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ProductInformation

Thermolysin
from *Bacillus thermoproteolyticus rokko*

Product Number **T7902**
Storage Temperature -0 °C

Product Description

Molecular Weight: 34.6 kDa (calculated)[1]
CAS Number: 9073-78-3
Enzyme Commission (EC) Number: 3.4.24.27
λ_{max}: 280 nm
Extinction coefficient: $E^{1\%}$ = 17.65
pI: 4.45
Synonyms: Protease from *Bacillus thermoproteolyticus rokko*, Thermophilic-bacterial protease

This product is cell culture tested and is suitable for use in cell culture experiments.

Thermolysin is a thermostable extracellular metalloendopeptidase that binds one zinc ion and four calcium ions as cofactors. It has a low substrate specificity, and thus produces a number of short fragments suitable for sequencing. Thermolysin is used to do limited proteolysis for peptide mapping and studies of protein structure and conformational changes.[2,3,4,5,6]

Thermolysin hydrolyzes protein bonds on the N-terminal side of hydrophobic amino acid residues, with preferential cleavage as follows:

X-(cleavage site)-Y-Z
X = any amino acid
Y = Leu, Phe, Ile, Val, Met, Ala
Z = any amino acid other than Pro

Cleavage N-terminal to Leu is preferred over cleavage N-terminal to Phe, which in turn is preferred over cleavage N-terminal to the other amino acids. The pH optimum of the reaction is 8.0 and the optimal temperature for activity is 70 °C. Thermolysin has considerable stability over the pH range 5 - 9.5.

The crystal structure of thermolysin to 1.6 Å resolution has been reported.[7] The isolation, cloning, and expression in *Bacillus subtilis* of the gene coding for thermolysin from *Bacillus thermoproteolyticus Rokko* has been published.[8]

Precautions and Disclaimer

For Laboratory Use Only. Not for drug, household or other uses.

Preparation Instructions

This product is soluble in acetate buffer, pH 7.5 (0.2 mg/ml), yielding a clear to very slightly hazy, colorless solution.

References

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3. Gwizdek, C., et al., Proteolytic mapping and substrate protection of the *Escherichia coli* melibiose permease. Biochemistry, **36(28)**, 8522-8529 (1997).
4. Fontana, A., et al., Probing the conformational state of apomyoglobin by limited proteolysis. J. Mol. Biol., **266(2)**, 223-230 (1997).
5. Alonso, M. C., et al., Proteolytic mapping of kinesin/ncd-microtubule interface: nucleotide-dependent conformational changes in the loops L8 and L12. EMBO J., **17(4)**, 945-951 (1998).
6. Burgess, R. R., et al., Mapping protein-protein interaction domains using ordered fragment ladder far-Western analysis of hexahistidine-tagged fusion proteins. Methods Enzymol., **328**, 141-157 (2000).
7. Holmes, M. A., and Matthews, B. W., Structure of thermolysin refined at 1.6-Å resolution. J. Mol. Biol., **160**, 623-639 (1982).
8. O'Donohue, M. J., et al., Cloning and expression in *Bacillus subtilis* of the npr gene from *Bacillus thermoproteolyticus Rokko* coding for the thermostable metalloprotease thermolysin. Biochem. J., **300(Pt 2)**, 599-603 (1994).

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