PRODUCT INFORMATION

Trypsin Sequencing Grade, modified — Cat. No. 37283

Product description:

General

Trypsin Sequencing Grade, modified 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 Sequencing Grade, modified is specially designed for digestion of proteins prior to mass spectrometric analysis

Features

- Source: porcine pancrease
- Purity: > 90 %
- Tryptic activity: > 6000 U/g*
- No chymotryptic activity detectable
- Modified by reductive methylation

Stability

Trypsin Sequencing Grade, modified is more resistant towards autolysis even at pH values in weakly basic range (table 1). Therefore the enzyme can be used in high concentrations in the digestion assay.

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

Incubation time [h]	Activity [%]	
	Trypsin Sequencing Grade, modified (Cat. No. 37283)	Trypsin native, not modified
0	100	100
3	100	43
5	87	30
7	84	25
22	46	5

Storage conditions

Trypsin Sequencing Grade, modified should be stored in a dry state at -15 °C to -25 °C

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Instructions for use:

General	Trypsin is routinely used in proteomics for mapping and protein sequencing due to its highly specific cleavage resulting in a limited number of tryptic peptides.
Digestion of proteins in solution	Lyophilized Trypsin Sequencing Grade, modified 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 to a final protease:protein ratio of 1:100 to 1:20 (w/w).
In-gel protein digestion	Lyophilized Trypsin Sequencing Grade, modified 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 solution 1:2.5 with 25 mM NH4HCO3, pH 8 and use 10 to 20 µl for rehydration / digestion of each gel piece. 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.

[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

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

Version 08/23

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