Endoproteinase AspN

Applications:
- Digestion of peptides of 5 kDa or less for proteomic analysis by Mass Spectrometry
- Protein and peptide identification

Reaction Conditions: 1X AspN Reaction Buffer. Incubate at 37°C.

Reagents Supplied with Enzyme:
2X AspN Reaction Buffer.

1X AspN Reaction Buffer:
50 mM Tris-HCl
2.5 mM Zinc Sulfate
pH 8.0 @ 37°C

Specific Activity: ~25 µmol/min/mg
Molecular Weight: 40,089.9 daltons

Reconstitution: Endoproteinase AspN should be reconstituted by the addition of 50–500 µl of high purity water. Rapid autolysis is a function of enzyme concentration.

Storage Conditions: Supplied in lyophilized form. 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: Endoproteinase AspN is free of glycerol and detergents which may interfere with Matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) or Electrospray Ionization (ESI) Mass Spectrometry (MS), or liquid chromatography (LC) methods.

Quality Controls
Endoproteinase AspN Activity:
Measured in two assays:
1. Peptide digestion and analysis by MALDI-TOF MS or ESI-TOF MS
2. Fluorometric substrate digestion and specific activity determination in digestion buffer:
   50 mM Tris-HCl (pH 8.0) and 2.0 mM Zinc Acetate.

50 µg Lot: 0021512
Store at −20°C Exp: 12/16

P8104S

Description: Endoproteinase AspN (flavastacin) is a zinc metalloendopeptidase which selectively cleaves peptide bonds N-terminal to aspartic acid residues (1).

Source: Purified from Flavobacterium meningosepticum.

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Endoproteinase AspN Protein Sequence:

1 TIVSSFIKTWPATVYTLPSQSLSTQAYNTFLTNINKAFDMISSKTSVKFVQRTNQTE
61 YITFTYSGNSPLGWKVRVNGIKIYNTTPAIIAHEIMHSMGIMHEQCRPDRQYIV
121 DTNRAODGTRHPYFVGVKSMDAKDIVTGDSTFGSKQRD
181 GLSAGDYAGINHLYGVNVTSTANGTYLTTSLGDKNIDTSGSTATGVDILVYATTG
241 NQKFIRKSEHYTFIJKSLDSTKVLTVRNGGTANQTVELRNTADDAKMLLFNLGN
301 EGFGFAPKNAKPLREVKDGLTNPLPVIDGSTQTLOPYTKQRFITLKV

Notes: Aspartic acid residues are strongly favored by Endoproteinase AspN in all buffer conditions we have examined (Tris-HCl, ammonium bicarbonate and potassium phosphate) (2).

Endoproteinase AspN is recommended for cleavage of peptides only. The cleavage rate of protein is much slower.

Endoproteinase AspN contains a O-linked carbohydrate on the protein. The protein appears as a single band by SDS-PAGE analysis. The protein sequence is also available at www.neb.com.

References: