Masaki (三島 正規) 1Pos011
Mishima, Masanori (三嶋 将紀) 2Pos157
Mitaku, Shigeki (美宅 成樹) **1SDA-03**
Mitomi, Tatsuya (三富 達矢) **1Pos250**
Mitsui, Hiromasa (三井 広大) **2Pos228**
Mitsui, Toshiyuki (三井 敏之) 1Pos157
1Pos158
1Pos162
2Pos307
2Pos317
3Pos311
Mitsuoka, Kaoru (光岡 薫) 3Pos003
3Pos006
Mitsutake, Ayori (光武 亜代理) 2Pos034*
2Pos037
Miura, Daisuke (三浦 大輔) 2Pos078
Miura, Kota (三浦 宏太) 2Pos228
Miyachi, Yoshiyuki (宮地 義之) 2Pos078
Miyadai, Toshiaki (宮台 俊明) 2Pos238
Miyafusa, Takamitsu (宮房 孝光) 2Pos306
Miyagi, Hiraku (宮城 拓) **3Pos202**
Miyakawa, Takeshi (宮川 毅) **2Pos014**
Miyakawa, Takuya (宮川 拓也) 2Pos001*
2Pos063
Miyakoshi, Shota (宮越 昭太) **2Pos317**
Miyanaga, Yukihiro (宮永 之寛) 3Pos071
3Pos192
Miyanoiri, Yohei (宮ノ入 洋平) **3Pos143**
Miyao, Yukiko (宮尾 有貴子) 3Pos184
Miyasaka, Go (宮坂 豪) **2Pos024**
Miyashiro, Daisuke (宮代 大輔) **1Pos314**
Miyashita, Hideaki (宮下 英明) 3Pos254
3Pos257
Miyashita, Osamu (Miyashita Osamu) 2Pos011
2Pos046*
Miyashita, Osamu (宮下 治) **3SCA-08**
2Pos009
3Pos036
Miyata, Kaede (宮田 楓) 1Pos297*
1Pos318*
Miyata, Makoto (宮田 真人) 1SDP-03
3SDA-05
2Pos162*
2Pos166*
Miyata, Masato (宮田 真人) 2Pos164
Miyata, Takaki (宮田 卓樹) 2Pos132*
Miyata, Tomoko (宮田 知子) **2Pos150**
3Pos035
3Pos144
Miyatake, Takumi (宮武 拓未) **1Pos172***
Miyauchi, Seiji (宮内 正二) 1Pos242
2Pos254
3Pos095
Miyawaki, Atsushi (宮脇 敦史) **2SAP-01**
Miyazaki, Makito (宮崎 牧人) **2SBA-05**
1Pos147
1Pos183
2Pos170
Miyazaki, Masaya (宮崎 真哉) **3Pos089**
Miyazaki, Naoyuki (宮崎 直幸) **2SGA-06**
Miyazaki, Satoru (宮崎 智) 3Pos270
Miyazaki, Shota (宮崎 翔太) **2Pos134**
Miyazaki, Tsuyoshi (宮崎 剛) 3Pos105
3Pos106
Miyazaki, Yusuke (宮崎 裕介) **2Pos203**
Miyazawa, Atsuo (宮澤 淳夫) 2Pos175
Miyazawa, Sanzo (宮澤 三造) **2Pos276**
Miyazono, Yuya (宮園 侑也) 2Pos315*
Mizuguchi, Takaha (水口 高翔) **2Pos314***
3Pos307
Mizuhara, Yukinobu (水原 志暢) 3Pos039
3Pos111
Mizukami, Shin (水上 進) 3Pos304
Mizukami, Taku (水上 卓) **2Pos128**
3Pos084
Mizukami, Takuya (水上 琢也) 1Pos052
Mizumoto, Kenta (水本 健太) **1Pos177**
Mizuno, Atsushi (水野 敦) 3Pos051
Mizuno, Daisuke (水野 大介) 1Pos159
1Pos161*
1Pos165
2Pos159
Mizuno, Katsutoshi (水野 克俊) 2Pos001*
Mizuno, Misao (水野 操) 1Pos236
Mizutani, Kenji (水谷 健二) 2Pos143*
2Pos254
3Pos058
3Pos141
Mizutani, Masaki (水谷 雅希) **2Pos162***
Mizutani, Takeomi (水谷 武臣) **2Pos188**

Mizutani, Yasuhisa (水谷 泰久) 1Pos236
Mochizuki, Atsushi (望月 敦史) 3Pos202
Mochizuki, Hideki (望月 秀樹) 1Pos059
Mochizuki, Kazushi (望月 一志) **2Pos059**
Mochizuki, Yuki (望月 佑樹) 3Pos068
Moens, Pierre (Moens Pierre) **2SCP-02**
Mogami, George (最上 譲二) 2Pos121
3Pos040
3Pos042
3Pos108
3Pos134
Mohamed, Mahmoud Shaaban (モハメッド マーモウド)
1Pos306*
Momiyama, Kyoko (籾山 京子) 3Pos072
Moniruzzaman, Md. (Moniruzzaman Md.) **2Pos205**
Mori, Hikari (森 ひかり) 1Pos180
Mori, Hiroyuki (森 博幸) 3SCA-06
Mori, Kazuo (森 一夫) **3Pos305**
Mori, Koichiro (森 広一郎) 3Pos144
Mori, Tadashi (森 直) 2Pos070*
Mori, Takaharu (森 貴治) 3SCA-06
3SCA-07
2Pos060
3Pos096
Mori, Tetsuya (盛 徹也) 1Pos287*
Mori, Toshifumi (森 俊文) 1Pos010
2Pos019
Mori, Yoshiharu (森 義治) **1Pos029**
2Pos075*
Morice, Maximilien (Morice Maximilien) 3Pos162
Morigaki, Kenich (森垣 憲一) 1SFP-06
Morigaki, Kenichi (森垣 憲一) 1SFP-99
2Pos201*
2Pos214*
2Pos218
3Pos209
Morii, Hisayuki (森井 尚之) 1Pos040
2Pos063
Morii, Takashi (森居 隆史) 1Pos307
Moriizumi, Yoshiki (森泉 芳樹) 1SGP-01
2Pos257*
2Pos259
Morikawa, Manatsu (森川 真夏) 1Pos133
Morikawa, Ryota (森河 良太) 2Pos014
Morikawa, Yoshitomi (森川 善富) **1Pos302**
Morimoto, Daichi (森本 大智) **2SDP-05**
Morimoto, Gentaro (Morimoto Gentaro) 2Pos016
Morimoto, Nobuyuki (森本 展行) 3Pos040
Morimoto, Yasumasa (森本 康幹) 1Pos024
3Pos067
Morimoto, Yusuke (森本 雄祐) 3SCA-06
3Pos144
Morimoto, Yusuke V. (森本 雄祐) **2SCP-03**
2Pos182
3Pos173
3Pos180
Morishita, Ryo (森下 了) 2Pos103*
Morita, Kohei (森田 康平) 1Pos210
Morita, Masamune (森田 雅宗) 1Pos121
1Pos124
1Pos213
Morita, Shin-ichi (盛田 伸一) **2SBP-02**
Morita, Yuki (森田 雄貴) 2Pos156
Moritake, Ryota (森竹 亮太) **1Pos115**
Moritsugu, Kei (森次 圭) 1Pos042
2Pos030
2Pos035
3Pos029
Moriwaki, Yoshihito (森脇 義仁) 1Pos009
Moriwaki, Yoshitaka (森脇 由隆) **1Pos080**
2Pos012
Moriyama, Katsuhiko (森山 克彦) 1Pos132*
Morizumi, Takefumi (森住 威文) 2Pos227
Moro, Kazuyo (茂呂 和世) 1Pos297*
1Pos318*
Morozumi, Akihico (両角 明彦) **2Pos297***
Motegi, Kentarou (茂木 健太郎) 1Pos076
Motegi, Toshinori (茂木 俊憲) 1Pos210
2Pos217
3Pos086
3Pos089
Motoike, Ikuko (元池 育子) **3Pos238**
Mouri, Kazunari (毛利 一成) **3Pos191**
Mukai, Yuri (向井 有理) 2Pos024
3Pos010
Mukaiyama, Atsushi (向山 厚) **1Pos286**
3Pos013
3Pos031
Muneyuki, Eiro (宗行 英朗) 1Pos131
3Pos141
Murai, Yuka (村井 裕佳) 3Pos086
Murakami, Aya (村上 彩) 1Pos307
Murakami, Hironori (村上 博則) 1SEP-02
Murakami, Kiyotaka (村上 清貴) 3Pos263
Murakawa, Takeshi (村川 武志) 1Pos027
Muraki, Yasushi (村木 靖) 2Pos168
Muramoto, Kazumasa (村本 和優) 3SFA-02
1Pos090
1Pos093
1Pos096
Murata, Agato (村田 崇人) 1Pos110*
1Pos113*
Murata, Kazuyoshi (村田 和義) 3Pos001
3Pos004
Murata, Masayuki (村田 昌之) 1Pos104
2Pos202
Murata, Satoshi (村田 智) 3SGA-04
1Pos124
2Pos309
2Pos316*
3Pos310
3Pos315
Murata, Takeshi (村田 武士) 1Pos100
1Pos132*
2Pos143*
2Pos148
2Pos254
3Pos001
3Pos058
3Pos097
3Pos136
3Pos138
3Pos141
3Pos142
Murate, Motoshide (村手 源英) 3Pos211
Murayama, Kazutaka (村山 和隆) **3Pos011**
Murayama, Tomo (村山 知) **3Pos213**
Murayama, Yoshihiro (村山 能宏) 1Pos120
1Pos179*
2Pos119
3Pos275
Murayama-Kato, Miyuki (村山-加藤 美幸) 3Pos011
Muta, Hiroya (牟田 寛弥) **1Pos063***
2Pos065
Mutoh, Masato (武藤 昌図) 3Pos265
Mutoh, Risa (武藤 梨沙) 1Pos023
2Pos244
2Pos246
2Pos249
3Pos210
Naekura, Kazuaki (苗村 和明) 2Pos133
Nag, Preetom (Nag Preetom) **2Pos187**
Nagadoi, Aritaka (長土居 有隆) 1Pos009
Nagae, Takayuki (永江 峰幸) 2Pos005
Nagai, Kan (永井 敢) **1Pos011**
Nagai, Ken (永井 健) **1SDP-05**
1Pos190*
1Pos285
Nagai, Masako (長井 雅子) 3Pos078
Nagai, Rina (永井 里奈) 1Pos180
Nagai, Takashi (永井 貴士) **1Pos248***
Nagai, Takeharu (永井 健治) **1SBA-06**
2SDA-03
1Pos309
2Pos175
2Pos263
2Pos296
2Pos298*
3Pos175
3Pos302
Nagai, Tetsuro (永井 哲郎) 1Pos056
1Pos061
2Pos268
Nagamine, Toshihiro (永峰 俊弘) 3Pos087
Naganathan, Sundar (Naganathan Sundar) 3Pos198
Nagao, Hidemi (長尾 秀実) 1Pos043
2Pos010
2Pos014
2Pos021
3Pos205
Nagao, Ryo (長尾 遼) 1Pos250
1Pos257
Nagao, Satoshi (長尾 聡) 1Pos094*
3Pos080
Nagao, Takashi (永尾 隆) 2Pos220
Nagaoka, Masataka (長岡 正隆) 2Pos031
Nagasaka, Arata (長坂 新) **2Pos132***
Nagasaki, Akira (長崎 晃) 3SDA-07
Nagasawa, Hiromichi (長澤 寛道) 3Pos087
Nagasawa, Hirotomo (永澤 廣知) **3Pos260**
Nagashima, Hiroki (長嶋 宏樹) **1Pos254**
1Pos256
3Pos249
Nagashima, Kenji (永島 賢治) 2Pos242
Nagashima, Toshio (長島 敏雄) **1Pos055**
Nagata, Koji (永田 宏次) 3Pos087
Nagata, Takashi (永田 崇) 2Pos067*
2Pos103*
2Pos234
3Pos023
Nagatomo, Shigenori (長友 重紀) **3Pos078**
Nagatsuka, Hideyuki (長塚 秀幸) **1Pos206**
Nagayama, Kazuaki (長山 和亮) **1Pos167**
Naito, Akira (内藤 晶) 1Pos057*
1Pos243*
2Pos215*
2Pos220
3Pos247
Naito, Yasutaka (内藤 靖貴) **2Pos121**
Nakabayashi, Ryo (中林 亮) 2Pos186
Nakabayashi, Seiichiro (中林 誠一郎) 1Pos166*
1Pos175*
Nakabayashi, Takakazu (中林 孝和) 2Pos193*
Nakae, Setsu (中江 摂) 2Pos029
Nakagawa, Atushi (中川 敦史) 1Pos216
Nakagawa, Koh (中川 恒) **2Pos223**
Nakagawa, Masaki (中川 正基) **3Pos277**
Nakagawa, Satoshi (中川 聖) **1Pos043**
2Pos021
3Pos205
Nakagawa, Shogo (中川 省吾) **1Pos092**
Nakagawa, Taro (中川 太郎) 1Pos022
2Pos029
Nakagawa, Yoshiko (中川 幸姫) **2Pos038**
Nakagawa, Yoshiyuki (中川 善之) 1Pos088
Nakagawa, Yuta (中川 裕太) 3Pos230
Nakahara, Naoya (中原 直哉) 3Pos124
3Pos128
Nakahata, Masaki (中畑 雅樹) 3Pos286
Nakai, Junichi (中井 淳一) **1SAA-05**
Nakai, Kenta (中井 謙太) 1SFA-00
Nakajima, Akihiko (中島 昭彦) 1SAP-04
2SFA-04
1Pos171
2Pos183
2Pos186
3Pos194
Nakajima, Daisuke (中嶋 大祐) 1Pos026
2Pos032
Nakajima, Tetsuo (中島 徹夫) **3Pos262**
Nakajima, Yoshiki (中島 芳樹) 1SEP-02
Nakamoto, Kazuya (中本 和哉) 2Pos143*
3Pos138
Nakamura, Akihiko (中村 彰彦) 3Pos162
3Pos166
Nakamura, Chikashi (中村 史) 1Pos298
3Pos073
Nakamura, Haruki (中村 春木) 1Pos019
1Pos142*
2Pos086
3Pos008
3Pos019
3Pos052

Nakamura, Kengo (中村 建五) **1Pos077**
Nakamura, N. (中村 夏紀) 2Pos241
Nakamura, Naohiko (中村 直彦) 1Pos169
Nakamura, Naotoshi (中村 直俊) 3Pos300
Nakamura, Satoshi (中村 聡) 2Pos141
Nakamura, Shigeyoshi (中村 成芳) 1SGA-07
2Pos061*
Nakamura, Shigeyoshi (中村 茂芳) 3Pos066
Nakamura, Shin (中村 伸) **1Pos253**
Nakamura, Shugo (中村 周吾) 3Pos268
Nakamura, Shuichi (中村 修一) 3Pos146
3Pos173
3Pos177
3Pos178
3Pos180
3Pos181
3Pos182
Nakamura, Shun (中村 駿) 1Pos097
Nakamura, Shunta (中村 駿太) **2Pos232***
2Pos236
Nakamura, Takuto (中村 拓人) 1Pos169
Nakamura, Teruya (中村 照也) **2Pos027**
Nakamura, Yasuyuki (中村 泰之) 3Pos312
Nakane, Daisuke (中根 大介) 1SDP-03
2Pos154
2Pos163
3Pos179
3Pos187
Nakane, Yuko (中根 優子) **2Pos283**
Nakanishi, Atsuko (中西 温子) 1SDP-02
2Pos145*
3Pos003
3Pos006
Nakanishi, Waka (中西 和嘉) 3SGA-04
Nakano, Akihiko (中野 明彦) 1Pos314
Nakano, Kentaro (中野 賢太郎) 3Pos149
3Pos153
Nakano, Masahiro (中野 雅裕) **2SDA-03**
3Pos302
Nakano, Miki (中野 美紀) **2Pos009**
Nakano, Yasuhisa (中野 靖久) 2Pos015
Nakao, Hidenobu (中尾 秀信) 1Pos178
Nakao, Kimiko (中尾 公子) 2Pos099
Nakao, Mitsuyuki (中尾 光之) 3Pos238
Nakaoka, Hidenori (中岡 秀憲) 2Pos301*
Nakasako, Masayoshi (中迫 雅由) 1Pos319
1Pos320
2Pos082
2Pos127
2Pos303
3Pos243
Nakashima, Satoru (中島 聡) 1Pos087
1Pos088
Nakashima, Satoru (中島 聡) 2Pos079
Nakashima, Takuto (中島 拓飛) **1Pos272**
Nakasone, Yusuke (中曽根 祐介) 2Pos071
2Pos080
3Pos248
Nakata, Ayako (中田 彩子) **3Pos105**
Nakata, Shoki (中田 翔貴) 1Pos247
3Pos032
Nakata, Yoshiki (中田 吉紀) **2Pos287**
Nakata, Youichi (中田 庸一) 1SFA-05
Nakatani, Naoki (中谷 真規) **1Pos188***
3Pos204
Nakatani, Yoichi (中谷 陽一) **1Pos260**
Nakatsui, Masahiko (中津井 雅彦) 2Pos084
Nakayama, Kazuhisa (中山 和久) 3Pos028
3Pos196
Nakayama, Koji (中山 浩次) 2Pos002
2Pos161
Nakayama, Rinako (中山 莉奈子) 2Pos144
Nakayama, Takayuki (中山 貴之) **2Pos307**
Nakayama, Yohei (中山 洋平) 1Pos131
Nakayama, Yoshitaka (中山 義敬) **1Pos215**
Nakazato, Masataka (仲里 正孝) 3Pos254
3Pos257
3Pos258
Nakazawa, Akiko (中澤 晶子) 2Pos061*
Nakazono, Kohei (中園 孝平) 2Pos134
Nam, Daewoon (Nam Daewoong) 2Pos009
Nam-Joon, Cho (Nam-Joon Cho) 2Pos281
Namba, Kei-ichi (難波 啓一) 1Pos006
Namba, Keichi (難波 啓一) 3Pos122
Namba, Keiichi (難波 啓一) 2SDP-05
2Pos150
2Pos177
3Pos007
3Pos035
3Pos144
3Pos172
3Pos174
3Pos176
3Pos180
Namba, Toshinori (難波 利典) **2Pos264**
Namise, Takahiro (浪瀬 貴弘) **1Pos007**
Namiuchi, Yoshiki (波内 良樹) 1Pos041
Nanba, Daisuke (難波 大輔) **1SAP-05**
Nanbo, Asuka (南保 明日香) 3SAA-03
Nara, Masayuki (奈良 雅之) **2Pos063**
Nara, Toshifumi (奈良 敏文) 1Pos242
Naraoka, Misaki (奈良岡 岬) 1Pos305
Narita, Akihiro (成田 哲博) 1Pos299
2Pos140
2Pos177
2Pos179
Narita, Hiroka (成田 宏夏) **1Pos131**
Naruse, Yasushi (成瀬 康) 3Pos185
Natsume, Ryo (夏目 亮) 3Pos087
Natsume, Yuhei (夏目 雄平) **3Pos206**
Natsume, Yuno (夏目 ゆうの) 3Pos206
Nawa, Ryota (名和 良太) 2Pos028*
Negami, Tatsuki (根上 樹) **1Pos045**
Nemoto, Naoto (根本 直人) 1Pos034
1Pos200
Nemoto, Sayaka (根本 紗也加) **3Pos058**
Nemoto, Wataru (根本 航) **2SEA-05**
1Pos266
1Pos268
Nenoi, Mitsuru (根井 充) 3Pos262
Ng, Kwokhoi (吳 國愷) **1Pos166***
Ngo, Kien (ンゴー キエン) 3SDA-07
Ngo, Kien X. (Ngo Kien X.) 3Pos153
Ngo, Kien Xuan (Ngo Kien Xuan) **1Pos146**
Nihei, Chiho (二瓶 千穂) **3Pos133**
Nii, Daisuke (二井 大輔) 3Pos253
Niimura, Nobuo (新村 信雄) 2Pos078
Niina, Toru (新稲 亮) **1Pos109**
Niitani, Yamato (新谷 大和) 2SCA-03
1Pos138
Niki, Hironori (仁木 宏典) 2Pos048
Ninomiya, Yasuharu (二宮 康晴) 3Pos262
Nishi, Hafumi (西 羽美) **2Pos271**
3Pos029
Nishibe, Nobuyuki (西部 伸幸) 2Pos304
Nishida, Takuma (西田 拓真) 3Pos075
Nishide, Shinya (西出 真也) 3SAA-03
Nishigaki, Takehiko (西垣 岳彦) 3Pos167
Nishigami, Hiroshi (西上 博士) **3Pos024**
Nishiguchi, Shigetaka (西口 茂孝) **1Pos174**
Nishiguchi, Tatsuhito (西口 達人) 1Pos087
2Pos079
Nishihara, Yasutaka (西原 泰孝) 2Pos151
Nishijima, Masaki (西島 政樹) 2Pos050*
Nishijima, Masaki (西嶋 政樹) 2Pos070*
Nishikata, Toru (西片 亨) 2Pos222
Nishikawa, Kaori (西川 香里) 1Pos145*
1Pos290*
Nishikawa, Koji (西川 幸志) 1Pos067
Nishikawa, Masatoshi (西川 正俊) 1Pos199
3Pos184
3Pos198
Nishikawa, Naohiro (西川 直宏) **2Pos075***
Nishikawa, Shoji (西川 翔士) 3Pos167
Nishikawa, Yosuke (西河 洋祐) 1Pos003*
Nishikino, Tatsuro (錦野 達郎) **2Pos152**
Nishima, Wataru (二島 渉) 2Pos306
Nishimasu, Hiroshi (西増 弘志) 1Pos116
Nishimori, Hiraku (西森 拓) 1Pos173
2Pos110
Nishimoto, Etsuko (西本 悦子) 3Pos060
3Pos061
Nishimoto, Etuko (西本 悦子) 1Pos066
Nishimura, Chiaki (西村 千秋) **2Pos087**
Nishimura, Nao (西村 尚) **1Pos236**
Nishimura, Naoto (西村 直人) **1Pos021**
Nishimura, Satoshi (西村 智) **1SBA-02**
Nishimura, Takahiro (西村 隆宏) 2Pos312
Nishimura, Tomoya (西村 知也) **3Pos114**
Nishimura, Yoshifumi (西村 善文) 1Pos009
1Pos042
Nishino, Yuuki (西野 優紀) 1Pos004
3Pos143
Nishio, Izumi (西尾 泉) 1Pos074
Nishiya, Koki (西谷 宏輝) **1Pos241**
Nishiyama, Masahiro (西山 雅祥) 2Pos005
Nishiyama, Masayoshi (西山 雅祥) **2SEP-01**
2Pos151
2Pos165
Nishiyama, Shunsuke (西山 俊介) 2Pos266
Nishiyama, So-ichiro (西山 宗一郎) **3Pos186**
Nishizaka, Takayuki (西坂 崇之) 1SDP-03
2Pos144
2Pos146*
2Pos154
2Pos162*
2Pos163
2Pos174*
3Pos160
3Pos179
Nishizawa, Hiroaki (西澤 宏晃) **3Pos048**
Nishizawa, Kenji (西澤 賢治) 1Pos159
1Pos161*
1Pos165
Nishizawa, Tomohiro (西澤 知宏) 1Pos002
Nisihigami, Hiroshi (西上 博士) 1Pos112
Nitani, Ayame (二谷 綾愛) **2Pos065**
Nitta, Ryo (仁田 亮) 1Pos133
Nitta, Takahiro (新田 高洋) **1Pos150**
Niwa, Fumihiro (丹羽 史尋) 2SBP-06
Niwa, Tatsuya (丹羽 達也) 2SDP-03
Nobe, Takuro (野辺 卓郎) **2Pos053**
Noguchi, Hiroshi (野口 博司) **1Pos202**
2Pos223
Noguchi, K. (Noguchi K.) 3Pos027
Noguchi, Takumi (野口 巧) 1Pos250
1Pos251
1Pos253
1Pos257
1Pos258
Noguchi, Taro (野口 太郎) 3SDA-07
Noguchi, Yoshifumi (野口 良史) 1Pos249
Noji, Hiroyuki (Noji Hiroyuki) 3Pos137
Noji, Hiroyuki (野地 博行) 1SGP-01
2SCA-03
1Pos084
1Pos103
1Pos130*
1Pos132*
1Pos135
1Pos301
2Pos081
2Pos094
2Pos148
2Pos192*
2Pos257*
2Pos259
2Pos280
2Pos285
2Pos293
3Pos074
3Pos112
3Pos114
3Pos139
3Pos140
3Pos175
Nojima, Tatsuya (野島 達也) 2SDP-02
Nomura, Fumimasa (野村 典正) 1Pos277
Nomura, M. Shin-ichiro (野村 M. 慎一郎) 1Pos124
Nomura, Mami (野村 真未) **2Pos181**
Nomura, Masashi (野村 匡志) **2Pos025**
Nomura, Saki (野村 紗希) 1Pos223
Nomura, Shin-Ichiro (野村 慎一郎) 3Pos315

Nomura, Shin-ichiro (野村 慎一郎) 2Pos309
2Pos316*
Nomura, Shin-ichiro M. (野村 M.慎一郎) 3Pos310
Nomura, Tadashi (野村 真) **2SDA-06**
Nomura, Takashi (野村 高志) **3Pos075**
Nomura, Takeshi (野村 健) 2Pos043
Nomura, Yasutomo (野村 保友) 1Pos305
Nomura, Yoshihiro (野村 芳弘) 2Pos164
Nomura, Yurika (野村 祐梨香) 1Pos240*
1Pos263*
1Pos264
2Pos256
Nomura, Yuta (野村 勇太) 2Pos144
Nonaka, Shigenori (野中 茂紀) **2SAP-06**
Nonaka, Yuta (野中 雄太) 2Pos123*
Norisada, Kazushi (則定 和志) 2Pos220
Nosaka, Michiko (野坂 通子) **2Pos097**
Noshiro, Daisuke (能代 大輔) **2Pos290**
Nozaki, Shohei (野崎 梢平) 3Pos196
Nozaki, Tadasu (野崎 慎) 1Pos315*
2Pos263
Nozawa, Yosuke (野沢 陽佑) **1Pos258**
Nukazuka, Genki (糠塚 元気) 2Pos078
Numata, Osamu (沼田 治) 3Pos149
3Pos153
Numoto, Nobutaka (沼本 修孝) **1Pos022**
1Pos040
Nureki, Osamu (濡木 理) 1Pos002
1Pos095*
1Pos116
Oasa, Sho (大浅 翔) **2Pos100**
2Pos114*
Oba, Yojiro (大場 洋二郎) 3Pos033
Ochiai, Yuki (落合 由貴) 2Pos121
3Pos042
Oda, Akiyoshi (小田 彰克) 2SDP-02
Oda, Arisa (小田 有沙) 2Pos301*
Oda, Hiroki (小田 広樹) 1Pos174
Oda, Masayuki (織田 昌幸) 1Pos033*
1Pos036*
1Pos040
Oda, Soutaro (尾田 颯太郎) **1Pos293**
Oda, Takashi (小田 隆) 1Pos023
Oda, Toshiyuki (小田 賢幸) 1SEA-06
1SEA-99
2SGA-03
Odahara, Takayuki (小田原 孝行) 1Pos186
Odaka, Masao (尾高 正朗) 2Pos287
2Pos318
3Pos130
3Pos229
3Pos308
3Pos309
Ode, Koji L. (大出 晃士) **2SGP-01**
Oestreicher, Zachery (Oestreicher Zachery) 2Pos289
Ogasawara, Naoki (小笠原 直輝) 2Pos123*
2Pos268
Ogawa, Hiroto (小川 宏人) 1Pos232
Ogawa, Shuhei (小川 修平) 1Pos040
Ogawa, Tadayuki (小川 覚之) **2SGP-06**
Ogawa, Taisaku (小川 泰策) 1Pos147
Ogawa, Wataru (小川 渉) 3Pos279
Ogawa, Yoshikatsu (小川 昌克) **3Pos199**
Ogawa, Yuya (小川 雄也) 3Pos172
Oguchi, Yusuke (小口 祐伴) 3Pos113
Ogura, Takashi (小倉 尚志) 1Pos087
1Pos088
2Pos079
3Pos078
3Pos082
Ogura, Toshihiko (小椋 俊彦) **3Pos303**
Ogura, Yoshitoshi (小椋 義俊) 2Pos135
Ogura, Yusuke (小倉 裕介) 2Pos312
Oh-oka, Hirozo (大岡 宏造) **2Pos246**
2Pos249
Ohara, Masayuki (大原 正行) 1Pos108
1Pos213
2Pos311*
3Pos313
Ohara, Osamu (小原 收) 1Pos297*
1Pos318*
3Pos115
Ohashi, Kaoru (大橋 郁) **3Pos279**
3Pos284
Ohba, Yasunori (大庭 裕範) **3Pos065**
Ohba, Yusuke (大場 雄介) **3SAA-03**
Ohgita, Takashi (扇田 隆司) **3Pos072**
Ohira, Ayaka (大平 彩花) **1Pos257**
Ohki, Mio (大木 規央) 3Pos076
Ohkoshi, A. (大越 慧) 2Pos241
Ohkura, Masamichi (大倉 正道) 1SAA-05
Ohmachi, Masashi (大町 優史) 3Pos151
Ohnishi, Kohei (大西 晃平) **1Pos274**
Ohnishi, Yuki (大西 裕季) 2Pos007*
Ohno, Naohito (大野 尚仁) 1Pos024
Ohno, Takashi (大野 隆) 2Pos242
2Pos248
Ohno, Tetsuo (大野 哲生) 3Pos124
3Pos127
3Pos128
Ohnuki, Jun (大貫 隼) 2Pos124
3Pos039
3Pos062
Ohnuki, Taichi (大貫 多一) **3Pos182**
Ohnuma, Kiyoshi (大沼 清) 2Pos134
Ohsawa, Noboru (大沢 登) 1Pos242
Ohta, Akane (太田 茜) 1Pos228*
Ohta, Kunihiro (太田 邦史) 2Pos301*
Ohta, Noboru (太田 昇) 1Pos014
3Pos043
Ohta, Taeko (太田 太恵子) 1Pos185
Ohta, Yoshihiro (太田 佳宏) 1SFA-05
Ohta, Yoshihiro (太田 善浩) 1Pos192
1Pos201
1Pos219
2Pos197
2Pos198
2Pos199
2Pos200
Ohtomo, Hideaki (大友 秀明) 1Pos009
2Pos062
Ohtsuka, Jun (大塚 淳) 3Pos087
Ohuchi, Hideyo (大内 淑代) 2Pos233
Ohue, Masahito (大上 雅史) **2Pos269**
Ohwada, Tomohiko (大和田 智彦) 2Pos020
Oide, Mao (大出 真央) **3Pos243**
Oikawa, Hiroyuki (小井川 浩之) **2SDP-01**
1Pos083*
2Pos052*
2Pos095*
Oiki, Shigetoshi (老木 成稔) 1Pos217
1Pos225
1Pos226
Oiwa, Kazuhiro (大岩 和弘) 1Pos129
1Pos141
1Pos143
1Pos285
2Pos173
2Pos313
3Pos161
3Pos183
3Pos185
Oka, Kohei (岡 浩平) **3Pos214**
Oka, Kotaro (岡 浩太郎) **2SBP-01**
1Pos227*
1Pos230*
1Pos232
2Pos194*
2Pos277*
Oka, Toshihiko (岡 俊彦) **1Pos208**
2Pos235
Oka, Toshihiko (岡 敏彦) **3SFA-05**
Oka, Yoshiki (岡 芳樹) 1Pos054*
1Pos078*
2Pos089
Okabe, Kohki (岡部 弘基) **1SBP-04**
2SDA-00
2SDA-03
1Pos317
2Pos195*
2Pos196
2Pos279
2Pos284
2Pos292
3Pos297
3Pos302
Okada, Masato (岡田 真人) **2SFP-01**
3Pos185
Okada, Rio (岡田 璃生) **2Pos070***
Okada, Takaharu (岡田 峰陽) **2SFA-05**
Okada, Takao (岡田 孝夫) 1Pos307
Okada, Takumi (岡田 卓巳) **3Pos233**
Okada, Tomoko (岡田 知子) 3Pos073
3Pos303
Okada, Yasushi (岡田 康志) **2SCA-05**
1Pos133
1Pos137*
2Pos160
2Pos191
2Pos263
3Pos155
3Pos191
Okagaki, Tsuyoshi (岡垣 壮) 3Pos129
Okai, Masahiko (岡井 公彦) 2Pos001*
Okajima, Koji (岡島 公司) 1Pos320
2Pos303
3Pos243
3Pos248
Okajima, Takaharu (岡嶋 孝治) 2Pos277*
2Pos281
Okamoto, Akihiro (岡本 章玄) 1Pos102*
Okamoto, Kenichi (岡本 賢一) **1SAA-04**
Okamoto, Kenji (岡本 憲二) **2Pos278**
Okamoto, Yuko (岡本 祐幸) 1Pos030
1Pos069
2Pos075*
Okamura, Hideyo (岡村 英世) 3Pos062
Okamura, Yasushi (岡村 康司) 1Pos216
1Pos224
2Pos298*
3Pos094
Okano, Keiko (岡野 恵子) 2Pos228
2Pos229*
2Pos238
Okano, Toshiyuki (岡野 俊行) 2Pos228
2Pos229*
2Pos238
Okawa, Keisuke (大川 啓介) 1Pos242
Okazaki, K. (岡崎 航大) 2Pos241
Okazaki, Kei-ichi (岡崎 圭一) **1Pos127**
Okazaki, Shigetoshi (岡崎 茂俊) 1Pos304
Okazaki, Susumu (岡崎 進) 2Pos203
Okazaki, Taku (岡崎 拓) **1SBA-03**
Okimoto, Noriaki (沖本 憲明) 3Pos084
Okimura, Chika (沖村 千夏) **1Pos160**
1Pos163
1Pos164
Okita, Nana (沖田 奈々) 2Pos126*
Okitsu, Takashi (沖津 貴志) 1Pos237
1Pos238*
1Pos243*
3Pos247
Okuda, Satoru (奥田 覚) **1Pos281**
Okumoto, Atsushi (納本 淳) 1Pos205
Okumura, Hisashi (奥村 久士) 1Pos025
1Pos029
2Pos075*
3Pos048
Okuno, Daichi (奥野 大地) 1Pos191
Okuno, Takashi (奥野 貴士) 2Pos225*
Okuno, Yasushi (奥野 恭史) 1Pos019
2Pos084
Okuta, Akiko (奥田 明子) 1Pos002
Oldenburg, Rudolf (Oldenburg Rudolf) 1SAA-03
Omagari, Katsumi (尾曲 克己) **2Pos006**
Omi, Tsuyoshi (近江 毅志) 1Pos138
Omori, Satoshi (大森 聡) **1Pos042**
Onami, Shuichi (大浪 修一) 1Pos316
2Pos137
3Pos280
Ono, Koji (小野 晃司) **1Pos044**
Ono, Takahito (小野 崇人) **2SDA-01**
Ono, Yukiko (小野 友紀子) 2Pos250*

Onoue, Yasuhiro (尾上 靖宏) 2Pos152
3Pos143
Ooi, Atsusi (大井 淳史) 3Pos129
Ooiwa, Kazuhiro (大岩 和弘) 3Pos158
Oono, Kodai (大野 広大) 3Pos173
Oosaki, Marika (大埼 麻里加) 3Pos085
Oosawa, Kenji (大澤 研二) 3Pos173
Ootaki, Masanori (大瀧 正訓) 2Pos008
Orii, Ayano (織井 彩乃) **2Pos229***
Oroguchi, Tomotaka (苙口 友隆) 1Pos319
1Pos320
2Pos082
2Pos127
2Pos303
3Pos243
Osaki, Toshihisa (大崎 寿久) 2Pos157
Osanai, Makoto (小山内 実) **3Pos235**
Osawa, Masanori (大澤 匡範) 3SCA-04
1Pos095*
Oshima, Atsunori (大嶋 篤典) 3Pos088
Ostuka, Takao (大塚 教雄) 3Pos105
Osuka, Saki (大須賀 彩希) 1Pos116
Ota, Chiaki (太田 千晶) **2Pos066**
Ota, Motonori (太田 元規) 1Pos273
2Pos140
3Pos271
Ota, Sadao (太田 禎生) 1Pos301
2Pos293
Otaki, Tatsuro (大瀧 達朗) **2SEP-04**
Otani, Yuko (尾谷 優子) **2Pos020**
Otani, Yuto (大谷 優人) **1Pos243***
Otomo, S. (大友 征宇) **2Pos241**
2Pos243
Otomo, Seiu (大友 征宇) 2Pos248
Otomo, Sheiu (大友 征宇) 2Pos242
Otosu, Takuhiro (乙須 拓洋) 1Pos212
Otsuka, Takao (大塚 教雄) 3Pos084
3Pos106
Oura, Makoto (大浦 真) **2Pos282***
Oura, Shusuke (大浦 秀介) **3Pos100**
Oyama, Kotaro (大山 廣太郎) 1Pos229
Oyama, Ryo (小山 糧) **1Pos245***
Ozaki, Mamiko (尾崎 まみこ) 1Pos014
Ozawa, Shoichi (小澤 翔一) 2Pos238
Ozeki, Kouhei (小関 康平) **3Pos249**
Pack, Chan-Gi (Pack Chan-Gi) **2SCP-05**
Park, Sam-Yong (朴 三用) 3Pos076
Parkin, Dan (パーキン 暖) 1Pos128
3Pos111
Parvez, Farliza (パーベツ、ファーリザ、) **1Pos198**
Peet, Daniel (Peet Daniel) 2Pos073
Perrett, Sarah (Perrett Sarah) **1SCP-03**
Peterman, Erwin (Peterman Erwin) 2Pos112
Peutz, Willem (Peutz Willem) 2Pos112
Piehler, Jacob (Piehler Jacob) **1SFP-03**
Pollard, Thomas (ポラード トーマス) 2Pos139
Portelli, Stefanie S. (Portelli Stefanie S.) **3Pos117**
Qi, Hao (斉 浩) 2Pos315*
Ramanujam, Kumaresan (ラマヌジャム クマレサン)
1Pos307
Rashid, M Harunur (Rashid M Harunur) **2Pos219**
Robertson, Elizabeth N. (Robertson Elizabeth N.)
3Pos117
Roome, Christopher J. (Roome Christopher J.) 1SAA-06
Ross, Jennifer L. (Ross Jennifer L.) 3Pos156
Rumi, Farhana (Rumi Farhana) 2Pos068
Rzeznicka, Izabela (Rzeznicka Izabela) 1Pos075
S.S, Ashwin (S.S. Ashwin) **3Pos116**
Sada, Kazuki (Sada Kazuki) 3Pos154
3Pos157
Sada, Kazuki (佐田 和己) 1Pos152*
1Pos289*
2Pos158
Sadakane, Kei (貞包 慧) 1Pos139
Sadakane, Koichiro (貞包 浩一朗) 1Pos122
Sagawa, Takashi (佐川 貴志) 1Pos137*
2Pos313
3Pos183
Saido, Takaomi C. (西道 隆臣) 2Pos076*
Saigo, Naoki (西郷 直記) **2Pos274**
Saiki, Toshiharu (斎木 敏治) 2Pos314*
3Pos307
Saito, Ai (齋藤 あい) **1Pos152***
Saito, Azusa (齋藤 梓) 1Pos074
Saito, Hiroaki (齋藤 大明) **3Pos084**
Saito, Hiroyuki (斎藤 博幸) 3Pos072
Saito, Kazuya (齋藤 一弥) 3Pos078
Saito, Ken (齊藤 健) 3Pos289
Saito, Mami (齋藤 真美) **1Pos118**
Saito, Mari (斉藤 真理) 2Pos114*
Saito, Masataka (齊藤 雅嵩) **2Pos052***
Saito, Mineki (齊藤 峰輝) 2Pos168
Saito, Minoru (斎藤 稔) 3Pos232
3Pos289
Saito, Natsumi (齋藤 夏美) 1SFP-06
Saito, Shinji (斉藤 真司) 1Pos010
2Pos019
2Pos251*
Saito, Takami (斎藤 崇己) 1Pos026
2Pos032
Saito, Takashi (斉藤 貴志) 2Pos076*
Saito, Takashi (斉藤 隆) **2SFA-02**
Saito, Toru (齋藤 徹) 2Pos015
Saito, Yohei (斎藤 陽平) **1Pos278**
Saitoh, Noriko (斉藤 典子) 1Pos313
Saitou, Hideo (齋藤 英夫) **3Pos195**
Sakabe, Kakeru (坂部 翔) **1Pos112**
3Pos024
Sakae, Yoshitake (榮 慶丈) 1Pos069
Sakaguchi, Kazuhiko (坂口 一彦) 3Pos279
Sakaguchi, Miyuki (坂口 美幸) **2Pos051**
Sakai, Atsushi (酒井 淳) **1Pos179***
Sakai, Hironori (酒井 博則) 2Pos023
Sakai, Kouichi (坂井 晃一) 3Pos126
Sakai, Masato (酒井 将人) 2Pos272
Sakai, Nobuaki (酒井 信明) 1Pos174
Sakai, Takahiro (酒井 貴弘) 1Pos256
Sakai, Tatsuya (堺 立也) **2Pos168**
Sakai, Tomofumi (酒井 智史) **3Pos144**
Sakai, Yuji (境 祐二) **3Pos276**
Sakajiri, Tetsuya (坂尻 徹也) 2Pos237
Sakajiri, Yuko (坂尻 由子) **2Pos237**
Sakakibara, Hitoshi (榊原 斉) **2Pos180**
Sakamoto, Naoaki (坂本 尚昭) 2Pos233
3Pos269
Sakamoto, Seiji (坂本 清志) 1Pos083*
2Pos095*
Sakamoto, Yuki (坂本 雄貴) **3Pos314**
Sakashita, Naoto (坂下 直人) 2Pos307
3Pos311
Sakata, Mikiya (坂田 樹哉) **1Pos140**
Sakata, Souhei (坂田 宗平) 1Pos224
Sakata-Sogawa, Kumiko (十川 久美子) 1Pos310
1Pos312
1Pos313
2Pos295
2Pos300
3Pos295
3Pos296
3Pos299
Sakatani, Yoshihiro (酒谷 佳寛) **2Pos258***
Sako, Yasushi (佐甲 靖志) 1Pos104
1Pos107
2Pos130
2Pos202
2Pos278
3Pos091
3Pos202
Sakuma, Kou (佐久間 昂) 2Pos126*
Sakuma, Kouya (佐久間 航也) **2Pos096**
Sakuma, Mayuko (佐久間 麻由子) 1Pos004
3Pos167
3Pos170
3Pos171
Sakuma, Morito (佐久間 守仁) **2Pos192***
Sakuma, Yosuke (佐久間 洋介) 1Pos052
Sakuraba, Shun (桜庭 俊) 2Pos115
3Pos099
Sakuraba, Shun (櫻庭 俊) 2Pos086
2Pos270
Sakurai, Kazumasa (櫻井 一正) 1Pos033*
1Pos063*
Sakurai, Minoru (櫻井 実) 1Pos271*
2Pos045
Sakurai, Takashi (櫻井 貴志) 2Pos273
3Pos312
Sakuta, Hiroki (作田 浩輝) 1Pos294
Salawu, Emmanuel (艾 曼紐) 2Pos108
Sano, Hitomi (佐野 瞳) 3Pos127
Sano, Kuniaki (佐野 訓明) 3Pos024
Sano, Masaki (佐野 雅己) 1Pos290*
2Pos185
Sano, Yuto (佐野 雄図) **1Pos183**
Sansom, Mark S.P. (Sansom Mark S.P.) 2Pos212
Sao, Yasushi (佐甲 靖志) 2Pos299
Saotome, Tomonori (早乙女 友規) 1Pos062
2Pos061*
Sarkar, Bidyut (Sarkar Bidyut) **1Pos126**
Saruta, Moeko (猿田 萌子) **1Pos212**
Sasai, Masaki (Sasai Masaki) 1Pos276*
1Pos279
Sasai, Masaki (笹井 理生) 1SFA-04
1Pos275
2Pos113
3Pos051
3Pos116
3Pos152
Sasaki, Akira (佐々木 章) 2Pos100
3Pos054
Sasaki, Kazuo (佐々木 一夫) **1Pos134**
Sasaki, Syogo (佐々木 将伍) 1Pos241
Sasaki, Tohru (佐々木 徹) 2Pos124
Sasaki, Yasuhiko (佐々木 寧彦) 1Pos149
Sasaki, Yuji (佐々木 裕次) **2SFP-04**
1Pos303
2Pos050*
2Pos070*
3Pos026
Sasaki, Yuji C. (佐々木 裕次) 1SEP-04
1Pos231*
3Pos070
Sato, Akihiko (佐藤 秋彦) 2Pos157
Sato, Akimasa (佐藤 皓允) **2Pos261***
Sato, Ayana (佐藤 文菜) 3Pos077
Sato, Chika (佐藤 千夏) **1Pos020***
Sato, Chikara (佐藤 主税) **3Pos002**
3Pos300
Sato, Daisuke (佐藤 大輔) **1SGA-05**
2Pos062
Sato, Hayao (佐藤 駿) 2Pos238
Sato, Hiroshi (佐藤 博司) 1Pos311*
Sato, Hiroyuki (佐藤 博之) **2Pos083**
Sato, Hiroyuki (佐藤 宏之) 2Pos269
Sato, Keiko (佐藤 啓子) 2Pos002
Sato, Keita (佐藤 恵太) 2Pos233
Sato, Ken (佐藤 健) **1Pos175***
Sato, Kento (佐藤 健人) **3Pos146**
Sato, Kyosuke (佐藤 恭介) **3Pos104**
Sato, Mamoru (佐藤 衛) 1Pos023
Sato, Mari (佐藤 真理) 3Pos002
3Pos300
Sato, Masaaki (佐藤 正晃) 1SAA-05
Sato, Masahiko (佐藤 昌彦) 1Pos009
Sato, Miwa (佐藤 美和) 2Pos084
Sato, Nobuhiro (佐藤 信浩) 3Pos033
Sato, Ryuma (佐藤 竜馬) **3Pos102**
Sato, Shiho (佐藤 志帆) **1Pos294**
Sato, Shizuma (佐藤 玄実) **2Pos118**
Sato, Shota (佐藤 翔太) **3Pos316**
Sato, Shunichi (佐藤 俊一) **2SAP-04**
Sato, Shunsuke (佐藤 俊輔) **3Pos047**
Sato, Takaaki (佐藤 孝明) **3Pos234**
Sato, Takato (佐藤 昂人) 1Pos115
2Pos124
3Pos062
Sato, Takeshi (佐藤 毅) 3Pos091
Sato, Wataru (佐藤 航) 1Pos086
1Pos089*
Sato, Yuki (佐藤 有紀) **1SBP-05**
Sato, Yusuke (佐藤 佑介) **2Pos316***
Sato, Yusuke (佐藤 雄介) **2SBP-05**
Sato-Tomita, Ayana (佐藤 文菜) **1SEP-04**
Satoh, Tadashi (佐藤 匡史) 3Pos014

Satou, Hana (佐藤 花) 2Pos313
Sawada, Ikutaro (沢田 幾太郎) 2Pos233
Sawada, Taihei (澤田 泰平) 1Pos078*
Sawada, Yasuyuki (澤田 康之) **2Pos043**
Sawai, Satoshi (澤井 哲) **1SAP-04**
2SFA-04
1Pos171
1Pos284
2Pos183
2Pos186
3Pos194
Schiessel, Helmut (Schiessel Helmut) 2Pos117
Seita, Akihisa (清田 晃央) **3Pos201**
Seki, Arashi (関 嵐) 2Pos176
Seki, Hayato (関 隼斗) 1Pos237
Seki, Yasutaka (関 安孝) **3Pos066**
Sekiguchi, Hiroshi (関口 博史) **1SEP-03**
1Pos231*
1Pos303
2Pos050*
2Pos070*
3Pos026
3Pos070
Sekiguchi, Tetsushi (関口 哲志) 2Pos273
2Pos315*
3Pos312
Sekiguchi, Yuki (関口 優希) 1Pos319
1Pos320
2Pos303
Sekino, Hideo (関野 秀男) 1Pos205
Sekiya, Yusuke (關谷 悠介) **1Pos101***
2Pos274
Senda, Toshiya (千田 俊哉) 3Pos087
Senga, Yukako (千賀 由佳子) **1Pos065**
Seno, Keiji (妹尾 圭司) 1SFP-06
Seo, Daisuke (瀬尾 悌介) **1Pos064**
Setoguchi, Yuuji (瀬戸口 裕二) **1Pos184**
Setou, Mitsutoshi (瀬藤 光利) 2Pos174*
3Pos160
Seyama, Kaho (背山 佳穂) 2Pos049
Shajaripourjaberi, Seyedehnoorolhoda (Shajaripourjaberi Seyedehnoorolhoda) **3Pos148**
Shaji, Divya (シャジ ディビヤ) 3Pos271
Sharmin, Sabrina (シャーミン サブリナ) 2Pos207
Sharmin, Sabrina (シャーミン、サブリナ、) **1Pos194**
Shen, Jian-Ren (沈 建仁) 1SEP-02
1Pos255*
Shew, Chen-Yang (Shew Chen-Yang) 1Pos188*
Shew, Chwen-Yang (Shew Chwen-Yang) 1Pos293
3Pos204
Shi, Beini (時 ベイニ) 1SBP-04
2Pos195*
Shi, Hengyu (史 恒宇) **2Pos295**
Shi, Xiaolei (シ シャオレイ) 2Pos199
Shiba, Kogiku (柴 小菊) **1SEA-03**
2Pos169
2Pos181
Shibata, Akihiro (柴田 明宏) 1Pos093
Shibata, Kaoru (柴田 薫) 1Pos059
3Pos077
3Pos123
Shibata, Kosei (柴田 耕生) **2Pos071**
Shibata, Kotomi (柴田 琴実) 3Pos150
Shibata, Mikihiro (柴田 幹大) **3SCA-05**
1Pos244
1Pos264
1Pos308
3Pos093
3Pos292
Shibata, Naoki (柴田 直樹) 3Pos080
Shibata, Satoshi (柴田 敏史) **3SDA-06**
2Pos161
Shibata, Takahiro (柴田 貴弘) **1Pos219**
2Pos197
Shibata, Tatsuo (柴田 達夫) 1Pos282
Shibata, Yohei (芝田 洋平) 1Pos246
1Pos247
3Pos032
Shibata, Yutaka (Shibata Yutaka) 2Pos288
Shibata, Yutaka (柴田 穣) 3Pos260
Shibata, Yutaka (柴田 穣) 1Pos075
Shibayama, Naoya (柴山 修哉) 1SEP-04
3Pos076
3Pos077
Shibly, Sayed Ul Alam (Shibly Sayed Ul Alam) **2Pos216**
Shibly, Sayed Ul Alam (シブリー サイエッド ウル アラム) 1Pos196
Shibly, Sayed Ul Alam (シブリー、 サイエッド ウル アラム、) 1Pos194
Shibuya, Risa (渋谷 理紗) **2Pos306**
Shichida, Yoshinori (七田 芳則) **2SAA-07**
1Pos104
2Pos230
2Pos233
2Pos235
Shidara, Hisashi (設樂 久志) **1Pos227***
1Pos230*
Shigematsu, Hideki (重松 秀樹) **2SGA-01**
Shigematsu, Taiki (重松 大輝) **2Pos210**
Shigeri, Yasushi (茂里 康) 1Pos065
Shigeta, Arisu (重田 安里寿) 1Pos237
1Pos238*
1Pos243*
Shigeta, Yasuteru (重田 育照) 1Pos027
2Pos036
2Pos261*
3Pos102
Shigeyoshi, N. (Shigeyoshi N.) 3Pos027
Shigihara, Eiji (鴫原 永之) **2Pos273**
Shihoya, Wataru (志甫谷 渉) **1Pos002**
Shiina, Masayuki (椎名 真之) **3Pos160**
Shima, Tomohiro (島 知宏) 1Pos116
Shima, Tomohiro (島 知弘) **1Pos133**
1Pos140
3Pos155
Shimabukuro, Katsuya (島袋 勝弥) **2SGA-05**
1Pos164
Shimada, Atsuhiro (Shimada Atsuhiro) 2Pos077
Shimada, Atsuhiro (島田 敦広) 3SFA-02
1Pos085
1Pos096
Shimada, Ichio (嶋田 一夫) 3SCA-04
1Pos095*
Shimada, Satoru (島田 悟) **1Pos085**
3Pos085
3Pos088
Shimazaki, Shunta (島崎 俊太) 3SFA-05
Shimba, Keigo (榛葉 啓悟) **1Pos035**
1Pos038*
Shimizu, Hirofumi (清水 啓史) **1Pos098**
Shimizu, Kentaro (清水 謙多郎) 1Pos045
2Pos012
3Pos067
3Pos268
Shimizu, Kohei (清水 幸平) 3Pos251
Shimizu, Masahiro (清水 将裕) **2Pos098***
Shimizu, Tatsuki (清水 達貴) 1Pos251
Shimizu, Yosuke (清水 洋輔) 2Pos180
Shimizu, Yuki (清水 悠生) **1Pos083***
2Pos095*
Shimojo, Hideaki (下條 秀朗) 1Pos009
Shimomura, Harunobu (下村 陽信) 3Pos085
Shimomura, Suguru (下村 優) **2Pos312**
Shimomura, Takumi (下村 拓海) **3Pos017**
Shimono, Kazumi (下野 和実) 1Pos242
2Pos254
3Pos095
3Pos141
Shimozawa, Togo (下澤 東吾) 1Pos147
Shin, Chanyoung (申 纂暎) 3Pos295
Shindo, Asako (進藤 麻子) **1SDP-06**
Shinoda, Tomoyasu (篠田 友靖) 2Pos132*
Shinoda, Toshiyuki (篠田 稔行) **3Pos253**
Shinoda, Wataru (篠田 渉) 2Pos203
Shinohara, Kyosuke (篠原 恭介) **1SEA-02**
2Pos176
2Pos177
2Pos179
Shinohara, Miho (篠原 美帆) 3Pos028
Shinozaki, Reina (篠崎 玲奈) 2Pos054
Shinozaki, Reina (篠﨑 玲奈) **1Pos051***
Shinozawa, Tomoki (篠沢 智伎) **2Pos069**
Shintaku, Hirofumi (新宅 博文) 2Pos156
3Pos156
Shintani, Seine (新谷 正嶺) 3Pos195
Shintani, Seine A. (新谷 正嶺) **3Pos132**
Shinzawa-Itoh, Kyoko (伊藤-新澤 恭子) 1Pos085
Shinzawa-Itoh, Kyoko (伊藤ー新澤 恭子) 1Pos096
Shinzawa-Itoh, Kyoko (伊藤（新澤）恭子) **3Pos085**
3Pos088
Shinzawa-Itoh, Kyoko (新澤- 伊藤 恭子) 3SFA-02
Shinzawa-Itoh, Kyoko (新澤-伊藤 恭子) 1Pos087
Shinzawa-Itoh, Kyoko (新澤ー伊藤 恭子) 1Pos089*
2Pos079
Shionyu, Masafumi (塩生 真史) 1Pos269
3Pos267
Shiraga, Misaki (白髪 美咲) **3Pos158**
3Pos161
Shirai, Nobu C. (白井 伸宙) **3Pos053**
Shirai, Tomohiro (白井 智宏) **2SEP-05**
Shirai, Tsuyoshi (白井 剛) 1SFA-99
1Pos267
1Pos269
2Pos152
3Pos143
Shiraishi, Kenji (白石 賢二) 2Pos261*
Shiraiwa, Yoshihiro (白岩 善博) 3Pos254
Shirakawa, Masahiro (白川 昌宏) 2SDP-05
Shirakwa, Masahiro (白川 昌宏) 3SCA-02
Shirasaki, Yoshitaka (白崎 善隆) **3SAA-05**
1Pos297*
1Pos318*
3Pos113
3Pos115
Shiro, Noritsugu (城 宜嗣) 1Pos093
Shiro, Yoshitsugu (Shiro Yoshitsugu) 2Pos033
Shiro, Yoshitsugu (城 宜嗣) 1Pos091
3Pos075
3Pos096
Shiroguchi, Katsuyuki (城口 克之) **1SBA-07**
Shirota, Matsuyuki (城田 松之) 2Pos044
3Pos022
Shirouzu, Mikako (白水 美香子) 1Pos242
3Pos011
3Pos095
3Pos141
Shohda, Koh-ichiroh (庄田 耕一郎) **2Pos222**
Shoji, Mitsuo (庄司 光男) **1Pos027**
2Pos261*
Shoji, Shuichi (庄司 習一) 2Pos315*
Shoji, Shuichi (庄子 習一) 2Pos273
3Pos312
Shoji, Yuya (荘司 憂也) 2Pos017
Shojima, Tomoki (生島 智樹) **2Pos001***
Shozui, Shunsuke (祥瑞 俊介) 3Pos292
Shribak, Michael (Shribak Michael) 1Pos315*
Sikdar, Arunima (Sikdar Arunima) 3Pos014
Sirai, Tsuyoshi (白井 剛) 2Pos029
Sitters, Gerrit (Sitters Gerrit) 2Pos112
So, Masatomo (宗 正智) 1Pos063*
2Pos064*
2Pos065
Soga, Kohei (曽我 公平) **2SAP-02**
Soga, Naoki (曽我 直樹) **1Pos103**
2Pos149
Sokabe, Masahiro (曽我部 正博) **1SDA-02**
1Pos170
2Pos043
Song, Changyong (Song Changyong) 2Pos009
Song, Chihong (ソン チホン) 3Pos001
3Pos004
Sonoda, Ayane (薗田 綾音) 1Pos163
Sonoyama, Masashi (園山 正史) 1Pos073
1Pos076
1Pos210
2Pos217
3Pos086
3Pos089
Sorimachi, Yuhta (反町 優太) 3Pos257
Sowa, Yoshiyuki (曽和 義幸) 1Pos199
2Pos153*
3Pos145
3Pos183

3Pos184
3Pos185
3Pos189
Srivastava, Arpita (Srivastava Arpita) **2Pos046***
Srivastava, Ashutosh (Srivastava Ashutosh) 3SCA-01
3Pos009
Stark, Holger (Stark Holger) 3Pos088
Stevens, M M (Stevens M M) 2Pos219
Su, Guan-Chin (蘇 綸勤) 1Pos117*
Su, Xiao-Dong (Su Xiao-Dong) 2Pos105
Subagyo, Agus (スバギョ アグス) 3Pos056
Subekti, Dwiky Rendra Graha (Subekti Dwiky Rendra Graha) **1Pos113***
Suda, Ayaka (須田 綾香) 3Pos080
Sudhaharan, Thankiah (Sudhaharan Thankiah) 2SCP-06
Sudo, Yuki (須藤 雄気) 2SAA-01
2SAA-04
1Pos237
2Pos253
2Pos254
3Pos058
3Pos209
3Pos240
3Pos245
3Pos247
Suegara, Masaaki (末柄 祐明) 3Pos160
Suematsu, Ayumi (末松 安由美) 3Pos109
Suematsu, Yuma (末松 佑麿) **1Pos032***
Suenaga, Atsushi (末永 敦) 3Pos150
Sueoka, Kazuhisa (末岡 和久) 3Pos056
Suetaka, Shunji (季高 駿士) **2Pos089**
Suetsugu, Shiro (末次 志郎) **1SFP-05**
3SBA-03
Sueyoshi, Noriyuki (末吉 紀行) 1Pos065
Suga, Michi (菅 倫寛) **3SCA-03**
Suga, Michihiro (菅 倫寛) 1SEP-02
Sugano, Eriko (菅野 江里子) 2Pos237
Sugano, Yasunori (菅野 泰功) 3SCA-06
Sugase, Kenji (菅瀬 謙治) 2SDP-05
Sugawa, Mitsuhiro (須河 光弘) 1Pos127
1Pos140
1Pos148
2Pos146*
2Pos178
Sugawara, Ko (菅原 皓) **2Pos292**
3Pos297
Sugawara, Takeshi (菅原 武志) **2Pos262**
2Pos264
3Pos269
Sugawara, Yoko (菅原 洋子) 2Pos008
Sughiyama, Yuki (杉山 友規) 1Pos278
1Pos280
Sugi, Takuma (杉 拓磨) 1Pos285
Sugiki, Toshihiko (杉木 俊彦) 3Pos059
Sugimori, Daisuke (杉森 大助) 3Pos011
Sugimori, Kimikazu (杉森 公一) 2Pos014
Sugimoto, Asako (杉本 亜砂子) **1SBP-06**
Sugimoto, Hiroshi (杉本 宏) 3Pos075
Sugimoto, Yasunobu (杉本 泰伸) 2Pos305
Sugimoto, Yuki (杉本 雄生) 1Pos192
Sugimura, Kaoru (杉村 薫) **1SAP-02**
Sugita, Masatake (杉田 昌岳) 2Pos129*
3Pos044
3Pos110
Sugita, Yuji (Sugita Yuji) 2Pos033
Sugita, Yuji (杉田 有治) 3SCA-06
3SCA-07
2Pos018
2Pos060
3Pos096
Sugiura, Kazunori (杉浦 一徳) **1Pos079**
Sugiura, Shinji (杉浦 慎治) 1Pos166*
Sugiyama, Hiroshi (杉山 弘) 2Pos315*
Sugiyama, Kohta (杉山 航太) 2Pos102
Sugiyama, Masaaki (杉山 正明) 3Pos033
Sugiyama, Shogo (杉山 翔吾) **1Pos287***
Sumi, Mariko (角 真理子) **2Pos206**
Sumi, Takuya (鷲見 拓哉) **1Pos046**
Sumi, Tomonari (墨 智成) **1Pos205**
2Pos029
Sumikama, Takashi (炭竈 享司) **1Pos217**
1Pos226
Sumikoshi, Kazuya (角越 和也) 3Pos268
Sumino, Ayumi (角野 歩) **1Pos226**
Sumino, Yutaka (住野 豊) 1Pos285
Suminokura, Makoto (墨野倉 誠) 1Pos024
Sunagawa, Naho (砂川 奈穂) **1Pos204**
Sunami, Tomoko (角南 智子) **1Pos119**
Supawich, Kamonprasertsuk (Supawich Kamonprasertsuk) 2Pos052*
Susaki, Moe (須﨑 萌) **3Pos073**
Suwa, Makiko (諏訪 牧子) 2Pos272
Suyama, Akira (陶山 明) 2Pos222
Suyama, Mikita (須山 幹太) **1SFA-02**
Suzuki, Haruo (鈴木 春男) 1Pos026
2Pos032
Suzuki, Hirofumi (鈴木 博文) **3Pos019**
Suzuki, Hiromi (鈴木 博実) **1Pos016**
Suzuki, Kano (鈴木 花野) 2Pos143*
3Pos141
Suzuki, Kazushi (鈴木 和志) 2Pos298*
Suzuki, Kazuya (鈴木 和也) **2Pos170**
Suzuki, Kenichi (鈴木 健一) 1SFP-00
Suzuki, Kenichi G.N. (鈴木 健一) 3Pos196
3Pos293
Suzuki, Kenshiro (鈴木 研士郎) 1Pos195
Suzuki, Madoka (鈴木 団) **2SDA-02**
Suzuki, Makoto (鈴木 誠) 2Pos121
3Pos040
3Pos042
3Pos108
3Pos134
Suzuki, Motoshi (Suzuki Motoshi) 3Pos009
Suzuki, Nobutake (鈴木 信勇) 1Pos297*
1Pos318*
3Pos115
Suzuki, Rika (鈴木 李夏) **2Pos194***
Suzuki, Ryo (鈴木 亮) 1Pos176
Suzuki, Ryo (鈴木 量) **3Pos287**
Suzuki, Ryuhei (Suzuki Ryuhei) 3Pos157
Suzuki, Ryuhei (鈴木 隆平) 1Pos152*
Suzuki, Ryuuhei (鈴木 隆平) 2Pos158
Suzuki, Sayaka (鈴木 沙弥香) 2Pos264
Suzuki, Seiichi (鈴木 誠一) **1Pos296**
Suzuki, Shoko (鈴木 翔子) 3Pos095
Suzuki, Takahiro (鈴木 崇弘) 1Pos176
3Pos041
Suzuki, Takayoshi (鈴木 孝禎) 1Pos069
Suzuki, Tatsuya (鈴木 達也) 3Pos087
Suzuki, Toshiharu (鈴木 俊治) 3SFA-00
3SFA-04
2Pos149
3Pos140
Suzuki, Yui (鈴木 由衣) **1Pos190***
Suzuki, Yuki (鈴木 勇輝) **3SGA-02**
1Pos203
1Pos213
2Pos309
Suzuki, Yuta (鈴木 悠太) 1Pos115
Suzuki, Yutaka (鈴木 穣) 2Pos135
Suzuki, Yuya (鈴木 裕也) **3Pos173**
Swadling, Jacob (Swadling Jacob) **1Pos123**
Sy, Redding (Sy Redding) 3Pos163
Tabata, Kazuhito (田端 和仁) **1SGP-01**
1Pos084
2Pos094
2Pos192*
3Pos074
3Pos175
Tabata, Kazuhito V. (田端 和仁) 2Pos257*
2Pos259
2Pos285
3Pos139
Tabata V., Kazuhito (田端 和仁) 2Pos081
Tachibanaki, Shuji (橘木 修志) **2Pos231**
Tachikawa, Masashi (立川 正志) **1Pos181**
3Pos276
Tachikawa, Takashi (立川 貴士) 1Pos010
1Pos256
Tada, Seiichi (Tada Seiichi) 1Pos081
Tadakuma, Hisashi (多田隈 尚史) **2SBA-04**
1Pos317
2Pos099
2Pos315*
Tagawa, Yoh-ichi (田川 陽一) **3SEA-05**
Taguchi, Hideki (田口 英樹) 2SDP-03
2Pos038
Taguchi, Mika (田口 美香) 3Pos124
Taguchi, Takahisa (田口 隆久) 3Pos230
Tahara, Hajime (田原 孟) **3Pos177**
Tahara, Tahei (Tahara Tahei) 1Pos126
Tahara, Tahei (田原 太平) 3SBA-01
2Pos051
Tahara, Yuhei (田原 悠平) 2Pos166*
Tai, Hulin (太 虎林) **1Pos067**
Taiji, Makoto (Taiji Makoto) 2Pos016
Taiji, Makoto (泰地 真弘人) 3Pos084
3Pos106
Tajima, Takuya (田島 拓也) 3Pos211
Takabe, Kyosuke (高部 響介) **3Pos178**
Takada, Shoji (Takada Shoji) 3Pos037
Takada, Shoji (高田 彰二) 1Pos044
1Pos109
1Pos118
1Pos144
2Pos059
2Pos098*
2Pos104
2Pos107
2Pos109
3Pos052
3Pos212
Takagi, Hiroaki (高木 拓明) **3Pos237**
Takagi, Jun (高木 潤) 2Pos170
Takagi, Junichi (高木 淳一) 3Pos001
3Pos005
Takagi, Maki (高木 眞希) 1Pos296
Takagi, Shin (高木 新) 2Pos254
Takagi, Toshiyuki (高木 俊之) 1Pos166*
1Pos210
2Pos217
3Pos086
3Pos089
Takaharu, Okajima (岡嶋 孝治) 1Pos300
Takahashi, Daiki (髙橋 大輝) 3Pos010
Takahashi, Daisuke (高橋 大輔) 2SDP-04
1Pos048*
Takahashi, Hiroki (高橋 弘喜) 2Pos106
2Pos135
Takahashi, Hiroshi (高橋 浩) 1Pos073
1Pos207
1Pos210
2Pos217
3Pos086
3Pos089
Takahashi, Hiroto (高橋 泰人) 3Pos188
3Pos193
Takahashi, Keisuke (高橋 慶祐) 2Pos233
Takahashi, Ken-ichi (高橋 健一) 2Pos029
Takahashi, Kosuke (高橋 孝輔) 3Pos043
Takahashi, Kosuke (髙橋 孝輔) **3Pos207**
Takahashi, Masayuki (高橋 正行) 1Pos154
Takahashi, Megumi (高橋 めぐみ) 2Pos254
Takahashi, Naoki (高橋 直樹) **3Pos130**
Takahashi, Nozomu (高橋 望) **3Pos079**
Takahashi, Ryoko (高橋 涼子) 3Pos085
Takahashi, Satoshi (高橋 聡) 1Pos083*
1Pos110*
1Pos113*
2Pos052*
2Pos095*
Takahashi, Shin-Ichiro (高橋 伸一郎) 1Pos166*
Takahashi, Takuya (高橋 卓也) 1Pos056
2Pos008
2Pos123*
2Pos126*
2Pos268
Takahashi, Takuya (高橋 拓哉) 1Pos201
Takahashi, Yohei (高橋 洋平) 3Pos186
Takahashi, Yuka (高橋 優嘉) **3SDA-02**
2Pos153*
Takahashi, Yusuke (髙橋 裕輔) 2Pos001*
Takahiro, Watanabe-Nakayama (中山 隆宏) 1Pos306*

Takai, Madoka (高井 まどか) 2Pos052*
Takaine, Masak (高稲 正勝) 3Pos149
3Pos153
3Pos294
Takaki, Kotaro (高木 航太郎) 1Pos197
Takamiya, Kazunori (高宮 一徳) **1Pos173**
Takamori, Aya (高森 綾) 2Pos164
Takamuku, Yuuki (高椋 勇樹) 3Pos097
Takano, Hiroshi (高野 宏) 2Pos034*
2Pos037
Takano, Mitsunori (高野 光則) 1Pos115
1Pos128
2Pos124
3Pos039
3Pos062
3Pos111
Takano, Yu (鷹野 優) **1SEP-01**
2Pos015
2Pos044
3Pos083
Takano, Yuta (高野 勇太) 3Pos073
Takao, Daisuke (高尾 大輔) **1SEA-05**
Takao, Keizo (高雄 啓三) **2SEA-02**
Takao, Masato (高尾 真登) 3Pos170
Takaoka, Katsuyoshi (高岡 勝吉) 2Pos176
Takaori, Akifumi (高折 晃史) 2Pos103*
Takaramoto, Shunki (宝本 俊輝) **2Pos080**
Takasato, Minoru (高里 実) **3SEA-04**
Takashima, Akito (高嶋 明人) 1Pos074
Takashima, Ichiro (高島 一郎) 3Pos234
Takashima, Yoshinori (高島 義徳) 3Pos286
Takasu, Masako (高須 昌子) 2Pos014
Takata, Takumi (高田 匠) 3Pos033
Takauchi, Hiroki (高内 大貴) 2Pos296
Takauchi, Hiroki (高内 広貴) 1Pos309
Takayama, Jun (高山 順) **2Pos137**
3Pos280
Takayama, Yuki (高山 裕貴) 3Pos243
Takayanagi, Masayoshi (高柳 昌芳) 2Pos031
Takebayashi, Kazutoshi (竹林 和俊) **3Pos071**
Takeda, Hanae (武田 英恵) **1Pos091**
Takeda, Hironori (竹田 弘法) 1Pos095*
Takeda, Hiroyuki (武田 洋幸) 1SEA-06
Takeda, Itaru (竹田 至) 3Pos073
Takeda, Kimitoshi (武田 公利) **3Pos246**
Takeda, Mitsuhiro (武田 光広) 1Pos039*
1Pos311*
2Pos076*
Takeda, Seiji (武田 晴治) **3Pos056**
Takeda, Shunsuke (武田 駿介) **2Pos279**
Takeda, Shuuichi (武田 修一) 3Pos125
Takeda, Tetsuya (竹田 哲也) 1Pos233
2Pos049
Takeda, Yusuke (武田 友祐) 1Pos012
Takei, Kengo (武井 健吾) **3Pos265**
Takei, Kohji (竹居 孝二) 1Pos233
2Pos049
Takei, Toshiki (武居 俊樹) 2Pos054
Takekawa, Norihiro (竹川 宜宏) **3Pos171**
Takemori, Shigeru (竹森 重) 3Pos124
3Pos128
Takemura, Akihiro (竹村 明洋) 2Pos238
Takemura, Kazuhiro (竹村 和浩) **3SBA-03**
Takenaka, Koshi (竹中 康司) 1Pos073
1Pos076
Takenaka, Yoshiko (武仲 能子) **3SGA-03**
Takeoka, Shinji (武岡 真司) 3Pos211
Takeshita, Kohei (竹下 浩平) 1Pos216
Takeshita, Naohiro (竹下 真広) 1Pos209
Takeuchi, Nao (竹内 奈央) **3Pos006**
Takeuchi, Nobuto (竹内 信人) **2Pos260**
Takeuchi, Saki (竹内 早希) 1Pos167
Takeuchi, Shoji (竹内 昌治) 2Pos157
Takigaura, Shohei (滝ヶ浦 尚平) **3Pos093**
Takiguchi, Kingo (滝口 金吾) 1Pos182
Takiguchi, Kingo (瀧口 金吾) 1Pos188*
2Pos226*
Takimoto, Yuki (瀧本 祐樹) **2Pos126***
Takinami, Yoshihiko (瀧浪 欣彦) 2Pos076*
Takinoue, Masahiro (瀧ノ上 正浩) **3SEA-03**
1Pos121
1Pos124
1Pos203
1Pos213
1Pos291*
1Pos292*
Takubo, Naoko (田久保 直子) **2Pos133**
Tama, Florence (Tama Florence) 3SCA-01
2Pos009
2Pos011
2Pos046*
3Pos009
Tama, Florence (タマ フロハンス) 3Pos036
Tamai, Nobutake (玉井 伸岳) **1Pos209**
3Pos208
Tamaru, Shoya (田丸 翔也) **1Pos255***
Tambi, Richa (Tambi Richa) **2Pos016**
Tamegai, Misato (為我井 美聡) **2Pos177**
Tamogami, Jun (田母神 淳) 1Pos241
1Pos242
Tamotsu, Akihiro (保 皓大) **3Pos313**
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Yajima, Junichiro (矢島 潤一郎) 1Pos140, 1Pos148, 2Pos157, 2Pos178
Yajima, Kaho (矢嶋 香歩) 3Pos095
Yakushiji, Fabiana L. (ヤクシジ ファビアナリカ) 3Pos141
Yakushiji, Fabiana Lica (薬師寺 Lica Fabiana) 3Pos001
Yakushiji, Fabiana. Lica (薬師寺 ファビアナ・リカ) 2Pos143*
Yakushiji, Lica Fabiana (薬師寺 リカ ファビアナ) 1Pos132*
Yamada, Daichi (山田 大智) 2Pos101, **3Pos273**
Yamada, Hiroshi (山田 浩司) **1Pos233**, 2Pos049
Yamada, Kazunori (山田 和範) 2SEA-03, **3Pos274**
Yamada, Keisuke (山田 啓祐) **3Pos275**
Yamada, Kenta (Yamada Kenta) 2Pos033
Yamada, Kenta (山田 健太) 1Pos249, **3Pos096**
Yamada, Masafumi (山田 正文) 3Pos153
Yamada, Masami (山田 雅巳) 2Pos175
Yamada, Takahiro (山田 崇広) 3Pos201
Yamada, Yasuyuki (山田 康之) 1Pos130*
Yamada, Yurika (山田 有里佳) **3Pos122**
Yamada, Yutaro (山田 祐太郎) 2SGA-05
Yamagata, Yuriko (山縣 ゆり子) 2Pos027
Yamagishi, Akihiko (山岸 明彦) 1SGA-03
Yamagishi, Ayana (山岸 彩奈) 1Pos298, 3Pos073
Yamagishi, Mai (山岸 舞) 1Pos297*, 1Pos318*, 3Pos113, **3Pos115**
Yamaguchi, Erika (山口 絵里花) 3Pos171
Yamaguchi, Hiroshi (山口 博史) **1SEA-06**
Yamaguchi, Hiroshi (山口 博司) 1Pos046
Yamaguchi, Hitomi (山口 瞳) 2Pos076*
Yamaguchi, Maki (山口 眞紀) 3Pos124
Yamaguchi, Naoya (Yamaguchi Naoya) 2Pos187
Yamaguchi, Shin (山口 真) 2Pos157, 2Pos178
Yamaguchi, Shoichi (山口 祥一) 1Pos212
Yamaguchi, Takaya (山口 貴也) 3Pos040, **3Pos134**
Yamaguchi, Tomoyuki (山口 智之) 3Pos200
Yamaguchi, Toshio (山口 敏男) 2Pos120
Yamaguchi, Yuki (山口 雄輝) 3Pos299
Yamahara, Kazuaki (山原 一晃) 3Pos093
Yamakoshi, Daiki (山越 大希) 1Pos128
Yamamori, Yu (山守 優) **3Pos045**
Yamamoto, Akihisa (山本 暁久) 3Pos286
Yamamoto, Ayumi (山本 愛弓) 1Pos195
Yamamoto, Daisuke (山本 大輔) 3Pos038, **3Pos210**
Yamamoto, Eiji (山本 詠士) **2Pos212**
Yamamoto, Johtaro (山本 条太郎) **2SEP-02**, 2Pos100, 2Pos282*
Yamamoto, Jotaro (山本 条太郎) 1Pos295
Yamamoto, Junpei (山元 淳平) 1Pos068, 2Pos101, 3Pos250, **3Pos251**
Yamamoto, Kentaro (山本 健太郎) 1Pos199, 3Pos184
Yamamoto, Mami (山本 真実) **2Pos088**
Yamamoto, Masaki (Yamamoto Masaki) 2Pos077
Yamamoto, Masaki (山本 雅貴) 1SEP-02, 1Pos320, 2Pos303, 3Pos243
Yamamoto, Masamichi (山本 正道) 2Pos294
Yamamoto, Naoki (山本 直樹) **2Pos122**
Yamamoto, Reiko (山本 れいこ) 2Pos099
Yamamoto, Ryoma (山本 龍眞) **1Pos139**
Yamamoto, Ryota (山本 良太) **1Pos013***
Yamamoto, Sakiyo (山本 祥世) 3Pos095
Yamamoto, Takamasa (山本 貴柾) **2Pos110**
Yamamoto, Takashi (山本 卓) 2Pos233
Yamamoto, Tetsuya (山本 哲也) **2Pos117**, 3Pos289
Yamamoto, Tsubasa (山本 翼) 2Pos055*
Yamamoto, Yuki (山本 悠生) 2Pos269
Yamamoto, Yuta (山本 悠太) 2Pos134
Yamanaka, Akihiro (山中 章弘) 2Pos253
Yamanaka, Masaru (山中 優) 2Pos051
Yamanaka, Shin'nosuke (山中 信之介) 1Pos308, **3Pos291**, 3Pos292
Yamane, Tsutomu (山根 努) **1Pos009**
Yamanishi, Yoshihiro (山西 芳裕) 1Pos268
Yamano, Megumi (山野 めぐみ) 2Pos068
Yamano, Nami (山野 奈美) **1Pos252***
Yamano, Yu (山野 優) 2Pos173
Yamano, Yumiko (山野 由美子) 2Pos230
Yamaoki, Yudai (山置 佑大) 2Pos067*
Yamasaki, Kazuhiko (山崎 和彦) 3Pos024, 3Pos278
Yamasaki, Takashi (山崎 孝史) 2SDP-02
Yamashiro, Shunsuke (山城 竣介) **2Pos116**

Yamashita, Daisuke (山下 大輔) **2Pos114***
Yamashita, Eiki (山下 栄樹) 3SFA-02
1Pos085
1Pos096
1Pos286
2Pos003
Yamashita, Hayato (山下 隼人) **3Pos094**
Yamashita, Hiroaki (山下 宏明) 3Pos126
Yamashita, Hironobu (山下 弘展) **2Pos081**
Yamashita, Kazuto (山下 和人) 1Pos013*
Yamashita, Keitaro (Yamashita Keitaro) 2Pos077
Yamashita, Keitaro (山下 恵太郎) 1SEP-02
Yamashita, Saki (山下 紗季) **2Pos197**
Yamashita, Takahiro (山下 高廣) 1Pos104
2Pos230
2Pos233
2Pos235
Yamashita, Yoshiki (山下 良樹) 1Pos005
3Pos034
Yamashita, Yuki (山下 雄己) 2Pos042
Yamashita-Kawano, Emi (山下（川野）絵美) 2Pos234
Yamato, Ichiro (山登 一郎) 3Pos141
Yamato, Masayuki (大和 雅之) 1Pos300
Yamauchi, Marie (山内 真梨江) **2Pos251***
Yamauchi, Shun (山内 舜) 2Pos079
Yamauchi, Yumeka (山内 夢叶) **2Pos239**
2Pos252
Yamazaki, Jun (山崎 純) 2SDA-05
Yamazaki, Masahito (山崎 昌一) 1Pos196
2Pos205
2Pos207
2Pos208
2Pos209
2Pos216
Yamazaki, Masahito (山崎、昌一、) 1Pos194
1Pos198
Yamazaki, Megumi (山崎 萌) 1Pos199
Yamazaki, Toshio (山崎 俊夫) 1Pos055
Yamazaki, Yoichi (山崎 洋一) **1Pos246**
1Pos247
2Pos066
3Pos028
3Pos032
Yamazaki, Yousuke (山崎 陽祐) 1SEA-06
Yamazawa, Toshiko (山澤 徳志子) 3Pos124
Yamazawa, Toshiko (山澤 德志子) 3Pos002
3Pos300
Yan, Jie (Yan Jie) 3Pos164
Yan, Xiyun (Yan Xiyun) **1SCP-05**
Yan, Zhiqiang (Yan Zhiqiang) 1Pos095*
Yanagawa, Fumiki (柳川 史樹) 1Pos166*
Yanagawa, Masataka (柳川 正隆) **1Pos104**
2Pos202
2Pos230
Yanagi, Takashi (柳 昂志) **2Pos196**
Yanagida, Ryota (柳田 亮太) 2Pos144
Yanagida, Toshio (柳田 敏雄) 3Pos151
3Pos165
Yanagisawa, Miho (柳澤 実穂) 1Pos121
1Pos124
1Pos179*
1Pos213
2Pos211
Yanaka, Saeko (谷中 冴子) **3Pos055**
Yang, Huiran (楊 恵然) 2Pos049
Yang, Lee-Wei (楊 立威) **2Pos108**
Yang, Pinfen (Yang Pinfen) 2Pos180
Yang, Zhuohao (楊 倬皓) 2Pos069
Yano, Kouichi (矢野 晃一) 2Pos048
Yano, Naomine (矢野 直峰) 3SFA-02
1Pos096
Yano, Shunsuke (矢野 俊介) **1Pos076**
Yano, Yoshiaki (矢野 義明) **3Pos090**
Yao, Min (姚 閔) 2Pos007*
Yarovsky, I (Yarovsky I) 2Pos219
Yasuda, Kenji (安田 賢二) 1Pos277
2Pos287
2Pos318
3Pos130
3Pos229
3Pos308
3Pos309
Yasuda, Satoshi (安田 哲) 3Pos092
Yasuda, Satoshi (安田 賢司) **1Pos100**
3Pos058
3Pos097
Yasugi, Fumitaka (八杉 文隆) 1Pos035
Yasuhara, Kazuma (安原 主馬) 1Pos204
3Pos203
Yasui, Masato (安井 真人) **2Pos299**
Yasunaga, Takuo (安永 卓生) 2SGA-04
1Pos007
1Pos153
1Pos156
2Pos175
2Pos302
3Pos159
3Pos301
Yasuoka, Kenji (泰岡 顕治) 2Pos212
Yata, Haruna (矢田 はる奈) **1Pos251**
Yawo, Hiromu (八尾 寛) **2SAA-02**
2Pos041
Ye, Jing Yong (イエ ジンヤン) 2Pos118
Yoda, Takao (依田 隆夫) 1Pos267
Yogo, Rina (與語 理那) 3Pos055
Yohda, M. (Yohda M.) 3Pos027
Yokawa, Satoru (横川 慧) 1Pos176
Yoko-o, Takehiko (横尾 岳彦) 1Pos155
Yokogawa, Mariko (横川 真梨子) 3SCA-04
Yokokawa, Ryuji (横川 隆司) 2Pos156
3Pos156
Yokosuka, Tadashi (横須賀 忠) **1SFP-04**
Yokota, Hiroaki (横田 浩章) **1Pos111**
1Pos223
Yokota, Homare (横田 誉) 1Pos012
Yokota, Kiyonori (横田 恭宣) 2Pos220
Yokota, Ryo (横田 亮) 2Pos186
3Pos282
3Pos317
Yokota, Ryuichi (横田 龍一) **2Pos144**
Yokoyama, Hiroyuki (横山 弘之) **2SAP-03**
Yokoyama, Ken (横山 謙) 1SDP-02
2Pos145*
3Pos003
3Pos006
3Pos088
3Pos135
3Pos290
Yokoyama, Shigeyuki (横山 茂之) 1Pos242
3Pos095
3Pos141
Yokoyama, Yasunori (横山 泰範) 1Pos073
1Pos076
Yonamine, Yusuke (与那嶺 雄介) **3SGA-04**
Yoneda, Hironori (米田 博紀) **2Pos048**
Yoneda, Shigetaka (米田 茂隆) 1Pos026
2Pos032
3Pos047
Yonekura, Koji (米倉 功治) 1Pos005
3Pos034
Yoneyama, Yosuke (米山 鷹介) 1Pos166*
Yonezawa, Tomoko (米澤 智子) 1Pos224
Yonezawa, Yasushige (米澤 康滋) 2Pos044
Yoon, Dong H. (Yoon Dong H.) 3Pos312
Yoon, Dong Hyun (尹 棟鉉) 2Pos273
Yoon, Tae-Young (Yoon Tae-Young) **2SCA-02**
Yoon, Tae-Young (尹 兌榮) 2SCA-00
Yoshida, Aoi (吉田 葵) 1Pos185
Yoshida, Hikaru (吉田 光) 1Pos131
Yoshida, Hiroshi (吉田 寛) **2Pos136**
Yoshida, Kazuaki (吉田 一章) 1Pos007
Yoshida, Kazuho (吉田 一帆) **2Pos255**
Yoshida, Kazunari (吉田 一也) 1Pos074
Yoshida, Keito (吉田 佳人) 1Pos247
Yoshida, Keito (吉田 桂人) 3Pos028
3Pos032
Yoshida, Kenta J. (吉田 謙太) 3Pos196
Yoshida, Kentaro (吉田 健太郎) **2Pos135**
Yoshida, Koji (吉田 亨次) **2Pos120**
Yoshida, Kotaro (吉田 浩太郎) **1Pos120**
Yoshida, Masaki (吉田 昌樹) 3Pos258
Yoshida, Masasuke (吉田 賢右) 1Pos129
2Pos149
Yoshida, Norio (吉田 紀生) 2Pos090
3Pos239
Yoshida, Ryo (吉田 亮) 2Pos133
3Pos228
Yoshida, Satoshi (吉田 知史) 3Pos294
Yoshida, Shin (吉田 慎) 3Pos316
Yoshida, Shintaro (吉田 晋太郎) **2Pos076***
Yoshidome, Takashi (吉留 崇) **1Pos319**
2Pos085*
Yoshihara, Toshitada (吉原 利忠) 2Pos297*
Yoshikaie, Kunihito (吉海江 国仁) 3SCA-06
3SCA-07
Yoshikawa, Hiroshi (吉川 洋史) 1Pos166*
1Pos175*
Yoshikawa, Kenichi (吉川 研一) 1Pos122
1Pos169
1Pos185
1Pos188*
1Pos293
1Pos294
2Pos310*
3Pos118
3Pos204
3Pos266
Yoshikawa, Shinya (Yoshikawa Shinya) 2Pos077
Yoshikawa, Shinya (吉川 信也) 3SFA-02
1Pos085
1Pos086
1Pos087
1Pos088
1Pos089*
1Pos096
2Pos079
Yoshikawa, Yuko (吉川 祐子) 2Pos310*
3Pos118
Yoshikawa, Yuko (吉川 裕子) 1Pos122
3Pos266
Yoshimatu, Daiki (吉松 大輝) 1Pos201
Yoshimura, Hideyuki (吉村 英恭) 3Pos306
Yoshimura, Kazuki (吉村 一輝) **3Pos026**
Yoshimura, Midori (吉村 翠) **1Pos164**
Yoshinaga, Sosuke (吉永 壮佐) 1Pos039*
1Pos311*
2Pos076*
Yoshino, Masaru (吉野 賢) 3Pos086
Yoshino, Ryunosuke (吉野 龍ノ介) 2Pos012
Yoshioka, Daisuke (好岡 大輔) **1Pos191**
Yoshizaki, Satoru (吉崎 慧) **1Pos058**
Yoshizawa, Keiko (慶澤 景子) 2Pos005
Yoshizawa, Ryo (吉澤 亮) **2Pos202**
Yoshizawa, Susumu (吉澤 晋) 2Pos253
3Pos240
3Pos245
You, Huijuan (游 慧娟) **3Pos164**
Yu, Isseki (優 乙石) **2Pos060**
Yu, Kai (Yu Kai) 2Pos105
Yu, L.-J. (于 龍江) 2Pos243
Yumiba, Takahiro (弓場 貴広) 3Pos250
Yumoto, Tenji (湯本 天嗣) **1Pos155**
Yunoki, Kaori (柚木 芳) 1Pos039*
Yura, Kei (由良 敬) 3Pos273
Yura, Yuki (由良 優季) 2Pos248
Yuxin, Ye (Yuxin Ye) 2Pos007*
Yuzuriha, Naoya (杠 直哉) **1Pos311***
Zakharian, Eleonora (Zakharian Eleonora) 2SDA-05
Zamani, Alemeh (Zamani Alemeh) **2Pos041**
Zarco - Zavala, Mariel (Zarco - Zavala Mariel) **3Pos140**
Zhang, Mingfeng (Zhang Mingfeng) 1Pos095*
Zhang, Yi (張 翼) **1Pos084**
2Pos094
3Pos074
3Pos112
3Pos114
Zhang, Yubai (張 羽白) 1Pos233
Zhang, ZhiYuan (張 致遠) **2Pos142***
Zhu, Tong (朱 とん) **1Pos189**
Zierep, Paul (Zierep Paul) 1Pos010
Zinchenko, Anatoly (Zinchenko Anatoly) 1Pos190*

第54回 日本生物物理学会年会

浜松ホトニクス株式会社
ランチョンセミナー

◇ プログラムNo.1LA
◇ 日時:2016年11月25日(金) 11:45 ～ 12:35
◇ 会場:A会場 (中ホール200)

演題1

高速蛍光一分子イメージング法とその応用

「Visualization of single fluorescent molecules in living cells with milliseconds exposure.」

岡田 康志 先生

国立研究開発法人理化学研究所
生命システム研究センター 細胞極性統御研究チーム

演題2

浜松ホトニクスの最新イメージング製品

伊東 克秀

浜松ホトニクス株式会社 システム事業部

浜松ホトニクス株式会社 URL: http://www.hamamatsu.com
システム事業部 システム営業推進部
〒431-3196 静岡県浜松市東区常光町812
TEL:(053)431-0150 FAX:(053)433-8031
E-mail: sales@sys.hpk.co.jp

第54回日本生物物理学会年会

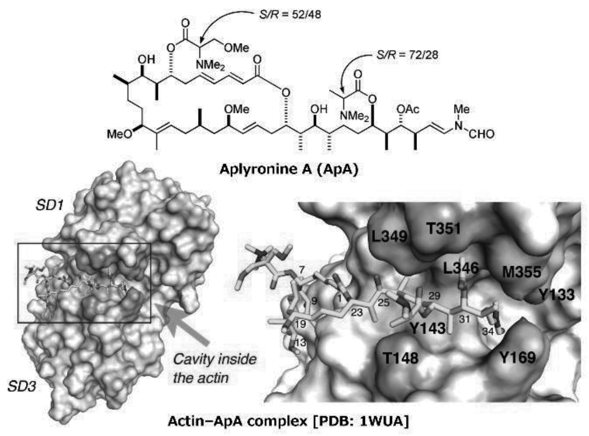
株式会社菱化システム ランチョンセミナー
分子間相互作用の解析と応用

日時： １１月２５日（金）　11:45 ～ 12:35
場所： Ｂ会場（中会議室２０２）

タンパク質間相互作用を誘導する有機小分子

筑波大学 数理物質系 化学域 准教授　**北 将樹**

近年、タンパク質間相互作用を制御する有機小分子が、天然物および合成化合物ライブラリから多数見いだされており、新たな作用機序を有する医薬品などへの応用が期待されています。海洋天然物アプリロニンＡ は、２つの細胞骨格タンパク質、アクチンとチューブリン間の相互作用を誘導し、微小管ダイナミクスを阻害するユニークな抗腫瘍性物質です。本ランチョンセミナーでは、ケミカルプローブ法および統合計算化学システム MOE によるシミュレーションを組み合わせたアプリロニン Ａ の結合様式解析、および新規活性リガンドの創出を目指した分子設計と構造活性相関研究などについて、これまでの取り組みを発表します。



タンパク質間相互作用の解析

株式会社菱化システム 科学技術システム事業部 **木村 嘉朗**

タンパク質間相互作用（PPI）は、免疫系、シグナル伝達、酵素阻害などにおいて、重要な役割を果たしており、生命科学分野における主要な研究対象の一つです。統合計算化学システム MOE には、タンパク質同士のドッキングシミュレーション、タンパク質表面の特徴づけ、タンパク質間の水素結合の検出など、PPI 解析に有用なさまざまな機能を搭載しています。本セミナーでは、これらの機能の応用例を紹介します。

Ryoka Systems Inc.

株式会社菱化システム
科学技術システム事業部
〒131-0045　東京都墨田区押上 1-1-2
東京スカイツリーイーストタワー

E-mail: support@rsi.co.jp
URL: http://www.rsi.co.jp/
TEL: 03-6830-9724
FAX: 03-5610-1161

Luncheon Seminar at the 54rd Annual Meeting of Biophysical Society of Japan

November 25 (Fri), 11:45 ～ 12:35, Room 20I（C）

1. New issues in the wwPDB and the PDBj

wwPDB と PDB における新しい話題について

Haruki Nakamura, Institute for Protein Research, Osaka University

The PDBj (PDB Japan, http://pdbj.org/) is the representative archive of macromolecular structural data by X-ray crystallography, NMR and cryo-EM, processing the deposited data from researchers in Asian and Middle-east regions, as one of the four members of the wwPDB (worldwide PDB, http://wwpdb.org/). In order to promote the recent “Data Science” , wwPDB now starts several new policies: (i) Usage of ORCID (Open Researcher and Contributor ID: http://orcid.org/) as the standard, (ii) Retiring the flat PDB format, (iii) Making the validation report more informative for NMR and cryo-EM data, (iv) Introduction of a versioning system, and (v) Change of the current 4-characters PDBID. Those issues will be introduced at the Luncheon Seminar.

2. Electron microscopy data in PDB and EMDB - deposition and browsing

EMDB と PDB における電子顕微鏡データ – 登録と閲覧

Hirofumi Suzuki, Institute for Protein Research, Osaka University

The number of structure data analyzed by electron microscopy (EM) is increasing. Atomic models and density maps by EM are stored in PDB and EM data bank (EMDB), respectively.

3. New generation of NMR analysis assisted by Deep Learning and highly sophisticated Web-tools developed by PDBj-BMRB

PDBj-BMRB により開発された深層学習により支援された次世代 NMR 解析と高度化された Web ツール

Naohiro Kobayashi, Institute for Protein Research, Osaka University

NMR is a very unique and useful technique to analyze structure, dynamics and interaction with ligand of biomolecules in an atomic resolution easily and quickly. One of the largest burden on the NMR analysis would be identification and assigning very few signals among a huge number of noises and artifacts. Several programs are available to automatically assign the NMR signals, however none of them can start from NMR signal identification. Although NMR signal identification in multi-dimensional NMR spectra has been known to be a key job, many NMR scientists still prefer to spend a lot of time for preparing NMR signal tables manually. We have developed new generation of program MagRO which can discriminate weak but important NMR signals with huge number of artifacts and noises assisted by image recognition technology of Deep Neural Networks. We are going to demonstrate that our program has finally got “eyesight” like a human to enable highly automated, correct and fast calculation for sequence specific backbone signal assignments of protein.

We also will show our new web-tool that we have developed recently and released. The web-tool allows users easily and intuitively to search for a wide variety of information related to the studies on biomolecules: free keyword and sequence search to provide rich information such as NMR data (BMRB), structure coordinates (PDB), homologues and functions (UniProt), interactors (IntAct). The new NMR data page is designed to represent detail of the NMR parameters such as chemical shifts and dynamics data by highly sophisticated graphical interface as user can visually inspect interested domain region easily and straightforwardly.

Protein Data Bank Japan
http://pdbj.org/

Institute for Protein Research, Osaka University,
3-2 Yamadaoka, Suita, Osaka 565-0871, Japan
TEL: +81-(0)6-6879-4311

第54回 日本生物物理学会年会
The 54th Annual Meeting of the Biophysical Society of Japan

オリンパス(株)ランチョンセミナー
プログラムNo:1LD

オートファジータンパク質群の動的相互作用と分子集合形態の解析

日時 11月25日[金] 11:45〜12:35

会場 つくば国際会議場 中ホール 300(D会場)

演者 山本 林 先生

東京大学大学院 医学系研究科 分子生物学分野 講師

TIRF顕微鏡による偏射蛍光イメージングや高速原子間力顕微鏡を用いて酵母におけるオートファジーメカニズム解明に取り組む山本 林先生。本ランチョンセミナーでは山本先生を講師にお迎えし、今年7月に「Developmental Cell」で発表された研究成果などを中心に、オートファジー研究における最新のイメージングについてご紹介します。

- 近年における代表論文 -

1. Yamamoto, H*, Fujioka Y*, Suzuki SW*, Noshiro D, Suzuki H, Kondo-Kakuta C, Kimura Y, Hirano H, Ando T, Noda NN, Ohsumi Y. The Intrinsically Disordered Protein Atg13 Mediates Supramolecular Assembly of Autophagy Initiation Complexes. Dev Cell. 2016 Jul 11;38(1):86-99. *co-first authors

2. Yamamoto, H*, Shima T, Yamaguchi M, Mochizuki Y, Hoshida H, Kakuta S, Kondo-Kakuta C, Noda NN, Inagaki F, Itoh T, Akada R, Ohsumi Y*. The Thermotolerant Yeast Kluyveromyces marxianus Is a Useful Organism for Structural and Biochemical Studies of Autophagy. J Biol Chem. 2015 Dec 4;290(49):29506-18. *correspondence

3. Suzuki SW, Yamamoto, H*, Oikawa Y, Kondo-Kakuta C, Kimura Y, Hirano H, Ohsumi Y*. Atg13 HORMA domain recruits Atg9 vesicles during autophagosome formation. Proc Natl Acad Sci U S A. 2015 Mar 17;112(11):3350-5. *correspondence

4. Fujioka, Y.*, Suzuki, S.W.*, Yamamoto, H.*, Kondo-Kakuta, C., Kimura, Y., Hirano, H., Akada, R., Inagaki, F., Ohsumi, Y., and Noda, N.N. Structural basis of starvation-induced assembly of the autophagy initiation complex. Nat Struct Mol Biol. 2014 Jun;21(6):513-21. *co-first authors

5. Yamamoto, H., Kakuta, S., Watanabe, T.M., Kitamura, A., Sekito, T., Kondo-Kakuta, C., Ichikawa, R., Kinjo, M., and Ohsumi, Y. Atg9 vesicles are an important membrane source during early steps of autophagosome formation. J Cell Biol. 2012 Jul 23;198(2):219-33.

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日時: 2016年 11月26日（土）11:45～12:35
場所: A会場（つくば国際会議場 中ホール200）

■超高感度カメラ キヤノン ME20F-SH 紹介
キヤノンマーケティングジャパン株式会社

■超高感度・超高視野・超低ノイズカメラを用いたバイオイメージング
永井 健治 先生 (大阪大学 産業科学研究所 生体分子機能科学研究分野)

蛍光タンパク質や化学発光タンパク質で構築されたプローブを用いたバイオイメージングにより、細胞や組織、個体内で繰り広げられている様々な生命現象を可視化し、研究することが可能になっている。バイオイメージングは色々な機器を駆使して行われるが、被写体の像を取得するカメラはその中でも要のユニットである。蛍光や化学発光などの暗い試料のイメージングでは CMOS カメラや EMCCD カメラがよく使われるが、最近では、従来の科学計測用カメラメーカー以外から、独自の技術を生かした CMOS カメラでバイオイメージング市場へ新規参入するケースが散見されるようになった。このようにバイオイメージング用カメラが多様化しつつあるものの、重要なスペックである感度、画素数、素子サイズ、フレームレート等がすべてベストであるカメラは存在しないため、イメージングの実施においては観察目的に即した特性のカメラを選択することが肝要である。本セミナーでは、キャノン社製超高感度・超高視野・超低ノイズカメラのパフォーマンスについて紹介するとともに、我々の研究室で開発した蛍光タンパク質プローブや化学発光タンパク質プローブを用いたイメージングの実施例から、蛍光タンパク質標識生体分子や化学発光の高感度イメージング、そして細胞の高精細・広視野イメージングを取り上げ、観察対象の可視化に要求されるスペックを解説する。

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2LB：第54回日本生物物理学会年会 FEIランチョンセミナー

The Revolution of Cryo-TEM

- 日時: 2016年11月26日（土）11:45 – 12:35
- 会場: B会場（中会議室202）
- 講演者: 重松　秀樹　先生
  （理化学研究所ライフサイエンス技術基盤センター）
- 演題: クライオ電子顕微鏡法における革新と我々の取り組み
- 要旨:

クライオ電子顕微鏡（cryo-EM）という単語がトップジャーナルを賑わすようになってそろそろ3年がすぎようとしている。構造データベースへの登録数はうなぎ登りという表現がピッタリの上昇曲線を描いており、今後も分野としての成長が期待される。クライオ電子顕微鏡研究者により、モデリングが可能な分解能の構造が発表される状況が続き、現在ではX線結晶構造解析で著名な研究者がクライオ電子顕微鏡を導入し、結晶化が困難な試料や分解能の伸びなかった試料の構造を発表する状況がはじまってきている。
本講演では、結晶化を必要とせず、必要量も比較的少量ですむクライオ電子顕微鏡単粒子解析での高分解能構造取得へ向けた我々の試みを紹介する。2014年に理化学研究所ライフサイエンス技術基盤研究センター横浜キャンパスに導入されたFEI Tecnai ArcticaとFalcon2の組み合わせにより長時間かつ高画質の画像取得の自動化が可能となり、巨大分子複合体の高分解能構造を取得することに成功している。

Human Frontier Science Program

«開催趣旨»
ＨＦＳＰ（ヒューマン・フロンティア・サイエンス・プログラム）は、ライフサイエンス分野における革新的な国際共同研究を推進するため、1989年に創設された国際プロジェクトです。本プログラムでは、基礎的な生物機能を解明するため、広く異なった分野の専門知識を融合することに重点をおいています。研究対象とする生物機能としては、細胞構造における詳細な分子状態から、神経システム科学における複雑な相互作用にまで及びます。ＨＦＳＰがサポートするフロンティアサイエンス分野には、情報生命科学者や生物物理学者らの参画も次第に一定規模を占めるようになっている中で、物理学分野からの参画もまた多く見られます。

今回の講演の中では、ＨＦＳＰの共同研究グラントについて、応募申請時におこしがちな失敗や、採択されやすい事例についての説明を交えながら意見交換をします。課題解決のための様々な専門領域や科学文化による「新規性」や革新的発想を備えた研究チームが、好結果を挙げています。また、若手研究者を支援するＨＦＳＰポスドク・フェローシップについてもご紹介いたします。

日時 ： 2016年11月26日（土）11：45－12：35

場所 ： 第54回日本生物物理学会年会 ランチョンセミナー C会場

演者 ： Dr. Geoff RICHARDS

Director of Research Grants The International Human Frontier Science Program Organization (HFSPO), Strasbourg, FRANCE

演題 ： “Broadening your horizons – international collaboration in the life sciences"

説明会プログラム

11：45～11：50	開会挨拶、趣旨説明	日本医療研究開発機構 国際事業部国際連携研究課
11：50～12：00	HFSP機構の概要説明、講演者紹介	水間　英城 国際ヒューマン・フロンティア・サイエンス・プログラム機構　事務局次長
12：05～12：25	講　演	Dr. Geoff RICHARDS 国際ヒューマン・フロンティア・サイエンス・プログラム機構 研究グラント部門部長
12：25～12：35	参加者との意見交換、質疑応答	

＊ヒューマン・フロンティア・サイエンス・プログラム（HFSP）とは
１９８７年のヴェネツィア・サミットにて日本政府が提唱した国際プロジェクトで、生体が持つ精妙かつ優れた機能の解明を中心とする基礎研究を国際的に推進し、その成果を広く人類全体の利益に供することを目的としています。以下の研究グラント、フェローシップを支給しています。
１）プログラムグラント：独立した科学者のチームによる、生命科学分野を基礎研究とした革新的な研究プロジェクト
２）若手研究者グラント：独立した研究室を与えられて５年以内、もしくは博士課程取得後１０年以内の研究者から成るチーム
３）長期フェローシップ：博士号取得後３年以内、海外の優れた研究機関で現在の研究分野とは別の新しい研究分野へ移ることを目指す研究者対象
４）学際的フェローシップ：生命科学分野以外（物理学、化学、数学、工学等）の若手研究者が、さらに生命科学分野の研究経験を積もうとする場合
※20015年開催のHFSP説明会ポスターより　参考）http://jhfsp.jsf.or.jp/about-us/index.html

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=Luncheon Seminar by Leica Microsystems K.K.=

「共焦点レーザー顕微鏡を用いたイメージングの最前線」

日時：11 月 26 日（土）11：45－12：35

会場：F 会場（つくば国際会議場 中会議室 406）

演者: **加藤 薫** 先生　　産業技術総合研究所 バイオメディカル研究部門

演者: **上条 桂樹** 先生　　東北医科薬科大学医学部 解剖学教室

演者: **岡部 弘基** 先生　　東京大学大学院薬学系研究科 生体分析化学教室

最先端のライフサイエンス研究において、より微細に、より正確に、細胞の動態や挙動を観察するためには、観察のためのツールである、“顕微鏡”システムの進化が重要な要素となります。近年、顕微鏡においても、共焦点レーザー顕微鏡を中心に次々と新しい技術開発が進み、“蛍光イメージング”の応用範囲が飛躍的に広がっています。ライカマイクロシステムズにおいても、ハイエンド共焦点レーザー顕微鏡 Leica TCS SP8 をベースとしたライカ独自技術に基づく製品を数多く開発しています。本セミナーでは、誘導放出抑制法（STED）による超解像イメージング、および、蛍光寿命イメージング顕微鏡法（FLIM）を題材に、学術的観点、実務的な観点よりご講演いただく予定です。

第54回日本生物物理学会
ランチョンセミナー　プログラムＮｏ．３ＬＡ

共焦点顕微鏡の光学系を応用した超解像顕微鏡法の原理と応用

日 時　2016年11月27日（日）　12：30～13：20

場 所　Ａ会場　（つくば国際会議場　中ホール200）

岡田 康志 先生

東京大学大学院理学系研究科 物理学専攻/
理化学研究所 生命システム研究センター細胞極性統御研究チーム

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第54回 日本生物物理学会年会
オプトライン ランチョンセミナー

ライトシート・ライトフィールド顕微鏡が実現する細胞・組織レベルの超高速3Dイメージング

日　時　11月27日（日）　12：30 ～ 13：20

会　場　B 会場（中会議室 202）

演　者

野中 茂紀 先生
自然科学研究機構　基礎生物学研究所
イメージングサイエンス研究領域 時空間制御研究室　准教授

細胞や組織といった数十～数百μmレベルの厚みを持った生物試料を観察する手法として、ライトシート顕微鏡は、低褪色・低光毒性とともに高速性が大きな特長である。
しかし現在、大手顕微鏡メーカーから市販されているタイプのライトシート顕微鏡はZ方向に試料を動かしながらXY像を撮影することで立体像を得るため、細胞運動などの解析のため高速性を追求すると、この移動自体が足かせとなる場合がある。試料の代わりにシート光と焦点面を動かせばこの問題は回避でき、より高速な画像取得が可能になる。
これに対して、ライトフィールド顕微鏡はZスキャンによってではなくマイクロレンズアレイによって複数焦点の像を作り1回の撮影で立体情報を取得するため、さらなる高速化が可能である。
本セミナーではそれぞれの方法の長所と限界について議論する。またこれらの顕微鏡観察を実現するPhaseView社のラインナップについて紹介する。

演　者

岩井　亮一　（株式会社オプトライン）

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本誌記事の動物実験における実験動物の扱いは, 所属機関のルールに従っています.

生物物理　**SEIBUTSU BUTSURI**

日本生物物理学会

THE BIOPHYSICAL SOCIETY OF JAPAN

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