Neutral protease, Paenibacillus polymyxa

Cat. No.: HY-131577

CAS No.: 42613-33-2

Target: Endogenous Metabolite

Pathway: Metabolic Enzyme/Protease

Storage:

Powder	-20°C	3 years
	4°C	2 years
In solvent	-80°C	6 months
	-20°C	1 month

Neutral protease, Paenibacillus polymyxa

SOLVENT & SOLUBILITY

In Vitro	H_2O : 25 mg/mL (Need ultrasonic)

BIOLOGICAL ACTIVITY

Description	Neutral protease, Paenibacillus polymyxa (Dispase II, Dispase) is a neutral protease and potent fibronectinase and type IV collagenase. Neutral protease, Paenibacillus polymyxa can be used to separate the intact epidermis from the dermis and intact epithelial sheets in culture from the substratum[1][2].
In Vitro	Protocol 1. Reconstitute Dispase 1) Dissolve the non-sterile enzyme in Dulbecco's Phosphate-Buffered Saline (DPBS) without calcium and magnesium to 10 mg/mL. Filter sterilize through a 0.22 μm filter membrane. 2) Further dilute with DPBS without calcium and magnesium to a final concentration of 0.6-2.4 U/mL. Note: Concentrations higher than 2.4 U/mL are not recommended. 2. Dissociate Tissue 1) Mince tissue into 3-4 mm pieces with a sterile scalpel or scissors. 2) Wash the tissue pieces several times in sterile PBS without calcium and magnesium. 3) Submerge tissue fragments in Dispase solution (0.6-2.4 U/mL) and incubate at 37°C. 4) Stir slowly at 37°C until the tissue is sufficiently dissolved. Note: For compact tissues, we recommend incubating for 1 hour. Cells will not be adversely affected even after several hours in Dispase. 5) If necessary, separate the dispersed cells from residual tissue by passing the mixture through a sterile stainless steel or nylon mesh or simply decant the cells after larger fragments have settled. Fresh Dispase solution may be added to the remaining tissue fragments if further disaggregation is required. 6) Pellet cells by centrifugation and decant the enzyme solution. 7) Seed cells into culture vessels containing appropriate culture medium and incubate under predetermined conditions. 3. Subculture Cells 1) Aspirate culture medium and cover the cells with Dispase solution, prewarmed to 37°C. Incubate for 5 minutes at 37°C. 2) Decant the Dispase solution and incubate the cells for an additional 10 minutes at 37°C. 3) Monitor cell detachment using an inverted microscope. If necessary, incubate for an additional 15 minutes or until detachment is complete.

4) Suspend the cells in culture medium and pellet by centrifugation.
5) Resuspend the cells in a fresh culture medium. Plate the cells as usual.
MCE has not independently confirmed the accuracy of these methods. They are for reference only.

REFERENCES

[1]. Stenn KS, et al. Dispase, a neutral protease from Bacillus polymyxa, is a powerful fibronectinase and type IV collagenase. J Invest Dermatol. 1989 Aug;93(2):287-90.

[2]. Calvo B, et al. Dissociation of neonatal and adult mice brain for simultaneous analysis of microglia, astrocytes and infiltrating lymphocytes by flow cytometry. IBRO Rep. 2020 Jan 13;8:36-47.

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