

Potential of Various HPLC/MS Modes in Comprehensive Lipidomics Approach

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Lipidomics, as a subgroup of metabolomics, is the characterization of maximum number of lipid classes and class species with a great importance in current bioanalytical and medical research. Our lipidomics approach is based on the combination of various chromatographic methods coupled to mass spectrometry to obtain complementary information with a different level of analytical details. Reversed-phase HPLC is used for the characterization of phospholipid classes, while non-aqueous reversed-phase (NARP) mode is more suitable for non-polar lipids [1,2]. Hydrophilic interaction (HILIC) chromatography offers the possibility of class separation of lipids, while silver-ion normal-phase chromatography provides the best resolution for lipids differing in the number of double bonds including regioisomers [3]. In NARP mode, the retention depends on the equivalent carbon number, while the retention in silver-ion mode increases with the increasing double bond number. The combination of orthogonal separation mechanisms in NARP and silver-ion modes in off-line 2D HPLC provides the most detailed characterization of neutral lipid samples ever reported, while UPLC brings a significant reduction of analysis time while maintaining the separation selectivity achieved under optimized chromatographic conditions. Finally, the extensive amount of analytical data is processed with modern statistical methods, e.g., PCA. The comparison of different HPLC modes, ionization techniques (ESI, APCI and APPI), polarity modes (positive vs. negative) and mass analyzers (ion trap and QqTOF) is illustrated on examples of plant and animal tissues and human serum samples.

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