

MeCAT: Applications for absolute Quantification of Proteines and Peptides using HPLC/ICP MS

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The ability to quantify properly very low amounts of proteins and peptides plays an important role in medical and biological research. This need lead to the development of several methods (1), (2) for comparative studies of a proteom within a biological system. These methods allow quantitative analyses of proteins and peptides but provide only a low dynamic range and relative high limits of detection. On the other hand the MeCAT-approach (3) developed by our group us works within an increased dynamic range and limits of detection in attomol range. The MeCAT-method uses lanthanoid chelate complexes which are covalently bound to proteins and peptides. These labeled molecules may be sequenced by MALDI and ESI MS and MS/MS and finally quantified in an absolute manner by ICP MS.

Complex mixture of MeCAT labeled Proteins require a prior separation before a quantitation by ICP MS is feasible Therefore we have established a HPLC/ICP MS setup to demonstrate the abilities of the MeCAT-Method. Using external calibration it is possible to quantify proteins and peptides in attomol range and at the same time over a dynamic range of 5 orders of magnitude.

The development of the MeCAT-procedure and its applications will be demonstrated on the quantification of Insulin and the quantification of mixture of proteins. Several instrumental setups are used and examined.

- (1) S.P. Gygi, et al, "Correlation between protein and mRNA abundance in yeast", Mol. Cell. Biol., 1999, 19, 1720-30.
- (2) P.L. Ross, et al, "Multiplexed Protein Quantitation in Saccharomyces cerevisiae Using Amine-reactive Isobaric Tagging Reagents", Mol. Cell. Proteomics, 2004, 1154-69.
- (3) R. Ahrends, S. Pieper, et al, "MeCAT – A Novel Approach for Quantitative Proteomics", submitted