

Dialysis with CelluSep membranes

The CelluSep product line has been discontinued (20190101).

See alternative products in the BioScience catalog [BB000d,8] (or p.B16(2-B52))

CelluSep	® T1 Tubings	(3500-40	000 MWCO)			
Flat width	Ø dry	Vol/cm	Cat. #	Length	Alternative: S/P 3 (or SP6, Biotech RC) <u>Tech Sheet S/P</u>	<u>1-7</u>	
19 mm	12.1 mm	1.15	T1-19-15 15 m * SP3 #132720 (18mm fw, 11.5mm Ø, 1ml/cm, 15m)				
46 mm	29.3 mm	6.74	T1-46-15	15 m *	SP3 #132724 (45mm fw, 29mm Ø, 6.4ml/cm, 30m)		
55 mm	35.0 mm	9.62	T1-55-15	15 m			
2	Tech Sheet S/P Biotech SP Biotech RC 3.5-5KDa #133192 (10mm fw, 6.4mm Ø, 0.32ml/cm 5m						
	SP Biotech RC 3.5-5KDa #133198 (16mm fw, 10mm Ø, 0.8ml/cm 5m)						

CelluSep® T2 Tubings (6000-8000 MWCO)

Flat width	Ø dry	Vol/cm	Cat. #	Length	Alternative: S/P 1 (or SP6/wet, SP7/wet&cleaned, Biotech RC/puriest)		
10 mm	6.37 mm	0.32	T2-10-15	15 m *	SP1 #132645 (10mm fw, 6.4mm Ø, 0.32ml/cm, 15m)		
23 mm	14.6 mm	1.67	T2-23-15	15 m *	SP1 #132650 (23mm fw, 14.6mm Ø, 1.7ml/cm, 30m)		
25 mm	15.9 mm	1.98	T2-25-15	15 m *			
32 mm	20.4 mm	3.27	T2-32-15	15 m *	SP1 #132655 (32mm fw, 20.4mm Ø, 3.3ml/cm, 30m)		
40 mm	25.5 mm	5.10	T2-40-15	15 m *	SP1 #132660 (40mm fw, 25.5mm Ø, 5.1ml/cm, 30m)		
50 mm	31.8 mm	7.94	T2-50-15	15 m *	SP1 #132665 (50mm fw, 32mm Ø, 8.0ml/cm, 30m)		
100 mm	63.7 mm	31.8	T2-100-15	15 m	SP1 #132670 (100mm fw, 64mm Ø, 32ml/cm, 15m)		
					SP1 #132675 (120mm fw, 76mm Ø, 46ml/cm, 15m)		

CelluSep® T3 Tubings (12000-14000 MWCO)							
Flat width	Ø dry	Vol/cm	Cat. #	Length	Alternative: S/P2 (or SP4/slow kinetic; SP5/reinforced, SP6/wet, SP7/wet&cleaned, Biotech RC/puriest) Tech Sheet S/P1-7		
10 mm	6.37 mm	0.32	T3-10-15	15 m *	SP2 #132685 (6mm fw, 3.8mm Ø, 0.1ml/cm, 15m) SP2 #132676 (10mm fw, mm Ø, 0.32ml/cm, 15m)		
25 mm	15.9 mm	1.98	T3-25-15	15 m *	SP2 #132678 (25mm fw, 6.4mm Ø, 2.0ml/cm, 15m)		
33 mm	21.0 mm	3.46	T3-33-15	15 m *			
45 mm	28.6 mm	6.42	T3-45-15	15 m *	SP2 #132680 (45mm fw, 16.0mm Ø, 6.4ml/cm, 15m)		
76 mm	48.4 mm	18.4	T3-76-5	5 m	SP2 #132645 (50mm fw, 32mm Ø, 8.0ml/cm, 30m)		
					SP2 #132682 (105mm fw, 29mm Ø, 34ml/cm, 15m) SP2 #132684 (120mm fw, 67mm Ø, 46ml/cm, 15m)		

CelluSep ®	CelluSep® T4 Tubings (12000-14000 MWCO):								
Flat width	Ø dry	Vol/cm	Cat. #	Length	Alternative: S/P 2	Tech Sheet S/P1-7			
10 mm	6.37 mm	0.32	T4-10-30	30 m	SP2 #132676 (10mm fw,	, mm Ø, 0.32ml/cm, 15m)			
25 mm	15.9 mm	1.98	T4-25-30	30 m	SP2 #132678 (25mm fw,	, 6.4mm Ø, 2.0ml/cm, 15m)			
33 mm	21.0 mm	3.46	T4-33-30	30 m					
45 mm	28.6 mm	6.42	T4-45-30	30 m	SP2 #132680 (45mm fw,	, 16.0mm Ø, 6.4ml/cm, 15m)			
75 mm	47.7 mm	17.9	T4-75-15	15 m	SP2 #132682 (105mm fv	w, 29.0mm Ø, 34ml/cm, 15m)			

Tech Sheet S/P Biotech

SBiotech RC 8-10KDa SBiotech RC 8-10KDa SBiotech RC 20KDa SBiotech RC20KDa

#133164 (10mm fw, 6.4mm Ø, 0.32ml/cm 5m) #133270 (16mm fw, 10mm Ø, 0.8ml/cm 5m) #133336 (10mm fw, 6.4mm Ø, 0.32ml/cm 5m) #133342 (16mm fw, 10mm Ø, 0.8ml/cm 5m)



This product line has been discontinued (20190101).

Introduction to Cellu-Sep Dialysis Products

CelluSep® Dialysis tubings are the best choice for dialysis, notably in Biology applications, for proteins and nucleic acids. They are regenerated cellulose membranes manufactured with high quality process for optimal and reproducible results:

T1, T2, T3 and T4 CelluSep® Dialysis Membranes

suit classical dialysis applications, especially when attention is paid to very low adsorption of proteins, low impurities levels, good compatibilities, and nice cost.

H1 CelluSep® Dialysis Membranes

suit most exigent applications (minimal contaminants content, ready-touse individual packages); great notably for DNA desalting.

CelluSep membrane closure clamps Macro Filler funnels

the right accessories for easier handling.

Technical tips:

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- See Instructions for use
 - See Technical Information
 - . principle and basics of dialysis
 - .sterilization of membranes
 - . cleaning membranes from heavy metals and sulfides
 - . concentrating sample directly in your dialysis bag !



CelluSep® membranes Performances

<u>Protein adsorption</u> (bovine serum albumin)

<=1mg/cm²

Retention

 (T=10 hours, with T2 Membranes)

 Cytochrome C (12,3 KDa)
 97%

 Vitamin B-12 (1355 Da)
 59%

Scientific and technical Information

Shall I rinse or pre-treat CelluSep membranes?

It is recommended to rinse briefly CelluSep membranes to remove the bulk ok preservative agents, especially in case of T1.2.3.4 membranes that contain glycerin and should be soaked. This can be omitted in most applications with H1 membranes, that contain 2% ethanol, although it is recommended as well. Only sensitive applications may require to treat membranes according protocol 2.

How long to dialyze - time, buffer volume and changes?

Low molecular weights salts and buffers (i.e. Tris or KPO₄) equilibrate within 3 hours with stirring. Equilibrium times for higher MW and viscous sample will be longer. Change the dialysis buffer as necessary. Usually, 2-3 dialysis buffer changes with 1000/100-fold excess over sample volume are sufficient.

More information in the <u>technical notice FT-CelluT</u> Quality/manufacturing, Principle of dialysis, Nominal filter rating, Performances, Compatibilities

See also the <u>technical notice about Dialysis</u>, <u>#NT-Dialya</u> Principle of dialysis | Advantages and main applications | FAQs



Products Descriptions

Cellu•Sep® T1, T2, T3 and T4 tubular dialysis membranes

Cellu•Sep® T1, T2, T3 et T4 membranes suit most dialysis applications

- Very low adsorption of proteins
- Pure regenerated cellulose for low impurities levels
- Nominal Filter ratings: 3500, 6000 and 12000 Daltons
- Economical

Each T1, T2, T3 package includes :

- a detailed instruction manual
- 2 clamps, scissors

CelluSep® membranes technical data

*CelluSep® T1-T4 membranes content:

Cadmium	< 0.1 ppm
Chromium	< 0.1 ppm
Copper	< 0.1 ppm
Iron	13.6 ppm
Nickel	< 0.1 ppm
Zinc	6.24 - 12.9 ppm
Lead	0.75 - 0.8 ppm

*CelluSep® T1, T2, T3 et T4 membranes contain glycerol to prevent brittleness, and are packaged with a desiccant to control humidity.

*CelluSep® T1, T2, T3 et T4 membranes have a symmetric pore structure to allows small molecule to migrate in either directions, and to improve separation accuracy.

(12-14000 MWCO)

Length

15 m *

15 m *

15 m*

15 m*

5 m

Cat. #

T3-10-15

T3-25-15

CelluSep® T1 Tubings Nominal Filter rating : 3 500 Da

CelluSep® T3 Tubings

Ø dry

6.37 mm

15.9 mm

Flat width

10 mm

25 mm

Nominal Filter rating : 12 000 Da

Vol/cm

0.32

1.98

			(3500-4000	MWCO)
Flat width	Ø dry	Vol/cm	Cat. #	Length
19 mm	12.1 mm	1.15	T1-19-15	15 m *
46 mm	29.3 mm	6.74	T1-46-15	15 m *
55 mm	35.0 mm	9.62	T1-55-15	15 m

* also exists in 30m length

CelluSep® T2 Tubings Nominal Filter rating : 6 000 Da

			(6-8000 MW	/CO)
Flat width	Ø dry	Vol/cm	Cat. #	Length
10 mm	6.37 mm	0.32	T2-10-15	15 m *
23 mm	14.6 mm	1.67	T2-23-15	15 m *
25 mm	15.9 mm	1.98	T2-25-15	15 m *
32 mm	20.4 mm	3.27	T2-32-15	15 m *
40 mm	25.5 mm	5.10	T2-40-15	15 m *
50 mm	31.8 mm	7.94	T2-50-15	15 m *
100 mm	63.7 mm	31.8	T2-100-15	15 m

33 mm 21.0 mm 3.46 T3-33-15 45 mm 28.6 mm 6.42 T3-45-15 76 mm 48.4 mm 18.4 T3-76-5

CelluSep® T4 Tubings Nominal Filter rating : 12 000 Da

Standard grade			(12-14000 MWCO)			
Flat width	Ø dry	Vol/cm	Cat. #	Length		
10 mm	6.37 mm	0.32	T4-10-30	30 m		
25 mm	15.9 mm	1.98	T4-25-30	30 m		
33 mm	21.0 mm	3.46	T4-33-30	30 m		
45 mm	28.6 mm	6.42	T4-45-30	30 m		
75 mm	47.7 mm	17.9	T4-75-15	15 m		



Cellu•Sep® H1 Membranes High Grade

Cellu•Sep® H1 are ideal for sensitive dialysis applications, with proteins and especially with DNA/RNA

- Pre-treated for minimal contaminants content
- Accurate MW separations
- Ready-to-use individual-use packaged
- MWCO from 1 000 to 25 000 Daltons



Cellu•Sep® H1 are ideal when glycerol, sulfur compounds, or small amounts of heavy metals will interfere with subsequent steps. Cellu•Sep® H1 membranes are EDTA pretreated and packaged wet in a preservative solution. Individual-use 50cm packages avoid waste, contamination and restorage problems.

Cellu•Sep® H1 provide an efficient tool for dialysis with accurate MWCO:

-For proteins, notably globular ones, Molecular Weight Cut-Off (MWCO) has become a convenient guide for determining which membrane to use. Ultimately, however the size and the three-dimensional shape of a molecule determine whether it pass or not through a membrane.

-With the important expansion of Molecular Biology and the subsequent need to desalt nucleic acids (DNA and RNA) samples, the behavior of dialysis membranes with DNA/RNA, and their high cleanness has become important.

Nucleic acids are routinely characterized by the length in base (single-stranded: ss) or pairs (double-stranded: ds), and they display far less tertiary structure than proteins. Choosing the appropriate membrane according to its retention of DNA, by length, provides a superior alternative to using the MW of the DNA fragment. The table below compares the nominal membrane MWCO to retention of various lengths of DNA (ss and ds).

Nominal Filter rating	15-mer	% of retention 35-mer	117-mer	
	2 000 Da	100 %	95 %	93 %
	5 000	95 %	93 %	90 %
% of retention -	10 000	50 %	82 %	82 %
Single stranded	15 000	50 %	82 %	82 %
	25 000	39 %	74 %	68 %
	2 000	94 %	100 %	100 %
% of retention -	5 000	93 %	100 %	100 %
Double stranded DNA	10 000	91 %	100 %	100 %
	15 000	90 %	95 %	100 %
	25 000	82 %	86 %	100 %

See Instructions for use

See Technical Information

MWCO of 2 000 and 5 000 permit desalting any size fragment (ss or ds). However, 10, 15 and 20 000 MWCO membranes provide a

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CelluSep H1 tubings in Lengths of 50cm

MWCO	Flat width	Ø	Vol/cm	Cat.number (5m)	Cat.number (10m)	Alternative: S/P 6 Rolls / or SP7 Wet&cleaned, or or S/P Biotech RC
						> 132636 (1KDa-18mmfw,1.1ml/cm) 13262 > 132638 (1KDa-38mmfw,4.6ml/cm) 13262 > 132640 (1KDa-45mmfw,6.4ml/cm) 13263
3 500 Da	19 mm	-	1.15	-	HR03-19-10	> 132590 (3.5KDa-18mmfw, 1.1ml/cm)
3 500	46 mm	-		-	HR03-46-10	> 132592 (2KDa-45mmfw, 6.4ml/cm)> 132594 (2KDa-54mmfw, 9.3ml/cm)
8 000	12 mm	-	0.46	HR08-12-5	HR08-12-10	> 132579 (8KDa-12mmfw, 0.45ml/cm) > 128058 (8KDa-18mmfw, 1.1ml/cm)
8 000	24 mm	-	1.67	HR08-24-5	HR08-24-10	>132580 (8KDa-24mmfw, 1.8ml/cm)
8 000	32 mm	-	3.27	HR08-32-5	HR08-32-10	> 132582 (8KDa-32mmfw, 3.3ml/cm)
8 000	40 mm	-	5.10	HR08-40-5	HR08-42-10	>132584 (8KDa-40mmfw, 5.1ml/cm)
8 000	50 mm	-	7.95	HR08-50-5	HR08-50-10	>132586 (8KDa-50mmfw, 7.9ml/cm)
10 000	12 mm	-	0.46	HR10-12-5	HR10-12-10	> 132570 (10KDa-12mmfw, 0.45ml/cm) > 128118 (10KDa-18mmfw, 1.1ml/cm)
10 000	24 mm	-	1.67	HR10-24-5	HR10-24-10	> 132572 (10KDa-24mmfw, 1.8ml/cm)
10 000	32 mm	-	3.27	HR10-32-5	HR10-32-10	> 132574 (10KDa-32mmfw, 3.3ml/cm)
10 000	45 mm	-	6.42	HR10-45-5	HR10-45-10	> 132576 (10KDa-45mmfw, 6.4ml/cm)
10 000	50 mm	-	7.94	HR10-50-5	HR10-50-10	> 132576
15 000	12 mm	-	0.46	HR15-12-5	HR15-12-10	> 132560 (15KDa-12mmfw, 0.45ml/cm) > 128158 (15KDa-18mmfw, 1.1ml/cm)
15 000	25 mm	-	1.98	HR15-25-5	HR15-25-10	> 132562 (15KDa-24mmfw, 1.8ml/cm)
15 000	34 mm	-	3.46	HR15-34-5	HR15-34-10	> 132564 (15KDa-32mmfw, 3.3ml/cm)
15 000	45 mm	-	6.42	HR15-45-5	HR15-45-10	>132566 (15KDa-45mmfw, 6.4ml/cm)
25 000						<pre>> 132550 (25KDa-12mmfw, 0.45ml/cm) > 128218 (25KDa-18mmfw, 1.1ml/cm) > 132552 (25KDa-28mmfw,2.5ml/cm) > 132554 (25KDa-34mmfw, 3.7ml/cm)</pre>
50 000	34 mm	-	3.46	HR50-50-5	HR50-50-10	> 132539 (50KDa-12mmfw, 0.45ml/cm) > 128240 (50KDa-18mmfw, 1.1ml/cm) > 132542 (50KDa-28mmfw, 2.5ml/cm) > 132544 (50KDa-34mmfw, 3.7ml/cm)

Tech Sheet S/P Biotech

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CelluSep H1 tubings Roll (10m Length)

MWCO	Flat width	Ø	Vol/cm	Cat. number (5U)	Cat. number (10U)	Alternative: S/P 6 Rolls ^{(or <u>SP123)</u> / or SP7 Wet&cleaned, or or S/P Biotech RC}
1 000 Da	18 mm	11.5 mm	1.04	H1-18-5	H1-18-10	> 132636 (1KDa-18mmfw,1.1ml/cm)
1 000	38 mm	24.2 mm	4.59	H1-38-5	H1-38-10	> 132638 (1KDa-38mmfw,4.6ml/cm)
1 000	45 mm	28.6 mm	6.42	H1-45-5	H1-45-10	> 132640 (1KDa-45mmfw,6.4ml/cm)
2 000	18 mm	11.5 mm	1.04	H2-18-5	H2-18-10	> 132620 (2KDa-18mmfw,1.1ml/cm) or SP3
2 000	38 mm	24.2 mm	4.59	H2-38-5	H2-38-10	> 132625 (2KDa-38mmfw4.6ml/cm)
2 000	45 mm	28.6 mm	6.42	H2-45-5	H2-45-10	> 132633 (2KDa-45mmfw,6.4ml/cm)
5 000	18 mm	11.5 mm	1.04	H5-18-5	H5-18-10	> 132645 (6/8KDa-10mmfw, 0.32ml/cm) SP
5 000	20 mm	12.7 mm	1.27	H5-20-5	H5-20-10	>132650 (6/8KDa-23mmfw, 1.7ml/cm) SP1
5 000	32 mm	19.1 mm	2.86	H5-32-5	H5-33-10	>132632 (6/8KDa-32mmfw, 3.3ml/cm) SP1
5 000	40 mm	25.5 mm	5.10	H5-40-5	H5-40-10	>132660 (6/8KDa-40mmfw, 5.1ml/cm) SP1
10 000	10 mm	6.37 mm	0.32	H10-10-5	H10-10-10	> 132570 (10KDa-12mmfw, 0.45ml/cm) > 128118 (10KDa-18mmfw, 1.1ml/cm) ^{or SP2}
10 000	25 mm	15.9 mm	1.98	H10-25-5	H10-25-10	> 132572 (10KDa-24mmfw, 1.8ml/cm)
10 000	32 mm	20.4 mm	3.27	H10-32-5	H10-32-10	> 132574 (10KDa-32mmfw, 3.3ml/cm)
10 000	40 mm	25.5 mm	5.10	H10-40-5	H10-40-10	> 132576 (10KDa-45mmfw, 6.4ml/cm)
15 000	10 mm	6.37 mm	0.32	H15-10-5 -	H15-10-10	> 132560 (15KDa-12mmfw, 0.45ml/cm) > 128158 (15KDa-18mmfw, 1.1ml/cm)
15 000	25 mm	15.9 mm	1.98	H15-25-5	H15-25-10	> 132562 (15KDa-24mmfw, 1.8ml/cm)
15 000	32 mm	20.4 mm	3.27	H15-32-5	H15-32-10	> 132564 (15KDa-32mmfw, 3.3ml/cm)
15 000	45 mm	28.6 mm	6.42	H15-45-5	H15-45-10	> 132566 (15KDa-45mmfw, 6.4ml/cm)
25 000	12 mm	7.62 mm	0.46	H25-12-5	H25-12-10	> 132550 (25KDa-12mmfw, 0.45ml/cm) > 128218 (25KDa-18mmfw, 1.1ml/cm)
25 000	25 mm	15.9 mm	1.98	H25-25-5	H25-25-10	> 132552 (25KDa-28mmfw,2.5ml/cm)
25 000	35 mm	22.3 mm	3.91	H25-35-5	H25-35-10	> 132554 (25KDa-34mmfw, 3.7ml/cm)
25 000	40 mm	25.5 mm	5.10	H25-40-5	H25-40-10	> 132554
50 000	28 mm		2.50	H50-34-5	H50-34-10	> 132539 (50KDa-12mmfw, 0.45ml/cm) > 128240 (50KDa-18mmfw, 1.1ml/cm) > 132542 (50KDa-28mmfw, 2.5ml/cm)
50 000	34 mm		3.46	H50-28-5	H50-28-10	> 132544 (50KDa-34mmfw, 3.7ml/cm)
						Tech Sheet S/P1 7

Tech Sheet S/P1-7 Tech Sheet S/P Biotech

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Accessories

CelluSep® membrane closure clamps



These clamps seal the tubing by a simple pressure and remain locked during dialysis until the user opens it. They avoid the hassle of making and checking knots, reduce the risk of loosing sample.

The clamps are made of resistant and inert polyamide 6.6 (NYLON), and do not float, for an optimal dialysis.

Description - max membrane widht	Length	Cat. #	Qty
Closure clamp - 46 mm	65 mm	CB-1050	10U
Closure clamp - 65 mm	90 mm	CB-1070	10U
Closure clamp - 105 mm	130 mm	CB-10100	10U
Clamp 35mm #142936	55mm #142	2937 75mm	#1429

Blue color. Exist in Organge, White, Green ; Weighed, Magnetic. See <u>Dialysis catalog p.24</u> [BB000d,24]

Blue colored Clamps

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interchim

35mm #142936 55mm #142937 75mm #142938

Macro filler funnels

Cut tubular membrane to length and clamp at one end, slip the open end onto the polypropylene funnel and secure with the silicone loop fastener.

Open snap cap, fill with sample, and hang in container. Funnels are supported by a threenotched universal hanger that fits over the edge of common vessels.



Description	Cat. #	Qty	
Macro filler funnels	P58480	5U	
For 23 mm, 25 mm, 32 mm, 40 mm and 45 mm flat widths. Package of five funnels and two silicone loops.			
Mini filler funnel	P58490	5U	
For 10 mm and 19 mm flat widths. Package of five funnels and two silicone loops			
Silicone loops	P58500	10U	



Directions for Use

For most applications, CelluSep membranes can be used directly after a brief rinse (protocol 1). For demanding applications, the membrane can be prepared as described by protocol 2, or also with more conveniency, Cellu-Sep H1 pre-treated, highly clean_wet membranes are recommended. The membranes can be sterilized (protocol 3).

Protocol 1: common use instructions of CelluSep tubings

This protocol suits most classic applications with CelluSep membranes.

-It is recommended to wear gloves (fingers may leave traces of proteins).

For <u>CelluSep T1.2.3.4 membranes</u>	For <u>CelluSep H1 membranes</u> (ready to use, sterile, and wet membranes)
 -Cut the desired length of tubing from the roll. -Soak and rinse briefly the inside with distilled water then dialysis buffer. -Close the tubing at one end (bottom): put a clamp (or make a knot and secure it). 	-Remove as many H1 membranes from the container as needed. Rem: It is particularly important to store the unused membranes at
	-Carefully cut the individual pouch at one end and remove preservative solution. Rinse briefly with distilled water.
	-Remove the membrane from its pouch and rinse with sterile distilled water.
	-Secure a clamp to one end of the tubing. If a 50cm length is too long, cut down to appropriate length.

-Eventually check the integrity of the membrane:

fill the tubing with dialysis buffer and hold vertically. Pour out test solution.

-Introduce the sample to dialyze though the open tubing end.

Use pipette, of fill if large container/tubings (see below funnel accessory)

-Close the tubing at upper end: put a clamp (or make a knot and secure it).

Note: Avoid trapping much air in the tube. For concentrated samples (i.e. salts), leave space enough in the tubing to allow net flow of dialysis buffer (osmosis) and to prevent tubing from bursting.

-Immerse the tubing in the dialysis bath (200-1000 times the sample volume) preferably under constant stirring at room temperature (or $+4^{\circ}$ C if temperature-sensitive sample).

-Change the bath buffer as necessary. As general guideline,

perform a first-time dialysis period of 30min-1H, a second one of 1-4 hours, then a third overnight at 4°C. *Note:* The number of changes, the dialysis duration and the volumes of bath/sample may be optimized depending on operating conditions (temperature, agitation, sample volume and type, membrane type...) and desired dialysis efficiency / application. Optimally, dialysis should reach the equilibrium state, but it usually requires a too long dialysis period. It is advantageous to shorten at least the first dialysis period(s). Viscous samples and rather hydrophobic molecules require longer dialysis time. For critical applications, one could monitor undesired compound absence/presence in the bath buffer and sample. For less exigent applications, sufficient desalting can typically be obtained with only 2 (large bath) buffer changes. For rapid estimation if dialysis volumes and changes number may be acceptable, it is useful to calculate the dilution factor of the sample in the bathes; i.e. a 5ml sample is dialyzed 3 times in 1liter, the dilution factor is at best (1000/5)³= 8.10⁷; so an initial 100mM concentration of a salt (i.e. Tris) could ideally reach down 12pM Tris under complete dialysis.

-Recover the sample from the tube.

Remove the tubing from dialysis bath, and if necessary wipe off it briefly (use an absorbent paper).

Place one end into a container (tube, vial...). Open the clamp, or cut with clean scissors just above the knot.

Note: For small tubing lengths, hold the dialysis tubing vertically by the other end.

For large volumes, pose the dialysis tubing on a clean bench, to avoid the sample flows too much rapidly. Allow the sample to drain out completely (raise the unopened end of the dialysis tube).

Press the tubing between 2 fingers from the upper end to the lower end.

Note: For good recovery (important for small volume samples), rinse the interior of the tube with a small volume of buffer, which will be added to the dialyzed sample.

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The sample can be controlled (final volume, protein concentration) and stored or used for downstream applications.



Protocol 2: pre-treatment of CelluSep membranes (sensitive applications)

Although CelluSep T1-4 series are pure materials, simple rince as in protocol 1 may leave traces of preservatives and other compounds than can be critical to sample further steps (glycerol, ethanol, sulfur compounds, or small amounts of heavy metals).

For most demanding applications, choose pre-treated H1 series for ready-to-use membrane, or treat CelluSep T1-4 and H1 membranes as follows:

Wearing gloves, cut the desired length of tubing from the roll.

Soak for 15min in distilled water.

Heat to 80°C in 10mM sodium bicarbonate and incubate for 30minutes while stirring gently.

Incubate the tubing in a 10mM Na₂. EDTA solution for 30 minutes.

You may repeat up 3 times a 30min incubation in fresh 10mM Na₂. EDTA soln

Incubate for 30min at 80°C in distilled water under constant stirring;

Cool, and store in a refrigerator in water (short term), or if possible with 0.5% sodium azide solution, or 0.1% sodium benzoate, or alternatively in a 20-50% ethanol solution. The tubing should always remain submersed.

The membrane is ready to use (protocol 1). If necessary, tubing may be sterilized (see protocol 3).

Protocol 3: Sterilization of Cellu-Sep membranes

The common method of sterilization is exposure to ethylene oxide gaz. Alternative methods are gamma irradiation and steam autoclaving. Suggested preparation of membrane before sterilization is to soak it for 30min in distilled water.

Protocol 3a: Chemical sterilization with ethylene Oxide

Place the soaked membrane in an open polyethylene bag in a vacuum oven.

Evacuate and fill the oven with a gas mixture of 20% Ethylene Oxide + 80% CO2 by a total pressure of 1 atmosphere. Treat the membrane for 5 hours at 40°C. Evacuate the sterilizing gas and admit 50% of humidity of air. A slight reduction, approximately 10% permeability characteristic, has been reported with the use of this Ethylene Oxide method

Protocol 3b: Gamma-Irradiation sterilization

Seal the membrane in a polyethylene bag

Expose the bag to gamma ray source for a total dose of 2.5 Megarads. The temperature should not go beyond 10°C. The permeability characteristic after the treatment is approximately 75%.

Protocol 3c: Autoclaving sterilization

The membrane is treated with boiling water or steam autoclaving (121°C at 100kPa (41bar) for 10min in distilled water; The length of the cycling should be as short as possible). Autoclaved membrane should not be dried. Uptima does not recommend this method, because temperature over 90°C will change the structure of the membrane. The permeation characteristics / performances should be re-characterized. Dry heating over a period of 48hours at 80°C drops the permeability to about 50%.

Additional Information

Reagent for R&D in vitro use only Ordering information at <u>www.interchim.com</u>

Related products

<u>PBS (powder pack, or tabs)</u> UP68723 <u>FastDialyser</u> (re-usable dialysis device) <u>GebaFlex Dialysers</u> (dialysis/electroelution device) <u>BC Assay Protein quantitation kit</u> UP40840A <u>IMAplate</u> (for UV-determination of nucleic acid concentration)

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Contact Uptima – Interchim for any question

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