SpectraPor[®] Tube-A-Lyzer[®] Dialysis Device

User Guide



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Abbreviations

CE	Cellulose Ester Membrane
DI	Deionized water
kD	Kilodalton
kGy	Kilogray
KR1	KR1 Peristaltic Pump
MWCO	Molecular Weight Cut-Off

1. Introduction

The SpectraPor® Tube-A-Lyzer® Dialysis Device from Repligen (Figure 1) is a ready-to-use, gentle separation device that combines convenience and disposability with the efficiency of dynamic dialysis to significantly increase the mass transfer rate and reduce the over-all dialysis from days to hours. Available in 2 sample volume sizes (8 - 10 ml and 25 - 30 ml), this self-contained, disposable device incorporates a semi-permeable dialysis membrane tubing inside a clear reservoir housing to separate the sample chamber from the surrounding buffer solution chamber. The glycerinated, dry Cellulose Ester (CE) Membrane is available in 6 molecular weight cut-offs (MWCO) ranging from 0.1 to 100 kD. The membrane defined sample chamber has a female Luer-Lok™ port on top that provides easy access for loading, withdrawing or in-process testing. The housing is designed to be a flow-through dialysate chamber and is equipped with a hose barb inlet and outlet ports for connecting to a buffer source and a waste collection vessel, or for discharging directly to drain.

Mounted onto the support rack of the KR1 Peristaltic Pump, the SpectraPor Tube-A-Lyzer Dialysis Device can be operated as a single unit or in parallel when multiple devices are combined using a coupling bracket (included in the Loading Kit). A second pump head can be mounted to the pump for operating coupled devices in parallel. The instructions that follow pertain to operating a single SpectraPor Tube-A-Lyzer Device.



Figure 1. SpectraPor Tube-A-Lyzer Dialysis Device

Single SpectraPor Tube-A-Lyzer Device set-up on a KrosFlo[®] KR1 Peristaltic Pump for buffer re-circulation.

Coupled SpectraPor Tube-A-Lyzer Devices with second pump head set-up for parallel operation in single-pass mode.

2. Specifications

Table 1. SpectraPor Tube-A-Lyzer Dialysis Device Specifications

	2 Volume Sizes	
	8 - 10 ml	25 - 30 ml
Buffer chamber volume	50 - 55 ml	120 - 130 ml
Total length	23 cm	50 cm
Total diameter	2.2 cm	2.2 cm
Membrane effective length	14 - 16 mm	36 - 38 mm
Membrane diameter	1.0 mm	1.0 mm

Table 2. SpectraPor Tube-A-Lyzer Dialysis Device MWCOs

6 MWCO's	
0.1 - 0.5 kD	
3.5 - 5 kD	
8 - 10 kD	
20 kD	
50 kD	
100 kD	

Table 3. Materials of Construction

Part Description	Materials
Membrane	Biotech grade Cellulose Ester membrane (CE) with glycerin
Sample Chamber	Polycarbonate top and bottom piece Polyurethane potting Female Luer-lok sample port Polypropylene male Luer-lok cap
Buffer Chamber	Polycarbonate housing Polycarbonate end caps ¼ in (6.3mm) hose-barb inlet/outlet ports

Table 4. Accessory Items and Kits

Part Nu	umber and Description
137100	Sample Loading Kit , includes 30 cc syringes (x3), dispensing tips (x3) and coupling brackets (x2)
137110	Replacement dispensing tips, 12 per package
163051	Extended Life Silicone Tubing, Size 17, ¼ in (6.3mm) ID, 10 ft (3m)

Check more product specifications on <u>www.repligen.com</u> or in the <u>Repligen E-Store</u>.

2.1 Applications

- Purification of labile proteins
- Buffer and pH change for protein refolding
- Protein prep for electrophoresis
- Removal of contaminants
- Isolation of delicate macromolecules and virus
- Tissue culture extract purification
- Desalting
- Concentration of antibodies
- Enzyme activity and ligand binding studies
- Temperature regulated dialysis
- Removal of oligosaccharides

3. Set-up instructions

- 1. Open the packaging and remove one device from packaging. Safely store the unused units in a dry environment at room temperature.
- 2. Cut two pieces of silicon pump tubing long enough to reach the preferred feed buffer source and return vessel or drain. The flexible tubing should be ¼ inch ID (163051). Connect a piece of pump tubing to both the bottom inlet hose-barb and the upper (side) outlet hose-barb of the unit's dialysate chamber.
- 3. Mount the SpectraPor Tube-A-Lyzer Device onto a standard lab stand or clamping mechanism in a vertical position with the sample port on the top and the dialysate inlet port on the bottom.
- 4. Thread the bottom inlet pump tubing (feed-line) through the peristaltic pump head.
- 5. Direct/connect the bottom inlet tubing to the dialysate feed source and the upper outlet tubing to the return vessel.

Note: In buffer recirculation mode, the buffer source also serves as the return vessel. For single pass mode, the upper outlet line can be directed to drain.

4. Pre-treating the membrane

- 1. Fill membrane and buffer chamber with 10 15% ethanol or isopropanol solution. Allow to sit at room temperature for 20-30 minutes until membrane appears translucent to clear.
- 2. Drain buffer and sample chamber.
- 3. Flush sample chamber thoroughly with deionized water (DI) water and drain. Repeat 2 more times.
- 4. Circulate DI water through buffer chamber and then drain. Repeat 2 more times.
- 5. Membrane is now pre-treated and ready for sample loading.

5. Recommended sample loading: Using syringe with dispensing tip

Allow air to escape while filling the sample chamber with a dispensing tip on the syringe.

- 1. Connect a flexible dispensing tip to a syringe that is sufficiently sized to hold your sample volume. For example, use at least a 10 cc syringe for 8-10 ml samples and at least 30 cc syringe for 25 30 ml samples. Fill syringe with sample.
- 2. Remove the male Luer-Lok cap of the sample chamber and direct the dispensing tip of the filled syringe into the sample port.

Note: Avoid occluding of sample port and preventing air from being able to escape.

- 3. Slowly press down on the syringe plunger to dispense the sample into the sample chamber. As the sample chamber fills, air will simultaneously be expelled around the dispensing tip. Make sure not to occlude the sample port. Do not overfill the sample chamber.
- 4. Remove the syringe and secure the male Luer-Lok cap on the sample port to seal the sample chamber.

6. Alternative Sample Loading: Using Only Syringe

Warning: Connecting a syringe directly to the sample port may rupture the membrane if all the air is not removed prior to loading the sample.

1. Draw sample into a syringe that has at least twice the capacity as the sample volume. For example, use a 20 or 30 cc syringe for 8 - 10 ml samples and 60 cc syringe for 25 - 30 ml samples. Invert and expel all air from leaving the syringe at most half-filled with sample only (no air).

Note: Before loading sample, first remove the air from the sample chamber to avoid rupturing the membrane.

- 2. Connect the half-filled sample syringe to the sample chamber port. Pull the plunger to draw all the air out of the sample chamber into the syringe, collapsing (flattening) the membrane.
- 3. After the air has been removed from the sample chamber, slowly press syringe plunger down to load the sample as the membrane opens back into a tubular shape. If the membrane is completely opened up and there is still sample in the syringe, do not force excess sample into the sample chamber. If there is still air in the sample chamber, pull back on the plunger to remove it and then press the plunger to replace it with remaining sample. If there is no air left to remove, do not attempt to load more sample. The maximum volume of sample has been loaded.
- 4. Remove the syringe and secure the male Luer-Lok cap on the sample port to seal the sample chamber.

7. Priming, Continued Operation and In-process Testing

- Using the KR1 Pump or a similarly equipped peristaltic pump, slowly begin pumping the buffer (dialysate) solution into the SpectraPor Tube-A-Lyzer[®] buffer chamber via the bottom feed (inlet) line. Flow rates between 10 - 20 ml/min are preferable to conserve the dialysate solution. Priming will expel the air from the dialysate chamber. Establish and maintain the desired dialysate flow rate based on the application.
- 2. Continue with the dynamic dialysis process until the desired separation endpoint is achieved.
- 3. Before doing in-process testing of the sample, first stop the pump, then use a syringe to withdraw a small volume of sample from the sample chamber.

8. Harvesting the Sample After Dialysis

- 1. Turn off the peristaltic pump and remove the feed line from the pump head while leaving it connected or directed to the feed source.
- 2. Drain the buffer out of the dialysate chamber by disconnecting the upper outlet line from the return vessel and raising it above the level of the SpectraPor Tube-A-Lyzer Device. You may also need to lower the feed source below the level of the unit (or alternatively raise the unit above the level of the feed source). This will drain the dialysate buffer back into the feed source reservoir.
- 3. Remove the sample chamber cap and connect an appropriate volume size syringe. Remove the SpectraPor Tube-A-Lyzer Device from the lab stand and clamping mechanism. Invert the unit so that the syringe is on the bottom and slowly pull back on the plunger to harvest the sample from the chamber, collapsing the membrane.
- 4. After the sample volume has been recovered, discard the used SpectraPor Tube-A-Lyzer Device.

9. Shelf-Life and Storage

Storage: Store new and unused Tube-A-Lyzer[®] Devices in a dry place at room temperature. Care must be taken to avoid humid environments. If the membrane becomes wet or moist and then subsequently dries out, it can crack and cause leaks during use.

Shelf Life: Approximately two years depending on storage conditions.

10. Sterilization

The following methods are approved for sterilizing the Tube-A-Lyzer® Device.

Note: Autoclaving is not recommended.

- Gamma Irradiation at 25 kGy
- Ethylene Oxide Gas exposure

Table 5. Tolerance

Temperature Limits	4 - 37° C
pH Limits	2 - 9
Organic Tolerance	Fair (refer to compatibility table <u>online</u> for specific tolerance)

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