Peptide mapping

GUIDELINES FOR SAMPLE SUBMISSION

Verification of protein sequence and identification of the nature and precise location of any structural modification in a protein is carried out by using mass mapping strategies based on MALDI analysis. The approach consists in the direct MALDIMS analysis of peptide mixtures generated by enzymatic or chemical digests of the protein without the need of any purification steps. Spectra are obtained and the recorded mass signals can be mapped onto the anticipated protein sequence. Confirmation of the assignments can be achieved by submitting selected mass signals to MS/MS analysis. Although some components may not give protonated molecular ions, most of the protein sequence can be mapped in a single experiment. In most cases, the entire protein structure can be screened by combining data from two or more maps. If unexpected signals are observed, errors of translation, deletion, insertion, point mutation [except inversions in the sequence], post-translational modifications or processing and the S-S bridge pattern can be detected and assigned using this method.

Conditions for Protein Mapping

Protein mapping can be performed on proteins either in solution or proteins separated by monodimensional electrophoresis (SDS-PAGE).

A) Sample in solution or lyophilized:

Single purified proteins are preferentially provided in aqueous solution or organic solvents (i.e. Acetonitrile, TFA). Lyophilized proteins are also acceptable.
For mapping analysis, the protein sequence must be provided.
Please, indicate the last step in sample preparation.
Provide information on protein concentration and buffer composition.

B) Samples from SDS-PAGE:

Protein bands must be stained with colloidal Coomassie.
Please provide at least three to six well stained protein bands. Alternatively, load several lanes with the protein samples and, if possible, send the intact gel, otherwise cut the bands and send them in vials with H_2O MilliQ.
Please send a copy of the gel image identifying the bands of interest.

Requested amount

Indicative amounts of proteins for mapping analysis consists of:

- 1-2 nmol of lyophilized or soluble protein
- three-six well visible Coomassie stained bands.

Important for sample submission

Please avoid the presence of high concentration of detergents and glycerol.
A minimum of 90% of protein sequence coverage is guaranteed.

Analysis Time

2-3 weeks from the arrival of the samples.