Trypsin-ultra™, Mass Spectrometry Grade

100 µg Lot: 0071609 Exp: 9/17
5 x 20 µg Store at −20°C

Description: Trypsin-ultra, Mass Spectrometry Grade is a serine endopeptidase. It selectively cleaves peptide bonds C-terminal to lysine and arginine residues (1). Trypsin-ultra is treated with L-(tosylamido-2-phenyl) ethyl chloromethyl ketone (TPCK) to inactivate any remaining chymotryptic activity. It is modified by acetylation of the ε-amino groups of lysine residues to prevent autolysis. Trypsin-ultra cleaves at Lys-Pro and Arg-Pro bonds at a much slower rate than other amino acid residues (2). Trypsin-ultra is a serine endopeptidase. It selectively cleaves peptide bonds C-terminal to lysine and arginine residues (1). Trypsin-ultra is treated with L-(tosylamido-2-phenyl) ethyl chloromethyl ketone (TPCK) to inactivate any remaining chymotryptic activity. It is modified by acetylation of the ε-amino groups of lysine residues to prevent autolysis. Trypsin-ultra cleaves at Lys-Pro and Arg-Pro bonds at a much slower rate than other amino acid residues (2).

Source: Isolated from bovine (Bos taurus) pancreas

Applications:
• Digestion of proteins for proteomic analysis by Mass Spectrometry
• Protein and peptide identification

Reaction Conditions: 1X Trypsin-ultra, Reaction Buffer. Incubate at 37°C.

Reagents Supplied with Enzyme: 2X Trypsin-ultra, Reaction Buffer.

1X Trypsin-ultra, Reaction Buffer:
50 mM Tris-HCl
20 mM CaCl₂
pH 8.0 @ 25°C

Note: Substrate must be in phosphate-free buffer to prevent calcium precipitation with both reconstituted enzyme and enzyme buffer.

Molecular Weight: 23,675 daltons

Reconstitution: Trypsin-ultra, Mass Spectrometry Grade should be reconstituted by the addition of 20–200 µl of high purity water. Rapid autolysis is a function of enzyme concentration.

Storage Conditions: Supplied freeze-dried from a sodium acetate and calcium chloride buffer. Store at −20°C.

Can be stored frozen in solution at −20°C for up to 2 weeks. A decrease in activity will occur if stored in solution. Use only freshly reconstituted protease for best results.

Quality Assurance: Trypsin-ultra, Mass Spectrometry Grade is free of glycerol and detergents which may interfere with Matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) Mass Spectrometry (MS) or liquid chromatography (LC) methods.

Quality Controls
Functional Test (Calcitonin Peptide Digestion): A 20 µl reaction in Trypsin-ultra, Mass Spectrometry Grade Reaction Buffer containing 2 µg of human calcitonin peptide and 0.1 µg of Trypsin incubated for 16 hours at 37°C results in the expected digestion products determined by MALDI-TOF MS analysis.

Functional Test (Cytochrome C Digestion): A 20 µl reaction in Trypsin-ultra, Mass Spectrometry Grade Reaction Buffer containing 2 µg of Cytochrome C and 0.1 µg of Trypsin incubated for 16 hours at 37°C results in the expected digestion products determined by MALDI-TOF MS analysis.

Specific Activity: The specific activity of Trypsin-ultra, Mass Spectrometry Grade is between 1.8 µmol min⁻¹ mg⁻¹ and 3.0 µmol min⁻¹ mg⁻¹.

Note: Trypsin-ultra, Mass Spectrometry Grade is acetylated on multiple lysine residues. This protein appears as a single band on SDS-PAGE. This sequence is also available at www.neb.com.

(see other side)
Trypsin-ultra, Mass Spectrometry Grade Protein Sequence:

1 IVGGYTCAENSVPYQVSLANYHFCGGGLINDQWVSAAHCYQYHIQVRLGYNID
61 VLEGGEQFIDASKIIHRHPKYYSSWTLDNDILLIKLSTPAVINARVSTLLLPSACASA
121 GTECLISGWNTLSSGVNYDPLLQCLVPALLSHADCEASYPGQITNNMICAGFLEG
181 GKDSCQGDSGGPVACNGQLQGIVSWGYGCAQKGPYTVKCNVYDVWIQETIAANS

References:


MALDI-TOF MS: *Issatchenka orientalis* Cytochrome c subjected to digestion by Trypsin-ultra, Mass Spectrometry Grade for 16 hours, dried and subjected to MALDI-TOF MS.