BIO BASIC
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Product information

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Blue Gravity Column

Catalog #: BSP089
Size: 1 UNIT
Storage: 4~8°C*

*: Product will be shipped at ambient temperature. Check storage conditions.

Product Description:

The carbohydrate nature of the agarose base of Blue sefinose™ 6 fast flow provides a hydrophilic and chemically favorable environment for coupling and excellent chromatographic properties. The dye ligand, Cibacron Blue F3G-A, is covalently attached to the Blue sefinose™ 6 fast flow matrix via the triazine part of the dye molecule. Cibacron Blue F3G-A shows certain structural similarities to naturally occurring molecules like the co-factors NAD+ and NADP+ which enables it to bind strongly and specifically a wide range of proteins, including kinases, dehydrogenases and other enzymes requiring adenylyl-containing substances. Of the enzymes currently catalogued, approximately a third require a nucleotide coenzyme, suggesting the potential application of ligands such as Cibacron Blue F3G-A is extremely wide. The use of immobilised dyes in place of immobilised nucleotides has a number of advantages. Firstly, dye ligands have greater versatility, which allows the purification of several proteins without tedious preparation of a range of gels. Secondly, the chemical stability of the adsorbent allows its continued re-use. Thirdly, loss of activity through storage is not a problem.

We also offer pre-packed, ready-to-use 1 ml gravity columns for preparative affinity chromatography. Fast, simple and easy separations are provided by the combination of a specially designed column and a high performance affinity medium. The resin is very versatile and particularly suitable for the isolation and purification of albumin, interferon, a broad range of nucleotide requiring enzymes, a2-macro-globulin, coagulation factors, and nucleic acid binding proteins. The removal of albumin is a particularly important application since excess amounts of albumin can disturb the results of several tests.

Features:

Resin Volume	1ml/column
Ligand	Cibacron Blue F3G-A
Ligand Concentration	3 mg/ml gel or 5umol cibacron blue F3G-A/ml drained gel
Binding Capacity	10 mg HSA/ml gel
Particle Size Range	45-165 µm
Bead Structure	Highly cross-linked spherical agarose: 6%
Max. Flow Rate	4 ml/min (1 ml)
Recommended Flow Rate	1 ml/min (1 ml)
pH Stability	4–12(long time), 3–13(short time)
Temperature Stability	+4 °C to room temperature.
Compatible Reagents	8M urea, 6M guanidine hydrochloride and so on.
Incompatible Reagents	Tween-20 and similar detergent in binding buffer will reduce binding capacity for albumin.
Storage Temperature	+4 to +8°C
Storage Buffer	20% ethanol

Storage:

Upon receipt, store Blue Gravity Column at 4 to 8°C in 20% ethanol buffer.

Purification Procedures:
A) Column Preparation

1. Mix the slurry by gently inverting the bottle several times to completely suspend the resin.

2. Use a pipette to transfer an appropriate volume of resin slurry to the column. Allow the resin to settle and the storage buffer to drain from the column.

3. Equilibrate the column with four bed volumes of binding/wash buffer or until A280 is stable.

> **NOTE:** When using the pre-packed column, please wash the resin with 5 bed volume of 20% ethanol, then wash the resin with 5 bed volume of 2M NaCl, followed by 5 bed volume of final wash in binding/wash buffer. This way, any uncoupled Cibacron Blue F3G-A residues are removed thoroughly.

B) Purification Protein

1. Bind the protein to the resin. We recommend that the sample pH and conductivity be the same as that of the binding/wash buffer, so you can mix the sample and the binding/wash buffer with the volume ratio 1/10. In the meantime you can filter the sample through a 0.22–0.45 µm filter to prolong the working life of the resin. Apply the above mixed solution to the column with a flow-rate of 0.5-1 ml per minute. Collect and save the flow-through for analysis.

2. Wash the column with eight bed volumes of binding/wash buffer or until A280 is stable at the flow-rate of 1 ml per minute.

3. Elute the protein of interest with five bed volumes of Elution buffer. Collect the elute and dialyze it against appropriate buffer, according to the specific application of the target protein.

> **NOTE:** Elution conditions have to be optimized for different samples in order to obtain maximum purity and throughput.

Restore the binding property of old Blue sefinose™ 6 fast flow:

The following procedure is to remove any precipitated or denatured substances generated in previous production runs. A suggested protocol is as follows:

a) Wash the packed column with 0.1 M NaOH.

b) Wash with 3–4 bed volumes of 2 M potassium thiocyanate.

c) Wash immediately with at least 5 column volumes of sterile filtered binding buffer at pH 8.

d) For longer periods of storage, e.g. several weeks, we recommend that the resin be stored at +4 to +8°C in 20 % ethanol, 0.1 M KH2PO4, pH 8.0.

PRODUCTS ARE INTENDED FOR BASIC SCIENTIFIC RESEARCH ONLY.
NOT INTENDED FOR HUMAN OR ANIMAL USE.

