



A Geno Technology, Inc. (USA) brand name

# **Immobilized Pepstatin**

For Purification of Cathepsins, Pepsin & Other Pepstatin Binding Molecules

(Cat. # 786-789)



## INTRODUCTION

G-Biosciences Immobilized Pepstatin is for the purification of cathepsins, pepsin, bacterial aspartic proteases, HIV proteases and other molecules that bind pepstatin. Pepstatin is isovaleryl-Val-Val-AHMHA-Ala-AHMHA where AHMHA= (3S, 4S)-4-amino-3-hydroxy-6-methyl-heptanoic acid and is a potent inhibitor of various aspartic proteases, including cathepsin D, renin, pepsin, bacterial aspartic proteases and HIV proteases. The resin consists of 6% beaded agarose that is covalently coupled to pepstatin through a diaminodipropylamine 23Å spacer arm and has a binding capacity of 1-2mg pepsin per millimeter of settled resin.

## KIT COMPONENTS

Cat. #	Description	Size
786-789	Immobilized Pepstain	5ml

Supplied as a 50% slurry in 0.5M NaCl and 0.02% thimerosal

## STORAGE CONDITIONS

Shipped at ambient temperature. Upon receipt store at 4°C, do NOT freeze.

## **IMPORTANT**

Activity: 1-2mg pepsin /ml of resin

• Support: 6% Beaded Agarose

## ADDITIONAL COMPONENTS

• Binding Buffer: 0.1M citrate, 0.5M sodium chloride, pH3.0

• Wash Buffer: 0.5M sodium chloride

Elution buffer: 0.1M Sodium bicarbonate, 0.5M sodium chloride, pH8.7

Gravity flow columns (see related products)

Sample dialyzed against Binding Buffer

## **PROTOCOL**

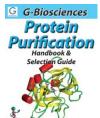
- 1. Aliquot 2ml slurry into an appropriate gravity column.
- 2. Allow the storage buffer to drain out and discard.
- 3. Equilibrate the resin with 5ml Binding Buffer.
- 4. Add the prepared sample to the resin and allow to pass through under gravity.
- 5. Wash the column with 4 times with four resin bed volumes of Binding Buffer.
- 6. Wash the column with4 times with four resin bed volumes of Wash Buffer.
- 7. Elute the bound protein with 6 times with four resin bed volumes of Elution Buffer and collect appropriate size fractions (0.5-1ml). Some proteins may require additional Elution Buffer volumes.
- 8. Monitor the elution of proteins by reading the absorbances at 280nm.
- 9. Store column by first washing with five resin volumes of water containing 0.05% sodium azide. Store upright at 4°C in deionized water with 0.05% sodium azide.

# TROUBLESHOOTING

Issue	Possible Reason	Suggested Solution
Protein of interest does not bind the resin	The protein does not binding pepstatin	Research pepstatin binding capability and optimal binding conditions. Ensure binding buffer ph is pH3.0
	Protein is inactive	Use extraction conditions to maintain proteins activity. We recommend our PE LB Systems.
Proteins fail to	Protein did not bind	Research pepstatin binding capability and optimal binding conditions Ensure binding buffer ph is pH3.0
elute	Above elution not appropriate	Use stronger elution method; use higher salt concentration or higher alkaline conditions

## RELATED PRODUCTS

Download our Protein Purification Handbook.



http://info.gbiosciences.com/complete-protein-purification-handbook

For other related products, visit our website at www.GBiosciences.com or contact us.



www.GBiosciences.com