

PD-10 Desalting Columns, PD MidiTrap™ G-25, PD MiniTrap™ G-25, PD SpinTrap™ G-25, and PD MultiTrap™ G-25

PD-10 Desalting Columns, PD MidiTrap G-25, PD MiniTrap G-25, PD SpinTrap G-25, and PD MultiTrap G-25 (Fig 1) are prepacked, single-use columns and 96-well filter plates for buffer exchange and cleanup of biological samples, for example proteins and carbohydrates. These columns and 96-well filter plates are members of the Trap platform, which addresses the need for flexible, small-scale preparation of protein samples prior to downstream analytical techniques such as gel electrophoresis, liquid chromatography, and LC-MS. Gel filtration, based on Sephadex™ G-25 Medium, enables group separation of biomolecules (with a molecular weight above 5000) from contaminants such as salts, dyes, and radioactive labels. This range of columns and plates covers the sample volume range from 70 µl to 2.5 ml. The protocols for the gravity columns (PD-10 Desalting Columns, PD MidiTrap G-25, and PD MiniTrap G-25) are also optimized to enable centrifugation, which results in less dilution of the eluted sample (Table 1).

Key benefits include:

- Convenient and rapid cleanup with high reproducibility and the potential for high throughput by running multiple samples in parallel
- A wide range of applications including desalting, buffer exchange, and the removal of low-molecular weight compounds
- High desalting capacity
- The use of centrifugation with all gravity columns



Fig 1. PD protein sample preparation products are prepacked with Sephadex G-25 Medium. The PD G-25 product range includes (from top to bottom), PD MultiTrap G-25, PD MiniTrap G-25 (two columns shown), PD MidiTrap G-25 (two columns shown), PD-10 Desalting Column, and PD SpinTrap G-25.

Characteristics

PD-10 Desalting Columns, PD MidiTrap G-25, PD MiniTrap G-25, PD SpinTrap G-25, and PD MultiTrap G-25 cover a sample volume range from 2.5 ml down to 70 µl (Table 1). They are prepacked with Sephadex G-25 Medium, which is one of the most useful and widely documented gel filtration matrices backed with comprehensive technical and regulatory support for production applications. (For more information, see the application note "Desalting and buffer exchange with Sephadex G-25," code number 18-1127-73). The columns and 96-well plates are manufactured from biocompatible polypropylene. Special column and plate frits protect the medium from running dry during use. The main characteristics of Sephadex G-25 Medium and the PD products are listed in Tables 2 and 3.



Table 1. Sample volume ranges of PD products

	Sample volume (gravity)	Sample volume (spin)
PD MultiTrap G-25		70–130 µl
PD SpinTrap G-25		70–130 µl
PD MiniTrap G-25	0.1–0.5 ml	0.2–0.5 ml
PD MidiTrap G-25	0.5–1.0 ml	0.75–1.0 ml
PD-10 Desalting Columns	1.0–2.5 ml	1.75–2.5 ml

Table 2. Characteristics of Sephadex G-25 Medium

Matrix	Cross-linked dextran
Separation mechanism	According to size
Particle size range	85–260 µm
Exclusion limit (M_r)	5000
Chemical stability	All commonly used buffers
Working pH range	2–13

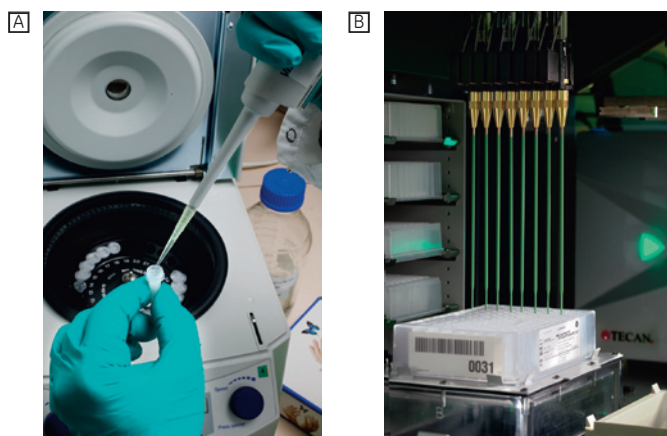
Table 3. Characteristics of PD products prepacked with Sephadex G-25

	PD MultiTrap G-25	PD SpinTrap G-25	PD MiniTrap G-25	PD MidiTrap G-25	PD-10 Desalting Columns
Volume of prepacked medium	500 µl/well	500 µl	2.1 ml	3.45 ml	8.3 ml
Packed bed dimensions			0.97 × 2.8 cm	1.3 × 2.6 cm	1.45 × 5.0 cm
Well/column volume	800 µl	800 µl	5 ml	8.5 ml	13.5 ml
Void volume	~ 150 µl	~ 150 µl	~ 0.5 ml	~ 1.0 ml	2.5 ml
Maximum sample volume	130 µl	130 µl	0.5 ml	1.0 ml	2.5 ml
Volume of eluted sample (gravity)			1.0 ml	1.5 ml	3.5 ml
Volume of eluted sample (spin) ¹	130 µl	130 µl	0.5 ml	1.0 ml	2.5 ml
Recovery ²	70% to 90%	70% to 90%	70% to 95%	70% to 95%	70% to > 95%
Desalting capacity	> 85%	> 85%	> 90%	> 90%	> 90%
Plate/column material	polypropylene and polyethylene	polypropylene and polyethylene	polypropylene and polyethylene	polypropylene and polyethylene	polypropylene and polyethylene
Storage solution	20% ethanol	0.15% Kathon™	0.15% Kathon	0.15% Kathon	0.15% Kathon
Storage temperature	4°C to 30°C	4°C to 30°C	4°C to 30°C	4°C to 30°C	4°C to 30°C

¹ Applied volume = eluted volume² Biomolecule dependent

Small-scale cleanup

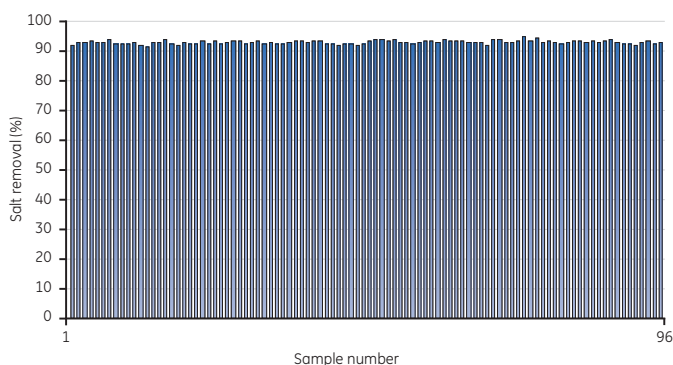
PD SpinTrap G-25 and PD MultiTrap G-25 are designed for small-scale cleanup and are valuable tools for screening purposes and high-throughput applications. SpinTrap columns only require a standard microcentrifuge (Fig 2A). MultiTrap 96-well filter plates allow cleanup by centrifugation, either manually or automated with robotics (Fig 2B).

**Fig 2.** (A) PD SpinTrap G-25 sample preparation. (B) PD MultiTrap G-25 sample automated preparation in a robotic system.

Typical desalting capacity is above 85% with recoveries between 70% and 90% (biomolecule dependent).

In a run of 96 parallel wells with bovine serum albumin (BSA) in 1 M NaCl, the salt removal capacity varied by 1% (relative standard deviation, Fig 3) and the recovery of total loaded material varied by 3%.

Trap product: PD MultiTrap G-25
 Sample: 1000 µg/ml bovine serum albumin (BSA) in 1 M NaCl
 Sample volume: 130 µl in each well
 Equilibration buffer: Milli-Q™ water

**Fig 3.** Removal of NaCl from BSA on a PD MultiTrap G-25 96-well plate showed highly reproducible results. The average desalting capacity was 93% and the well-to-well variation was 1% (relative standard deviation).

Columns with both gravity and spin protocols

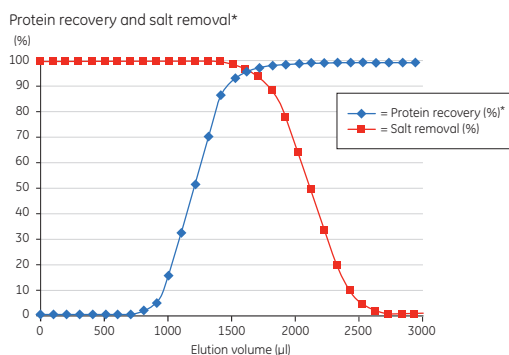
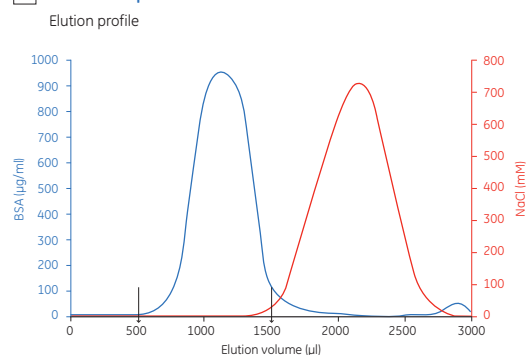
PD-10 Desalting Columns, PD MidiTrap G-25, and PD MiniTrap G-25 provide two possible application protocols, gravity and spin. The typical desalting capacity, for both protocols, is above 90% with recoveries between 70% and 95% (biomolecule dependent).

Gravity protocol

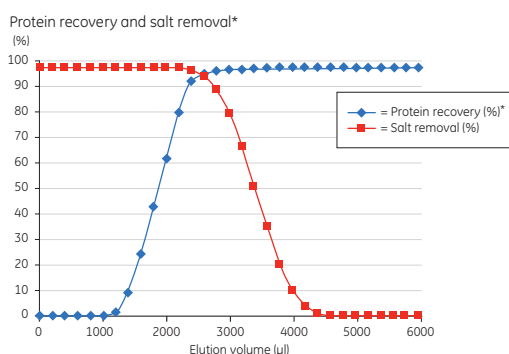
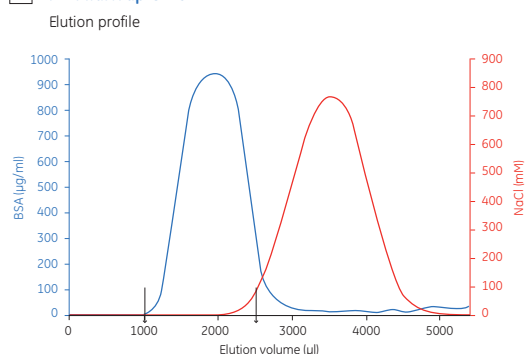
By using the gravity protocol, a simple cleanup of the sample is done on the lab bench with one or several columns in parallel without any need for a purification system. To simplify the use of PD-10 Desalting Columns with the gravity protocol, the LabMate™ PD-10 Buffer Reservoir may be used (see "Ordering information"). Using buffer reservoir, wash and equilibration buffers can be applied in one step. Elution profiles for BSA using the gravity protocol are shown in Figure 4.

Trap products: A) PD MiniTrap G-25 B) PD MidiTrap G-25, and C) PD-10 Desalting Column
Sample: 1000 µg/ml bovine serum albumin (BSA) in 1 M NaCl
Sample volumes: A) 500 µl, B) 1000 µl, and C) 2.5 ml
Equilibration buffer: Milli-Q water

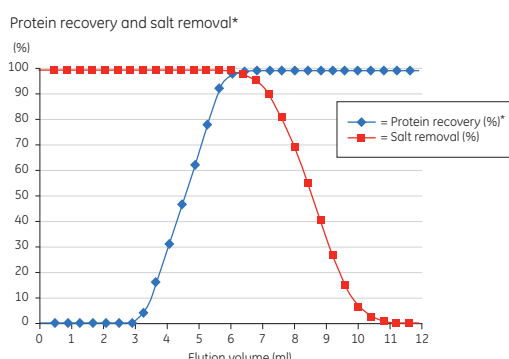
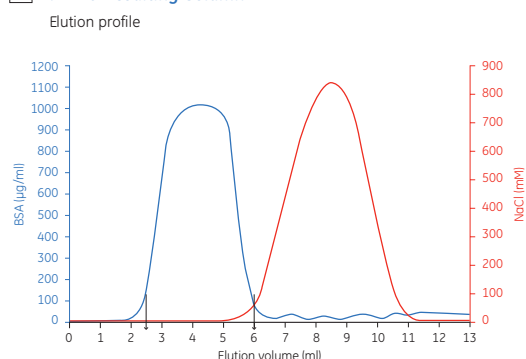
A PD MiniTrap G-25



B PD MidiTrap G-25



C PD-10 Desalting Column



Spin protocol

By using the spin protocol, the samples are run in parallel in a standard centrifuge. The spin protocol gives minimal dilution of the eluted sample. Four adapters are included in each product package to enable easy use of a standard centrifuge; see Figure 5. To facilitate increased throughput, a 10-pack package of adapters can be ordered separately.

Fig 5. Spin adapters are used together with PD-10 Desalting Columns, PD MidiTrap G-25, and PD MiniTrap G-25 to enable use in a standard centrifuge.

Fig 4. Removal of NaCl from BSA using the gravity protocol. The protein recovery (between arrows) was 95% for PD MiniTrap G-25 and PD MidiTrap G-25 (A & B) and > 95% for PD-10 Desalting Column (C). The desalting capacity was > 98% for PD MidiTrap G-25 (B) and > 99% for PD MiniTrap G-25 and PD-10 Desalting Column (A & C). The images to the right illustrate the protein recovery and salt removal versus the total elution volume on the used column.

* The recovery curves are normalized against the total amount of loaded sample.

Efficient cleanup of carbohydrates before enzymatic cleavage

The PD products prepacked with Sephadex G-25 Medium are excellent for desalting biomolecules other than proteins, for example carbohydrates. To demonstrate this, bovine intestinal ^3H -labeled heparan sulfate was eluted from a DEAE column using a high salt concentration. The eluate was then run on a PD MidiTrap G-25 column before enzymatic cleavage. Due to high peak resolution, the sample was collected with high recovery in a very low concentration of salt. The elution profile is shown in Figure 6.

Trap product: PD MidiTrap G-25
 Sample: 18 600 cpm bovine intestinal ^3H -labeled heparan sulfate (^3H)HS in 1.5 M NaCl
 Sample volume: 0.5 ml
 Equilibration buffer: Distilled water

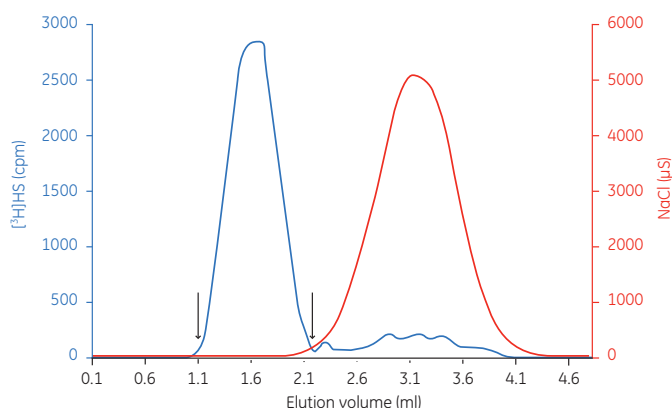


Fig 6. Removal of NaCl from ^3H HS on a PD MidiTrap G-25 column. The fractions were analyzed with regard to conductivity and radioactive content. Fractions 11 through 21 (between arrows) contained 87% of the total recovery of ^3H HS. The recovery volume was 1.1 ml with a NaCl concentration of 20 mM, corresponding to > 98% salt removal.

Ordering information

Products	Quantity	Code no.
PD-10 Desalting Columns	30 columns	17-0851-01
PD SpinTrap G-25	50 columns	28-9180-04
PD MultiTrap G-25	4 × 96-well plates	28-9180-06
PD MiniTrap G-25	50 columns	28-9180-07
PD MidiTrap G-25	50 columns	28-9180-08
MiniSpin Adapter	10	28-9232-43
MidiSpin Adapter	10	28-9232-44
PD-10 Spin Adapter	10	28-9232-45
Collection plate 500 µl V-bottom	5 × 96-well plates	28-4039-43
(for collection of fractions from MultiTrap)		
LabMate PD-10 Buffer Reservoir	10	18-3216-03

Related products

PD MiniTrap G-10	50 columns	28-9180-10
PD MidiTrap G-10	50 columns	28-9180-11
HiTrap™ Desalting	5 × 5 ml	17-1408-01
HiTrap Desalting ¹	100 × 5 ml	11-0003-29
HiPrep™ 26/10 Desalting	1 × 53 ml	17-5087-01
HiPrep 26/10 Desalting	4 × 53 ml	17-5087-02

¹ Pack size available by special order

Literature

Gel Filtration Principles and Methods, Handbook	18-1022-18
Gel Filtration Columns and Media, Selection Guide	18-1124-19
Sample preparation for analysis of proteins, peptides and carbohydrates, Selection Guide	18-1128-62
Protein Purification, Handbook	18-1132-29
Desalting and Buffer Exchange with Sephadex G-25, Application Note	18-1127-73

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