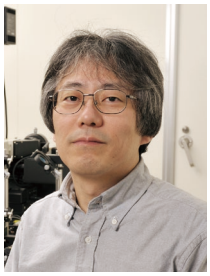
【Grant-in-Aid for Scientific Research (S)】

Broad Section G



Title of Project : Comprehensive approach toward understanding cell surface receptor functions coupled with membrane structure and lipid composition

SAKO, Yasushi
(RIKEN, Cluster for Pioneering Research, Chief Scientist)

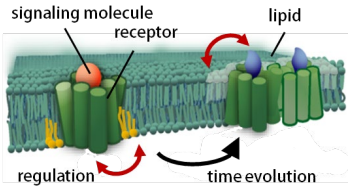
Research Project Number : 19H05647 Researcher Number : 20215700
Keyword : Receptor, Cell membrane, Cell signaling, Single-molecule measurement

【Purpose and Background of the Research】

The cell membrane, which is the boundary of cells and environment, has the fundamental structure of lipid bilayer. Receptor proteins embedded in the cell membrane are responsible for the acceptance, processing and transduction of extracellular information into the cells.

Lipid bilayer is a two-dimensional fluid, in which several hundred species of lipid molecules inhomogeneously distributed dynamically changing their assembly. Interactions with membrane domain structure and specific lipids regulate functions of membrane receptors. Vice versa, activities of membrane receptor affect compositions of boundary lipids and membrane structure. Such interactive communications produce self-organization of cell membrane for signal processing and transduction.

In this research, by applying cutting edge single-molecule technologies, we study behavior-function relationships of the most species of the major human membrane receptors to elucidate general mechanism of regulation of membrane receptor functions by the self-organization of membrane structures.



【Research Methods】

We have developed method of single-fluorescent molecule measurement of membrane proteins. This method allows quantification of molecular movements, dimerization, oligomerization, and interaction with extra- and intra-cellular proteins of membrane proteins on the living cell surface. It also allows measurements of structural dynamics and reactions of purified receptor molecules in artificial membranes. This project will use this method for the study of membrane receptors.

G protein-coupled receptors (GPCRs) and receptor tyrosine kinases (RTKs) are the targets of this study. We will compare the single molecule behaviors and functions about 300 species of GPCRs (excluding odorant receptors) and 60 species of RTKs in human cells to obtain general mechanism of signal processing and transduction of membrane receptors. We will focus on the diversification of the signaling pathways, signal bias, and crosstalk between different species of receptors.

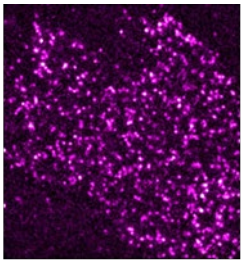
We also study dynamics of membrane domain structure and composition of boundary lipids of receptors. By using protein probe for specific lipid molecules, we can achieve super resolution imaging of 10~100 nm-scale lipid domains. Biochemical analysis of the boundary lipids of membrane receptors will be done in nanodiscs.

Major items of our project are as follows:

1. Comprehensive single-molecule measurement of membrane receptors
2. Super-localization imaging of membrane receptors and lipids
3. Analysis of the boundary lipids of membrane receptors
4. Measurement of molecular dynamics and lipid regulation in artificial membrane

【Expected Research Achievements and Scientific Significance】

In this study, we wish to understand meanings of the transient spatiotemporal dynamics of membrane structure and lipid compositions in the expression of signal processing and transduction functions of membrane receptors. Comprehensive single-molecule measurement is first enabled by our recent development of the automated imaging system. Since GPCRs and RTKs are the major targets medical drugs, this study will contribute to medical science and pharmacology.


Single-molecule imaging of membrane receptors

【Publications Relevant to the Project】

・Yanagawa M, Hiroshima M, Togashi Y, Yamashita T, Shichida Y, Murata M, Ueda M, Sako Y. Single-molecule diffusion-based estimation of GPCR activity. Sci. Sig. 11, eaaao1917 (1-16) (2018)
・Hiroshima M, Pack C-g, Kaizu K, Takahashi K, Ueda M, Sako Y. Transient acceleration of epidermal growth factor receptor dynamics produces higher-order signaling clusters. J. Mol. Biol. 430, 1386-1401 (2018)

【Term of Project】 FY2019-2023

【Budget Allocation】 117,700 Thousand Yen

【Homepage Address and Other Contact Information】

http://www2.riken.jp/cell-info/
sako-lab@riken.jp