



Product Information **Desalting Gravity Column**

Product information for BSP090:

Product Code: BSP090

Product size: 1 column

Introductions:

Bio Basic's Desalting Gravity Columns is a ready-to-use desalting columns for separating larger molecules from small molecules and buffer components. More specifically, molecules greater than 5,000 MW will elute in the void volume, separating them from smaller molecular weight buffer salts and reagents. Buffer-Exchange can also be achieved by first equilibrating a column with the desired buffer, then applying sample. Molecules greater than 5,000Da will then be exchanged into the desired buffer. These columns are pre-packed with cross-linked dextran and display excellent chromatographic properties and have good rigidity for easy handling and good flow properties. The Gel is stable, easy to use, and provides excellent sample recoveries. The gel can withstand water, salt solutions, organic solvents, alkaline and weakly acidic solutions. It is stable to heat and can be autoclaved dry or heated at 120°C in solution of neutral pH for 30 minutes without affecting its chromatographic properties. The desalting process or buffer exchange can therefore be performed in less than one hour. The column displays unique stop-flow characteristics which prevents drying of the gel and loss of precious samples. Recoveries of 90% or greater can be expected, assuming that the desalting buffer does not enhance nonspecific binding between the protein and the matrix.

Characteristics:

Total Column volume	10.0 ml
Recommended Sample	about 0.25-0.3 ml
Gel bed volume	2.5 ml
Void Volume (~1/3 gel volume)	0.9 ml
Exclusion Limit (for globular proteins)	5,000 daltons
Wet Bead Diameter	50-150 microns

Procedures for Buffer Exchange/Desalting:

1. Remove the top cap from the desalting column and decant the storage solution containing a preservative.
2. Remove the bottom cap and position the column upright in a suitable collection tube, Equilibrate the column using the buffer of your choice (DD water or exchange buffer) with approximately 3 resin-bed volumes.

Note: DeSalting Columns are supplied in DD H₂O containing preservative.



3. Place the column in a new collection tube, apply the sample to be desalted or for buffer exchange on to the column (add to the centre of the column. Sample volume no more than 10-15% of the gel-bed volume (~ 0.2-0.3ml) should be used.
4. Allow the sample to enter the gel. The column will stop flowing when the sample has entered the gel. This prevents the column from drying out. Equilibration buffer volume equal to the sample volume will drip out from the column tip
5. Place the tip of the column in a new collection tube and apply a volume of buffer equal to the Void volume you wish to collect(e.g. 0.5-1ml).
6. Allow the buffer to soak into the resin bed and collect the buffer that emerges from the column tip.
7. Repeat steps 5 and 6 until your protein is eluted from the column.
Note: Sample elution can be monitored by measuring the absorbance of each fraction at 280 nm. The first peak in absorbance will generally appear when the first or second void volume of buffer has been added after the sample is applied. This peak is the protein. Molecules smaller than the exclusion limit of the gel (i.e., buffer salts) will elute from the column in subsequent fractions. These fractions can be discarded after confirming that all fractions containing protein have been collected.
8. Desalting columns can be regenerated by washing with 10 column volumes of PBS or other buffer. For storage, wash the column with 5 column volumes of ultrapure water containing 0.02% sodium azide and cap the bottom then the top of the column when approximately 3 ml of solution remains above the gel.
9. Store the columns at 4°C.