Invited Lecture
3rd Asian Conference on Chemosensors and Imaging Probes (AsianChIP – 2019)

Super-resolution imaging enabled by super-photostable fluorescent organelle markers

Masayasu Taki
Institute of Transformative Bio-Molecules, Nagoya University; JST PRESTO
E-mail: taki@itbm.nagoya-u.ac.jp

Abstract:

The new technologies in fluorescence microscopy have been contributing much to various areas of life sciences. The biological applications of these new technologies often require the development of fluorescent chemical probes. To visualize biological phenomena in detail for a long period of time under microscopic conditions, fluorophores with high photostability are strongly desired. Toward this goal, we have focused on the development of novel π-conjugated skeletons involving a main group element and demonstrated their practical applications for fluorescence imaging. I'd like to highlight how we designed photostable dyes and what we can do with these molecular tools.

We have developed fluorescent xanthene dyes contain an electron-withdrawing phosphine oxide (P=O) moiety in the place of oxygen in classic xanthene fluorophore. Among a series of P=O-containing xanthene dyes, rhodamine derivatives can absorb and emit light in the near-infrared (NIR). PREX 710 (PREX means Photo-Resistant Xanthene dye), one of the P=O-containing rhodamines, exhibits excellent photo-stability and chemical-stability under physiological conditions.[1] We found that the fluorescent signals from PREX 710 in single molecule imaging could be detected at least for 2 min, whereas half of the signals disappeared within 20 s with a cyanine-based dye. Moreover, we performed deep imaging of blood vessels in mice brain using the bright NIR-emitting PREX 710-dextran conjugate and successfully reconstructed a 3D image of blood vessels with z-stack images.

As another type of fluorophore, we are progressing in the development of super photostable fluorescent dyes,[2–4] which enable enables ultrastructural imaging of organelle dynamics with a high spatiotemporal resolution in living cells. MitoPB Yellow is a newly developed mitochondrial inner-membrane marker based on a structurally reinforced naphthophosphole fluorophore.[4] MitoPB Yellow allows us to selectively capture the ultra-structures of the mitochondrial cristae with a resolution of ~45 nm when depleted at 660 nm. It will be a promising new tool to understand the molecular mechanism that controls mitochondrial membrane dynamics.

References and Notes:

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(2) Wang, C.; Fukazawa, A.; Taki, M.; Sato, Y.; Higashiyama, T.; Yamaguchi, S. *Angew. Chem. Int. Ed.* **2015**, *54*, 15213-15217.
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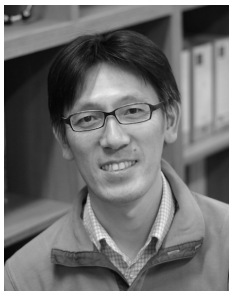
Bio-Sketch of the Speaker

Masayasu Taki
Associate Professor

Affiliation
Institute of Transformative Bio-Molecules, Nagoya University
JST PRESTO
Address
Furo, Chikusa, Nagoya, 464-8601 Japan
Webpage
http://orgreact.chem.nagoya-u.ac.jp/olddocs/en/Home.html



Masayasu Taki graduated from Doshisha University in 1997 and received his ph.D. in 2002 from Osaka University under the supervision of professor Shunichi Fukuzumi. He worked with professor Shinobu Itoh at Osaka City University as a JSPS research fellow from 2002 to 2004, during which he also joined the group of professor Thomas O'Halloran at Northwestern University. He became an assistant professor at graduate school of human and environmental studies, Kyoto University in 2004, and joined the group of professor Shigehiro Yamaguchi as a designated associate professor in 2014. His research interests are in the development of synthetic chemical tools to visualize specific biomolecules as well as biological phenomena in fluorescence.