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IDENTIFICATION AND CHARACTERIZATION OF ENDOGENOUS PHOSHOPEPTIDES IN THE PEPTIDOME OF HUMAN CEREBRAL SPINAL FLUID BY COLLISION CELL ICP-MS AND ESI MASS SPECTROMETRY

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Reversible protein phosphorylation of the amino acids serine, threonine and tyrosine are important dynamic processes present in all eukaryotes. The process affects the 3D structure of proteins and influences the catalytic activity of proteins during physiological processes. It is also important for protein-protein interactions and regulates the expression of various genes and protein synthesis

Neurodegenerative processes such as the disruption of the neuronal cytoskeleton and the accumulation of paired helical filaments (PHF), which can be found in connection with Alzheimers disease, show a strong correlation with protein hyperphosphorylation. This is especially true for the microtubule-associated protein “Tau”, the major protein subunit of PHF, which demonstrates abnormal hyperphosphorylation in the Alzheimer disease brain.

ESI-MSMS is the technique of choice in the identification and quantification of peptides. However, modified peptides often show poor ionization efficiencies in comparison to unmodified ones when using ESI. Also, large phosphorylated peptides often show poor fragmentation characteristics in MS/MS experiments that result in difficulties in identification.

ICP-MS does not replace other analytical techniques in proteomics, however it does offer a powerful complementary system to ESI-MSMS for characterizing phosphorylated proteins. The technique provides almost compound-independent ionization in comparison to ESI especially when using low-flow separation techniques. The inorganic technique also provides a rapid pre-screening of tryptic digests for phosphorylated peptides before their final characterization via ESI-MSMS or ESI-TOF-MS.

An understanding of biochemical mechanisms depends critically on the possibility of identification, characterization and quantification of the chemical forms of relevant elements involved in life processes. Biomolecules, especially their phosphorylation, can play a key role in the context of diverse diseases such as cancer, Alzheimers, Parkinsons, multiple sclerosis and others. Their identification and characterization may help diagnostic approaches.

A multi-dimensional analytical strategy using hyphenated systems with modern mass spectrometric methods for inorganic and organic trace analysis are discussed. Using ICP-MS as a powerful on-line detector, both chromatographic and electrophoretic separation have been employed for the characterization of various phosphorylated peptides and data correlated with results from ESI-MSMS.