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ProductInformation

Clostripain from Clostridium histolyticum

Product Number **C 7403** Storage Temperature -0 °C

Product Description

EC Number: 3.4.22.8 CAS Number: 9028-00-6 Molecular Weight: 50 kDa¹

 $pl = 4.8-4.9^2$

Synonym: Clostridiopeptidase B

Clostripain is a sulfhydryl protease isolated from the culture filtrate of *Clostridium histolyticum*. It is composed of two polypeptide chains with molecular weights of 45 kDa and 12.5 kDa.³

Clostripain cleaves peptides at the C-terminal side of arginine residues. Lysine bonds are also cleaved, but at a much slower rate. Peptide digestion may be conducted at an enzyme to substrate ratio (w/w) of 1:100 at 37 °C in a buffer containing 1 mM CaCl₂ and 1 mM 2-mercaptoethanol. The following K_M values have been reported in the literature for clostripain: N α -benzoyl-L-arginine ethyl ester (0.25 mM) and N α -benzoyl-L-lysine methyl ester (3.0 mM). The pH optimum for the hydrolysis of L-arginine methyl ester in phosphate buffer is 7.2, while the optimal activity with N α -benzoyl-L-arginine ethyl ester is in the pH range of 7.4-7.8 with Tris-HCl buffer.

Clostripain is activated by dithiothreitol, cysteine, and other sulfhydryl reducing agents. The enzyme is also activated by the presence of calcium. Clostripain is inhibited by iodoacetic acid, phenylmethanesulfonyl fluoride, diisopropyl fluorophosphate, leupeptin, antipain, EDTA, and Co²⁺, Cu²⁺, Cd²⁺, Na⁺, and K⁺. ^{1,6}

Precautions and Disclaimer

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

Preparation Instructions

This enzyme is soluble in 10 mM MOPS buffer, pH 7.4, with 2.5 mM DTT and 1 mM CaCl₂ (0.2 mg/ml), yielding a clear solution.

Storage/Stability

Clostripain must be activated in the previously described buffer for at least 2-3 hours before use.

References

- Mitchell, W. M., and Harrington, W. F., Purification and properties of Clostridiopeptidase B (Clostripain). J. Biol. Chem., 243(18), 4683-4692 (1968).
- 2. The Enzymes, Vol. III, Boyer, P. D., ed., Academic Press (New York, NY: 1971), pp. 699-719.
- Gilles, A. M., et al., α-Clostripain. Chemical characterization, activity, and thiol content of the highly active form of Clostripain. J. Biol. Chem., 254(5), 1462-1468 (1979).
- 4. Cole, P. W., et al., Specificity and mechanism of clostripain catalysis. Biochemistry, **10(23)**, 4246-4252 (1971).
- 5. Mitchell, W. M., and Harrington, W. F., Clostripain. Meth. Enzymol., **19**, 635-642 (1970).
- 6. Giroux, E., and Vargaftig, B. B., Clostridiopeptidase B inhibition by plasma macroglobulins and microbial antiproteases. Biochim. Biophys. Acta, **525(2)**, 429-437 (1978).

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