EUROSEQUENCE bv

The Art of Protein Sequence Analysis

PRODUCT INFORMATION *PEPTIDE MASS FINGERPRINTING BY MALDI-MS*

Method and General Remarks

Identification of your protein is performed by comparing observed peptide masses with theoretical protein digest mass numbers in data bases. In addition MS/MS may have been used to get sequence information from one or more internal peptides, to strengthen the evidence of identification, or generating a *de novo* sequence, enabling a very high search specificity for the correct identification of your protein.

To get the peptide masses, your protein band was reduced & alkylated and subjected to in-gel tryptic digestion. The resulting peptides were extracted from the gel and purified using a C18 cartridge. The peptide-containing fractions were collected and analyzed by Delayed Extraction-Matrix Assisted Laser Desorption Ionisation-Time of Flight Mass Spectrometry (DE-MALDI-TOF-MS or –TOF/TOF-MS/MS), or, in addition, we may have used nano-spray ESI-MS/MS.
In principle, peptide mass fingerprinting may provide information on N-terminal, mid-chain and C-terminal part of the sequence with equal probability.
Because of the identification is done by comparing the observed masses of the peptides with those of known proteins in a database search, one should be careful with chemical modifications and we should be informed about any chemical modification of your protein (for instance alkylation of Cys residues, biotinylation), or any relevant sample preparation history (please consult us in case of any doubt).
Please remember that identification by MS, especially with MALDI-MS peptide mapping, will fail if the protein is unknown, and might fail if:
- not enough peptide masses are found,
- the protein is (severely) contaminated with another protein,
- (severe) unknown post-translational or (unwanted) chemical modifications (have occurred),
- strong heterogeneity (polymorphism).

Explanation of the interpretation report

The scientific interpretation of the results is summarized in a report signed by the responsible scientist. On this report you will find our product code and order number (if chromatograms are delivered with the report, the same product number is also printed at the top of the corresponding chromatogram).
The relevant MS data produced from analysis of a portion of the peptide-containing fraction are tabulated, as are their masses (m/z signals), and peaks consistent with keratin (very well known contamination) or auto-digestion products of trypsin are either left out or are noted and assigned as such. Further, observed parts of the identified protein are indicated in bold and underlined in the complete sequence, as was listed in the data base. It should be noted that (mass) modified peptides will not easily be recognized; this implies glycosylated and other, uncommon modified peptides.

We would like to emphasize that the identification of your sample is based on the best possible interpretation of the observed mass data. However, contamination of a sample, especially with other protein material than keratin, as well as chemical modifications prior to analysis, may lead to misinterpretations. Therefore, information about the sample history is essential for obtaining the best possible interpretation.

Literature

[1] Kussmann M, Lassing U, Sturmer CAO, Przybylski M, Roepstorff P: Matrix-assisted Laser desorption/ionization mass spectrometric peptide mapping of the neural cell adhesion protein neurolin purified by sodium dodecyl sulfate polyacrylamide gel electrophoresis or acidic precipitation. *J Mass Spectrometry* 1997, 32:483-493.