

New functional looks into the proteomes using LC/MS and Co-fractionation mass spectrometry

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Abstract

The sensitivity, speed and reproducibility of modern mass spectrometers enable deep new function looks into the cellular proteome. Because the dynamics of the protein ensemble link genotype to phenotype, knowledge on protein complex assembly and localization is important for marker-assisted breeding for precision plant breeding. Co-Fractionation Mass Spectrometry (CFMS) enables systems-level analyses of protein complex dynamics. Our CFMS pipeline accepts soluble and membrane-associated cell fractions from agriculturally important cell types and uses orthogonal chromatographic separations, reproducibility filters, and correlation analyses to predict localization and composition based on experimental data. We have applied the CFMS method to analyze protein complex dynamics as a function of dark-induced metabolic stress. We discovered dozens of interesting protein complex rearrangements that likely reflect an adaptive response to reduced energy status. Similar assays have been used to analyze the system of protein complex rearrangements that are observed in a single gene knockout or those that occur in dark grown hypocotyls after short term treatment ethylene. A current project focuses on the protein multimerization and localization dynamics that occur during the development of unicellular cotton fibers. This methodology has great potential in gene discovery and systems-level phenotyping tool. We are organizing the data from our multi-omics studies so that it is findable and useful to the community with the goal of accelerating the genetic engineering of plant traits.

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