Native Bacillus polymyxa Neutral Protease (Dispase)

Cat. No. NATE-0482

Lot. No. (See product label)

Introduction

Description Neutral protease (Dispase) is a non-mammalian animal origin free (AOF) metallo, neutral protease. Its mild proteolytic action makes the enzyme especially suitable for the preparation of primary cells and secondary (subcultivation) cell culture, since it is gentle on cell membranes. This protease is also used as a secondary enzyme in cell isolation and tissue dissociation applications, commonly used with collagense.

Applications Tissue disaggregation and subcultivation; Prevention of unwanted cell clumping; Preparation of cells for culture; Separation of intact epidermis from dermis and intact epithelial sheet in culture from the substratum (Kurt et al. 1989); Harvest and transfer of normal, diploid cells and cell lines (Matsumura et. al 1975); Gentle and intact detachment of epidermal cells (Kitano and Okada 1983).

Synonyms Bacillolysin; EC 3.4.24.28; Bacillus metalloendopeptidase; Bacillus subtilis neutral proteinase; anilozyme P 10; Bacillus metalloproteinase; Bacillus neutral proteinase; megateriopeptidase; Neutral Protease (Dispase)

Product Information

Source Bacillus polymyxa

Form lyophilized powder

EC Number EC 3.4.24.28

CAS No. 9001-92-7

Molecular Weight 32.5 kDa

Purity Chromatographically purified

Activity > 4 units per mg dry weight

Isoelectric point 5.14 (Theoretical)

Optimum pH 5.9-7.0 (Fogarty and Griffin 1973)

Composition The enzyme is known to contain 1g-atom of zinc per g-mol of purified enzyme. If this zinc component is removed by chelating agents such as EDTA or EGTA, an inactive apoenzyme is obtained. Calcium has been detected in the purified protein and is believed to play a role in maintaining the structure and configuration, and preventing autolysis (Griffin and Fogarty 1973 and Alvarez 2006).

Specificity Neutral protease is a non-specific metalloprotease. It cleaves fibronectin, collagen IV, and to a lesser extent collagen I, but it does not cleave collagen V or laminin. It hydrolyzes N-terminal peptide bonds of non-polar amino acid residues and may preferentially attack denatured and intercellular proteins with exposed hydrophobic amino acid residues. It is believed to bind one zinc ion and four calcium ions per subunit. Unlike other Bacillus species that produce neutral, alkaline, or a mixture of both proteases, Paenibacillus polymyxa is one of three species that produces only a neutral protease (Fogarty and Griffin 1973).

Activators Ca^{2+}, Mg^{2+}, Mn^{2+}, Fe^{2+}, and Al^{3+} Manganese has a greater activating effect in the case of

Activators	Ca2+, Mg2+, Mn2+, Fe2+, and Al3+ Manganese has a greater activating effect in the case of Paenibacillus polymyxa than other Bacilli neutral proteases (Griffin and Fogarty 1973).
Inhibitors	EDTA, EGTA, Hg2+; Other heavy metals (Griffin and Fogarty 1971); Not serum
Unit Definition	One Unit releases one micromole of Folin positive amino acids, measured as tyrosine, at 37°C, pH 7.5, using casein as the substrate.

Storage and Shipping Information

Storage	Store at 2-8°C
Stability	Stable at 2-8°C for 12 months. Aliquot and Store at -20°C after reconstitution with water or commonly used balanced salt solutions or media.