

**BIOGRAPHICAL SKETCH**

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NAME: Takanari Inoue

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POSITION TITLE: Associate Professor of Cell Biology, Cell Dynamics, Pharmacology and Molecular Sciences, Biological Chemistry, and Biomedical Engineering

**EDUCATION/TRAINING**

INSTITUTION AND LOCATION	DEGREE (if applicable)	Completion Date MM/YYYY	FIELD OF STUDY
University of Tokyo (Tokyo, Japan)	B.S.	05/98	Pharmaceutical Sciences
University of Tokyo (Tokyo, Japan)	Ph.D.	03/03	Chemical & Cell Biology
Stanford University (Palo Alto, CA)	Postdoctoral	03/08	Chemical & Systems Biology

**A. Personal Statement**

Synthetic Cell Biology – Signaling events in cells are localized and rapid, often complicated by the presence of feedback loops and crosstalk. Conventional tools used to probe cell signaling are often limited due to its speed and global effect. My scientific research career, to date, has focused on the development of alternative visualization and perturbation strategies for deconstructing and reconstructing spatiotemporally dynamic signaling events observed in nature. These strategies include series of molecular sensors and actuators based on chemically-induced dimerization techniques that allow for the induction of specific activity at different subcellular localizations of live cells in the order of seconds. In particular, these techniques have been used to manipulate activities and levels of second messengers such as small GTPases and membrane lipids, shedding light on several important, previously unanswered questions, including the regulatory mechanisms of potassium ion channels, membrane targeting mechanisms of small GTPases and positive feedback machinery of neutrophil migration. Our studies have had a strong impact on the community as indicated by over 500 reagents requests; several patents filings; multiple press releases and interviews; and various invited lectureships. More recently, we have extended these techniques to reconstitute cellular functions such as chemotaxis and phagocytosis. Ultimately, the reconstitution of these behaviors, collectively, can be used to endow cells with important therapeutic function with applications in the treatment of cancer and Alzheimer's disease.

**B. Positions and Honors****Positions and Employment**

1994 – 1998 Undergraduate Student, Pharmaceutical Sciences, University of Tokyo (Tokyo, Japan)  
 1998 – 2003 Graduate Student, Chemical & Cell Biology, University of Tokyo (Tokyo, Japan)  
 2003 – 2008 Postdoctoral Fellow, Chemical & Systems Biology, Bio-X Program, Stanford University  
 2008 – 2013 Assistant Professor of Cell Biology and Cell Dynamics, Johns Hopkins University  
 2013 – Associate Professor, Cell Biology, Cell Dynamics, Pharmacology and Molecular Sciences, Biological Chemistry, and Biomedical Engineering, Johns Hopkins University

**Other Experience and Professional Memberships**

1998- Member, Japanese Society for Pharmaceutical Sciences  
 2005- Member, American Society of Cell Biology  
 2008- Ad hoc grant reviewer: National Science Foundation, Swiss National Science Foundation, Lowe Trust Foundation, North Carolina Biotechnology Center, National Kidney Foundation of Maryland, Biotechnology and Biological Sciences Research Council  
 2010- Member, Biophysical Society

- 2010-11 Organizer, Campus-wide scientific meeting on lipids (Lipid Club@Hopkins), Baltimore, MD
- 2011 Session Chair, POSTECH International Symposium on "Bioimaging", Pohang, Korea
- 2011- Member, American Chemical Society
- 2011- Founder and organizer, Japanese Science Seminar in Baltimore (JSSB), Baltimore, MD
- 2013 Organizer, ASCB Annual Meeting, Subgroup Session, New Orleans, LA
- 2014 Committee member, R.R. Bensley Award, American Association of Anatomists
- 2014 Committee member, Young Investigator's Day Award, Johns Hopkins University, MD
- 2014 Co-organizer, ASCB Annual Meeting, Subgroup Session, Philadelphia, PA
- 2015 Organizer, BPS International Thematic Meeting, Taipei, Taiwan
- 2015 Organizer, Experimental Biology Meeting Symposium, Boston, MA
- 2015 Editorial Board Member, *Scientific Reports*
- 2015 Organizer, ASCB Annual Meeting, Subgroup Session, San Diego, CA
- 2016 Organizer, AAA Annual meeting at EB2016, Symposium, San Diego, CA
- 2016 Co-organizer, ASCB Annual Meeting, Min-Symposium, San Francisco, CA

### Honors

- 1999-2000 Japan Student Services Organization First Class Scholarship for *magna cum laude* students
- 2000-2003 Young Scientist Scholarship from Japanese Society for Promotion of Science
- 2003-2004 Postdoctoral Fellowship from Japanese Society for Promotion of Science
- 2004-2006 Ruth L. Kirschstein National Research Service Award, Quantitative Chemical Biology Program
- 2010-2012 NIDDK Polycystic Kidney Center Pilot Project Investigator
- 2012-2015 PRESTO investigator, Japanese Science and Technology
- 2013 Young Investigator Award from Pharmaceutical Society in Japan
- 2014 NIDDK Silvio O. Conte Digestive Disease Research Core Center Investigator
- 2014 The Young Scientists' Prize, Commendation for Science and Technology by the Minister of Education, Culture, Sports, Science and Technology in Japan
- 2014 R.R. Bensley Award in Cell Biology, American Association of Anatomists
- 2015 Breakthrough Award in Breast Cancer Research Program, Department of Defense
- 2015 Catalyst Award, Johns Hopkins University, Office of the Provost
- 2015 Discovery Award, Johns Hopkins University, Office of the Provost
- 2016 SPIE's Systems Biology Pioneer Award, International Society for Optics and Photonics
- 2016 Mirowski Discovery Award, Johns Hopkins University
- 2017 Hopkins-Allegheny Health Network Cancer Research Award
- 2017 Hamilton O. Smith Award for Innovative Research

### Patents

- 2000 "IP<sub>3</sub> Receptor Ligands" JPN Patent 4226245, **Inoue T.**, Kikuchi K. Hirose K., Iino M., and Nagano T.
- 2003 "IP<sub>3</sub> Receptor Ligands" U.S. Patent 6,656,927, **Inoue T.**, Kikuchi K. Hirose K., Iino M., and Nagano T.

### **C. Contribution to Science**

#### **Peer-Reviewed Original Science Research**

(#First Author, \*Corresponding author)

#### **Complete List of Published Work in MyBibliography:**

<http://www.ncbi.nlm.nih.gov/myncbi/browse/collection/43813035/?sort=date&direction=ascending>

1. **Inoue T**<sup>#</sup>, Kikuchi K, Hirose K, Iino M, Nagano T. "Synthesis and evaluation of 1-position-modified inositol 1,4,5-trisphosphate analogs." *Bioorg. Med. Chem. Lett.* 1999;9: 1697-1702.
2. **Inoue T**<sup>#</sup>, Kikuchi K, Hirose K, Iino M, Nagano T. "Small molecule-based laser inactivation of inositol 1,4,5-trisphosphate receptor." *Chem. Biol.* 2001;8:9-15.
3. Nakanishi W, Kikuchi K, **Inoue T**, Hirose K, Iino M, Nagano T. "Hydrophobic modifications at 1-phosphate of inositol 1,4,5-trisphosphate analogues enhance receptor binding." *Bioorg. Med. Chem. Lett.* 2002; 12: 911-913.
4. **Inoue T**<sup>#</sup>, Kikuchi K, Hirose K, Iino M, Nagano T. "Spatiotemporal Laser Inactivation of Inositol 1,4,5-Trisphosphate Receptors Using Synthetic Small-molecule Probes" *Chem. Biol.* 2003;10:503-509.

**Note:** Selected for *Cover Article*

5. Yogo T, Kikuchi K, **Inoue T**, Hirose K, Iino M, Nagano T. "Modification of intracellular Ca<sup>2+</sup> dynamics by laser inactivation of inositol 1,4,5-trisphosphate receptor using membrane-permeant probes." **Chem. Biol.** 2004;11:1053-1058.
6. **Inoue T**<sup>\*\*</sup>, Heo WD, Grimley JS, Wandless TJ, Meyer T. "Inducible translocation strategies to rapidly activate and inhibit small GTPase signaling pathways." **Nature Methods.** 2005;2:415-418.
7. Heo WD, **Inoue T**, Park WS, Kim ML, Park BO, Wandless TJ, Meyer T. "PI(3,4,5)P<sub>3</sub> and PI(4,5)P<sub>2</sub> lipids target Ras, Rho, Arf and Rab GTPases to the plasma membrane." **Science.** 2006;314:1458-1461.
8. Suh BC<sup>#</sup>, **Inoue T**<sup>#</sup>, Meyer T, Hille B. "Rapid chemically-induced changes of PtdIns(4,5)P<sub>2</sub> gate KCNQ ion channels." **Science.** 2006;314:1454-1457.  
**Note:** "Perspectives" (*Science* 314, 1402-1403 (2006)), "Editor's Choice" (*Science STKE* 364, tw410 (2006)), "Spotlight" (*ACS Chem. Biol.* 1, 608 (2006)), "Research Highlights" (*Nature Methods* 4, 7 (2007))
9. Liou J, Fivaz M, **Inoue T**, Meyer T. "Live-cell imaging reveals sequential oligomerization and local PM targeting of STIM1 following Ca<sup>2+</sup> store depletion." **Proc. Natl. Acad. Sci.** 2007;104:9301-9306.  
**Note:** "Research Roundup" (*J. Cell Biol.* 177 (6), (2007))
10. Aoki. K, Nakamura T, **Inoue T**, Meyer T, Matsuda M. "An essential role for the SHIP2-dependent negative feedback loop in neuritogenesis of nerve growth factor-stimulated PC12 cells." **J. Cell Biol.** 2007;177:817-827.
11. Yogo T, Urano Y, Mizushima A, Sunahara H, **Inoue T**, Hirose K, Iino M, Kikuchi K, Nagano T., "Selective photoinactivation of protein function through environment-sensitive switching of singlet oxygen generation by photosensitizer." **Proc. Natl. Acad. Sci.** 2008;105:28-32.
12. Fivaz M, Bandara S, **Inoue T** Meyer T. "Robust neuronal symmetry breaking by Ras-triggered local positive feedback." **Current Biology.** 2008;18:44-50.
13. Abe N, Klein L, **Inoue T**, Galvez T, Meyer T. "Dissecting the role of PI(4,5)P<sub>2</sub> in transferrin receptor endocytosis and recycling." **J. Cell Sci.** 2008;121:1488-1494.
14. **Inoue T**<sup>\*\*</sup> Meyer T.\* "Synthetic activation of endogenous PI3K and Rac identifies an AND-gate switch for cell polarization and migration." **PLoS ONE.** 2008;3(8): e3068.
15. Rahdar M, **Inoue T**, Meyer T, Zhang J, Vazquez F, Devreotes PN. "A phosphorylation-dependent intramolecular interaction regulates the membrane association and activity of the tumor suppressor PTEN." **Proc. Natl. Acad. Sci.** 2009;106(2):480-5.
16. **Komatsu T**, Kukelyansky I, McCaffery JM, **Ueno T**, Varela LC, **Inoue T**.\* "Organelle-Specific, Rapid Induction of Molecular Activities and Membrane Tethering." **Nature Methods.** 2010;7:206-208.  
**Note:** "Breaking News" (*Genetic Engineering and Biotechnology News* "Hopkins researchers put proteins right where they want them"), Selected for 2013 calendar by ibidi
17. **Umeda N**, **Ueno T**, **Pohlmeier C**, Nagano T, **Inoue T**.\* "A photocleavable rapamycin conjugate for spatiotemporal control of small GTPase activity." **Journal of American Chemical Society.** 2011;133(1):12-14.  
**Note:** Press release: "Scientists Use Light to Move Molecules Within Living Cells" ("*Science News*" at *ScienceDaily*)
18. **Komatsu T**, Johnsson K., Okuno H., Bito H., **Inoue T**, Nagano T, Urano Y. "Real-time measurements of protein dynamics using fluorescence activation-coupled protein labeling (FAPL) method." **Journal of American Chemical Society.** 2011;133(17):6745-6751.
19. **Ueno T**, Falkenburger BH, **Pohlmeier C**, **Inoue T**.\* "Triggering Actin Comets Versus Membrane Ruffles: Distinctive Effects of Phosphoinositides on Actin Reorganization." **Science Signaling.** 2011;4(203): ra87.  
**Note:** "Perspective", (*Science Signaling*, 5, pe7 (2012), "How Actin Gets the PIP"), "Editorial Guides" (*Science Signaling*, 5, eg3 (2012), "A Sense of Direction"), Press release: "Rearranging the Cell's Skeleton" (*Biochemist e-VOLUTION* at Biochemical Society), Featured in F1000, Selected for Cover Article
20. DeRose R, **Pohlmeier C**, **Umeda N**, **Ueno T**, Nagano T, Kuo S, **Inoue T**.\* "Moving molecules by light; Spatio-temporal manipulation of small GTPase activity at subcellular level and on timescale of seconds in living cells." **Journal of Visualized Experiments.** 2012; e3794 (<http://www.jove.com/video/3794/>).
21. **Miyamoto T**, DeRose R, **Suarez A**, **Ueno T**, **Chen M**, Sun T.-p, Wolfgang MJ, Mukherjee C, Meyers D, **Inoue T**.\* "Rapid and Orthogonal Logic Gating with a Gibberellin-induced Dimerization System." **Nature Chemical Biology.** 2012;8:465-470.

**Note:** “Tools in Brief”, (*Nature Methods*, 9, 534 (2012), “Rapamycin’s Sister”), “Interview” by TV Science Channel, Karga7 Pictures, “This Changes Everything” (2012), Press Release: “Researchers Turn Living Cells Into Logic Gates, Moving Toward Cell Circuits” (“TRENDING”, at *SLATE.com*) (<http://www.youtube.com/watch?v=bHdF4kIZYYE>), “News from the Field”, (*National Science Foundation*, “Training cells to perform Boolean functions? It’s logical”)

22. Phua SC, Pohlmeier C, Inoue T.\* “Rapidly Relocating Molecules Between Organelles to Manipulate Small GTPase Activity.” *ACS Chemical Biology*. 2012;7:1950-1955. **Note:** “Podcast”, (*ACS Chemical Biology*), F1000
23. Sample V, Ni Q, Inoue T, Zhang J “Controlling Enzymatic Action in Living Cells with a Kinase-dependent Bimolecular Switch.” *ACS Chemical Biology*. 2013;8:116-21.
24. Lin B, Wang J, Ueno T, Harwell A, Inoue T\*, Levchenko A.\* “Synthetic spatially graded Rac activation drives directed cell polarization and locomotion.” *PNAS* 2012;109:E3668-E3677.

**Note:** “Rac Gets Cells on Track” Introductory article by *Cell Migration Gateway*, News and Views by “*Biofutur*”, Press release “Microchoreography: Researchers use synthetic molecule to guide cellular dance”

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**Note:** “Research on cilia heats up: Implications for hearing vision loss and kidney disease” Press release, Featured in “ChemBioVault”, “New cilia research could have implications for kidney disease” (American Society of Nephrology, In The Loop), “New clues about molecular composition of cilia” MIGMS Latest News, “Cilia Admit Large Cytosolic Proteins”, Kidney News, “Signaling: Shifting at Ciliary Base” News & Views, *Nature Chemical Biology*

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**Note:** “Tandem Orthogonal Chemically Induced Dimerization” Highlights in *ChemBioChem* (DOI: 10.1002/cbic.201300446)

27. Thevathasan JV, Tan E, Hui Z, Lin YC, Li Y, Inoue T\*, Fivaz M.\* “The small GTPase HRas shapes local PI3K signals through positive feedback and regulates persistent membrane extension in migrating fibroblasts.” *Molecular Biology of the Cell*. 2013;24:2228-2237.

**Note:** “Self-Perpetuating Signaling Circuit in Cells May Drive Cancer Spread” Press release

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**Note:** “Author File” *Nature Methods*, 10, 1039 (2013)

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31. Onuma H, Komatsu T, Arita M, Hanaoka K, Ueno T, Terai T, Nagano T, Inoue T.\* “Rapidly rendering cells phagocytic through a cell-surface display technique and concurrent Rac activation.” *Science Signaling*. 2014;7:rs4, 1-7.

**Note:** “Cover Story” *Science Signaling*, “Turning cells into garbage collector” Editor’s Choice in *Science*, Interviewed and broadcast by FOX45 NEWS (<https://drive.google.com/file/d/0B-wUSlcBaaEsSmpmQmhlblMzTXc/view?usp=sharing>), Interview for “Thought Leaders” series by *Medical-News* (<http://www.news-medical.net/news/20140903/Artificial-cells-to-devour-undesirables-an-interview-with-Dr-Takanari-Inoue.aspx>), Press release: MOLECULAR “EAT NOW” SIGNAL MAKES CELLS DEVOUR DYING NEIGHBORS, “Training Cells to Devour Dying Neighbors” highlighted in *NIGMS Biomedical Beat Blog*, Highlighted in 5<sup>th</sup> Annual Henrietta Lacks Memorial Lecture, “Designer Immune Cells” highlighted in 2014 Research Highlights from Johns Hopkins Medicine (<https://www.youtube.com/watch?v=Lb58GUNP5Qk>)

32. Razavi S, Su S, Inoue T.\* “Cellular Signaling Circuits Interfaced with Synthetic, Post-Translational, Negating Boolean Logic Devices.” *ACS Synthetic Biology*. 2014;3(9):676-685.

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**Note:** Press release: “A digital field guide to cancer cells”
35. Miyamoto T, Rho E, Sample V, Akano H, Magari M, Ueno T, Chen M., Tokumitsu H, Zhang J, **Inoue T**\* “Compartmentalized AMPK Signaling Illuminated by Genetically Encoded Molecular Sensors and Actuators.” **Cell Reports**. 2015;11:657-670.  
**Note:** Press release: “Molecular spies sabotage a protein’s activities in specific cellular compartments”
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37. Meier EL, **Razavi S**, **Inoue T**, Goley ED. “A novel membrane anchor for FtsZ is linked to cell wall hydrolysis in *Caulobacter crescentus*.” **Mol Microbiol**. 2016;101(2):265-80
38. Garcia-Gonzalo FR, **Phua SC**, Roberson EC, Galo Garcia III, Abedin M, Schurmans S, **Inoue T**\*, Reiter JF.\* “Ciliary Phosphoinositides Modulate Hedgehog Signaling.” **Developmental Cell**. 2015;34:400-409.  
**Note:** “Phosphoinositide Code for Primary Cilia” *Preview by Developmental Cell*, “Ciliary phosphoinositides regulate Hedgehog signaling” *Highlight by Nature Reviews Molecular Cell Biology*
39. Antonczak AK, Mullee L, Wang Y, Comartin D, **Inoue T**, Pelletier L, Morrison CG. “Opposing effects of pericentrin and microcephalin on the pericentriolar material regulate CHK1 activation in the DNA damage response.” **Oncogene** 2016;30:2003-2010
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**Note:** “Some Cells Need a ‘Haircut’ Before Duplicating” Press release, “Study Shows How and Why Hairlike Structures on Cells are Lost” PKD News Blog
41. **Miao YC**, Bhattacharya S, Edwards M, Cai HQ, **Inoue T**, Iglesias PA, Devreotes PN “Altering the threshold of an excitable signal transduction network changes cell migratory modes.” **Nature Cell Biology** 2017;19:329-340

#### **Publications: Under Peer-Review**

1. Hideki Nakamura, Albert A. Lee, Ali Sobhi Afshar, Yu-Chun Lin, Makoto Tanigawa, Allison Suarez, Shiva Razavi, Robert DeRose, Diana Bobb, William Hong, Sandra Beatriz Gabelli, John Goutsias, Takanari Inoue  
 “Intracellular production of hydrogels and synthetic RNA granules by multivalent enhancers”  
 (Under second revision at *Nature Materials*)  
**bioRxiv preprint:** <https://doi.org/10.1101/117572>

#### **Review Articles (Peer-reviewed)**

1. Miyamoto T, Razavi S, DeRose R, **Inoue T**\* “Synthesizing Biomolecule-based Boolean Logic Gates.” **ACS Synthetic Biology**. 2013;2(2):72-82.  
**Note:** “Podcast” (*ACS Synthetic Biology*), “Introducing Authors” *ACS Synthetic Biology*
2. DeRose R, Miyamoto T, **Inoue T**\* “Manipulating signaling at will: chemically-inducible dimerization (CID) techniques resolve problems in cell biology.” **Pflügers Archiv** 2013;465:409-417.
3. Lin YC, Su S, **Inoue T**\* “Visualizing Molecular Diffusion through Passive Permeability Barriers in Cells: Conventional and Novel Approaches.” **Current Opinion in Chemical Biology**. 2013;17:663-671.
4. **Phua SC**, Lin YC, **Inoue T**\* “An Intelligent Nano-Antenna: Primary cilium harnesses TRP channels to decode polymodal stimuli.” **Cell Calcium**. 2015;5:415-422.
5. Kim AK, DeRose R, Ueno T, Lin B, Komatsu T, Nakamura H, **Inoue T**\* “Toward total synthesis of cell function: Reconstituting cell dynamics with synthetic biology.” **Science Signaling**. 2016;9:414, re1, 1-7.
6. Coutinho K., **Inoue T**\* “Deconstructing and constructing innate immune functions using molecular sensors and actuators” **Proc. SPIE** 9871, Sensing and Analysis Technologies for Biomedical and Cognitive Applications 2016, 987102 (May 19, 2016); doi:10.1117/12.2225185

7. Niu J., Johny M.B., Dick I.E., **Inoue T.**\* “Following Optogenetic Dimerizers and Quantitative Prospects” *Biophysical Journal*. 2016;111(6):1132–1140

### **Invited Review Articles**

1. **Inoue T.**, Kazuya K, Nagano T. (2002) *Pharmacia*\*38, 131-134. “Spatiotemporal regulation of protein inactivation” (\*Japanese Review of Pharmaceutical Sciences)
2. **Miyamoto T.**, **Inoue T.**\* (2012) *Life Science Review*\* 4376. “Synthetic Generation of Intracellular Boolean Logic Gates” (\*Japanese online review on latest high profile papers)
3. **Inoue T.**\* “Unraveling molecular mechanism of cell migration using novel perturbation tools” *Yakugaku Zasshi*\* 134 (5), 647-654 (2014) (\*Japanese Review of Pharmaceutical Sciences)
4. Chiba S, **Inoue T.**\* “How to Image Calcium Inside Cellular Compartments” *Jikken Igaku*\*, 32 (9) 1275-1280 (2014) (\*Japanese monthly journal on general life science)

### **Editorials**

1. **Miyamoto T.**, **Rho E.**, **Inoue T.**\* “Deconvoluting AMPK Signaling.” *Oncotarget*. 2015;6:30431-30432.
2. **Komatsu T.**, and **Inoue T.**\* “Japanese Scientists in Science Signaling 2014”, *Science Signaling* 2015
3. Gaus K., **Inoue T.**\* “New Biological Frontiers Illuminated by Molecular Sensors and Actuators” *Biophysical Journal*. 2016;111(6):E01–E02
4. **Nakamura H.**, and **Inoue T.**\* “Japanese Scientists in Science Signaling 2016”, *Science Signaling* 2017 (In press)

### **Book Chapters and Science Essays**

1. **Inoue T.** *Biology and Physics*\* 52, (3), 164-165 (2012). “Letter from abroad - One ordinary day of young investigator in US” (\*Japanese Society Proceedings in Biophysics)
2. **Inoue T.** *Yakuji Nippo*\*, Vol. 11254, 2013 (\*Japanese daily newspaper on Pharmaceutical Sciences)
3. **Inoue T.** *Jikken Igaku*\*, Vol. 31, Dec. 2013 (\*Japanese monthly journal on general life science)
4. **Komatsu T.**, **Inoue T.** Vol. 1174, 1231-245, 2014 “Organelle specific induction of molecular activities and membrane tethering” *Methods in Molecular Biology (MMB)* by Humana Press, “Exocytosis and Endocytosis II”
5. **Inoue T.**\* “Toward Total Synthesis of Cell Functions” *Gendai Kagaku*\*, 518, 25-30 (2014) (\*Japanese monthly magazine of general chemistry)