INSTRUCTIONS



ZebaTM Spin Desalting Columns

1729.8

Number	Description
89889	Zeba Spin Desalting Columns, 2mL, 5 each, for 200-700µL samples
89890	Zeba Spin Desalting Columns, 2mL, 25 each, for 200-700µL samples
89891	Zeba Spin Desalting Columns, 5mL, 5 each, for 500-2000µLsamples
89892	Zeba Spin Desalting Columns, 5mL, 25 each, for 500-2000µL samples
89893	Zeba Spin Desalting Columns, 10mL, 5 each, for 700-4000µL samples
89894	Zeba Spin Desalting Columns, 10mL, 25 each, for 700-4000μL samples
	Note: Columns are recommended for processing compounds > 7000Da. The resin slurry is supplied in 0.05% sodium azide.

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

Introduction

The Thermo ScientificTM ZebaTM Spin Desalting Columns contain a high-performance resin that offers exceptional desalting or buffer-exchange for protein samples. Samples containing as low as $20\mu g$ of protein/mL can be processed with unsurpassed protein recovery and $\geq 95\%$ retention of salts and other small molecules (< 1000Da). These columns require no chromatography system or cumbersome preparation or equilibration. The spin-column method eliminates waiting for samples to emerge by gravity flow and the subsequent monitoring of fractions for protein recovery, enabling the processing of multiple samples in ~ 8 minutes.

Procedure for Desalting or Buffer Exchange

A. Additional Materials Required

- Variable-speed centrifuge
- 15mL conical collection tubes or equivalent for the 2mL and 5mL spin columns
- 50mL conical collection tubes for the 10mL spin columns
- Buffer for exchange

B. Protein Desalting Spin Column Preparation

- 1. Twist off the column's bottom closure and loosen cap. Place column in a collection tube.
- Centrifuge column at 1000 × g for 2 minutes to remove storage solution. Place a mark on the side of the column where
 the compacted resin is slanted upward. Place column in centrifuge with the mark facing outward in all subsequent
 centrifugation steps.

Note: Resin will appear compacted after centrifugation.

- 3. Add buffer or ultrapure water to the column.
 - For 2mL columns use 1mL of buffer.
 - For 5mL columns use 2.5mL of buffer.
 - For 10mL columns use 5mL of buffer.
- 4. Centrifuge at $1000 \times g$ for 2 minute to remove buffer or ultrapure water.
- 5. Repeat Steps 3 and 4 two or three additional times, discarding buffer from the collection tube.



C. Sample Loading

1. Place column in a new collection tube, remove cap and slowly apply sample to the center of the compact resin bed.

Note: See Table 1 for recommended sample volumes for each column size.

2. (Optional) To ensure maximal protein recovery from low-volume samples, apply a stacker of ultrapure water or buffer to the resin bed after the sample has fully absorbed.

Note: See Table 1 for sample volumes for which a stacker is recommended.

3. Centrifuge at $1000 \times g$ for 2 minutes to collect the sample. Discard column after use.

Table 1. Recommended sample and stacker volumes to use with the Thermo Scientific Zeba Spin Desalting Columns.

Column	Sample Volume	Optional Stacker Volume*
2mL	$200\text{-}700\mu L$	$40\mu L$ stacker for samples $< 350\mu L$
5mL	500-2000μL	$100\mu L$ stacker for samples $< 750\mu L$
10mL	$700\text{-}4000\mu L$	$200\mu L$ stacker for samples $< 1500\mu L$

^{*} To ensure maximal protein recovery, apply a stacker to the resin after sample has entered the resin bed.

Troubleshooting

Problem	Possible Cause	Solution
Sample or buffer does not	Centrifugation problem	Ensure that centrifuge is in proper working condition
flow through resin		Ensure bottom closure is removed
		Ensure top cap is loosened
Sample contamination	Improper sample loading	Apply sample directly to center of the resin bed; touch tip to resin to expel all sample
		Avoid contact with sides of column
	Improper centrifugation	For fixed-angle rotors, place column in the same orientation each time
		Do not exceed recommended centrifugation speed or time
Low yield	No stacker used	Apply a stacker above sample
	Unstable protein	Equilibrate column in a suitable buffer

Related Thermo Scientific Products

23225	Pierce BCA Protein Assay Kit, for 20-2000µg/mL samples
23235	Pierce Micro BCA Protein Assay Kit, for 0.5-20µg/mL samples
89877	Zeba Micro Spin Desalting Columns, 25/pkg, for 2-12μL samples
89879	Zeba Micro Spin Columns (Empty), 50/pkg, 100µL gel-bed capacity
89882	Zeba Spin Desalting Columns, 0.5mL, 25/pkg, for 30-130μL samples



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