FT-WU6750

DSV Agarose

Product Information

Name: **DVS activated agarose gel**
for immobilizing biomolecules by binding to amino or hydroxyl groups

Part Number : **WU6750**, 5ml **WU6751**, 10ml **WU6752**, 25ml

Matrix : Sepharose® CL-4B
(Highly cross-linked spherical agarose, 4%; 40-165µm beads)
Max back pressure: 0.3 MPa, 3 bar
Max. flow rates: 15 ml/min/cm^2
Recommended flow rate: 8-12 ml/min/cm^2
Stability of the matrix: pH 2-11.

Graffting: Activation method: Divinyl Sulfone.
Binding capacity: 2-4mg of bovine serum albumin (BSA) per ml of gel

Form: Suspension in distilled water

Storage: +4°C (L) DO NOT FREEZE
stable for extended period of time when stored refrigerated and light-protected.

Introduction

Divinyl Sulfone (DVS) Activated Agarose is rigid Sepharose beads loaded with high density of reactive groups. The matrix is suitable for binding amino, or hydroxyl groups, present on biomolecules such as proteins and carbohydrates. Coupling reactions to biomolecules may be done between pH values of 8-10 pH.

Uptima DSV-Agarose is designed for various R&D applications *in vitro use*:

- immobilization of biomolecules conjugate for subsequent purifications
  typical useful molecules are antibodies, peptides, oligonucleotides...
- affinity purification, immunoprecipitation, depletion

Also available: **DVSA Activated agarose** #WU6740, for binding thiols.

Directions for use

Protocol: Immobilization of biotinylated ligand using Iminobiotiun-Agarose (r)

A. Buffers required for protein conjugation (using 10ml of DVS-A Activated Beads)

Please Scale up or down all values when conjugating more or less than 10 ml of beads

DVS-A Activated Beads: 10ml.
Double distilled water (100ml).
Neutral conjugation buffer (50ml): Phosphate Buffered Saline (PBS) pH 6-8
Conjugation buffer (50ml): 0.1N Sodium bicarbonate buffer (pH-8-10).

For any question,
contact your local distributor

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B. Coupling protocol

1. Suspend beads in storage solution and remove by pipette the desired amount of beads. Wash beads 3 times with double distilled H2O (ddH2O). Washing could be done either using a suitable filter funnel or by centrifugations/suspension cycles of 1 minute long each one, done at approximately 600-1,000xg. Do not exceed 1,000xg as beads may deform.

2. Prepare the protein/ peptide/ carbohydrate (biomolecules solution) in the conjugation buffer at 1-20mg/ml. Concentrations, between 5-20 mg/ml are preferred over concentrations between 1-5 mg/ml.

3. Add two volumes of biomolecules solution to one volume of washed DVS Activated Beads in polypropylene tube and mix gently.

4. Sample 100ul upper supernatant (without beads) as time zero reference sample and store in the refrigerator.

5. Mix slowly overnight at 4oC to 25oC (reaction is insensitive to temperature), preferably with the use of a rocker. Do not use magnetic stirrer for mixing

6. Sample 100ul upper supernatant (without beads) for conjugation efficiency determination.

7. Read both samples diluted to 0.1 to 0.5mg/ml protein using a spectrophotometer at OD 280.

8. Wash beads three times, 2 minutes each time with the conjugation buffer at room temperature, in order to remove unbound biomolecules.

9. Add approximately two gel volumes 0.1M ethanolamine or 0.1M Tris Base to block unconjugated DVS groups. Stir gently for 2 hours.

10. Wash gel three times 2 minutes each time, with 5 volumes of saline (0.7%w/v NaCl in ddH2O) added with 0.05% azide w/v, at room temperature, in order to remove unbound ethanolamine or Tris.

C. Storage

Store the gel refrigerated in any desired buffer added with 0.1% azide (w/v) until use.

Related Uptima products

DVSA Activated agarose #WU6740, for binding thiols groups.
Immobilized ligands: Monomeric avidin #UP29337 , iminobiotin UP88722A
Dialysis products: CelluSep tubings
Buffers, i.e. PBS #UP68723A

rev. J11E

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