IPR-IFReC Joint Seminar

June 7th, 2010, 13:00-14:00
Institute for Protein Research, First Floor Seminar Room, Osaka University

Membrane Targeting, Binding, and Deformation by the ENTH Domain

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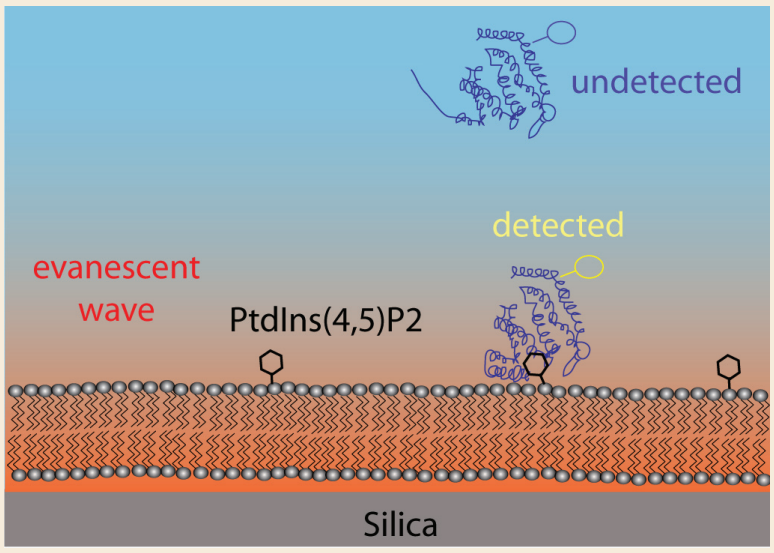
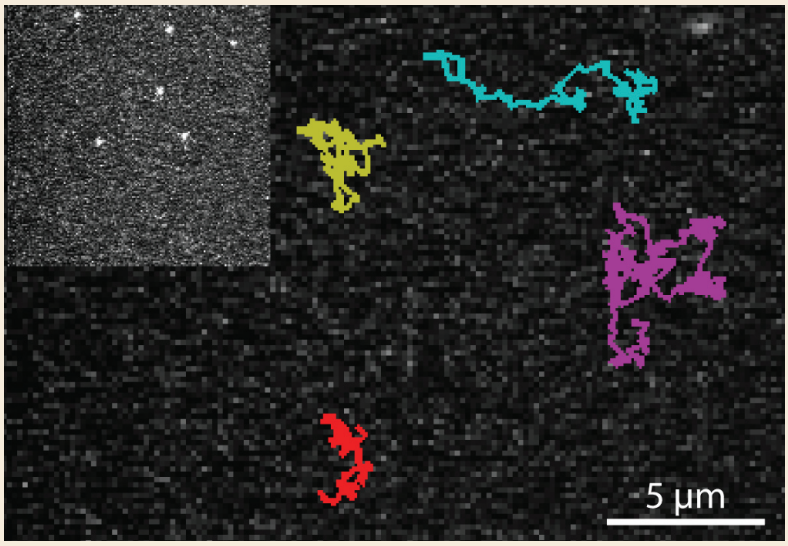
Abstract

Transient recruitment of proteins to membranes is a fundamental mechanism by which the cell exerts spatial and temporal control over protein localization and interactions. Thus the specificity and kinetics of peripheral proteins' membrane-residence are an attribute of their function and reactivity. While bulk measurements can quantify average properties of these important protein-membrane associations, they are unable to delineate the details of their underlying mechanism. In here, we report direct visualization and quantification of protein-membrane interactions using single molecule total internal reflection fluorescence microscopy. TIRFM allows recording of single binding events of proteins to simple model membranes (i.e. a fluid lipid bilayer deposited on a silica) by discriminating between membrane-bound and unbound protein. We record the kinetic behavior of the interfacial Epsin N-terminal homology domain. Subsequent single fluorophore tracking permits us to build up distributions of residence times and measure ENTH dissociation rates as a function of membrane composition. This report demonstrates the capability of this well-controlled model systems and single molecule detection to provide quantitative measurements of membrane protein interactions and assembly on the membrane surface. This is particularly notable since it allows access to weak binding interactions in the presence of physiological concentration of the target lipid. The observed diffusion indicates a multi process diffusion process.





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