# DVS Activated Beads

(Cat. 1007-5/10/25)

**DVS Activated Sepharose gel** **ver. 1.2 03.07**

## Introduction

**Adar Biotech's Divinyl Sulfone (DVS) Activated Beads** are 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. The DVS activated beads are stable for extended period of time when stored refrigerated and light-protected. Coupling reactions to biomolecules may be done between pH values of 8-10 pH.

## DVS Activated Beads characteristics.

Matrix: Sepharose CL-4B
Activation method: Divinyl Sulfone.
Binding capacity: 2-4 mg Bovine serum albumin (BSA) per ml.
Bead size: 40-165µm
Bead structure: Highly cross-linked spherical agarose, 4%
Max back pressure: 0.3 MPa, 3 bar
Max. flow rates: 15 ml/min/$cm^2$
Recommended flow rate: 8-12 ml/min/ $cm^2$
Storage: 4°C in DDW.

## Buffers required for protein conjugation (using 10ml of DVS Activated Beads)

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

DVS 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).

## A. Coupling protocol

1. Suspend beads in storage solution and remove by pipette the desired amount of beads. Wash beads 3 times with double distilled $H_2O$ ($ddH_2O$). Washing could be done either using a suitable filter funnel or by centrifugations/suspension cycles of 1 minute long each one, done at approximately 600-1,000*xg*. Do not exceed 1,000*xg* 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 4°C to 25°C (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 $ddH_2O$) added with 0.05% azide w/v, at room temperature, in order to remove unbound ethanolamine or Tris.

## C. Storage

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