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**Ordering Information**

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**70 ml Capacity Slide-A-Lyzer G2 Dialysis Cassettes**

- 6/pkg.

**12 ml Capacity Slide-A-Lyzer Dialysis Cassettes**

- 6/pkg.

**3 ml Capacity Slide-A-Lyzer Dialysis Cassettes**

- 8/pkg.

**0.5-3 ml Capacity Slide-A-Lyzer Dialysis Cassettes**

- 10/pkg.

**0.5-3 ml Capacity Slide-A-Lyzer Dialysis Cassettes**

- 10/pkg.

**0.2-0.5 ml Capacity Slide-A-Lyzer Dialysis Cassettes**

- 10/pkg.

**10-100 μl Capacity Slide-A-Lyzer Dialysis Cassettes**

- 50/pkg.

**10K MWCO Membrane Products**

- Dialysis Cassette
- Dialysis Cassette Kit
- Dialysis Unit
- Dialysis Unit Kit
- Plus Microtubes

**20K MWCO Membrane Products**

- 0.5-3 ml 6/pkg.
- 0.1-0.5 ml 10/pkg.
- 10-100 μl 250/pkg.
- 10-100 μl 50/pkg.
- 10-100 μl 250/pkg.

**15 ml Capacity Slide-A-Lyzer Dialysis Cassettes**

- 8/pkg.

**10-100 μl Capacity Slide-A-Lyzer Dialysis Cassettes**

- 10/pkg.
- 100 μl 250/pkg.
- 100 μl 50/pkg.
- 100 μl 250/pkg.

**Price**

- U.S.
- £
- €

**Contact Information**

- Thermo Fisher Scientific, Inc.
- Tel: +32 53 85 71 84
- Tel: 0800 50 82 15
- Tel: +32 53 85 71 84
- Tel: 0800 50 82 15
- Email: sales@thermoscientific.com
- www.thermofisher.com

Frequently Asked Questions

High-Performance Dialysis

3.5K MWCO Membrane Products

2K MWCO Membrane Products

SnakeSkin Dialysis Tubing

Protein Concentrators

Cell Lysis Solutions

Desalting Columns and Plates

Detergent Removal Columns

Concentrating with Slide-A-Lyzer

Teflon is a registered trademark of E.I. du Pont de Nemours & Co., Inc.

*Although slides are shipped liquid free, prior to opening a container, it is recommended that the Slide-A-Lyzer Dialysis Cassette be rinsed thoroughly. The Slide-A-Lyzer Dialysis Cassette is compatible with these additional material media.

To order, call 800-476-3720 or visit 855-660-0761. Outside the United States, contact your local distributor or distributor.
High-Performance Dialysis

Dialysis is a separation technique that gained popularity in life science laboratories during the 1950s. Research papers of that era described dialysis as a new, cutting-edge tool that scientists could use to unravel complex mixtures of biomacromolecules. Many of the dialysis theories established at that time are the cornerstones for contemporary products featured in this brochure. There are, however, two major differences between the dialysis tools of yesterday and today – preparation time and the amount of sample loss due to leaks. Early laboratory dialysis methods involved dedicating a significant amount of time to membrane preparation; Thermo Scientific Pierce Dialysis Products are essentially ready to use and resist sample leakage.

New developments in dialysis techniques were stagnant during the end of the 20th century, while ultrafiltration systems flourished fueled by advances in non-cellulose membranes and accessibility of bench-top centrifuges. Ultrafiltration via centrifugation was the established convention until we introduced the Slide-A-Lyzer Dialysis Cassette in 1994.

Products are essentially ready to use and resist sample leakage.

Enlarged view of Slide-A-Lyzer Cassette interior

Old dialysate discarded and replaced with 1,000 ml of 100 mM PBS, pH 7.6

IgG would diffuse out if it could, but it is too large to enter the pores in the membrane; therefore, the amount of IgG inside the Cassette remains constant. The Tris buffer concentration drops below 0.01 M inside the Cassette as the Tris buffer diffuses out and the PBS buffer diffuses in.

Enlarged view of Slide-A-Lyzer Cassette interior

Old dialysate discarded and replaced with 1,000 ml of 100 mM PBS, pH 7.6

IgG inside the Cassette remains constant. The Tris buffer inside the Cassette drops to near undetectable levels. The buffer inside the Cassette is 100 mM PBS, pH 7.6.

3 ml of 1 mg/ml IgG in 0.1 M Tris buffer, pH 7.4 inside a Slide-A-Lyzer Dialysis Cassette (10K MWCO) placed in 1,000 ml of 100 mM PBS, pH 7.6.

Enlarged view of Slide-A-Lyzer Cassette interior

Old dialysate discarded and replaced with 1,000 ml of 100 mM PBS, pH 7.6

IgG would diffuse out if it could, but it is too large to enter the pores in the membrane; therefore, the amount of IgG inside the Cassette remains constant. The Tris buffer concentration drops below 0.01 M inside the Cassette as the Tris buffer diffuses out and the PBS buffer diffuses in.

Enlarged view of Slide-A-Lyzer Cassette interior

Old dialysate discarded and replaced with 1,000 ml of 100 mM PBS, pH 7.6

IgG inside the Cassette remains constant. The Tris buffer inside the Cassette drops to near undetectable levels. The buffer inside the Cassette is 100 mM PBS, pH 7.6.
Dialysis is the separation of small and large molecules in a solution by selective diffusion through a semipermeable membrane. Typically a sample containing a protein or nucleic acid will contain unwanted small molecular weight (MW) compounds such as a buffer salt (Tris, PBS, etc.), a reducing agent (dithiothreitol (DTT), β-mercaptoethanol (BME), etc.) or a preservative (sodium azide, thimerosol, etc.).

The sample is contained inside the dialysis membrane. A dialysate 200 to 300 times the volume of the sample is outside the dialysis membrane, which creates and maintains a concentration differential across the membrane. Once the liquid-to-liquid interface (sample on one side of the membrane and dialysate on the other) is initiated, all molecules will try to diffuse in either direction across the membrane to reach equilibrium. Dialysis (diffusion) will stop when equilibrium is achieved. Generally the rate of dialysis slows as equilibrium approaches, requiring the dialysate be changed after several hours to re-create the concentration differential that drives the dialysis process.

The membrane is the key to dialysis. The semipermeable membrane contains pores of a known size range that are large enough to let small MW compounds pass through, but restrict large MW compounds (e.g., proteins and nucleic acids). The ideal membrane is thin, has numerous pores of uniform diameter, and does not bind proteins and nucleic acids. What scientists have been using for decades is an extruded regenerated cellulose membrane that is close to an ideal membrane.

However, most scientists often assume too much chromatographic resolution associated with the membrane’s molecular weight cutoff (MWCO).

For more information, or to download product instructions, visit www.thermo.com/pierce
We determine the MWCO of our dialysis membrane by using the rotating batch dialysis cell (see diagram above). In the rotating cell, the membrane to be tested is held in place between two circular cavities of equal size. One side of the cell is partially filled with a solution containing a molecule of known MW. The other side is filled with an equal volume of buffer or saline. The solutions are mixed and kept in contact with the membrane by rotating the cell at a constant speed. The MW standard concentration in each half of the cell is measured after a fixed period of time and the percent retention is calculated. This type of system provides a more accurate MWCO determination than using ultrafiltration methods that measure hydraulic permeability or volumetric flux vs. pressure using saline or buffer alone.

Other important variables are sample and dialysate volume. The ideal scenario is to have a small sample volume and a large dialysate volume to maximize the concentration differential. The sample volume is important because subsequent applications have certain minimum volume requirements. However, after the minimum volume requirements are met, it is not advantageous to dialyze more sample than is needed. Depending on the surface area of a given sample, a small volume sample will dialyze much faster than a large volume sample. Not only is expending additional time wasteful, it can result in sample loss because the longer a sample is in contact with solid-phase surfaces, the more likely proteins or nucleic acids will nonspecifically bind or denature.

Reference
Frequently Asked Questions About Dialysis

1) How precise is the MWCO?

The MWCO is reproducible, but not very precise. When choosing which MWCO membrane to use, it is advisable to have both the high MW compounds that you want to retain, and the low MW compounds that you want to diffuse out as far removed from the membrane's MWCO as possible.

Our dialysis products are available with 2K, 3.5K, 7K, 10K and 20K MWCO membranes. The retention profile exhibited is clearly distinct and reproducible for each MWCO membrane when testing compounds of known MW. We do not sell products with regenerated cellulose membranes with MWCOs below 2K and above 16K because they cannot be manufactured to our high-quality standards at this time.

2) Is stirring necessary?

Stirring significantly decreases the dialysis time. All membranes possess an inner skin, which experts have described as “seaweed-like,” and an outer skin. There are no channels of a fixed diameter extending from the sample side through to the dialysate side. Instead, low MW compounds from the sample diffuse into the inner skin pores then through the membrane interior. These low MW compounds exit through a pore in the outer skin of the membrane, to a micro-environment called the Nernst layer. In this layer, which is approximately 200-300 molecules thick, low MW compounds are at a higher concentration in relation to the rest of the dialysate. Stirring, which efficiently breaks up the macro-environment outside the Nernst layer, quickly restores the concentration differential needed to drive the diffusion process.

3) Is temperature important?

Temperature is somewhat important because molecules move and diffuse faster at higher temperatures; however, maintaining the viability of your sample is the priority. So the typical range for dialysis is from ambient to cold-room temperatures.

4) When is my dialysis finished?

There is no easily measured dialysis endpoint. The goal is to reduce the concentration of low MW compounds to a level that will not interfere with subsequent steps in your experiment.

Standard practice has been as follows:
1) Dialyze for 2 hours at room temperature (RT),
2) Change the dialysate before dialyzing for another 2 hours at RT, and
3) Change the dialysate again and dialyze for 1 hour to overnight in the cold room.

Thermo Scientific Pierce High-Performance Dialysis Products make the dialysis process faster than ever. The basic principle of the Slide-A-Lyzer MINI Dialysis Unit is to deposit a 10 μl sample (essentially a monolayer) on a dialysis membrane in contact with a dialysate that is 100,000 times larger than the sample volume. Small MW compounds have an extremely short (< 1 mm) migratory distance to exit the membrane. Also, with a gigantic concentration differential, the dialysis rate is fast (see page 4 Figures 1 and 2).

5) Is membrane pretreatment necessary?

A short hydration is necessary for some MWCO membranes in the Slide-A-Lyzer Dialysis Cassette product line. Otherwise, the regenerated cellulose membranes are clean and require no pretreatment. A very small amount of either glycerine or sulfur may be present. These low MW compounds will diffuse out of the membrane and into the dialysate during the normal dialysis process. If necessary, these compounds may be dialyzed ahead of time but this is usually unnecessary.

6) When sample is injected into the Slide-A-Lyzer Dialysis Cassette, the membrane sometimes folds. What causes this?

Because the dialysis membrane is manufactured as a tube, the regenerated cellulose polymer has “memory” and wants to return to that shape even though the tube was cut into a flat membrane. Therefore, when a membrane is hydrated and the Cassette is filled, the membrane will stretch or pull differently with respect to the X-axis or Y-axis. Although this does have minor implications relative to surface area, these Slide-A-Lyzer Dialysis Cassettes will function just fine.
To order, call 800-874-3723 or 815-968-0747. Outside the United States, contact your local branch office or distributor.

Highlights:
• 100% leak-tested
  Patented design does not permit “wicking” that can occur in homemade devices
• Very affordable
• Excellent sample recoveries
  The Slide-A-Lyzer MINI Dialysis Unit generally recovers 9-10 μl after dialysis of a 10 μl sample
• Time of dialysis drastically reduced
  Converts 100 μl of pH 2.8 buffer to pH 9.4 dialyzing against 1 L bicarbonate buffer, pH 9.4 in less than 10 minutes

The Slide-A-Lyzer MINI Dialysis Unit is a small disposable cup made of polypropylene and regenerated cellulose. Sample is added and removed easily using a standard laboratory pipette. A float (sold separately) holds the Slide-A-Lyzer MINI Dialysis Unit upright, floating on the dialysate surface with the membrane in contact with the dialysate. Although the device’s patented design is very simple, the easy-to-use Slide-A-Lyzer MINI Dialysis Unit is an invaluable tool for applications, like equilibrium competitive dialysis, for which only 10-100 μl samples are available.

Dialysis Rate and Sample Recovery

The 3.5K Slide-A-Lyzer MINI Dialysis Unit was used for salt reduction analysis. Samples of 5-100 μl of 1 M NaCl were placed in the Slide-A-Lyzer MINI Dialysis Unit and dialyzed against 1 L of water for 10 minutes. To recover the smallest (5 μl and 10 μl) volumes from the Slide-A-Lyzer MINI Dialysis Unit, the device was tilted and gently tapped on the bottom edge to pool the sample. NaCl standards and samples were diluted in 50 ml ultrapure water and conductivity was measured (Cole-Parmer). The Slide-A-Lyzer MINI Dialysis Unit dialyzes efficiently (Figure 1). Dialysis rate of 100 μl of 5 M NaCl was also analyzed by conductivity (Figure 2). In a third experiment, the rate of pH exchange in the Slide-A-Lyzer MINI Dialysis Unit was determined and is also rapid. In less than 10 minutes, 100 μl of IgG Elution Buffer, pH 2.8 is converted to pH 9.4 by dialysis against 1 L of BupH™ Carbonate-Bicarbonate Buffer, pH 9.4 (data not shown).

See ordering information on pages 9-12.
Thermo Scientific Slide-A-Lyzer G2 Dialysis Cassettes
New design for enhanced performance and ease-of-use

Highlights:
• Pipette-accessible for easy sample loading and retrieval
• Self-floating chambers for buoyancy and vertical orientation during dialysis
• Designed to maintain the highest possible sample integrity and protection
• Fast and consistent dialysis with maximum sample recovery
• Rigorous quality testing for maximum consistency
• Ideal for removing low-molecular weight contaminants, performing buffer exchange and desalting

Join the thousands of researchers worldwide who save time and preserve their valuable samples by using Thermo Scientific Slide-A-Lyzer Dialysis Cassettes. The new generation of Slide-A-Lyzer Cassettes are flexible and easy-to-use. They are pipette-accessible, making it easy to add and remove your samples!

Dialysis is the most commonly used method for removing low-molecular weight solutes from macromolecules in solution or for buffer exchange. Dialysis separates sample components based on selective diffusion across a porous membrane. The membrane’s pore size determines the molecular-weight cutoff (MWCO), which is characterized by the molecular weight at which 90% of the solute is retained. The permeability of a solute is dependent upon the shape of the molecule, the degree of hydration and its charge. Each of these characteristics may be influenced by the nature of the solvent, the pH and the ionic strength.

The new Slide-A-Lyzer G2 Cassette offers maximum efficiency, convenience and sample protection in one package. Sample loading and removal are easily accomplished by using a serological pipette or hypodermic needle (optional) attached to a syringe. The built-in air chamber provides sample buoyancy and vertical orientation of the cassette during dialysis. The cassette membrane is composed of low-binding regenerated cellulose for maximum sample recovery while maintaining maximum sample purity. The cassettes are available in five precise membrane MWCOs (2K, 3.5K, 7K, 10K and 20K) for dialyzing sample volumes from 100 μl up to 70 ml. The cassettes are manufactured using clean room conditions to ensure cassettes are contaminant-free.

See ordering information on pages 9-13.
Thermo Scientific Slide-A-Lyzer Dialysis Cassettes
The original

**Highlights:**

- **> 95% sample recovery**
  Sample volume remains visible throughout dialysis
- **No knots or clamps to loosen and leak**
  Secure design prevents sample loss due to leaks
- **Rigid frame permits smooth sample withdrawal**
  Removing every last drop is easy – even for scientists who have never before performed dialysis
- **High surface area/sample volume ratio will dialyze twice as fast as dialysis via conventional tubing**
  Patented Cassette design spreads the sample over a large surface area and the double membrane promotes fast dialysis

The Slide-A-Lyzer Dialysis Cassette effectively and quickly dialyzes sample volumes from 100 μl to 30 ml. The Cassette’s patented design, which provides a maximum surface area/sample volume ratio, allows for excellent sample recoveries. Unlike standard flat tubing, the innovative Cassette does not require the use of knots or clips that can lead to leaking and sample loss.

### Quantitative Sample Recovery

Three sample volume batches of water (0.5 ml, 1.7 ml and 3.0 ml) were loaded and recovered per the respective manufacturer’s instructions in a Slide-A-Lyzer Dialysis Cassette and conventional dialysis tubing to determine the volumes of recovery. Water volume recovered was determined gravimetrically. The following table summarizes the results:

<table>
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<tr>
<th>Sample Volume Loaded</th>
<th>Thermo Scientific Slide-A-Lyzer Dialysis Cassette % Volume Recovery</th>
<th>Traditional Dialysis Tubing % Volume Recovery</th>
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<tr>
<td>3.0 ml</td>
<td>99.47</td>
<td>92.32</td>
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<tr>
<td>1.7 ml</td>
<td>99.30</td>
<td>93.12</td>
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<tr>
<td>0.5 ml</td>
<td>98.76</td>
<td>87.51</td>
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*See ordering information on pages 9-13.*
Thermo Scientific SnakeSkin Dialysis Tubing
Avoid the hassles of large-sample dialysis using flat tubing

Tubing Specifications

Membrane Type:
Regenerated cellulose

Glycerol Content: Varies with MWCO membrane

Sulfur Content: 0.1%-0.15%

Heavy Metals Content: Trace

Tubing Nominal
Dry Thickness*
3.5K MWCO 1.0 mils
7K MWCO 1.2 mils
10K MWCO 0.9 mils

*1 mil = 25 microns

Traditional flat dialysis tubing is difficult to open and often requires a presoak in water or buffer before it can be used. Handling the tubing after the presoak step can be messy and awkward. Thermo Scientific SnakeSkin Dialysis Tubing was developed to simplify large-sample dialysis. SnakeSkin Dialysis Tubing is open, regenerated cellulose dialysis tubing that is pleated (compressed) into a hollow stick. It is supplied in eight-inch sticks containing 35 feet of 22 mm internal diameter (I.D.) tubing, equivalent to 10.5 meters of 34 mm dry flat width tubing. SnakeSkin Dialysis Tubing can be used for 15-100 ml samples. The hydrated tubing will hold ~3.7 ml of sample per centimeter of length.

The pleated format of SnakeSkin Dialysis Tubing makes it easy to open and ready to use, streamlining dialysis preparation. To use it, a researcher simply pulls out the required length of tubing, cuts it off and applies a closure. The sample is then added through the other end of the dry tubing and the second closure is applied.

We recommend closure using SnakeSkin Dialysis Tubing Clips (sold separately). To use the clips, cut the desired length of tubing, fold one end over twice and apply a clip. Add the sample through the second end of the tubing, fold over twice and attach the second clip.

As an alternative to these clips, SnakeSkin Dialysis Tubing can also be closed with knots. Dip two to three inches of one end of the tubing into water or buffer and tie a knot in the wet membrane. (Dipping is required to assure a good seal at the knot point.) Add the sample to the open, dry end and tie a knot at this end. Because the sample quickly hydrates the membrane, there is no need to pre-wet the second end of the tubing.

The pleating process does not change the tubing’s MWCO. Also, any low MW contaminants present are removed during the dialysis process. Because SnakeSkin Dialysis Tubing is made from the same type of regenerated cellulose as flat tubing, its dialysis performance matches that of conventional tubing.

SnakeSkin Dialysis Tubing is available in three MWCOs: 3.5K, 7K and 10K. The product is stored in its original packaging at room temperature, although refrigerated storage may also be used. Properly stored membrane is stable for at least one year.

Ordering Information

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<td>68035</td>
<td>SnakeSkin Dialysis Tubing</td>
<td>3.5K</td>
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<tr>
<td>68700</td>
<td>SnakeSkin Dialysis Tubing</td>
<td>7K</td>
<td>22 mm dry I.D. x 35 feet*</td>
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<tr>
<td>68100</td>
<td>SnakeSkin Dialysis Tubing</td>
<td>10K</td>
<td>22 mm dry I.D. x 35 feet*</td>
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*Equivalent to 10.5 meters of 34 mm dry flat width tubing.

Product Accessories

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<td>68011</td>
<td>SnakeSkin Dialysis Tubing Clips</td>
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Specifications

Membrane Composition: Regenerated cellulose synthesized by the Viscose method

Hydration Required Before Use: 2 minutes

Glycerol Content: None

Sulfur Content: 0.169%

Heavy Metals Content: Trace

Characterization of membrane pore size. Vitamin B₁₂, bacitracin, tyrosine kinase peptide 1, biotin-TPKs substrate, protein kinase Ce (PKCe) peptide substrate and insulin chain A model systems (0.5–1 mg/ml) in either saline or 0.2 M carbonate bicarbonate buffer pH 9.4 were dialyzed overnight (17 hours) at 4°C. The amount of retentate was estimated using either the Pierce BCA Protein Assay or absorption at 360 nm (for vitamin B₁₂).

Desalting rate of the membrane for salts. Sodium chloride (1 M) in water was dialyzed at 4°C and the rate of removal of NaCl was determined by measuring the conductivity of the retentate at different time intervals.

Ordering Information

New Advanced Design Slide-A-Lyzer G2 Dialysis Cassettes

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New Advanced Design Slide-A-Lyzer MINI Dialysis Units

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<td>Sufficient caps are included</td>
<td>10-100 μl</td>
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<tr>
<td>69553</td>
<td>Slide-A-Lyzer MINI Dialysis Unit</td>
<td>Sufficient caps are included</td>
<td>10-100 μl</td>
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Slide-A-Lyzer Dialysis Cassettes

<table>
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<td>0.5-3 ml</td>
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Product Accessories

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<tbody>
<tr>
<td>66430</td>
<td>Slide-A-Lyzer Buoy</td>
<td>Holds one 0.1-0.5 ml or 0.5-3 ml cassette.</td>
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<tr>
<td>66431</td>
<td>Slide-A-Lyzer Carousel Buoy</td>
<td>Holds ten 0.1-0.5 ml or 0.5-3 ml cassettes.</td>
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<tr>
<td>66432</td>
<td>Slide-A-Lyzer Buoy</td>
<td>Holds one 3-12 ml cassette.</td>
</tr>
<tr>
<td>66494</td>
<td>Slide-A-Lyzer Syringe (1 ml)*</td>
<td></td>
</tr>
</tbody>
</table>

* Can also be used with the new Slide-A-Lyzer G2 Dialysis Cassettes.

View or request our FREE Cell Lysis Technical Handbook today!

The 49-page, full-color handbook provides technical and product information related to protein extraction, cell fractionation, DNA extraction, detergents and protein refolding.

Log on to www.thermo.com/pierce or call 800-874-3723 or 815-968-0747 to request your free copy today! Outside the United States, contact your local branch office or distributor.
**Specifications**

Membrane Composition:
Regenerated cellulose synthesized by the Viscose method

Hydration Required Before Use: 30 seconds

Glycerol Content: Trace

Sulfur Content: 0.1%–0.15%

Heavy Metals Content: Trace

---

Sample retention by the 3.5K MWCO Thermo Scientific Slide-A-Lyzer Dialysis Cassette membrane. Known MW standards were dissolved at a concentration of 1 mg/ml in either 0.15 M sodium chloride or 0.2 M carbonate-bicarbonate buffer, pH 9.4 (Product # 28382). Rotating cells were assembled with the nominal 3.5K MWCO membranes. One half of the cell was filled with MW standard solution and the other half was filled with an equal volume of the plain diluent. Cells were rotated overnight at 100 rpm.

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### Ordering Information

#### New Advanced Design Slide-A-Lyzer G2 Dialysis Cassette

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Capacity</th>
<th>Pkg. Size</th>
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<tbody>
<tr>
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<td>Slide-A-Lyzer G2 Dialysis Cassette</td>
<td>0.1-0.5 ml</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>87723</td>
<td>Slide-A-Lyzer G2 Dialysis Cassette</td>
<td>0.5-3 ml</td>
<td>10/pkg.</td>
</tr>
</tbody>
</table>

#### Slide-A-Lyzer MINI Dialysis Units

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Capacity</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>69550</td>
<td>Slide-A-Lyzer MINI Dialysis Unit</td>
<td>0.5-100 μl</td>
<td>50/pkg.</td>
</tr>
<tr>
<td>69552</td>
<td>Slide-A-Lyzer MINI Dialysis Unit</td>
<td>10-100 μl</td>
<td>250/pkg.</td>
</tr>
</tbody>
</table>

#### Slide-A-Lyzer Dialysis Cassettes

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Capacity</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>66333</td>
<td>Slide-A-Lyzer Dialysis Cassette</td>
<td>0.1-0.5 ml</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>66335</td>
<td>Slide-A-Lyzer Dialysis Cassette Kit</td>
<td>0.1-0.5 ml</td>
<td>Kit</td>
</tr>
<tr>
<td>66330</td>
<td>Slide-A-Lyzer Dialysis Cassette</td>
<td>0.5-3 ml</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>66332</td>
<td>Slide-A-Lyzer Dialysis Cassette Kit</td>
<td>0.5-3 ml</td>
<td>Kit</td>
</tr>
<tr>
<td>66110</td>
<td>Slide-A-Lyzer Dialysis Cassette</td>
<td>3-12 ml</td>
<td>8/pkg.</td>
</tr>
<tr>
<td>66107</td>
<td>Slide-A-Lyzer Dialysis Cassette Kit</td>
<td>3-12 ml</td>
<td>Kit</td>
</tr>
</tbody>
</table>

#### SnakeSkin Dialysis Tubing

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>68035</td>
<td>SnakeSkin Dialysis Tubing</td>
<td>22 mm dry I.D. x 35 ft</td>
</tr>
</tbody>
</table>

#### Product Accessories

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>66494</td>
<td>Slide-A-Lyzer Syringe (1 ml capacity)*</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>68011</td>
<td>SnakeSkin Dialysis Tubing Clips</td>
<td>6/pkg.</td>
</tr>
</tbody>
</table>

* Can also be used with the new Slide-A-Lyzer G2 Dialysis Cassettes.
Specifications

Membrane Composition: Regenerated cellulose synthesized by the Viscose method

Hydration Required Before Use: 30 seconds for low-volume samples

Glycerol Content: 13%

Sulfur Content: 0.1%–0.15%

Heavy Metals Content: Trace

Sample retention by the 7K MWCO Thermo Scientific Slide-A-Lyzer Dialysis Cassette membrane. Known MW standards were dissolved at a concentration of 1 mg/ml in either 0.15 M sodium chloride or 0.2 M carbonate-bicarbonate buffer, pH 9.4 (Product # 28382). Rotating cells were assembled with the nominal 7K MWCO membranes. One half of the cell was filled with MW standard solution and the other half was filled with an equal volume of the plain diluent. Cells were rotated overnight at 100 rpm.

Ordering Information

New Advanced Design Slide-A-Lyzer G2 Dialysis Cassettes

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Capacity</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>87727</td>
<td>Slide-A-Lyzer G2 Dialysis Cassette</td>
<td>0.1-0.5 ml</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>87728</td>
<td>Slide-A-Lyzer G2 Dialysis Cassette</td>
<td>0.5-3 ml</td>
<td>10/pkg.</td>
</tr>
</tbody>
</table>

Slide-A-Lyzer MINI Dialysis Units

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Capacity</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>69560</td>
<td>Slide-A-Lyzer MINI Dialysis Unit</td>
<td>10-100 μl</td>
<td>50/pkg.</td>
</tr>
<tr>
<td>69562</td>
<td>Slide-A-Lyzer MINI Dialysis Unit</td>
<td>10-100 μl</td>
<td>250/pkg.</td>
</tr>
</tbody>
</table>

Slide-A-Lyzer Dialysis Cassettes

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Capacity</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>66373</td>
<td>Slide-A-Lyzer Dialysis Cassette</td>
<td>0.1-0.5 ml</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>66375</td>
<td>Slide-A-Lyzer Dialysis Cassette Kit</td>
<td>0.1-0.5 ml</td>
<td>Kit</td>
</tr>
<tr>
<td>66370</td>
<td>Slide-A-Lyzer Dialysis Cassette</td>
<td>0.5-3 ml</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>66372</td>
<td>Slide-A-Lyzer Dialysis Cassette Kit</td>
<td>0.5-3 ml</td>
<td>Kit</td>
</tr>
<tr>
<td>66710</td>
<td>Slide-A-Lyzer Dialysis Cassette</td>
<td>3-12 ml</td>
<td>8/pkg.</td>
</tr>
<tr>
<td>66707</td>
<td>Slide-A-Lyzer Dialysis Cassette Kit</td>
<td>3-12 ml</td>
<td>Kit</td>
</tr>
</tbody>
</table>

SnakeSkin Dialysis Tubing

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>68700</td>
<td>SnakeSkin Dialysis Tubing</td>
<td>22 mm dry</td>
</tr>
</tbody>
</table>

Product Accessories

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>66494</td>
<td>Slide-A-Lyzer Syringe (1 ml capacity)*</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>68011</td>
<td>SnakeSkin Dialysis Tubing Clips</td>
<td>6/pkg.</td>
</tr>
</tbody>
</table>

* Can also be used with the new Slide-A-Lyzer G2 Dialysis Cassettes.

To order, call 800-874-3723 or 815-968-0747. Outside the United States, contact your local branch office or distributor.
Specifications
Membrane Composition: Regenerated cellulose synthesized by the Viscose method
Hydration Required Before Use: 30 seconds
Glycerol Content: 21%
Sulfur Content: 0.05%
Heavy Metals Content: Trace

Sample retention by the 10K MWCO Thermo Scientific Slide-A-Lyzer Dialysis Cassette membrane. Known MW standards were dissolved at a concentration of 1 mg/ml in either 0.15 M sodium chloride or 0.2 M carbonate-bicarbonate buffer, pH 9.4 (Product # 28382). Rotating cells were assembled with the nominal 10K MWCO membranes. One half of the cell was filled with MW standard solution and the other half was filled with an equal volume of the plain diluent. Cells were rotated overnight at 100 rpm.

Ordering Information
New Advanced Design Slide-A-Lyzer G2 Dialysis Cassette
Product # Description Capacity Pkg. Size
87729 Slide-A-Lyzer G2 Dialysis Cassette 0.1-0.5 ml 10/pkg.
87730 Slide-A-Lyzer G2 Dialysis Cassette 0.5-3 ml 10/pkg.

Slide-A-Lyzer MINI Dialysis Units
Product # Description Capacity Pkg. Size
69574 Slide-A-Lyzer MINI Dialysis Unit Plus Microtubes Sufficient caps are included. 10-100 μl 10/pkg.
69570 Slide-A-Lyzer MINI Dialysis Unit Sufficient caps are included. 10-100 μl 50/pkg.
69572 Slide-A-Lyzer MINI Dialysis Unit Sufficient caps are included. 10-100 μl 250/pkg.
69576 Slide-A-Lyzer MINI Dialysis Unit Plus Float Sufficient caps are included. 10-100 μl Kit/10 units

Slide-A-Lyzer Dialysis Cassettes
Product # Description Capacity Pkg. Size
66383 Slide-A-Lyzer Dialysis Cassette 0.1-0.5 ml 10/pkg.
66384 Slide-A-Lyzer Dialysis Cassette 0.1-0.5 ml 5 x 10/pkg.
66385 Slide-A-Lyzer Dialysis Cassette Kit Contains 10 cassettes, 10 buoys and 10 syringes. 0.1-0.5 ml Kit
66380 Slide-A-Lyzer Dialysis Cassette 0.5-3 ml 10/pkg.
66381 Slide-A-Lyzer Dialysis Cassette 0.5-3 ml 5 x 10/pkg.
66382 Slide-A-Lyzer Dialysis Cassette Kit Contains 10 cassettes, 10 buoys and 10 syringes. 0.5-3 ml Kit
66810 Slide-A-Lyzer Dialysis Cassette 3-12 ml 8/pkg.
66811 Slide-A-Lyzer Dialysis Cassette 3-12 ml 5 x 10/pkg.
66807 Slide-A-Lyzer Dialysis Cassette Kit Contains 8 cassettes, 8 buoys and 10 syringes. 3-12 ml Kit

Irradiated 10K MWCO Membrane
Product # Description Capacity Pkg. Size
66454 Slide-A-Lyzer Dialysis Cassette 0.1-0.5 ml 10/pkg.
66455 Slide-A-Lyzer Dialysis Cassette 0.5-3 ml 10/pkg.
66453 Slide-A-Lyzer Dialysis Cassette 3-12 ml 8/pkg.
66456 Slide-A-Lyzer Dialysis Cassette Kit Contains 8 cassettes, 8 buoys and 10 syringes. 0.1-0.5 ml Kit

SnakeSkin Dialysis Tubing
Product # Description Capacity Pkg. Size
68100 SnakeSkin Dialysis Tubing Equivalent to 10.5 meters of 34 mm dry width. 12-30 ml 22 mm dry I.D. x 35 ft

Product Accessories
Product # Description Pkg. Size
66430 Slide-A-Lyzer Buoy Fits one 0.1-0.5 ml or 0.5-3 ml cassette. 10/pkg.
66431 Slide-A-Lyzer Carousel Buoy Fits ten 0.1-0.5 ml or 0.5-3 ml cassettes. 1/pkg.
66432 Slide-A-Lyzer Buoy Fits one 3-12 ml cassette. 8/pkg.
66494 Slide-A-Lyzer Syringe (1 ml capacity)* Each syringe comes with 18-gauge 1-inch beveled needles. 10/pkg.
68011 SnakeSkin Dialysis Tubing Clips 6/pkg.

* Can also be used with the new Slide-A-Lyzer G2 Cassettes.
Specifications

Membrane Composition:
Regenerated cellulose synthesized by the Viscose method

Hydration Required
Before Use: 2 minutes

Glycerol Content: None
Sulfur Content: 0.04%
Heavy Metals Content: Trace

Thermo Scientific Membrane Products – 20K MWCO

Model Systems

% Protein Retention

Insulin B Chain (3.5K MW)
Cytochrome C (12.5K MW)
Lysozyme (14K MW)
Myoglobin (17K MW)

Characterization of membrane pore size. Insulin B chain, cytochrome C, lysozyme and myoglobin were dialyzed overnight (17 hours) at 4°C in PBS pH 7.4. The amount of retentate was estimated using the Pierce BCA Protein Assay.

View or request our FREE Mass Spec Sample Prep Technical Handbook today!

This 48-page handbook breaks the Mass Spec process into five logical steps and then provides protocols and technical and product information to help maximize results. The handbook provides background, helpful hints and troubleshooting advice for cell lysis, 2D sample prep, detection, mass spec sample prep and downstream applications. Exciting new Thermo Scientific products include Zeba Micro Desalt Spin Columns, ProteoSeek Albumin and IgG Removal Kits, Imperial™ Protein Stain, The In-Solution Tryptic Digest and Guanidination Kit, and Deuterated (Heavy) Crosslinkers.

Log on to www.thermo.com/pierce or call 800-874-3723 or 815-968-0747 to request your free copy today! Outside the United States, contact your local branch office or distributor.

Ordering Information

New Advanced Design
Slide-A-Lyzer G2 Dialysis Cassette

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Capacity</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>87734</td>
<td>Slide-A-Lyzer G2 Dialysis Cassette</td>
<td>0.1-0.5 ml</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>87735</td>
<td>Slide-A-Lyzer G2 Dialysis Cassette</td>
<td>0.5-3 ml</td>
<td>10/pkg.</td>
</tr>
</tbody>
</table>

Slide-A-Lyzer Dialysis Cassettes

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Capacity</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>66005</td>
<td>Slide-A-Lyzer Dialysis Cassette</td>
<td>0.1-0.5 ml</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>66003</td>
<td>Slide-A-Lyzer Dialysis Cassette</td>
<td>0.5-3 ml</td>
<td>10/pkg.</td>
</tr>
</tbody>
</table>

Product Accessories

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Description</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>69588</td>
<td>Slide-A-Lyzer MINI Dialysis Unit Float</td>
<td>Holds one 0.1-0.5 ml or 0.5-3 ml cassette.</td>
<td>4/pkg.</td>
</tr>
<tr>
<td>66430</td>
<td>Slide-A-Lyzer Buoy</td>
<td>Holds one 0.1-0.5 ml or 0.5-3 ml cassette.</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>66431</td>
<td>Slide-A-Lyzer Carousel Buoy</td>
<td>Holds ten 0.1-0.5 ml or 0.5-3 ml cassettes.</td>
<td>1/pkg.</td>
</tr>
<tr>
<td>66432</td>
<td>Slide-A-Lyzer Buoy</td>
<td>Holds one 3-12 ml cassette.</td>
<td>8/pkg.</td>
</tr>
<tr>
<td>66494</td>
<td>Slide-A-Lyzer Syringe (1 ml capacity)*</td>
<td>Holds one 3-12 ml cassette.</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>66490</td>
<td>Slide-A-Lyzer Syringe (5 ml capacity)*</td>
<td>Holds one 3-12 ml cassette.</td>
<td>8/pkg.</td>
</tr>
<tr>
<td>66493</td>
<td>Slide-A-Lyzer Syringe (20 ml capacity)*</td>
<td>Each syringe comes with 18-gauge 1-inch beveled needles.</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>68011</td>
<td>SnakeSkin Dialysis Tubing Clips</td>
<td>Each syringe comes with 18-gauge 1-inch beveled needles.</td>
<td>6/pkg.</td>
</tr>
</tbody>
</table>

* Can also be used with the new Slide-A-Lyzer G2 Dialysis Cassettes.
Determining the extent to which a molecule binds to plasma proteins is a critical phase of drug development because it influences compound dosing, efficacy, clearance rate and potential for drug interactions. This determination is enabled by equilibrium dialysis, an accepted standard method for reliable estimation of the nonbound drug fraction in plasma. Although it is the preferred method, equilibrium dialysis is labor-intensive, time-consuming, cost-prohibitive and difficult to automate. The RED Device for rapid equilibrium dialysis was developed in close association with the pharmaceutical industry to specifically address these issues, accelerating lead optimization and reducing attrition rate.

The RED System consists of disposable tube inserts and a 96-well Teflon® Base Plate. The unique design of the base plate provides compatibility with automated liquid handling systems while the large dialysis surface area of the tube inserts accelerates equilibrium.

The RED Device has been extensively validated for plasma-binding assays producing results consistent with those reported in the literature (Table 1). Using the RED Device to measure Warfarin binding to plasma (human and rat) proteins at two concentrations of 1 and 10 μM, the RED Device produced results with minimal intra-experimental variability (Figure 1).

### The RED Device Enables:
- Determination of free vs. bound drug to plasma proteins
- Pharmacokinetics studies
- Formulation of drug dosage for *in vivo* studies
- Drug-to-drug interaction studies
- Selection criteria during drug lead optimization

### Highlights:
- **Ease of use**
  Disposable tubes require no presoaking, assembly or specialized equipment
- **Short incubation time**
  Equilibrium can be reached in as few as three hours as a result of the high membrane surface-to-volume ratio
- **96-well format**
  Suitable for automated liquid handlers
- **Flexible**
  Can be used for the desired number of assays (one to 48 assays/plate) without wasting the entire plate
- **Robust**
  Compartmentalized design eliminates potential for crosstalk or leakage
- **Reproducible and accurate**
  Validated for plasma-binding assays, producing results consistent with those reported in the literature (Table 1)
- **Versatile**
  The high-grade Teflon Base Plate is chemically inert, eliminating nonspecific binding and risk of contamination
- **Validated**
  Each lot is functionally tested in a protein-binding assay for guaranteed performance
- **Convenient**
  The RED Device membrane has a MWCO of 8K; other MWCO membranes are available upon request

### Table 1. Comparison of results obtained using the RED Device with values reported in the literature.

<table>
<thead>
<tr>
<th>Compound</th>
<th>Literature Value</th>
<th>RED Device</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ranitidine</td>
<td>10-19</td>
<td>17</td>
</tr>
<tr>
<td>Propranolol</td>
<td>87-96</td>
<td>84</td>
</tr>
<tr>
<td>Warfarin</td>
<td>99</td>
<td>99</td>
</tr>
<tr>
<td>Naproxen</td>
<td>99</td>
<td>99</td>
</tr>
</tbody>
</table>

Figure 1. The RED Device binds plasma proteins. More than 99% of Warfarin was consistently bound to plasma protein showing minimal intra-experimental variability. Three replicate RED Device inserts were set up for each tested time point. Warfarin solutions at 1 μM or 10 μM were made in the plasma of choice and added to the insert sample chamber. PBS was added to the buffer chamber. At each time point (1, 2, 3 and 4 hours), 50 μl was removed from the plasma and the buffer chambers and transferred to separate wells of a deep well plate. After all the time points were collected, 50 μl of blank plasma was added to every buffer sample and 50 μl buffer was added to every plasma sample. After precipitation buffer was added, vortexed and centrifuged, the supernatants were analyzed by LC/MS/MS (API4000). A standard curve of the drug of interest was prepared along with the samples. The concentration of each sample was determined from the standard curve.

Table 2. A comparison of critical attributes for equilibrium devices.††

<table>
<thead>
<tr>
<th>Device (Source)</th>
<th>Hours to reach Equilibrium</th>
<th>Leakage</th>
<th>Disposable</th>
<th>Labor Intensity</th>
<th>Automation Accessible</th>
<th>Volume Shift</th>
</tr>
</thead>
<tbody>
<tr>
<td>RED (Rapid Equilibrium Dialysis) Device (Thermo Scientific)</td>
<td>4</td>
<td>None</td>
<td>Yes</td>
<td>•</td>
<td>Yes</td>
<td>None</td>
</tr>
<tr>
<td>Multi-Equilibrium Dialyzer (Harvard Apparatus)</td>
<td>3-4</td>
<td>Minimum</td>
<td>No</td>
<td>• • •</td>
<td>No</td>
<td>Minimum</td>
</tr>
<tr>
<td>96-well Equilibrium DIALYZER (Harvard Apparatus)</td>
<td>16</td>
<td>20%</td>
<td>Yes</td>
<td>• • •</td>
<td>Possible</td>
<td>Yes</td>
</tr>
<tr>
<td>96-well Micro Equilibrium Dialysis Block (HTDialysis, LLC)</td>
<td>6</td>
<td>Some</td>
<td>No</td>
<td>• • •</td>
<td>Possible</td>
<td>Yes</td>
</tr>
<tr>
<td>24-Multwell Dialysis (BD Biosciences)</td>
<td>24</td>
<td>Not measured</td>
<td>Yes</td>
<td>• •</td>
<td>Possible</td>
<td>Not measured</td>
</tr>
</tbody>
</table>

†† Li, S., Xiong, B., Huang, T., Li, L., Donovan, J., Lee, F., Yu, S., Miwa, G., and Yang, H. Validation of a novel rapid equilibrium dialysis (RED) device for high throughput plasma protein binding determination. 1. DMPK/Drug Safety & Disposition; 2. Linden Bioscience, 35A Cabot Road, Woburn, MA 01801, USA; and 3. Process Technology, Millennium Pharmaceuticals, Inc., 40 Landsdowne Street, Cambridge, MA 02139 USA.

1. Place RED Device Inserts into the PTFE Plate.
2. Place 200 μl of sample (mixture of compound with plasma at appropriate concentrations) in the plasma chamber indicated by the red ring.
3. Add appropriate amount of buffer to the buffer chamber. Cover plate and incubate. Equilibrium is often reached in four hours or less.
4. After dialysis, remove equal volumes from both chambers. Proceed to sample preparation before LC/MS/MS analysis.

Figure 2. Schematic protocol for the RED Device.

Ordering Information

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>89809</td>
<td>RED Device Inserts</td>
<td>50/pack</td>
</tr>
<tr>
<td>89810</td>
<td>RED Device Inserts</td>
<td>250/case</td>
</tr>
<tr>
<td>89811</td>
<td>PTFE Base Plate</td>
<td>1 plate</td>
</tr>
<tr>
<td>89812</td>
<td>RED Device Insert Removal Tool</td>
<td>1 remover</td>
</tr>
</tbody>
</table>

† The RED Device is manufactured by Linden Bioscience. Patent Pending on RED Device by Linden Bioscience.
A critical phase of drug development is determining the extent to which a drug is distributed between plasma and specific tissues, which determines compound dosing, efficacy, clearance rate and the potential for drug interactions or tissue damage. The Thermo Scientific Competition Rapid Equilibrium Dialysis (Competition RED) Device is an expansion of our popular rapid equilibrium dialysis (RED) product line and was developed in association with pharmaceutical laboratories to better model in vivo drug-tissue interactions.

**Highlights:**

- **Easy to use** – disposable inserts are supplied ready to use (i.e., no presoaking, assembly or specialized equipment necessary)
- **Short incubation time** – equilibrium can be reached as fast as two to six hours
- **Flexible format** – variable well sizes enable small-molecule partitioning studies of two to 16 tissue or protein samples
- **Versatile** – base plate is composed of chemically inert high-grade PTFE, eliminating nonspecific binding and contamination risks
- **Validated** – each lot is functionally tested in a protein-binding assay for guaranteed performance

Because of time, high cost and regulation of animal dosing and testing, in vitro models are highly desired for pre-screening compounds. A wide variety of cell, membrane and tissue section or distribution and equilibrium dialysis-based pre-screening methods have been developed and implemented to varying degrees of success. Currently, tissue-plasma drug-binding studies using equilibrium dialysis examine the binding affinity of a small molecule between plasma and one tissue homogenate at a time. In an individual tissue-plasma binding study, a drug may result in significant binding but the binding profile will be different in a complex system.

Within the body, a drug distributes and reaches equilibrium based on competitive interactions with all encountered fluids and tissues. The Competition RED Device is specifically designed to mimic, in the best possible way, these multiple interactions in an in vitro assay. Although, competition RED screening does not replace animal testing, performing a pre-screen accelerates and improves drug candidate selection and minimizes cost by potentially identifying drugs with overly strong or weak binding to specific tissues. The Competition RED System consists of disposable dialysis tube inserts and a reusable PTFE base plate, which minimizes waste while providing experimental design flexibility. The unique base plate design allows placement of two to 16 dialysis chambers into a common well enabling researchers to perform several experiments simultaneously. The Competition RED System has a standard 96-well plate footprint with 9 x 9 mm well spacing. Additionally, the small volume and large dialysis surface area of the tube inserts allows rapid dialysis, achieving equilibrium in two to four hours with high levels of reproducibility and accuracy. The device inserts have a molecular-weight cutoff (MWCO) of 12,000.

**Applications:**

- Hit-to-lead selection of new chemical entities for preclinical studies
- Preliminary drug candidate screening in ADME-Tox studies – in vitro screening of drug partitioning between plasma and multiple tissues before in vivo studies
- Determining formulation of drug dosage for in vivo studies
- Competitive binding and dissociation constant determination for small molecules versus multiple targets

**Ordering Information**

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
<tr>
<td>90087</td>
<td>Competition RED Inserts</td>
<td>10/pkg.</td>
</tr>
<tr>
<td>90088</td>
<td>Competition RED Inserts</td>
<td>50/pkg.</td>
</tr>
<tr>
<td>90085</td>
<td>Competition RED Base Plate</td>
<td>1 unit</td>
</tr>
</tbody>
</table>

For more information, or to download product instructions, visit [www.thermo.com/pierce](http://www.thermo.com/pierce)
Thermo Scientific Slide-A-Lyzer Concentrating Solution

Thermo Scientific Slide-A-Lyzer Concentrating Solution is a proprietary, hygroscopic, high MW compound that pulls water through dialysis membrane quickly. Other concentrating solutions concentrate and contaminate samples with a compound of similar MW that is difficult to remove by dialysis or other means. These contaminants absorb strongly at 280 nm, distorting protein measurements using the tyrosine absorption method. The Slide-A-Lyzer Concentrating Solution special formulation is free of low MW compounds that could cross the membrane to contaminate the sample.

Many samples will take on water or buffer during the dialysis process. To return the sample to its original concentration, or to concentrate it even further, the Slide-A-Lyzer Concentrating Solution is ideal. To concentrate the sample, the Slide-A-Lyzer Dialysis Cassette containing the sample is placed in a small plastic bag containing the concentrating solution. By diffusion, water and other small molecules are drawn out of the cassette, into the bag. The large molecular size of the concentrating solution prevents it from crossing the membrane and entering the cassette. Therefore, a one-way flow of water and other small molecules out of the Cassette results in concentration of the sample.

The Slide-A-Lyzer Concentrating Solution quickly reduces a starting volume of 3 ml of sample inside the Slide-A-Lyzer Dialysis Cassette to 0.5 ml in about 50 minutes. This is comparable to other concentration methods such as centrifuge-driven membrane devices.

Highlights:
- Dialysis and concentration occur in one device
  Avoids protein loss by using a single device
- Faster concentration
  A starting volume of 3 ml is reduced to 0.5 ml in about 75-80 minutes
- Easy to use
  Just pour the Slide-A-Lyzer Concentrating Solution into the small plastic bag provided and drop in the Slide-A-Lyzer Dialysis Cassette containing the sample
- Improved formulation and protocols
  Improved product makes concentration easier with rocking-platform protocols
- The process can be monitored
  Because both the concentrating solution and the bag are clear, the sample concentration can be easily monitored, something that is not possible with closed-system centrifuge-type devices

Ordering Information

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
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<tbody>
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<td>Slide-A-Lyzer Concentrating Solution For use with 0.5-3 ml cassettes.</td>
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<tr>
<td>66529</td>
<td>Slide-A-Lyzer Concentrating Solution For use with 3-30 ml cassettes.</td>
<td>500 ml</td>
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<tr>
<td>66530</td>
<td>Slide-A-Lyzer Concentrating Solution For use with Slide-A-Lyzer MINI Dialysis Units</td>
<td>25 ml</td>
</tr>
</tbody>
</table>

Concentrating with the Thermo Scientific Slide-A-Lyzer MINI Dialysis Unit

Slide-A-Lyzer Concentrating Solution works on even very small samples using the Slide-A-Lyzer MINI Dialysis Unit. Samples from 10 to 100 μl are placed in the Slide-A-Lyzer MINI Dialysis Unit and then placed in a microcentrifuge tube that contains Slide-A-Lyzer Concentrating Solution at a minimum ratio of 3:1 (Concentrating Solution to sample).
Detergents or surfactants are important for solubilizing, stabilizing and disaggregating proteins; however, detergents interfere with many downstream analysis methods. Therefore, it is often crucial to remove non-bound detergents before using proteins samples for ELISA, isoelectric focusing or mass spectrometry (MS). Unfortunately, typical sample clean-up methods, such as dialysis and size-exclusion chromatography, are often ineffective at removing detergents. We developed an efficient and rapid spin-column method (Figure 1) for removing detergents from protein and peptide solutions. The Thermo Scientific Pierce Detergent Removal Resin efficiently removes high concentrations of detergents from 0.01-1 ml samples with minimal sample loss.

Results and Discussion
We processed protein samples containing a wide range of detergents with the Pierce Detergent Removal Resin. Detergents at concentrations from 1 to 5% were effectively removed with generally > 90% protein recovery (Table 1).

Table 1. Detergents are effectively removed with high protein recovery.*

<table>
<thead>
<tr>
<th>Detergent and Removable Concentration (%)</th>
<th>Detergent Removal (%)</th>
<th>BSA Recovery (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>SDS (2.5)</td>
<td>99</td>
<td>95</td>
</tr>
<tr>
<td>Sodium deoxycholate (5)</td>
<td>99</td>
<td>100</td>
</tr>
<tr>
<td>CHAPS (3)</td>
<td>99</td>
<td>90</td>
</tr>
<tr>
<td>Octyl glucoside (5)</td>
<td>99</td>
<td>90</td>
</tr>
<tr>
<td>Octyl thioglucoside (5)</td>
<td>99</td>
<td>95</td>
</tr>
<tr>
<td>Lauryl maltoside (1)</td>
<td>98</td>
<td>99</td>
</tr>
<tr>
<td>Triton X-100 (2)</td>
<td>99</td>
<td>87</td>
</tr>
<tr>
<td>Triton X-114 (2)</td>
<td>95</td>
<td>100</td>
</tr>
<tr>
<td>NP-40 (1)</td>
<td>95</td>
<td>91</td>
</tr>
<tr>
<td>Brij-35 (1)</td>
<td>99</td>
<td>97</td>
</tr>
</tbody>
</table>

* Samples (0.1 ml containing 100 µg BSA and detergent) were processed through 0.5 ml of Pierce Detergent Removal Resin and the percent detergent removed was determined. Similar results were produced for insulin (6.7 kDa), α-lactalbumin (14.2 kDa) and carbonic anhydrase (29 kDa) (data not shown).

Figure 1. Protocol summary for Thermo Scientific Pierce Detergent Removal Spin Columns (0.5 ml).
Detergent removal from peptide samples is a challenge, especially for MS analysis in which even low detergent concentrations contaminate instruments and interfere with column binding, elution and ionization. We used the Pierce Detergent Removal Resin to remove a variety of detergents from BSA and HeLa cell lysate tryptic digests followed by LC-MS/MS and MALDI-MS analysis (Figures 2 and 3).

Figure 2. Peaks corresponding to detergents are eliminated in processed samples, allowing reliable peptide/protein identification. BSA tryptic digest (0.1 ml, 100 µg) containing a detergent was processed through 0.5 ml of Pierce Detergent Removal Resin and subjected to MALDI-MS analysis on a MALDI-Orbitrap Mass Spectrometer. Similar results were produced for samples containing CHAPS, NP-40 and SDS (data not shown).

Figure 3. Effective detergent removal eliminates interference and allows high sequence coverage analysis of BSA. Tryptic digests (0.1 ml, 100 µg) containing detergent were each processed through 0.5 ml of Pierce Detergent Removal Resin and subjected to LC-MS/MS analysis. Top Row: Base peak LC-MS chromatograms. Bottom Row: Integrated mass spectra. Similar results were produced for Brij-35, octyl glucoside, octyl thioglucoide and SDS (data not shown).
After processing samples, the high baseline caused by detergents is reduced or eliminated. Analysis of digested HeLa cell lysates by LC-MS/MS resulted in an approximate four-fold increase of identified peptides compared to a contaminated sample and equivalent numbers of peptides compared to a control sample, indicating minimal losses of peptides (Figure 4).

Analysis of HeLa Cell Lysate

![Graph showing peptide identification](image)

Figure 4. Effective detergent removal enables greater peptide identification. A tryptic digest of HeLa cell lysate (0.1 ml, 100 μg) containing 1% SDS was processed through 0.5 ml of Pierce Detergent Removal Resin and subjected to LC-MS/MS analysis. The processed sample allowed similar numbers of identified peptides as digests containing no SDS. Peptide identification is greatly reduced in sample containing SDS.

**Method**

**Detergent removal analysis:** Protein samples (1 mg/ml) containing detergent in 0.15 M NaCl and 0.05% sodium azide were processed through 0.5 ml of Pierce Detergent Removal Resin. Residual SDS was measured using Stains-All (Sigma Aldrich); Triton X-100, Triton X-114 and NP-40 were measured by absorbance at 275 nm (protein absorbance was subtracted); sodium deoxycholate, CHAPS, octyl glucoside, octyl thioglucoisde and lauryl maltoside were measured using concentrated sulfuric acid and phenol. Removal of Brij-35 was monitored by LC-MS/MS and MALDI-MS analysis. Protein concentration was determined with the Thermo Scientific Pierce BCA Protein Assay (Product # 23225).

**LC-MS/MS and MALDI-MS analysis:** BSA and HeLa lysate (1 mg/ml) in 50 mM ammonium bicarbonate buffer, pH 8.0 were digested overnight with trypsin at 37°C (enzyme-to-protein ratio, 1:50) in the presence of 1% of each detergent except SDS, which was added after trypsin digestion. Each sample (0.1 ml) was processed through 0.5 ml of Pierce Detergent Removal Resin. Control samples (unprocessed) were not processed. Samples were diluted and loaded (~1.5 pmol) directly onto a C18 column and subjected to LC-MS/MS analysis using a Thermo Scientific LTQ Mass Spectrometer. For MALDI-MS analysis, samples were diluted 1:15 (1 pmol) and analyzed using a Thermo Scientific MALDI-Orbitrap Mass Spectrometer. The matrix was alpha-cyano 4-hydroxy cinnamic acid (5 mg/ml) with acetonitrile/water/0.1% TFA as a co-solvent.

**References**


**Ordering Information**

<table>
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<tr>
<th>Product #</th>
<th>Description</th>
<th>Pkg. Size</th>
</tr>
</thead>
<tbody>
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<td>87776</td>
<td>Pierce Detergent Removal Micro Spin Column</td>
<td>25 columns</td>
</tr>
<tr>
<td></td>
<td>125 ml settled resin/column</td>
<td></td>
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<tr>
<td>87777</td>
<td>Pierce Detergent Removal Spin Column, 0.5 ml</td>
<td>25 columns</td>
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<tr>
<td></td>
<td>0.5 ml settled resin/column</td>
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<tr>
<td>87778</td>
<td>Pierce Detergent Removal Spin Column, 5 ml</td>
<td>5 columns</td>
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<td>4 ml settled resin/column</td>
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<td>87780</td>
<td>Pierce Detergent Removal Resin</td>
<td>10 ml</td>
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</tbody>
</table>

For more information, or to download product instructions, visit www.thermo.com/pierce
Forced Dialysis for Sample Concentration
Concentrates 100 ml down to 12 ml in six hours

Small samples dialyze much faster than larger samples because the concentration differential is much higher and the migratory diffusion distance is shorter. With a 100 ml dilute sample, it is often prudent to concentrate down to 12 ml with forced dialysis using SnakeSkin Tubing before dialysis in a Slide-A-Lyzer Dialysis Cassette. The following forced dialysis SnakeSkin Tubing application has been adapted from the method described in:


Method
1) Cut off and discard the bottom of two pipette tips (2.5-5 ml) so SnakeSkin Tubing easily fits through the pipette tip.
2) Insert one pipette through the rubber stopper.
3) Cut off the desired length of SnakeSkin Dialysis Tubing (for larger volumes, the membrane will extend above the flask).
4) Thread the SnakeSkin Dialysis Tubing (dry) through the rubber stopper containing the pipette tip.
5) Clip or tie several knots in the lower end of the SnakeSkin Tubing.
6) Pour the sample to be concentrated through the top of the open end of the SnakeSkin Tubing. (Before you completely fill the SnakeSkin Tubing, place the second pipette tip inside the SnakeSkin Tubing to create a secure seal between the SnakeSkin Dialysis Tubing and the first pipette tip.) Fill with remaining sample.
7) Place 3-4 cm of buffer in the flask.
   NOTE: Most of the SnakeSkin Tubing will not be exposed to buffer.
8) Clip or tie the open end of the SnakeSkin Dialysis Tubing to ensure a closed vacuum system.
9) Connect the side arm to house vacuum.
10) Concentrate sample until desired volume is reached.

Sample Results
1) A 1 mg/ml solution of bovine serum albumin was prepared in phosphate-buffered saline, pH 7.4.
2) Approximately 30 cm of SnakeSkin Dialysis Tubing was used and assembled as described previously.
3) After six hours, the starting sample volume (100 ml) was concentrated to 12 ml with an estimated protein recovery of 65%.

Evaporation for Sample Concentration
Water inside a Slide-A-Lyzer Dialysis Cassette will evaporate. The cassette is ideally suited for sample concentration via evaporation because of the dual membranes and high surface area. Place a sample in the cassette, then withdraw the air inside. Let your sample evaporate on the bench top (using a fan to increase airflow across the membrane will speed up the process), making sure to check every 10 minutes or less to prevent evaporation to dryness. When concentrating by evaporating the water from your sample, the small molecules (buffer salts, reducing agents, etc.) will also be concentrated because no diffusion occurs.
Desalting Columns and Plates

Gel Filtration

Gel filtration involves the chromatographic separation of molecules of different dimensions based on their relative abilities to penetrate into a suitable stationary phase. A chromatographic resin, usually consisting of very small, uncharged porous particles in an aqueous solution, is packed into a column and then used for the separation. Different levels of separation can be achieved based on the pore size of the resin. The resin can be chosen to totally exclude proteins or large molecules, while still including small solutes. Large molecules are excluded from the internal pores of the resin and emerge first from the column in the “void volume.” The smaller molecules are able to penetrate the pores, then progress through the column at a slower rate. These smaller molecules emerge from the column after the target sample.

Desalting and buffer exchange are two of the most widely used applications of gel filtration chromatography.

Desalting

Desalting involves the chromatographic separation of macromolecules in the void volume from smaller molecules that penetrate the gel bed.

Applications:
- Removing salts from protein solutions
- Removing phenol from nucleic acid preparations
- Separating excess crosslinker from conjugate preparations
- Removing excess derivatizing agents from modified proteins
- Removing unreacted dye from fluorescent antibodies
- Removing free radiolabel from labeled proteins

Buffer Exchange

Buffer exchange is used to place a protein solution into a more appropriate buffer prior to applications such as electrophoresis, ion exchange or affinity chromatography. In both desalting and buffer exchange, the macromolecular components are recovered in equilibrium with the same buffer used to equilibrate the column. If water is used for equilibration, the components will be desalted. If another buffer is used, a buffer exchange will result.
Thermo Scientific Zeba Desalt Spin Columns

**Highlights:**
- Exceptional protein recovery
- Wide product offering accommodates your sample needs
- Easy to use with no cumbersome column preparation or equilibration
- No screening fractions for protein or waiting for protein to emerge by gravity flow
- Minimal sample dilution

Although numerous techniques and resins for desalting are available, most have many drawbacks, including significant sample loss, long processing times and the need to collect multiple fractions. Zeba Desalt Spin Columns provide excellent protein recovery without the limitations associated with other desalting methods. Zeba Desalt Spin Columns are available in micro, 0.5, 2, 5 and 10 ml formats and allow processing of samples ranging from 2 μl to 4 ml (Table 1).

### Table 1. Recommended sample volumes for Thermo Scientific Zeba Spin Columns.

<table>
<thead>
<tr>
<th>Resin Bed</th>
<th>Sample Volume</th>
</tr>
</thead>
<tbody>
<tr>
<td>75 μl (micro) column</td>
<td>2-12 μl</td>
</tr>
<tr>
<td>0.5 ml column</td>
<td>30-130 μl</td>
</tr>
<tr>
<td>2 ml column</td>
<td>200-700 μl</td>
</tr>
<tr>
<td>5 ml column</td>
<td>600-2,000 μl</td>
</tr>
<tr>
<td>10 ml column</td>
<td>1,500-4,000 μl</td>
</tr>
<tr>
<td>96-well</td>
<td>20-100 μl</td>
</tr>
</tbody>
</table>

The easy-to-use Zeba Spin-Column Format dramatically improves results over standard drip-column methodologies, eliminating the need to wait for samples to emerge by gravity flow and the need to monitor fractions for protein recovery. Zeba Desalt Columns require no chromatographic system, cumbersome column preparation or equilibration and they can process multiple samples in ~8 minutes.

Zeba Desalt Spin Columns contain a high-performance desalting resin that offers exceptional desalting and protein-recovery characteristics compared to other commercially available resins (Figure 1). Samples containing as low as 25 μg/ml of protein can be processed, providing exceptional protein recovery and ≥ 95% retention of salts and other small molecules (< 1,000 MW).

### Figure 1. Increased protein recovery with Thermo Scientific Zeba Desalt Spin Columns.

#### A.

<table>
<thead>
<tr>
<th>Sample Size</th>
<th>700 μl</th>
<th>200 μl + 50 μl stacker</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>97%</td>
<td>90%</td>
</tr>
<tr>
<td>Zeba Column</td>
<td>78%</td>
<td>56%</td>
</tr>
<tr>
<td>Brand B</td>
<td>79%</td>
<td>29%</td>
</tr>
</tbody>
</table>

#### B.

<table>
<thead>
<tr>
<th>Sample Size</th>
<th>250 μg/ml BSA (66 kDa)</th>
<th>25 μg/ml BSA (66 kDa)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>97% 0.19 0.45 0.73</td>
<td>90% 0.14 0.07 0.14</td>
</tr>
<tr>
<td>Zeba Column</td>
<td>78% 0.14 0.07 0.14</td>
<td>79% 0.14 0.07 0.14</td>
</tr>
<tr>
<td>Brand B</td>
<td>79% 0.73 0.45 0.19</td>
<td>78% 0.73 0.45 0.19</td>
</tr>
<tr>
<td>Brand G</td>
<td>79% 0.73 0.45 0.19</td>
<td>78% 0.73 0.45 0.19</td>
</tr>
</tbody>
</table>

Samples of bovine serum albumin (BSA) at Figure 1A. 250 μg/ml and Figure 1B. 25 μg/ml in 1 M NaCl were desalted with the 2 ml Zeba Desalt Spin Columns and other commercial desalting resins using similar formats. A portion of the recovered sample (10 μl) was analyzed by SDS-PAGE. The remaining sample was used for conductivity measurements and Pierce BCA Protein Assay (Product # 23225) was performed to determine protein concentration. Zeba Desalt Resin provides significantly greater protein recovery under all conditions tested. Conductivity and protein recovery values after desalting are indicated for 250 μg/ml samples.
<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Pkg. Size</th>
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<tbody>
<tr>
<td>89877</td>
<td>Zeba Micro Desalt Spin Columns' 7K MWCO For 5-14 µl samples.</td>
<td>25/pack</td>
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<tr>
<td>89878</td>
<td>Zeba Micro Desalt Spin Columns' 7K MWCO For 5-14 µl samples.</td>
<td>50/pack</td>
</tr>
<tr>
<td>89882</td>
<td>Zeba Desalt Spin Columns, 0.5 ml 7K MWCO For 7-200 µl samples.</td>
<td>25/pack</td>
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<tr>
<td>89883</td>
<td>Zeba Desalt Spin Columns, 0.5 ml 7K MWCO For 7-200 µl samples.</td>
<td>50/pack</td>
</tr>
<tr>
<td>89889</td>
<td>Zeba Desalt Spin Columns, 2 ml 7K MWCO For 200-900 µl samples.</td>
<td>5/pack</td>
</tr>
<tr>
<td>89890</td>
<td>Zeba Desalt Spin Columns, 2 ml 7K MWCO For 200-900 µl samples.</td>
<td>25/pack</td>
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<tr>
<td>89891</td>
<td>Zeba Desalt Spin Columns, 5 ml 7K MWCO For 300-2,000 µl samples.</td>
<td>5/pack</td>
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<tr>
<td>89892</td>
<td>Zeba Desalt Spin Columns, 5 ml 7K MWCO For 300-2,000 µl samples.</td>
<td>25/pack</td>
</tr>
<tr>
<td>89893</td>
<td>Zeba Desalt Spin Columns, 10 ml 7K MWCO For 1,000-4,000 µl samples.</td>
<td>5/pack</td>
</tr>
<tr>
<td>89894</td>
<td>Zeba Desalt Spin Columns, 10 ml 7K MWCO For 1,000-4,000 µl samples.</td>
<td>25/pack</td>
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</tbody>
</table>

For more information, or to download product instructions, visit [www.thermo.com/pierce](http://www.thermo.com/pierce)
Thermo Scientific
Zeba 96-Well
Desalt Spin Plates

Highlights:
• Desalt protein in one fraction with no dilution
• Exceptional protein recovery
• Easy to use with no cumbersome plate preparation or equilibration
• Minimal sample dilution

The new Thermo Scientific Zeba 96-Well Desalt Spin Plates provide high-throughput removal of salt and small molecules from samples, preparing them for downstream analysis, including mass spectrometry, HPLC, capillary electrophoresis, metabolite screening and assay development.

The Zeba 96-Well Desalt Spin Plates contain a high-performance resin that provides exceptional desalting and protein recovery characteristics. Process small (20-100 μl) sample volumes and achieve exceptional protein recovery (Table 1) and > 95% removal of salts and other small molecules (< 1,000 Da) such as DTT, biotin, FITC or biotin-FITC. The Zeba 96-Well Desalt Spin Plates require no resin dispensing or hydration. One plate of 96 samples can be processed in 5 minutes.

Table 1. Protein recovery and desalting efficiency.

<table>
<thead>
<tr>
<th>Protein</th>
<th>% Protein Recovery</th>
<th>% NaCl Removed</th>
</tr>
</thead>
<tbody>
<tr>
<td>BSA (66 kDa)</td>
<td>98.5</td>
<td>&gt; 95</td>
</tr>
<tr>
<td>α-Lactalbumin (14.1 kDa)</td>
<td>91.5</td>
<td>&gt; 95</td>
</tr>
<tr>
<td>Ubiquitin (8.6 kDa)</td>
<td>85</td>
<td>&gt; 95</td>
</tr>
</tbody>
</table>

Protein samples (1 mg/ml) were prepared in 1.0 M NaCl and 100 μl samples were desalted using Zeba Desalt Spin Plates. Results were analyzed by Pierce BCA Protein Assay and conductivity measurements.

Ordering Information

<table>
<thead>
<tr>
<th>Product #</th>
<th>Description</th>
<th>Pkg. Size</th>
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<tbody>
<tr>
<td>89807</td>
<td>Zeba 96-well Desalt Spin Plates</td>
<td>2 plates</td>
</tr>
<tr>
<td></td>
<td>Each well contains ~550 μl resin slurry and can process 20-100 μl samples. The package contains two wash plates and two collection plates.</td>
<td></td>
</tr>
<tr>
<td>89808</td>
<td>Zeba 96-well Desalt Spin Plates</td>
<td>4 plates</td>
</tr>
<tr>
<td></td>
<td>Each well contains ~550 μl resin slurry and can process 20-100 μl samples. The package contains two wash plates and four collection plates.</td>
<td></td>
</tr>
<tr>
<td>89934</td>
<td>Pierce Desalting Chromatography Cartridges</td>
<td>5 x 1 ml</td>
</tr>
<tr>
<td></td>
<td>Each cartridge is packed with 1 ml Zeba Desalting Resin. Recommended for processing compounds &gt; 7K MW.</td>
<td></td>
</tr>
<tr>
<td>89935</td>
<td>Pierce Desalting Chromatography Cartridges</td>
<td>5 x 5 ml</td>
</tr>
<tr>
<td></td>
<td>Each cartridge is packed with 5 ml Zeba Desalting Resin. Recommended for processing compounds &gt; 7K MW.</td>
<td></td>
</tr>
</tbody>
</table>

1. Remove the bottom seal and stack the desalt plate on top of a wash plate, then remove the top seal.
2. Centrifuge for 2 minutes at 1,000 x g to remove the storage buffer.
3. Stack the desalt plate on top of a sample collection plate. Apply sample.
4. Centrifuge for 2 minutes at 1,000 x g.
5. Recover the desalted samples.

Figure 1. Thermo Scientific Zeba 96-Well Desalt Spin Plates are easy to use.
Protein Concentration

Protein concentration is a commonly performed and essential procedure for sample preparations. There are a variety of devices available, all containing ultrafiltration membranes with a range of molecular-weight cutoffs (MWCO). The type of device used can significantly affect protein recovery, especially with low-concentration samples that are often lost or damaged in the process.

Thermo Scientific Pierce Protein Concentrators

The new Thermo Scientific Pierce Concentrators are disposable ultrafiltration centrifugal devices for concentration and diafiltration/buffer-exchange of biological samples such as enzymes, antigens or antibodies. Because of their unique design, the Pierce Concentrators avoid the problems commonly associated with protein concentration. These concentrators consist of a high-performance regenerated cellulose membrane welded to a conical device and are compatible with swinging-bucket and fixed-angle rotors. The design enables a high degree of concentration in a single centrifugation step, while minimizing polarization and adsorption at the membrane surface. Additionally, researchers can accurately control the dead-stop volume and final concentration factor for reliable and consistent sample processing. Concentration factors of > 110-fold are achieved in 45 minutes with the 20 ml devices (Table 1). The membranes are accurately rated and routinely provide > 90% recovery of proteins larger than the membrane MWCO, and > 85% with low-concentration samples (Tables 1 and 2).

Highlights:
• Superior protein concentration and recovery – achieve > 110-fold protein concentration in 45 minutes with > 90% protein recovery
• Convenient – concentrate 1-20 ml of sample in a fast spin format
• Instrument-compatible – use swinging-bucket or fixed-angle rotors; collect sample without invert spinning
• Versatile – the 150K MWCO concentrator is ideal for samples containing microorganisms such as viral particles

Table 1. Thermo Scientific Pierce Concentrators provide exceptional recovery with low-concentration samples.

<table>
<thead>
<tr>
<th>Pierce Concentrator</th>
<th>Protein†</th>
<th>Time (min)</th>
<th>Protein Concentration (mg/ml)</th>
<th>Recovery (%)</th>
<th>Fold Concentration‡</th>
</tr>
</thead>
<tbody>
<tr>
<td>20 ml/9K* Cytochrome c</td>
<td>45</td>
<td>0.2</td>
<td>100</td>
<td>121</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.01</td>
<td>96</td>
<td>117</td>
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</tr>
<tr>
<td>20 ml/20K** BSA</td>
<td>45</td>
<td>0.2</td>
<td>98</td>
<td>137</td>
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<tr>
<td></td>
<td></td>
<td>0.01</td>
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<td>118</td>
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<tr>
<td>7 ml/9K* Cytochrome c</td>
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<td>0.2</td>
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<td>137</td>
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<tr>
<td></td>
<td></td>
<td>0.01</td>
<td>88</td>
<td>129</td>
<td></td>
</tr>
<tr>
<td>7 ml/20K** BSA</td>
<td>35</td>
<td>0.2</td>
<td>97</td>
<td>81</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>0.01</td>
<td>87</td>
<td>65</td>
<td></td>
</tr>
</tbody>
</table>

† Protein samples were centrifuged at 3,000 x g for 45 minutes at 22°C using a starting volume of 20 ml with the 20 ml concentrators and 5 ml with the 7 ml concentrators.  
‡ Fold concentration was determined by dividing the starting volume by the recovered (retentate) volume.
*For the 9K concentrators, percent recovery was calculated by measuring the absorbance at 409 nm of the retentate adjusted with buffer to the original volume.
**For the 20K concentrators, percent recovery was determined using the Thermo Scientific Micro BCA Protein Assay (Product # 23235).

Table 2. Typical retention of proteins using the 150 MWCO concentrator.*

<table>
<thead>
<tr>
<th>Protein</th>
<th>Concentration (mg/ml)</th>
<th>Recovery (%)</th>
</tr>
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<tbody>
<tr>
<td>IgG (150 kDa)</td>
<td>0.5</td>
<td>93</td>
</tr>
<tr>
<td>Aldolase (160 kDa)</td>
<td>0.25</td>
<td>87</td>
</tr>
</tbody>
</table>

* Protein solutions (13.5 ml) in phosphate-buffered saline were concentrated in a fixed-angle rotor at 2,000 x g at 22°C. The concentration was determined by Pierce BCA Protein Assay.
Pierce Concentrators effectively combine speed, capacity and recovery for high performance concentration, purification and separation of proteins even with dilute samples. We compared the Pierce Concentrators to ultrafiltration centrifugal devices from the other suppliers. Using a variety of test proteins and starting concentrations, protein recovery was monitored by Thermo Scientific Pierce BCA Protein Assay (Product #23225). For samples above the rated MWCO, high levels of protein was recovered (approximately 90% or greater) with Pierce Concentrators with a starting concentration as low as 0.02 mg/ml (Table 3). Significantly lower or no protein was recovered with devices from supplier V, indicating inappropriate molecular weight ratings or significant binding of protein to the device membrane.

Applications:
• Protein concentration with tissue culture media, antiserum or monoclonal antibody preparations
• Concentration of protein peaks following gel-permeation chromatography
• Removal of unincorporated protein label
• Concentration and desalt/buffer-exchange after eluting protein from ion-exchange, hydrophobic interaction (HIC), metal-chelate or affinity-chromatography columns

Table 3. Thermo Scientific Pierce Concentrators perform better than units from other suppliers.*

<table>
<thead>
<tr>
<th>Concentrator (MWCO)</th>
<th>BSA (66,000 MW)</th>
<th>Lysozyme (14,000 MW)</th>
<th>Ubiquitin (8,700 MW)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pierce Concentrator (9K)</td>
<td>91</td>
<td>98</td>
<td>96</td>
</tr>
<tr>
<td>Supplier M (10K)</td>
<td>90</td>
<td>93</td>
<td>91</td>
</tr>
<tr>
<td>Supplier V (10K)</td>
<td>76</td>
<td>5</td>
<td>5</td>
</tr>
</tbody>
</table>

* Proteins samples (~0.02 mg/ml) were centrifuged in concentrators at 3,000 x g until a 15- to 25-fold decrease in sample volume was achieved. Samples were recovered without membrane washing. Recovery was determined using the Pierce BCA Protein Assay. Results were unaffected by centrifugation rate and similar for the 7 and 20 ml Pierce Concentrators. Similar or higher recovery values were obtained with the Pierce Concentrators at higher protein loads (data not shown).

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<td>Pierce Concentrators, 9K/7 ml</td>
<td>10/pkg.</td>
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<tr>
<td>89884A</td>
<td>Pierce Concentrators, 9K/7 ml</td>
<td>25/pkg.</td>
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<td>87749</td>
<td>Pierce Concentrators, 9K/20 ml</td>
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<td>89885A</td>
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<td>89921</td>
<td>Pierce Concentrators, 150K/20 ml</td>
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<td>89923</td>
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<td>B-PER Reagent^</td>
<td>Gram(-) bacteria, <em>S. aureus</em>, <em>H. pylori</em>, <em>E. coli</em> strains BL21(D3) &gt; JM109&gt; DH5 &lt;M15, Archaebacteria, nematodes and <em>Acinetobacter</em> sp.</td>
<td>Yes</td>
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<td>B-PER II Reagent (A 2X version of B-PER Reagent)</td>
<td>Gram(-) bacteria, <em>S. aureus</em>, <em>H. pylori</em>, <em>E. coli</em> strains BL21(D3) &gt; JM109&gt; DH5 &lt;M15, Archaebacteria, nematodes and <em>Acinetobacter</em> sp.</td>
<td>Yes</td>
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<td>B-PER PBS Reagent</td>
<td>Gram(-) bacteria, <em>S. aureus</em>, <em>H. pylori</em>, <em>E. coli</em> strains BL21(D3) &gt; JM109&gt; DH5 &lt;M15, Archaebacteria, nematodes and <em>Acinetobacter</em> sp.</td>
<td>Yes</td>
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<tr>
<td>Y-PER Reagent</td>
<td><em>S. cerevisiae</em>, <em>Schizo-saccharomyces pombe</em>, <em>C. albicans</em>, <em>B. subtilis</em>, <em>E. coli</em>, <em>P. pastoris</em>, <em>Strep. avidinii</em> and <em>Acinetobacter</em> sp.</td>
<td>No</td>
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<td>Y-PER Plus Reagent</td>
<td>Yeast (<em>S. cerevisiae</em>) and <em>Acinetobacter</em> sp.</td>
<td>Yes</td>
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<tr>
<td>M-PER Reagent</td>
<td><em>COS</em>-7, <em>Hela</em> 1-6, 293, <em>CHO</em>, <em>MDA</em>, <em>MB231</em> and <em>FM2</em></td>
<td>Yes</td>
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<tr>
<td>P-PER Plant Protein Extraction Reagent 89003, Kit</td>
<td>Multiple plant organs (leaf, stem, root, seed and flowers); multiple plant species (<em>Arabidopsis, tobacco, maize, soybeans, peas, spinach, rice and other plant tissues</em>); and fresh, frozen and dehydrated plant tissues</td>
<td>No</td>
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<tr>
<td>T-PER Reagent</td>
<td>Heart, liver, kidney and brain</td>
<td>Yes</td>
</tr>
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<td>I-PER Reagent</td>
<td>Baculovirus-infected insect cells grown in suspension or monolayer culture</td>
<td>No</td>
</tr>
<tr>
<td>NE-PER Reagent 78833</td>
<td>Tissue: calf liver. Tissue: mouse heart, kidney, lung and liver; Cultured cells: epithelial (HeLa), fibroid (COS-7), kidney (NIH 3T3), liver (Hepa 1) and brain (C6)</td>
<td>No (CER) ER (NER)</td>
</tr>
<tr>
<td>Mem-PER Reagent 89826</td>
<td>Cultured cells: brain (C6), epithelial (HeLa), fibroblasts (NIH 3T3) and yeast (<em>S. cerevisiae</em>)</td>
<td>Yes^</td>
</tr>
<tr>
<td>Subcellular Protein Fractionation Kit 78940</td>
<td>Cultured mammalian cells</td>
<td>Yes</td>
</tr>
<tr>
<td>Mitochondria Isolation Kit for Cultured Cells 89874</td>
<td>Mammalian cells</td>
<td>Yes^</td>
</tr>
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<td>Mitochondria Isolation Kit for Tissue 89081</td>
<td>Heart, liver, kidney and brain</td>
<td>Yes^</td>
</tr>
<tr>
<td>Pierce RIPA Buffer 89080, 100 ml 89081, 250 ml</td>
<td>Cultured mammalian cells and cytoplasmic, membrane and nuclear proteins</td>
<td>Yes</td>
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<td>Lyssosome Enrichment Kit for Tissues and Cultured Cells 89059</td>
<td>Tissues and cultured cells</td>
<td>N/A</td>
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<tr>
<td>Peroxixosome Enrichment Kit for Tissue 89040</td>
<td>Heart, liver, kidney and brain</td>
<td>N/A</td>
</tr>
<tr>
<td>Pierce IP Lysis Buffer 87757, 160 ml 87759, 250 ml</td>
<td>Cultured mammalian cells</td>
<td>Yes</td>
</tr>
<tr>
<td>Nuclei Enrichment Kit for Tissue 89841</td>
<td>Heart, liver, kidney and brain</td>
<td>N/A</td>
</tr>
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</table>

1. The detergent can be removed by dialysis
2. Immunoprecipitation
3. Halt Protease Inhibitor Cocktail, Product # 78425 (EDTA-free) and 78430
4. Samples prepared in Mem-PER Reagent can be dialyzed if the buffer contains detergent (e.g., CHAPS), otherwise use Pierce SDS-PAGE Sample Prep Kit (Product # 89888)
5. Slide-A-Lyzer MINI Dialysis Units
6. 2-D Sample Prep for Nuclear Proteins (Product # 89863) and 2-D Sample Prep for Membrane Proteins (Product # 89844) were designed using our NE-PER and Mem-PER Reagents.
7. Need to lyse mitochondria first.
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Protein Assay Compatibility Notes

Pierce BCA Assay and Coomassie Plus Assay

Pierce BCA Assay and Coomassie Plus Assay can be used for any protein or enzyme. Sample solutions must be filtered for Pierce BCA Assay with Filter Supports.

Pierce BCA Assay

Pierce BCA Assay is adequate for most protein determination. hatten agents and reducing agents may be added to the sample or reaction. Pierce BCA Assay with Filter Supports is recommended for Pierce BCA Assay. Sample solutions must be filtered through Pierce BCA Assay with Filter Supports.

Pierce BCA Assay, Pierce Mini on Protein Assay with Filter Supports

Pierce BCA Assay, Pierce Mini on Protein Assay is adequate for most protein determination. Protein samples may be treated as for standard Pierce BCA Assay, Pierce Mini on Protein Assay with Filter Supports. Sample solutions must be filtered through Pierce BCA Assay with Filter Supports.

Slide-A-Lyzer Concentrating Solution

Slide-A-Lyzer Concentrating Solution can be used for protein concentration and analysis. It is recommended for use with Slide-A-Lyzer Dialysis Unit. It is not recommended for use with Slide-A-Lyzer G2.

2X N A X X X

3X N A X X X

5X N A X X X

10X N A X X X

Protein Assay

Protein Assay may be used to determine protein degradation. It is recommended for use with Protein Assay. Solutions may be treated as for standard Pierce BCA Assay. Sample solutions must be filtered through Protein Assay.

Detecting the Protein Assay

Detecting the Protein Assay may be used to determine protein degradation. Solutions may be treated as for standard Pierce BCA Assay. Sample solutions must be filtered through Detecting the Protein Assay.

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<td>Slide-A-Lyzer MINI</td>
<td>0.2-0.5 ml</td>
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<td>Slide-A-Lyzer</td>
<td>0.5-3 ml Kit</td>
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<td>Slide-A-Lyzer</td>
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Contact Information:

Pierce and Europe:

City Media Trade Center
Tel: 0800 38 40 820
Tel: 0800 252 185
Germany
Tel: 0228 9125650
France
Tel: +32 53 85 71 84
Tel: 0800 50 82 15
United States
Tel: 800 56 31 40
Tel: 0800 56 31 40

Customer Assistance E-mail: Pierce.CS@thermofisher.com

Website: www.thermo.com/pierce

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Technical Handbook

Version 2

Thermo Scientific Pierce High-Performance Dialysis, Desalting and Detergent Removal

Featuring Thermo Scientific Slide-A-Lyzer Dialysis Cassettes