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ProductInformation

α-Chymotrypsin TLCK Treated from bovine pancreas

Product Number **C 3142** Storage Temperature -0 °C

Product Description

Enzyme Commission Number (EC): 3.4.21.1

CAS Number: 9004-07-3 Molecular Weight: 25 kDa¹

pl: 8.75²

Extinction Coefficient: E^{1%}: 20.4 (280 nm),

 E^{mM} : 5.2 (281 nm)³

This material is treated with TLCK to inactivate trypsin which is usually present in chymotrypsin. α-Chymotrypsin has 241 amino acid residues contained in three polypeptide chains (A chain-13 residues, B chain-131 residues, and C chain-97 residues) linked by disulfide bridges. It selectively catalyzes the hydrolysis of peptide bonds on the C-terminal side of tyrosine, phenylalanine, tryptophan, and leucine. A secondary hydrolysis will also occur on the C-terminal side of methionine, isoleucine, serine, threonine, valine, histidine, glycine, and alanine. ¹ Its activity in organic solvents has been reported. ⁴

 $\alpha\text{-Chymotrypsin}$ is both activated and stabilized by $\text{Ca}^{2^+}.$ The enzyme is active in the presence of 0.1% SDS and 2 M guanidine hydrochloride. It is a serine protease and is inhibited by diisopropyl fluorophosphate (DFP), phenylmethanesulfonyl fluoride (PMSF), N-p-tosyl-L-phenylalanine chloromethyl ketone (TPCK), chymostatin, aprotinin, $\alpha_1\text{-antitrypsin},$ and $\alpha_2\text{-macroglobulin}.$ $\alpha\text{-Chymotrypsin}$ is also completely inhibited by 10 mM Cu $^{2^+}$ and Hg $^{2^+}.^1$

Precautions and Disclaimer

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

Preparation Instructions

This material is soluble in 1 mM HCl (10 mg/ml), yielding a clear solution.

Storage/Stability

Solutions prepared in 1 mM HCl containing 2 mM CaCl₂ may be stored at -20 °C as frozen aliquots for approximately 1 week. Autolysis will occur when stored at a higher pH. Calcium ion serves as a stabilizer.¹

Procedure

For peptide digestion, use a ratio (w/w) of approximately 1:60 for chymotrypsin:peptide. Perform peptide digests in 100 mM Tris HCl containing 10 mM CaCl2, pH 7.8, at 30 °C. Self digestion may occur if temperatures above 37 °C are used.

References

- Enzymes of Molecular Biology, Vol. 16, Burrell, M. M., ed., Humana Press (Totowa, NJ: 1993), pp. 277-281.
- 2. Ui, N., Isoelectric points and conformation of proteins. II. Isoelectric focusing of α -chymotrypsin and its inactive derivative. Biochim. Biophys. Acta, **229(3)**, 582-589 (1971).
- Nakagawa, Y., and Bender, M. L., Methylation of histidine-57 in α-chymotrypsin by methyl p-nitrobenzenesulfonate. A new approach to enzyme modification. Biochemistry, 9(2), 259-267 (1970).
- 4. Zaks, A., and Klibanov, A. M., Enzymatic catalysis in nonaqueous solvents. J. Biol. Chem., **263(7)**, 3194-3201 (1988).

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