INSTRUCTIONS

Protein Desalting Spin Columns

89849 89862

1377.1

Number	Description
89849	**Protein Desalting Spin Columns**, 25 columns, each column contains ~700 µl of resin in 10 mM Tris, pH 7.5 with 0.02% sodium azide
89862	**Protein Desalting Spin Columns**, 50 columns, each column contains ~700 µl of resin in 10 mM Tris, pH 7.5 with 0.02% sodium azide

Storage: Upon receipt store at 4°C. Product shipped at ambient temperature.

Introduction

Protein Desalting Spin Columns are designed to desalt or exchange buffer of protein samples with volumes of 30-120 µl. These devices have exceptional desalting characteristics with ≥ 95% retention of salts and small molecules while providing excellent recovery of proteins greater than 7,000 Da. Multiple samples can be processed in less than 5 minutes without cumbersome column preparation steps.

Procedure for Protein Desalting

A. Additional Materials Required

- Variable-speed bench-top microcentrifuge
- 1.5-2.0 ml microcentrifuge collection tubes

B. Protein Desalting Spin Column Preparation

1. Invert column to suspend slurry.
2. Twist off bottom closure and loosen cap. Do not snap off bottom. To remove, twist slightly in one direction and then the other direction.
3. Place column in 1.5-2.0 ml microcentrifuge collection tube.
4. Centrifuge at 1,500 × *g* for 1 minute to remove excess liquid.
5. Blot bottom of column on a paper towel to remove any excess trapped liquid.

C. Sample Loading

Note: Sample yield and purity obtained largely depends on the sample loading volume. For best results, load from 30 to 120 µl of sample. If sample is of high ionic strength, such as 0.5-1 M $CaCl_2$, reduce maximum sample volume to 75 µl.

1. Place column in a new collection tube, remove cap and apply 30-120 µl of sample to the center of the compacted resin bed. Be careful not to disturb the resin or to allow sample to flow around the resin bed.
2. (Optional) To improve recovery percentage of low molecular weight proteins or for small sample volumes, add 20-40 µl of 10 mM Tris buffer, pH 7.5, on top of the resin. Do not exceed a total sample plus stacker volume of 120 µl.
3. Centrifuge at 1,500 × *g* for 2 minutes. The desalted sample is in the collection tube. Discard desalting column after use.

Procedure for Buffer Exchange

Additional Materials Required

- Variable-speed bench-top microcentrifuge
- 1.5-2.0 ml microcentrifuge collection tubes
- Buffer for exchange

A. Protein Desalting Spin Column Preparation

1. Invert column to suspend slurry.
2. Twist off bottom closure and loosen cap. Do not snap off bottom. To remove, twist slightly in one direction and then the other direction.
3. Place column in 1.5-2.0 ml microcentrifuge collection tube.
4. Centrifuge at 1,500 × *g* for 1 minute to remove excess liquid.
5. Add 400 µl of exchange buffer to the top of column.
6. Centrifuge the column at 1,500 × *g* for 1 minute to remove excess liquid.
7. Repeat steps 5-6 two to three additional times, discarding buffer from the collection tube.

B. Sample Loading

1. Place column in a new collection tube, remove cap, and apply 30-120 µl of sample to the center of the compacted resin bed. Be careful not to disturb the resin or to allow sample to flow around the resin bed.
2. (Optional) To improve recovery percentage of low molecular weight proteins or for small sample volumes, add 20-40 µl of exchange buffer on top of the resin. Do not exceed a total sample plus stacker volume of 120 µl.
3. Centrifuge at 1,500 × *g* for 2 minutes. The desalted sample is in the collection tube. Discard desalting column after use.

Troubleshooting

Problem	Cause	Solution
Sample or buffer does not flow through resin	Centrifugation problem	Ensure that centrifuge is in proper working condition
		Ensure bottom closure is removed
Contamination in sample	Improper sample loading	Load sample directly in center of the resin bed; tip touch to expel all sample; do not “blow out” the tips
		Avoid contact with sides of the column
	High molecular weight contaminate	If contaminant is 700-2,000 Da, lower sample volume applied to the column
	Centrifugation problem	Do not exceed recommended centrifuge times or speeds
Low yield	Centrifugation problem	Apply 20-40 µl buffer overlay on top of sample before centrifugation
		Increase sample concentration
		Increase load volume

Related Thermo Scientific Products

43240 **D-Salt™ Polyacrylamide Desalting Columns**, 5 × 5 ml

69550 **Slide-A-Lyzer® MINI Dialysis Unit 3.5K MWCO**, 10-100 µl capacity, 50/pkg

The Slide-A-Lyzer® MINI Dialysis Unit is protected by U.S. Patent # 6,039,871.

This product (“Product”) is warranted to operate or perform substantially in conformance with published Product specifications in effect at the time of sale, as set forth in the Product documentation, specifications and/or accompanying package inserts (“Documentation”) and to be free from defects in material and workmanship. Unless otherwise expressly authorized in writing, Products are supplied for research use only. No claim of suitability for use in applications regulated by FDA is made. The warranty provided herein is valid only when used by properly trained individuals. Unless otherwise stated in the Documentation, this warranty is limited to one year from date of shipment when the Product is subjected to normal, proper and intended usage. This warranty does not extend to anyone other than the original purchaser of the Product (“Buyer”).

No other warranties, express or implied, are granted, including without limitation, implied warranties of merchantability, fitness for any particular purpose, or non infringement. Buyer’s exclusive remedy for non-conforming Products during the warranty period is limited to replacement of or refund for the non-conforming Product(s).

There is no obligation to replace Products as the result of (i) accident, disaster or event of force majeure, (ii) misuse, fault or negligence of or by Buyer, (iii) use of the Products in a manner for which they were not designed, or (iv) improper storage and handling of the Products.

Current versions of product instructions are available at *www.thermo.com/pierce*. For a faxed copy, call 800-874-3723 or contact your local distributor.