

Imaging and controlling GPCR activities in live cells: Opto-bioanalysis

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A current trend of bioanalysis is to deeply understand life phenomena and practically apply the knowledge to medical and pharmaceutical fields. To analyze the biological events in living cells, technologies related to fluorescence and bioluminescence imaging have been advancing rapidly in the past decade. Remarkable advancement has been achieved in the monitoring of bioactive small molecules, protein conformational changes, protein localization and dynamics, and protein-protein interactions in real-time at the level of single living cells and organisms. I herein focus on a novel design of engineered luciferases for the analysis of GPCR activities in living cells; the principle is based on complementation and reconstitution of the split-luciferase fragments when they are brought sufficiently close together. I will show the results of direct monitoring of GPCR- β -arrestin interactions in living cells. In addition, an optogenetics approach was used for the analysis of GPCR- β -arrestin interactions, in which we found a unique role of β -arrestin in GPCR trafficking: Temporally controlling the interaction between β -arrestin and GPCR reveals that the duration time of the GPCR- β -arrestin interaction determines the trafficking pathway inside the cells. I will also focus on the screening of chemical libraries for discovering a specific GPCR inhibitors using the above technologies. I will show the inhibitory mechanism of G protein signaling by fluorescence imaging of GPCR and G protein at their single molecule level. I will discuss the significance of GPCR dimerization on the plasma membrane on the activation of intracellular signaling.



Takeaki OZAWA received his Ph.D. in Science from the University of Tokyo, in 1998. In 1998, he was appointed as Research Associate at Department of Chemistry, School of Science, the University of Tokyo and later promoted to Lecturer in 2002. In 2005, he was appointed as Associate Professor, Department of Molecular Structure, Institute for Molecular Science and later promoted to Professor in 2007. From 2014, he appointed as Professor, Department of Biological Sciences, Graduate School of Science. His current research interests: Analytical Chemistry, Protein Chemistry.

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