

Prepare and Use Slide-A-Lyzer Concentrating Solution (PEG-20 Solution)

TR0078.0

Introduction

Thermo Scientific™ Slide-A-Lyzer Concentrating Solution (Product Nos. 66528, 66529, 66530) was discontinued in November 2014. The product was a solution of poly(ethylene glycol), which could effectively decrease the volume of a sample by hygroscopically removing water by ordinary dialysis. Following are instructions for preparing and using this solution in your own laboratory.

Supplies Needed for 500mL of PEG-20 Solution

- Poly(ethylene glycol), average M_n 20,000 (PEG-20) [Sigma-Aldrich Product No. 81300 (1kg)]
- 500mL of ultrapure water
- 1L glass or plastic beaker
- Stir plate, hot plate or water bath
- Stir bar

Preparation

- 1. Add 250mL of ultrapure water to a glass or plastic container.
- 2. Turn on stir bar and slowly add 200g of PEG-20 to container while mixing.
- 3. If PEG-20 does not dissolve into solution, gently warm solution while continuing to mix. If not using a stir plate, mixture can be hand mixed in a warm water bath to help dissolve.
- 4. Bring total volume to 500mL with ultrapure water and continue to mix until PEG-20 is fully dissolved.
- 5. Cool to room temperature before use.
- 6. Store the solution at room temperature.

Using PEG-20 Solution with Thermo Scientific Slide-A-Lyzer Cassettes

Using this PEG-20 concentrating solution with a Thermo ScientificTM Slide-A-LyzerTM cassette can quickly reduce sample volume, returning the sample to its original concentration or further concentrating it to a desired volume. Concentration is achieved by diffusion, drawing water and other small molecules out of the cassette and into the solution contained in a plastic bag. The volume reduction rate is comparable to centrifugation methods for sample concentration. Using the Slide-A-Lyzer cassette, dialysis and concentration can occur in the same device, alleviating potential protein loss when switching from one device to another.

Important Product Information

- If pH control is critical for the sample, use a sample buffer concentration of 100mM or greater. The low buffer salts in the sample will equilibrate between the sample and the concentrating solution, reducing their concentration in the sample. For example, when using a 3mL sample in 100mM phosphate and 25mL of concentrating solution, the phosphate will be diluted to ~10mM.
- Do not use concentrating solution with 20K MWCO Slide-A-Lyzer cassettes.

Procedure for Sample Concentration using Slide-A-Lyzer Cassettes

1. Determine the amount of concentrating solution and resealable bag size to use based on the Slide-A-Lyzer cassette size being used (Table 1).

Table 1. Concentrating solution volume and bag size to use.

Cassette Size (mL)	Concentrating Solution (mL)	Resealable Bag Size
0.1-0.5	20	3" × 4"
0.5-3	25	3" × 4"
3-12	50	5' × 8"
12-30	75	5' × 8"



- 2. Use a graduated cylinder to measure the required amount of PEG-20 solution and pour it into the appropriate-sized resealable bag. Drain the graduated cylinder into the resealable bag for 1 minute to allow the cylinder to empty.
- 3. Place the sample-containing cassette into the concentrating solution inside the resealable bag and fold the bag edge over the cassette, if needed.
- 4. Lay the bag containing the cassette on the counter, allowing the solution flow to cover the bottom membrane of the cassette. Use the palm of your hand to push the concentrating solution over the top membrane of the cassette and continue upward to remove excess air from the bag. Make sure there are no air bubbles between the cassette membrane windows and the concentrating solution. Seal the bag.
- 5. If needed, fold the edges of the resealable bag to consolidate the solution around the cassette. Place the bag on a rocking platform. Set a rocking speed to ensure the cassette within the bag is slightly moving (~35rpm). Alternatively, periodically knead the bag containing the concentrating solution.
- 6. Note: Use a rotating or microplate mixer with sufficient motion to keep the cassette moving.
- 7. Carefully monitor the volume decrease because concentration will occur rapidly.

Note: Wear gloves for the next two steps.

- 8. When the sample reaches the desired volume, remove the cassette from the bag.
- 9. To avoid introducing concentrating solution into the sample, thoroughly wash the outside of the cassette with ultrapure water. Also, wash the cassette cavity by performing the following steps:
 - Fill a 5-10mL syringe with ultrapure water. Partially insert needle into the needle port without touching or penetrating the gasket located within the cassette.
 - Flush the cassette cavity to eliminate any residual concentrating solution that could contaminate the sample.
 - Continue flushing until the water in the cavity flows freely away from the needle port area. Observe the water flushing action through the transparent cassette.
 - Using a clean syringe and needle, carefully remove sample from the cassette without puncturing the membrane.

Procedure for Sample Concentration in Thermo ScientificTM Slide-A-LyzerTM MINI Devices (10-100μL capacity)

Note: Use a 1.5mL or 2mL microcentrifuge tube for each sample to be concentrated. Make sure the Slide-A-Lyzer MINI Device fits inside the microcentrifuge tube.

- 1. Add 1.1mL of concentrating solution to a 1.5mL tube. Alternatively, add 1.4mL of concentrating solution to a 2mL microcentrifuge tube.
 - **Note:** The volume of concentrating solution required may vary with tubes from different manufacturers. Determine the volume needed with water before adding the concentrating solution.
- 2. Using a pipette, add sample to the bottom of the unit.
- 3. Wearing gloves, place the unit into the centrifuge tube containing the concentrating solution. Avoid trapping air bubbles under the membrane when inserting the unit into the tube.
- 4. Allow concentration to occur at room temperature until desired volume is reached.
- 5. Wearing gloves, use a pipette to retrieve the sample.

Current versions of product instructions are available at <u>thermoscientific.com/pierce</u>. For a faxed copy, call 800-874-3723 or +1 815-968-0747 or your local distributor.

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