COBRE RESEARCH SEMINAR SERIES

Advances in Super-resolution Light Microscopy

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The super-resolution method of photoactivated localization microscopy (PALM) can be used to analyze the distribution and dynamics of single molecules within bigger structures, making it an ideal tool for mechanistic investigation of biological processes. This technique is particularly useful for the investigation of protein organization on the cell surface due to spatial and temporal resolution advantages over conventional fluorescence microscopy. However, because of photophysical properties of fluorescent molecules and the uncertainty of their localization, quantitative determination of oligomeric structures is challenging. To address this, we used a pair correlation (PC) approach and developed a new method to analyze the distribution of single molecules obtained with PALM. By separating contributions from stochastic clustering (corresponding to multiple appearances of a single protein) and protein clustering (corresponding to homo- and hetero-oligomers) we determine the size, density, and abundance of proteins in the clusters. We demonstrate distinct nanoscale organization of PM proteins with different membrane anchoring and lipid partitioning characteristics (including GPI-anchored, Lyn, Lat, and VSVG), and show dramatic changes in GPI-anchored protein arrangement under varying PM perturbations. PC-PALM is thus an effective tool with broad applicability for analysis of protein heterogeneity and function, adaptable to other single molecule strategies.

Friday, October 21, 2011 at 1:00 p.m.
John A. Burns School of Medicine, Kaka'ako
Medical Education Building Auditorium, Room 315
For further information, call 692-1654