Resolution of Sphingomyelins in Complex Lipid Extracts by Differential Ion Mobility Spectrometry (DMS)

SelexION® Technology

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A significant challenge in the field of lipidomics is the extensive isobaric and isomeric overlap of different lipid molecular species. Liquid chromatography is the method of choice to address this issue, but due to the wide variety of chemical structures among lipids, there is no one LC method that effectively resolves all lipid molecular species. An orthogonal separation method is needed.

The Challenge:

The characterization of sphingomyelins (SM) in complex lipid extracts is confounded by the fact that this class of lipids shares a common, diagnostic fragment with phosphatidylcholine (PC) - the phosphocholine head group (*m/z* 184, positive ion mode). High resolution mass spectrometry can resolve these two classes of lipids; however, subsequent product ion analysis relies on a low resolution precursor ion isolation step, so the resultant composite MS/MS spectra contain significant isobaric interferences

The Solution:

Using SelexION Technology (DMS), SM and PC are resolved from one another using lipid class-specific compensation voltages (CoV) prior to MS analysis that removes isobaric interference and generates class-homogenous ion spectra. Here, using DMS on a QTRAP® 5500 System, bovine heart extract was infused, and a precursor ion of *m/z* +184 scan was performed to identify both SM and PC molecular species (Figure 1A). Lipid class-specific DMS CoV values were applied during precursor ion analysis to generate spectra for SM (Figure 1B) and PC (Figure 1C) without isobaric interferences.

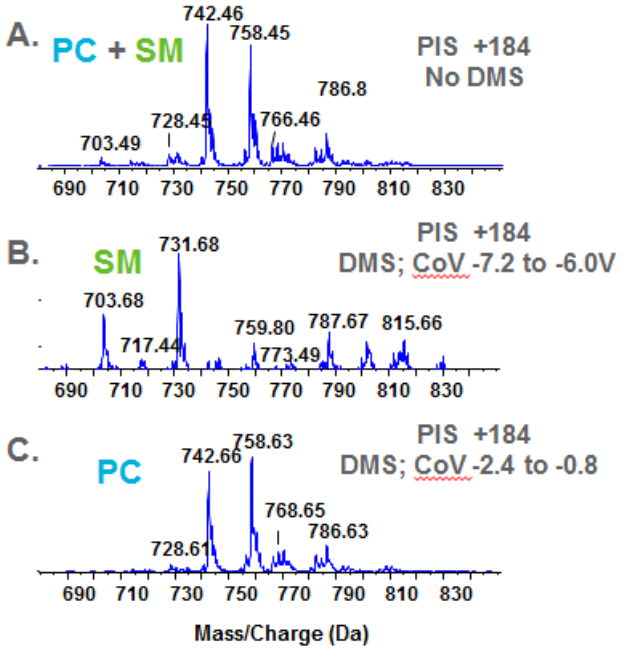

Figure 1. Resolution of Sphingomyelins and Phosphatidylcholines During Precursor Ion Analysis. Bovine heart extract was infused and analyzed by detecting molecules with a precursor ion of m/z 184 in the positive ion mode on a QTRAP® 5500 System equipped with SelexION Technology. Without the DMS, this experiment generates a spectrum containing both PC and SM due to both classes of lipid producing a phosphocholine fragment (+184) during fragmentation (A). Using the DMS and class specific CoVs to resolve these two classes prior to MS analysis generates clean spectra for SM (B) and PC (C). The two lipid classes are completely resolved from one another, facilitating identification. Subsequent MS/MS analyses of individual molecular species will not have cross-species interferences, enabling clear qualitative and quantitative data to be obtained.

SelexION Technology resolves sphingomyelin form phosphatidylcholine prior to MS analysis to provide simplified MS spectra and interference free MS/MS spectra.

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