

PRODUCT INFORMATION

Trypsin NB Premium Grade, MS approved

Cat.No. 37284

TP Standard Trypsin Peptide (TP) Standard is delivered in a separate vial and contains trypsin to generate masses m/z 842 and 2211. If standard peptides are needed for internal calibration add to the trypsin digestion solution in a ratio: 1:5.

Storage conditions Trypsin NB Premium Grade, MS approved and TP Standard should be stored in a **dry state** at -15 to -25 °C.

Instructions for use:

General Trypsin is used in proteomics for peptide mapping due to its highly specific cleavage resulting in a limited number of tryptic peptides.

Digestion of proteins in solution Lyophilized Trypsin NB Premium Grade is reconstituted in 25 µl 50 mM acetic acid to a final concentration of 1 µg/µl. For digestion of the target protein add Trypsin NB to a final protease:protein ratio of 1:100 to 1:20 (w/w). For a detailed protocol please see user manual for Trypsin NB Premium Grade, MS approved (Cat.No. 37284) at www.serva.de.

In-gel protein digestion Lyophilized Trypsin NB Premium Grade is reconstituted in 25 µl 50 mM acetic acid. Then add 475 µl 25 mM NH₄HCO₃, pH 8 to a concentration of 50 µg/ml. For the final use dilute Trypsin NB solution 1:2.5 with 25 mM NH₄HCO₃, pH 8 and use 10 to 20 µl for rehydration / digestion of each gel piece. For a detailed protocol please see user manual for Trypsin NB Premium Grade, MS approved (Cat.No. 37284) at www.serva.de.

Optional: To avoid clogging of the LC system clear the solution from the In-gel digest by centrifugation of extract the peptides, e.g. with acetic acid and acetonitrile.

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Product Description:

General Trypsin NB Premium Grade, MS approved is a serine endopeptidase which specifically cleaves at the carboxyl side of lysine, arginine and S-aminoethyl cysteine residues. There is little or no cleavage at arginyl-proline or lysyl-proline bonds. Cleavage may also be reduced when acidic residues are present on either side of a potentially susceptible bond [1].

Application Trypsin NB Premium Grade, MS approved is specially designed for digestion of proteins prior to mass spectrometric analysis.

- Features**
- Source: porcine pancreas
 - Purity: > 90 %
 - Tryptic activity: > 6000 U/g*
 - No chymotryptic activity detectable
 - Modified by reductive methylation
 - Each lot qualified by in-gel digestion and mass spectrometric analysis
 - Quantity: $\geq 25 \mu\text{g}/\text{vial}$, determined by measuring A_{280} .

*Unit definition: 1 U catalyzes the hydrolysis of 1 μmol N α -Benzoyl-L-arginine-4-nitroanilide hydrochloride (BAPNA) per minute at 30 °C, pH 8.0.

Quality control

Each lot of Trypsin NB Premium Grade, MS approved is qualified by in-gel digestion and mass spectrometric analysis. An example of a spectrogram is shown in figure 2. Lot specific generated spectrograms using bovine serum albumin (BSA) as substrate are available at tech.service@serva.de.

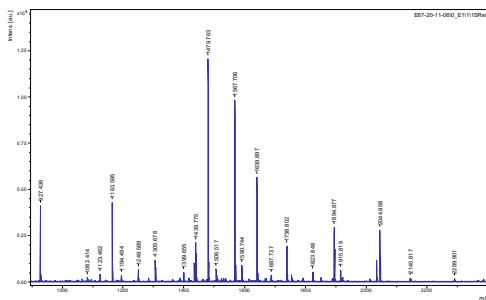


Fig. 2: Spectrogram of BSA digested with Trypsin NB Premium Grade, MS approved. 300 ng BSA were separated by gel electrophoresis and digested with 10 ng/ μl Trypsin NB Premium Grade in 50 mM NH_4HCO_3 at 37 °C overnight. The peptides generated were analysed in reflectron mode using the Bruker Ultraflex MALDI-TOF/TOF mass spectrometer. Indicated mass values were identified as BSA protein using the Mascot search engine (Score >300). No tryptic autocatalytic digestion signals were identified (Ref. A. Pich, unpublished, Medical School Hanover (MHH)).