

# **Analysis of Lipids at the Molecular Level by Ultra High Resolution Mass Spectrometry**

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Abnormalities in the synthesis, metabolism and catabolism of lipids result in disorders, which have grown to pandemic proportions in recent decades. Mass spectrometry plays a key role in analysis of lipids, particularly when it comes to quantitative issues at the level of molecular species. The mass spectrometric methods widely used to tackle this topic are either offline Nano ESI-MS or online LC-MS/MS. Both methodologies are well established, but both also have substantial drawbacks. Offline nano ESI-MS of lipids suffers from ion suppression phenomena distorting quantitative data, whereas in an online approach with MS/MS coupled to HPLC the set of ascertainable molecular species is relatively limited.

It is the aim of this project to overcome the shortcomings of the established technologies by development of an online method capable to determine up and down regulation of phospholipids, triacylglycerols, diacylglycerols, cholesterolesters, sphingomyelin and ceramides in one LC-MS run. The instrumental ingredients for such an ambitious project are highest possible chromatographic and even more so mass spectrometric resolution: A UPLC (Accela) supplying the needed chromatographic performance, and an ion cyclotron MS (LTQ-FT) with its unprecedented resolving power and mass accuracy. In spite of the high resolving power of the UPLC we are not able to get a baseline separation for all components of a lipid extract, particularly as mammalian samples get enormously complex, featuring hundreds, if not even thousands of different lipid species. At this point ultra high resolution mass spec enters the stage and provides the capability to detect lipids not resolved by chromatography separately. Key elements for identification are exact retention time and exact mass. Experimental data show retention time stability well below 1% and a mass accuracy of better than 2ppm. Future goals are implementation of a routine for peak identification, data alignment, lipid ID, data normalization and data visualization.