Getting a Grip on Greasy Molecules

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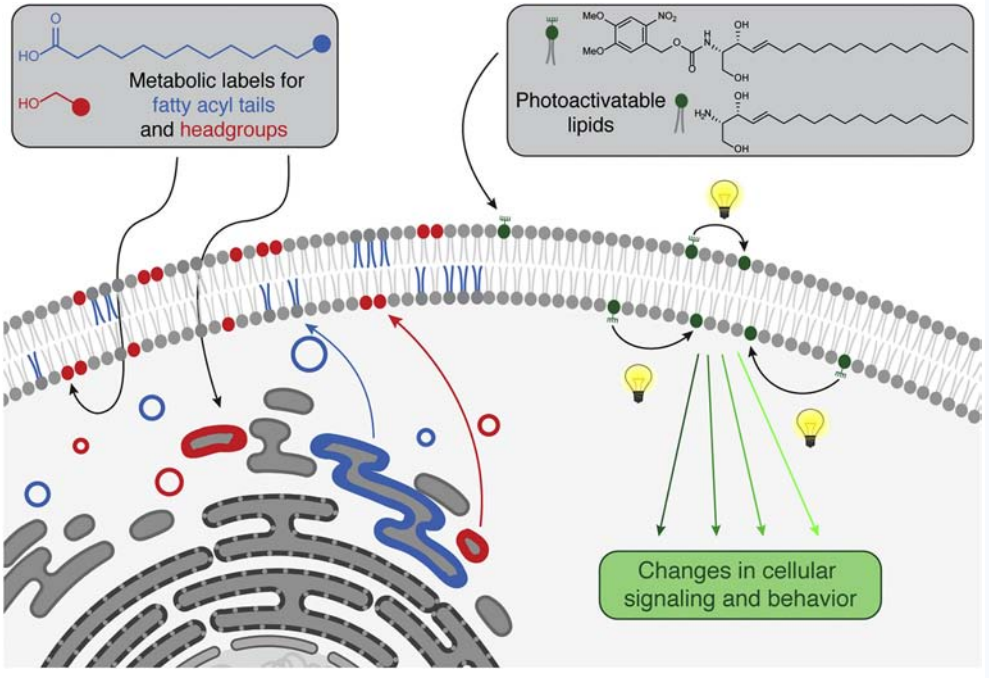
	Probe	Description and Application(s)
Bioorthogonal metabolic labels		Alkynyl fatty acid Broad lipid labeling and identification
		Diazirine fatty acid Identification of lipid-binding proteins
		Azidopropanol Imaging phospholipase D activity
		Propargylcholine Imaging choline-containing lipid biosynthesis
Photoactivatable lipids		Nitrobenzyl-caged sphingosine Rapid release of sphingosine
		Photoswitchable diacylglycerol (DAG) Reversible activation of DAG
Multifunctional lipids		Trifunctional diacylglycerol Spatiotemporal release of DAG capable of imaging and pulling down binding partners

Trends in Biochemical Sciences

Lipids are structurally and functionally diverse biological metabolites that have long proven difficult to study, particularly as targets for molecular imaging. This challenge arises from their small size, hydrophobicity, rapid diffusion and trafficking rates, and, perhaps most critically, their status as metabolites whose structures and biosynthesis are not directly encoded in the genome, making them inaccessible to direct fusion with fluorescent proteins.



Trends in Biochemical Sciences

As a result of these challenges, most analytical methods for lipids rely on measuring lipid levels after sample homogenization and lipid extraction. These studies are critical to our understanding of lipid biology, but bulk biochemical, *ex vivo* analysis carries no spatial information and thus obscures the role of localization in downstream biological outcomes. Recent advances have focused on new approaches to study lipids within intact cells and tissues to better elucidate the spatial component of the function of these critical molecules.

ADVANTAGES:

New analytical tools in lipid biology, such as metabolic labeling followed by bioorthogonal ligation, enable real-time observation of lipid signaling in living cells and link the spatial and temporal domains of signaling.

Photoactivatable lipids enable rapid release of signaling lipids with precise control of time and localization for highly targeted studies of signaling events in live cells.

Multifunctional lipids, which can be tailored to specific applications, couple the spatiotemporal precision of photoactivatable groups with photo-crosslinkers and bioorthogonal handles, which enable visualization or enrichment of lipids and their interaction partners.

CHALLENGES:

Metabolic labeling of lipids relies on native biosynthetic enzymes to incorporate unnatural groups. Tagged lipids are produced in the same membrane environment as their natural counterparts, which is desirable. Yet, because of their different chemical makeups, these lipids may be trafficked differently.

Different lipids can share biosynthetic enzymes, limiting the points at which metabolic probes may be introduced to maintain selectivity to a desired lipid.

When certain highly functionalized, exogenous lipid analogs are added to cells, they may be trafficked differently and incorporated into different membranes than their native counterparts. To partially mitigate against this problem, localization tags can direct unnatural lipids desired organelle membranes.

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