

Simultaneous flux analysis across all major lipid metabolic pathways by high-resolution mass spectrometry

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Advances in mass spectrometry and lipidomics have in the last decade enabled in-depth, quantitative characterization of the yeast lipidome [1,2]. This technology has so far been used to primarily monitor steady-state lipid levels and not the actual flux across the multitude of lipid enzymatic transitions that cells continuously use to carry out membrane-related processes and maintain cellular homeostasis [2]. In order to gain insights into the regulation of lipid metabolic flux we have recently developed an experimental framework that uses simultaneous pulse labeling with multiple stable isotope-labelled precursors, specific detection and absolute quantification of labeled and unlabeled lipid molecules by high-resolution mass spectrometry, and a dedicated algorithm that accounts for changes in the pool sizes of metabolically coupled lipid molecules. Through analysis of yeast strains with over-expression of lipid enzymes or sub-lethal enzyme inhibition we demonstrate that our novel framework supports accurate flux analysis across all major lipid metabolic pathways in yeast, and importantly, can identify regulatory cross-talk between seemingly independent lipid metabolic pathways. Future applications of our framework will serve to identify factors responsible for maintenance of lipid metabolic homeostasis at the systems-level.

[1] Ejsing CS, Sampaio JL, Surendranath V, Duchoslav E, Ekroos K, Klemm RW, Simons K, Shevchenko A. (2009) [Global analysis of the yeast lipidome by quantitative shotgun mass spectrometry](#). Proc Natl Acad Sci USA. 106(7):2136-41.

[2] Casanovas A, Sprenger RR, Tarasov K, Ruckerbauer DE, Hannibal-Bach HK, Zanghellini J, Jensen ON, Ejsing CS. (2015) [Quantitative analysis of proteome and lipidome dynamics reveals functional regulation of global lipid metabolism](#). Chem Biol. 22(3):412-25.