

INVITED LECTURE T5

Visualizing sphingolipids in membranes, cells and embryos

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The pathology of Alzheimer's disease and several other neurodegenerative diseases leads to alterations in the metabolism and storage of cholesterol and sphingolipids. Sphingolipids and cholesterol coalesce to form so-called "rafts" or microdomains in the plasma membrane, which serve as endocytic portals into the cell. Our lab is interested in observing the changes in behavior of sphingolipids and cholesterol in cellular models of neurodegenerative diseases. To do this, we have developed fluorescent peptide- and lipid-based probes that can be used to follow the trafficking and biophysical behaviors of sphingolipids and membrane microdomains in live cells. The fluorescent sphingolipid/raft-tracing probes are being used in two ways: 1) in cultured cells, we observe changes in trafficking pathways of the fluorescent probes under conditions that interfere with lipid metabolism, using quantitative colocalization with fluorescent transgenic markers; 2) the biophysical and binding behavior of the probes are being studied in artificial membranes and in live cells. Various techniques including fluorescence correlation spectroscopy (FCS), atomic force microscopy (AFM), and surface plasmon resonance (SPR) are used to gain insight into the rules of behavior of these sphingolipid interacting probes. Finally, in a *Drosophila* model, we show that FCS can be used in living embryos to determine the diffusion behavior in situ of various membrane and raft-localized probes