TECHNICAL NOTES SEI

Biochemicals

Electrophoresis Bioseparation Life Sciences Specials

Product Description

Trypsin NB Premium Grade, MS approved is designed for digestion of proteins prior to sequence analysis. Each lot is qualified for use with in-gel digestion and mass spectrometric analysis. Based on excellent and proprietary production procedures, Trypsin NB Premium Grade, MS approved is of unique stability due to exceptional low autocatalytic activity (Fig. 1).

Trypsin NB Premium Grade, MS approved is a serine endopeptidase which specifically cleaves peptide bonds 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 considerable reduced when acidic residues are present on either side of a potentially susceptible bond [1].

Outstanding performance is guaranteed by:

- · Each lot MS approved
- · Exceptional low autoproteolysis
- · Extreme stability
- High purity
- · High specificity
- · No chymotryptic activity

Trypsin NB Premium Grade, MS approved is supplied as lyophilisate in vials at 25 µg each. It is manufactured by the pharmaceutical plant of Nordmark Arzneimittel GmbH & Co. KG, Germany.

Incubation time (h)	Activity (%)		
	Trypsin NB Premium Grade, MS appoved	Trypsin native, not modified	
0	100	100	
3	100	43	
5	87	30	
7	84	25	
22	46	5	

Tab.1: Stability of Trypsin NB Premium Grade, MS approved and Trypsin native, not modified in 20 mM Tris-HCl, pH 8.0 at 37 °C.

Trypsin NB Premium Grade, MS approved

Extreme Stability

Trypsin NB Premium Grade, MS approved is modified by reductive methylation and purified by chromatography, yielding a highly active molecule that is extremely resistant to autolytic digestion (Tab. 1).

High Purity

Trypsin NB Premium Grade, MS approved is a highly purified enzyme preparation that is free of activity from other proteases. The absence of chymotryptic activity is verified by purity and function control which is carried out for each lot (Fig. 1, Fig. 2).



Fig.1: Purity of Trypsin NB Premium Grade, MS approved by Reversed Phase HPLC.

High Specificity

The specificity of Trypsin NB Premium Grade, MS approved is verified with the oxidized B chain of insulin (insulin B_{ox}) as substrate. 25 µg of insulin B_{ox} are incubated with 0.5 µg Trypsin NB Premium Grade, MS approved at 37 °C for 18 h to detect traces of impurities of chymotrypsin (Fig. 2).



[1] Wilkinson, J. M. (1986): Fragmentation of Polypeptides by Enzymatic Methods. In: Practical Protein Chemistry: A Handbook. A. Darbre, ed., John Wiley and Sons, New York, N.Y.

TECHNICAL NOTES

Electrophoresis

Trypsin NB Premium Grade, MS approved

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 3. Lot specific generated spectrograms using bovine serum albumin (BSA) as substrate are available at tech.service@serva.de.



Fig. 3: Spectrogram of BSA digested with Trypsin NB Premium Grade. 300 ng BSA were separated by gel electrophoresis and digested with 10 ng/µl Trypsin NB Premium Grade in 50 mM NH₄HCO₃ 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 trypsin petid at m/z 2211 was identified that indicated autocatalytical digestion of Trypsin NB Premium Grade. In contrast, other commercial available modified trypsins exhibited autocatalytical activity under identical conditions. Mascot scores for protein identification were significant higher using Trypsin NB Premium Grade than for other modified trypsins (Ref: A. Pich, unpublished, Medical School Hanover (MHH)).

Contact us!

If you require more detailed information, please contact us:

Technical Support

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Ordering Information

Product	Quantity	Cat. No.
Trypsin NB Premium Grade, MS approved	4x25 μg	37284.01

CRESCENT CHEMICAL COMPANY 2 Oval Drive, Islandia, NY 11749 Phone:631-348-0333 · Fax: 631-348-033 crescent@creschem.com · www.creschem.com



Trypsin Peptide (TP) Standard for internal calibration

Trypsin Peptide (TP) Standard is delivered with each package of Trypsin NB Premium Grade, MS approved. The standard contains trypsin to generate masses m/z 842 and 2211 and thus allows individual addition of these standard peptides if needed. It facilitates easy internal calibration to enhance mass accuracy in MS analysis and can be adjusted to any experimental conditions (Fig. 4).



Fig. 4: Peptide mass fingerprint of BSA using Trypsin NB Premium Grade and TP Standard in a ratio of 5:1. BSA digestion was carried out with Trypsin NB Premium Grade and TP Standard. Desired masses of m/z 842 and 2211 are indicated.

Features at a glance:

- 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
- Convenient packaging: 4 vials at 25 μg each, Trypsin Peptide (TP) Standard is delivered with each package.
- * Unit definition: 1 U catalyzes the hydrolysis of 1 μmol Na-Benzoyl-L-arginine-4-nitroanilide hydrochloride (BAPNA) per minute at 30 °C, pH 8.0.

Related Products

Product	Quantity	Cat. No.
Trypsin NB Sequencing Grade, modified	4x25 μg	37283.01

