



Iminobiotin Beads 6FF

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1. Product Description

Iminobiotin Beads 6FF are used to purify avidin, streptavidin and streptavidin tag protein. Avidin-biotin or streptavidin-biotin requires elution under more stringent conditions such as 6 M guanidine hydrochloride, pH1.5. **Iminobiotin Beads 6FF** can be eluted at pH4.0 and the elution condition is milder.

Table 1. Characteristics of **Iminobiotin Beads 6FF**

Item	Description
Matrix	Highly cross-linked 6% agarose
Binding capacity	> 20 mg Streptavidin
Particle size (μm)	45–165
Maximum pressure	0.3 MPa, 3 bar
Storage buffer	0.01 M phosphate, 0.5 M NaCl, 0.02% NaN ₃ , pH6.8
Storage	2-8℃

2. Purification Procedure

2.1 Buffer Preparation

Water and chemicals used for buffer preparation should be high purity. It is recommended filtering the buffers by passing them through a 0.22μm or 0.45 μm filter before use.

Binding /Wash Buffer: 50 mM boric acid, 0.5 M NaCl, pH11.0

Elution Buffer: 50 mM sodium acetate, 0.5 M NaCl, pH4.0

Note: SDS may affect the binding of samples.

2.2 Sample Preparation

The sample should be adjusted to the composition of the binding buffer. This can be done either by diluting the sample with binding buffer or by buffer exchange. The sample should be filtered through a 0.45 μm filter or centrifuged before it is applied to the column.

2.3 Packing Columns

1) Remove air from the column dead spaces by flushing the end-piece and adapter with packing buffer. Make sure no air has been trapped under the column net.

2) Close the column outlet leaving the net covered with packing buffer.

3) Resuspend the beads stored in its container by shaking (avoid stirring the sedimented medium). Pouring the slurry down a glass rod held against the column wall will minimize the introduction of air bubbles.

If using a packing reservoir, immediately fill the remainder of the column and reservoir with packing buffer. Mount the adapter or lid of the packing reservoir and connect the column to a pump. Avoid trapping air bubbles under the adapter or in the inlet tubing.

4) Open the bottom outlet of the column and set the pump to run at the desired flow velocity. Ideally, **Iminobiotin Beads 6FF** is packed at a constant pressure of approximately 3 bar (0.3 MPa). If the packing equipment does not include a pressure gauge, use a packing flow velocity of approximately 400 cm/h (10 cm bed height, 25℃, low viscosity buffer). If the recommended pressure or flow velocity can not be obtained, use the maximum flow velocity the pump can deliver. This should also give a reasonable well-packed bed. Do not exceed 75% of the packing flow velocity in subsequent chromatographic procedures.

5) When the bed has stabilized, close the bottom outlet and stop the pump.

If using a packing reservoir, disconnect the reservoir and fit the adapter to the column. If using the column, carefully place the top filter on top of the bed before fitting the adapter.





- 6) With the adapter inlet disconnected, push the adapter down, approximately 2 mm into the bed, allowing the packing solution to flush the adapter inlet.
- 7) Connect the pump, open the bottom outlet and continue packing. The bed will be further compressed at this point and a space will be formed between the bed surface and the adapter.
- 8) Close the bottom outlet. Disconnect the column inlet and lower the adapter approximately 2 mm into the bed. Connect the pump. The column is now ready to use.

2.4 Sample Purification

- 1) Fill the syringe or pump tubing with binding buffer. Remove the stopper and connect the column to the syringe (with the provided connector), or pump tubing, "drop to drop" to avoid introducing air into the column. Remove the snap-off end at the column outlet.
- 2) Wash the column with 10 column volumes of binding buffer.
- 3) Apply the sample, using a syringe fitted to the connector or by pumping it onto the column.
- 4) Wash with 5 to 10 column volumes of binding buffer or until no material appears in the effluent.
- 5) Elute with 5 column volumes of elution buffer. Other volumes may be required if the interaction is difficult to break.

2.5 Analysis

SDS-PAGE will be used to test the purification effect of the samples obtained from the purified product (including the effluents, eluted and eluted components) and the original samples.

3. Related Products

Product	Cat. No.	Size
Iminobiotin Beads 6FF	SA059005	5 ml
	SA059025	25 ml
	SA059100	100 ml
	SA059500	500 ml
	SA05901L	1 L
	SA05910L	10 L

