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A Geno Technology, Inc. (USA) brand name

# Immobilized Monomeric Avidin Resin

(Cat. # 786-595, 786-596)



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## INTRODUCTION

G-Biosciences Immobilized Monomeric Avidin Resin is designed for the simple affinity chromatography purifications of proteins, antibodies and other molecules with a biotin tag. The resin consists of monomeric subunits of avidin covalently coupled to 4% agarose, offering a stable, reusable resin for the purification of biotinylated molecules.

Monomeric avidin offers a distinct advantage over native avidin, a tetrameric molecule, and streptavidin as it has a much lower biotin binding affinity,  $K_d=10^{-7}$  as opposed to  $K_d=10^{-15}$  for native avidin. This lower binding affinity allows elution of molecules with mild elution buffers (2mM D-Biotin in PBS), as opposed to the strong denaturing buffers (8M Guanidine•HCl, pH 1.5) used with native avidin.

The covalent attachment of monomeric avidin to the agarose ensures no detectable leaching of the avidin during biotin purification and offers a wide tolerance to chemicals. This ensures the resin can be reused at least 10 times with no loss of function.

## ITEM(S) SUPPLIED

Cat. #	Description	Size*
786-595	Monomeric Avidin, Immobilized	5ml resin
786-596	Monomeric Avidin, Immobilized	10ml resin

\* Immobilized monomeric avidin resin is supplied as a 50% slurry with 0.02% azide as a preservative.

## STORAGE CONDITIONS

It is shipped at ambient temperature. Upon arrival, store refrigerated at 4°C, **DO NOT FREEZE**. This product is stable for 1 year at 4°C.

## SPECIFICATIONS

- Biotin Binding Capacity:  $\geq 1.2$ mg biotinylated BSA/ml resin
- Bead Structure: 4% agarose

## ADDITIONAL ITEMS REQUIRED

- Biotinylated protein, antibody or other molecules in solution (1-3mg biotinylated protein/ml packed resin)
- Columns (optional): G-Biosciences offers columns for a large range of resin volumes (Cat. # 786-718 to 786-724)
- Wash Buffer: PBS
- Blocking/Elution Buffer: 2mM D-Biotin in PBS
- Regeneration Buffer: 0.1M Glycine, pH2.8

## PROTOCOL

1. Allow the resin and reagents to equilibrate to room temperature.
2. Pack an appropriate volume of Monomeric Avidin Resin into a column.
3. Wash the column with 4 column volumes of Wash Buffer.
4. Wash the column with 3 column volumes of Blocking/Elution Buffer to block any residual multimeric avidin molecules.
5. Wash away the biotin from the Monomeric Avidin by adding 6 column volumes of Regeneration Buffer.
6. Wash the column with 4 column volumes of Wash Buffer.
7. Add the biotinylated antibody/protein/molecule to the column and allow it to enter the resin. Apply 100µl Wash Buffer to ensure the sample fully enters the resin bed. Place a stopper on the bottom of the column and then apply a cap to the top of the column.

**NOTE:** *If the volume of the sample is too large, then add appropriate amount, incubate for 10 minutes, drain column and repeat steps 7. Do not exceed resin's binding capacity.*

8. Incubate the column at room temperature for up to 60 minutes.

**NOTE:** *Binding is only slightly increased with incubation.*

9. Wash the column with 6 column volumes of Wash Buffer.

**Note:** *Monitor the absorbance at 280nm, when a baseline has been reached the non-bound protein has been removed.*

10. Elute the protein with 6-10 column volumes of Blocking/Elution Buffer. Collect in 0.5-1ml fractions. Monitor protein collection with a suitable protein assay or absorbance at 280nm.

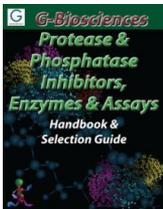
**NOTE:** *The Regeneration Buffer may be a better elution buffer for some proteins. If used, neutralize the pH with 0.1 volumes of 1M Tris, pH9 after elution.*

11. Regenerate the column with 4 column volumes of regeneration buffer. For long term storage, wash with 2.5 column volumes of Wash Buffer supplemented with 0.02% sodium azide. Store upright at 4°C.

**NOTE:** *The resin and columns can be regenerated at least 10 times without significant loss of performance.*

## RELATED PRODUCTS

Download our Protein Labeling & Conjugation Handbook.



<http://info.gbiosciences.com/complete-protein-labeling-coniugation-handbook/>

For other related products, visit our website at [www.GBiosciences.com](http://www.GBiosciences.com) or contact us.

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