GE Healthcare

PlusOne Mini Dialysis Kit

Product Specification Sheet

PlusOne™ Mini Dialysis Kit is designed for the dialysis of small samples with minimal handling and sample loss. Each dialysis tube consists of a sample tube with a cap that incorporates a dialysis membrane. Sample is placed in the tube, which is capped and inverted in a beaker containing the solution that the sample is to be dialyzed against. Salts and molecules smaller than the molecular weight cut-off of the dialysis membrane rapidly exchange through the membrane. Following dialysis, the tube is centrifuged briefly to recover the contents.

PlusOne Mini Dialysis Kit can be used for many research applications, but is particularly well-suited for de-salting protein samples prior to analysis by isoelectric focusing (IEF) and 2-D electrophoresis. The kit is available with a choice of molecular weight cut-off (either 1 kDa or 8k Da) and a choice of size (either 250 µl or 2 ml). Each kit supplies dialysis tubes and associated accessories sufficient for preparing 50 samples.

Warning
For research use only.
Not recommended or intended for diagnosis of disease in humans or animals.
Do not use internally or externally in humans or animals.

Storage

The kit should be stored at 4–8°C

Function testing

Each lot of the PlusOne Mini Dialysis Kit is tested to ensure that thedialysis tubes seal well and do not leak.

Safety warnings and precautions

All chemicals should be considered as potentially hazardous. We therefore recommend that this product is handled only by those persons who have been trained in laboratory techniques and that it is used in accordance with the principles of good laboratory practice. Wear suitable protective clothing such as laboratory overalls, safety glasses and gloves. Care should be taken to avoid contact with skin or eyes. In the case of contact with skin or eyes wash immediately with water. See material safety data sheet(s) and/or safety statement(s) for specific advice.

Components

Dialysis tubes: Tubes with caps that incorporate a dialysis membrane.

Caps: Standard tube caps to place on the tubes following dialysis.

Floats: Floating plastic sponges to suspend the inverted dialysis tubes in the dialysis solution.

Overview

Many macromolecular separation and analysis techniques require a sample that is largely free of salts and other small ionic molecules. Many current methods for de-salting are either complicated or result in sample loss or a substantial volume increase. Dialysis is a simple and straightforward technique that circumvents many of these problems, but dialysis of small samples can present handling problems. Sample can be lost during transfer in and out of dialysis bags.

PlusOne Mini Dialysis Kit offers a simple solution to the handling problems of small volume dialysis. The kit contains dialysis tubes, each of which consists of a sample tube with a cap that is adapted with a dialysis membrane. Sample is easily and quantitatively transferred into and out of the tube by pipet-ting. The capped tube is inverted in a stirred beaker containing the solution against which the sample is to be dialyzed. Salts and molecules smaller than the molecular weight cut-off of the dialysis membrane rapidly exchange through the membrane.

Following dialysis, the tube is centrifuged briefly. This forces the entire contents of the dialysis tube into the bottom of the tube, ensuring essentially 100% recovery. The dialyzing cap is replaced with a normal cap for storage of the dialyzed sample.

The first dimension isoelectric focusing (IEF) step of 2-D electrophoresis is particularly sensitive to low molecular weight ionic impurities. Even relatively low concentrations of salts (<5 mM) can slow the separation down, prevent sharp focusing or cause disturbances that result in a poor quality 2-D result. Low molecular weight ionic impurities can originate either as endogenous components of the sample source, or as salts and buffers introduced during preparation of the sample. In either case, the ability of a sample to be effectively separated by first dimension IEF, and the subsequent quality of the 2-D electrophoresis result can often be improved by dialyzing the sample prior to application. PlusOne Mini Dialysis Kit is well suited for this application because the capacity of the dialysis tubes (10–250 µl or 200 µl–2 ml) corresponds to a typical volume range for 2-D samples and because sample losses from the procedure are negligible. Dialysis times of a few hours to overnight are sufficient to reduce ionic contaminants to a level that does not interfere with first dimension IEF separation.

These products are covered by US patent 6217772.

Protocol

Introduction

PlusOne Mini Dialysis Kit is supplied in a choice of two different molecular weight cut-offs (1 kDa and 8 kDa) and a choice of two sizes (250 µl and 2 ml). The 1 kDa cut-off membrane will retain molecules with a nominal MW above 1000 and the 8 kDa cut-off membrane will retain molecules with a nominal MW above 8 000. The 8 kDa cut-off dialysis membrane allows faster dialysis than the 1 kDa cut-off membrane. The 250 µl tube accommodates samples in the volume range 10–250 µl and the 2 ml tube accommodates samples in the volume range 200 µl–2 ml.

Although the dialysis tubes have a capacity higher than indicated by their maximum volume designation, they should not be filled with more liquid than suggested. Dialysis becomes less efficient with a deeper layer of sample ovear the membrane.

Because some detergents, notably Triton X-100 and SDS, form high molecular weight micelles at low concentration, they cannot be effectively removed by dialysis. Other techniques, such as sample precipitation with PlusOne SDS-PAGE Clean-Up Kit or PlusOne 2-D Clean-Up Kit, must be used to remove these detergents.

Note: *The sample may lose or gain volume by osmosis during dialysis.*

General use instructions cover the use of the PlusOne Mini Dialysis Kit for all applications. Instructions also follow that cover the use of the kit specifically for preparing samples for 2-D electrophoresis.

Note: *Handle dialysis tubes and dialysis caps only with gloves. Do not touch or disturb the dialysis membrane.*

General use

1. The bag of dialysis tubes has a closure which allows it to be resealed after opening, and the bag must not be cut between the closure and the tubes. Open the bag above the reseal strip with scissors.
2. Remove one or more dialysis tubes from the bag and reseal it. The dialysis tubes are supplied in 0.05% (w/v) sodium azide storage solution which should be rinsed off prior to use with distilled or de-ionized water. Remove the dialysis cap and rinse each tube briefly with water from a squirt bottle. Decant away the rinse water. Place the dialysis cap in a clean beaker with the membrane side facing downward and cover with distilled or de-ionized water. Keep the cap in water until ready for use and do not allow them to dry.
3. Directly before use, remove each cap from the water. Remove excess water from the cap with a micropipette.
4. Place the sample into the dialysis tube. For 250 µl dialysis tubes, use 10–250 µl of sample. For 2 ml dialysis tubes, use 200 µl–2 ml of sample. Position the dialysis cap on the tube and tighten firmly.
5. Invert the dialysis tube ensuring that the entire sample rests on the dialysis membrane.

Note: *If the sample is viscous and does not initially rest on the dialysis membrane, the dialysis tube should be centrifuged briefly in the inverted position. This is done by placing the dialysis tube in a 50 ml centrifuge tube with the cap facing downward.*

Centrifuge at 10–100 × g for no more than 5–6 s. Spinning longer or faster may rupture the membrane. Check the tube to make sure that the entire sample has moved onto the dialysis membrane.

6. Secure each dialysis tube to one of the provided floats. Keeping the dialysis tube inverted, push the bottom of the tube through the hole in the float until the float stops against the dialysis cap.
7. Place the dialysis tube and float assembly, cap-end down, in a beaker of the solution to be dialyzed against. Several tubes may be placed in one beaker of solution if desired. Check that the dialysis membrane fully contacts the solution in the beaker and that there are no large air bubbles trapped beneath the dialysis membrane. Tilt the tube or squirt solution to remove air bubbles if necessary. Magnetically stir the solution in the beaker.

Note: *Optimal dialysis time will depend on several factors, including the nature and volume of the sample, the MW cut-off of the dialysis membrane and the temperature. Normally dialysis 2 h to overnight is sufficient. Dialysis may be conducted at 4–8°C to minimize sample degradation or modification, but this will slow dialysis. Dialysis can be conducted at room temperature if degradation or modification is not a concern.*

8. During dialysis, the contents of the dialysis tube should be mixed by inverting or tapping the tube once or twice. If necessary, repeat the centrifugation described in Step 5. The dialysis solution can also be replaced during dialysis.
9. Following dialysis, remove the dialysis tube from the float and immediately collect the sample in the bottom of the tube by centrifugation. Centrifuge the tube for 5–6 s at 500–1000 × g. Do not spin longer or faster as this might rupture the membrane.

Note: *if a centrifuge rotor that holds the dialysis tube snugly is unavailable, the dialysis tube may be placed inside a larger centrifuge tube for centrifugation. Since centrifugation is brief, and at relatively low speed, the tube does not need to fit snugly.*

10. Remove and discard the dialysis cap. Replace with a normal cap (provided in the kit) for storage.

Note: *Re-use of the dialysis tube is not recommended.*

Use in sample preparation for 2-D electrophoresis

A substantial reduction in interfering ions can be achieved by dialyzing 2-D samples against at least 40 × sample volume of solution for 2 h to overnight.

The 2-D sample should be prepared in a solution that will be compatible with first dimension IEF, including urea, CHAPS and DTT. For general guidelines and recipes, see "2-D Electrophoresis Using Immobilized pH Gradients: Principles and Methods."

Dialyze the sample against a solution that has the same concentrations of chaotropes (urea and thiourea) and DTT as the sample. Other, more expensive solution components such as CHAPS and carrier ampholytes do not need to be included in the dialysis solution. These components may be added back to their intended concentrations following dialysis.

Related products

Product	**Code No.**
Tris, 500 g	17-1321-01
Urea, 500 g	17-1319-01
CHAPS, 1 g	17-1314-01
Triton X-100, 500 ml	17-1315-01
Dithiothreitol (DTT), 1 g	17-1318-01
Bromophenol Blue, 10 g	17-1329-01
Sample Grinding Kit, 50 samples, up to 100 mg tissue or cell sample	80-6483-37
2-D Quant Kit, 500 assays, 1–50 µl and up to 50 µg	80-6483-56
2-D Clean-Up Kit, 50 samples, 1–100 µl	80-6484-51
SDS-PAGE Clean-Up Kit, 50 samples, 1–100 µl	80-6484-70
Mini Dialysis Kit 1 kDa cut-off, up to 250 µl	80-6483-75
Mini Dialysis Kit 1 kDa cut-off, up to 2 ml	80-6483-94
Mini Dialysis Kit 8 kDa cut-off, up to 250 µl	80-6484-13
Mini Dialysis Kit 8 kDa cut-off, up to 2 ml	80-6484-32
Handbook: *2-D Electrophoresis Using Immobilized pH Gradients, Principles & Methods*.	80-6429-60

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