Heparin-27A Use and Applications

1. Product Introduction

Heparin Beads is a highly cross-linked agarose gel matrix that uses an epoxy activation process to optimize the crosslinking of the matrix, providing good flow properties and high physicochemical stability. It utilizes a special extension arm and coupling technology to bond with high-purity heparin sodium ligand, which offers good capacity and selectivity. The ligand has low detachment and minimal non-specific adsorption, and the ligand is orientedly coupled to enhance the binding ability to antithrombin III. It is mainly used for the separation and purification of lactoferrin, antithrombin III, coagulation factors, lipoproteins, esterases, protein synthesis factors, hormones, steroid receptors, nucleic acid-binding enzymes, and interferons.

2. Product Information

Item	Specification
Ligand	Heparin Sodium
Matrix	Highly cross-linked agarose
Particle Size	45μm~165μm
Ligand Density	~5mg Heparin/mL Gel
Loading Capacity per mL	>15mg Lactoferrin
Maximum Flow Rate	100 to 500 cm/h (select appropriate flow rate based on column size)
Maximum Pressure Resistance	0.3 MPa
pH Stability	4 to 12 (Operational); 4 to 13 (Cleaning)
Chemical Stability	Stable in common aqueous buffers, 0.05M sodium acetate (pH 4.0), 20% ethanol, 4M sodium chloride; 8M urea, 6M guanidine hydrochloride, 0.1M sodium hydroxide
Storage Conditions	20% Ethanol + 0.05M Sodium Acetate solution at 2 to 8° C
Shelf Life	Stable storage at 2~8°C, with a shelf life of 3 years

3. Operating Instructions

1). **Buffer Preparation:**

Equilibrium Buffer: 20~50mM PB or Tris/HCl, pH 7.4~8.0, may add 0.15 M NaCl to inhibit non-specific adsorption.

Elution Buffer: 20~50mM PB or Tris/HCl + 1~2M NaCl, pH 7.4~8.0.

2). **Sample Preparation:** To avoid clogging the chromatography column, samples should be centrifuged or microfiltered (0.45μm). The feed amount is calculated based on the medium's capacity and the content of the target protein in the feed solution.

3). **Sample Purification:**

Equilibrium: Equilibrate the chromatography column with 5~10CV of equilibrium buffer until the conductivity and pH of the effluent remain unchanged (consistent with the equilibrium buffer).

Feeding: The sample buffer should be as consistent as possible with the equilibrium buffer. Solid samples can be dissolved and prepared with the equilibrium buffer; low-concentration sample solutions can be dialyzed with the equilibrium buffer; high-concentration sample solutions can be diluted with the equilibrium buffer.

Rinsing: Continue to rinse with the equilibrium buffer after loading until the baseline is stable.

Elution: Elute with the elution buffer (the NaCl concentration should be appropriately adjusted based on the binding strength of the target protein), and collect the effluent. Linear gradient or stepwise gradient elution can be used.

Regeneration: Flush with 5~10 CV of equilibrium buffer, then flush with 5~10 CV of pure water, and finally flush with 20% ethanol for 2~3 CV. Store at 2~8°C.

Important Note: If using pre-packed columns, the packing step can be omitted.

4. In-Place Cleaning

To avoid interference between different samples or when the medium is heavily contaminated (increased back pressure), the medium needs to be cleaned in place.

1). For proteins bound by ionic bonds, they can be cleaned with more than 2~3 CV of 2M NaCl and flushed with more than 3 CV of pure water.

2). For precipitated or denatured proteins, they can be cleaned with 0.1M NaOH (1~2h) and flushed with more than 3~10 CV of equilibrium buffer and more than 3 CV of pure water. They can also be cleaned with 6M Gua-HCl or 8M urea (0.5~1 h).

3). For proteins bound by hydrophobic interactions, they can be cleaned with 0.1~0.5% non-ionic detergent (1~2 h) and flushed with more than 3~10 CV of pure water.

Other Notes: To avoid column drying out, bubbles entering, and other issues, take care when packing, using, and storing the column.

5. Storage

Store in 20% ethanol containing 0.05M sodium acetate at 2~8°C