

## Automated system enables study of various processing trains for serum peptidome analysis

There has been increasing interest in the investigation of the serum peptidome, which consist of low molecular weight (<30kDa) endogenous peptides. They are products of in-vivo proteolytic cleavages throughout the body which “leak” in the serum and constitute an ideal source for biomarker candidates. We developed a modular automated processing system (MAPS) which enables in-line multidimensional processing of serum samples. MAPS comprises a supporting hardware infrastructure (miniaturized fittings, manifolds, pumps and microvalves) and a set of modules based on 10 $\mu$ l to 200  $\mu$ l volume cartridges mounted on a breadboard platform. The modular architecture and automation enables rapid and highly reproducible testing of customized processing trains.

We will present a comparison of protein and peptides identified in mouse serum samples using three methods. We packed two restricted access media-type materials, ProteoEnrich and BondElute, in our cartridges and investigated their capacity and selectivity towards small proteins/peptides. The third processing method assessed the capacity of semi-preparative size exclusion chromatography for selective fractionation of lower molecular weight and low abundance proteins and peptides. Size separation was followed by anion exchange fractionation of the serum sample. Each processing method revealed common and distinct proteins/peptides underlining the significant impact of sample processing path on the output data.