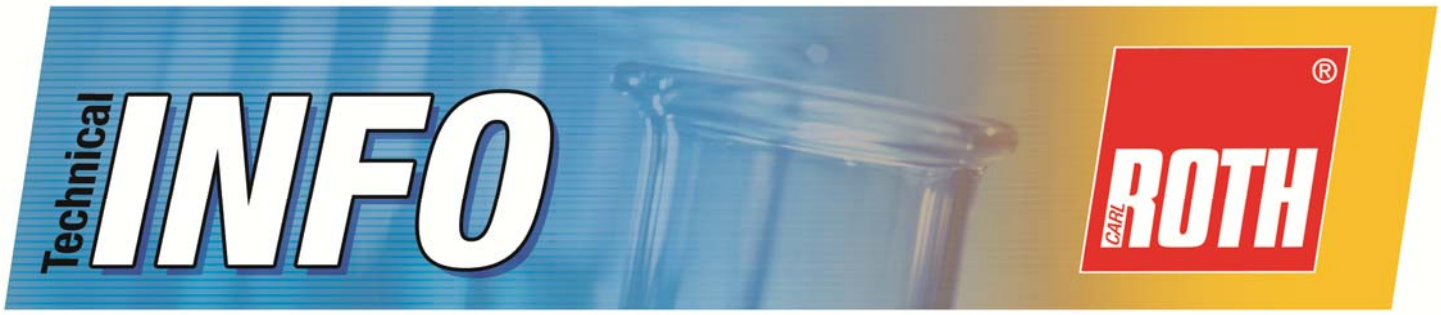


ZelluTrans/Roth Mini Dialyzer (MD)

Vol. 10-100 µl

MWCO	1 MD in microtube	12 MD in microtube	1 strip (= 8 MD)
3500 (3.5 kDa)	4765.1	4766.1	4767.1
6000-8000 (6-8 kDa)	4769.1	4770.1	4771.1
12000-14000 (12-14 kDa)	4772.1	4775.1	4776.1
Schwimmhilfe/Float			4768.1
Abdeckfolie/Sealing Film			4781.1

Technical Data Sheet and Applications

Content

Technical Data Sheet

ZelluTrans/Roth Mini Dialyzer

Specifications:

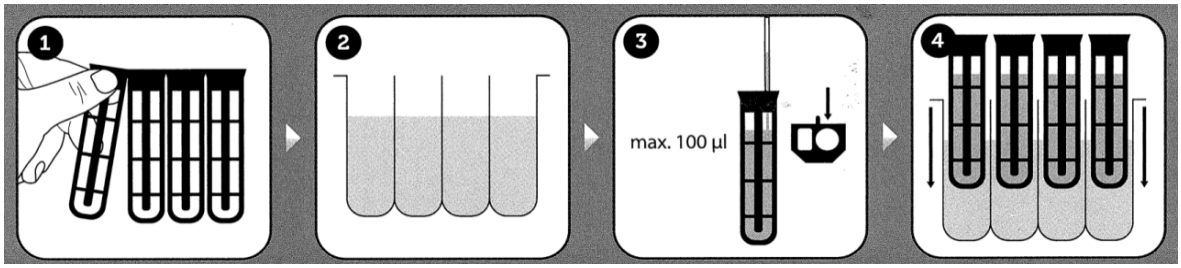
- max. sample volume 100 µl
- temperature 1-60 °C
- for aqueous solution
- incubation time up to 24 h
- pH 3-10
- cut off 3.5, 6-8, 12-14 kDa
- storage 4-22 °C
- strips with 8 samples, scalable from 1 to 96 samples
- conform to Microplate Standard (SBS)
- membrane: low binding regenerated cellulose, contain glycerole to prevent embrittlement and traces of elements like sulphides and heavy metals

Applications:

- Protein and peptide sample purification e.g. desalting before mass spectrometry
- Optimization of protein renaturation with different renaturation buffers and steps
- Removal of dyes after protein labeling
- Protein sample rebuffering
- Glycoprotein modification and engineering
- Protein in vitro translation
- Cell culture: studies of a cell line with a virus strain
- Enzyme activity assays
- Plasmid or primer purification

Manual – English Version

ZelluTrans/Roth Mini Dialyzer



Preparation

For a smaller number of samples, break carefully the desired number of single dialyzers from the dialyzer strip or use a single dialyzer in microcentrifuge tube. Please do not touch the membrane!

Buffer Preparation

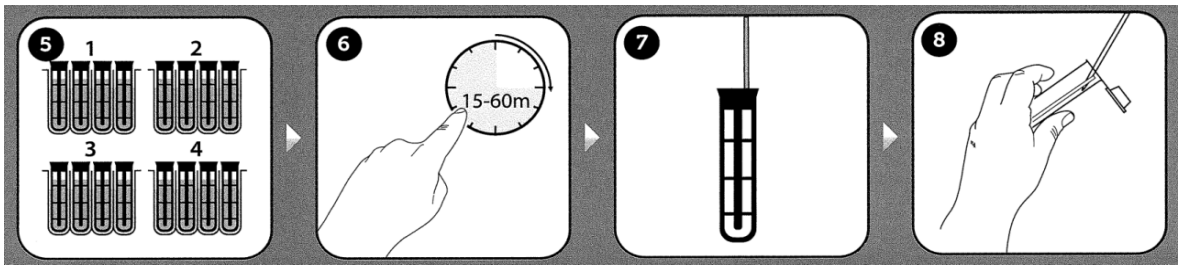
Pipette dialysis buffer into microcentrifuge tubes (max 1.4 ml/tube) or into a 96-deep well plate (max 1.8 ml/well).

Loading Sample

Load pipette (2-200µl) with sample (max 100µl). Insert the pipette tip upright into the round opening and press on slightly. Inject sample slowly.

Introduction

Place the single dialyzer into a prepared microcentrifuge tube (upright position) and the dialyzer strip into a prepared 96-deep well plate, respectively.



Dialysis

Dialysis can be done in one step. To accelerate the procedure change buffer solution repeatedly. For this purpose simply put the single dialyzer into a tube with fresh dialysis buffer or change the position of the dialyzer strip in the deep well plate.

Dialysis Time

The dialysis time depends on the compounds of the sample and the cut-off of the membrane. Typical dialysis times range from 30 min to 4 h, with repeated replacement of the dialysis buffer (every 15-60 min). For dialysis of salts change buffer every 30 min.

Sample Retrieval

Use pipette 2-200µl and set volume to 140µl. Press the piston to the first stop, hold it, put the pipette upright into the round opening. Aspirate sample carefully.

Storage

Finally, pipette the sample into a microcentrifuge tube or a microtest plate for further analytics.

Materials Required

Single dialyzer or dialyzer strip (consisting of 8 connected single dialyzers).

Deep well plate (2 ml) or microcentrifuge tubes (2 ml)

Pipette 2-200 µl (for sample), pipette 1-2 ml (für dialysis buffer).

Microcentrifuge tubes or microtest plates for sample storage

Forceps to use for disposal of single dialyzers out of microcentrifuge tube.

Suitable protection gloves and goggles

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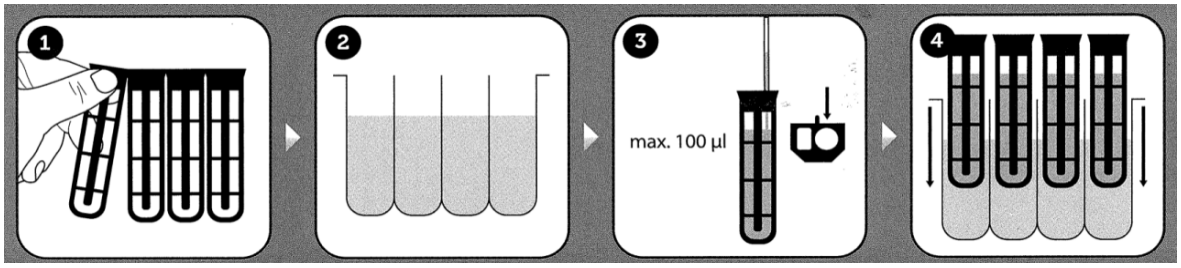
Manual – German Version

ZelluTrans/Roth Mini Dialyzer



Vorbereitung

Falls man nur einen oder mehrere Einzeldialyzer verwenden möchte, kann man diese vorsichtig vom Dialyzer-Streifen abbrechen. Bitte die Membran nicht berühren! Alternativ bieten wir Einzeldialyzer im Micro-zentrifugenröhrchen an.

Vorlegen

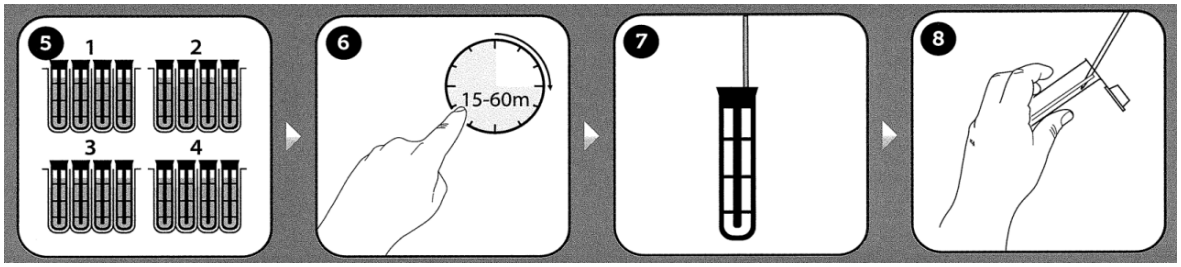
Dialyselösung in Mikro-zentrifugenröhrchen (max. 1,4 ml/Tube) oder in 96-Deepwellplatte (max. 1,8 ml/Well) pipettieren.

Probe einfüllen

Probe (max. 100 µl) mit Pipette 2-200 µl aufnehmen, Pipetten-spitze möglichst senkrecht in die runde Öffnung einführen und leicht andrücken. Probe langsam einfüllen.

Einsetzen

Einzeldialyzer in senkrecht stehendes Mikrozentrifugenröhrche n bzw. Dialyzer-Streifen in vorbereitete Deepwell-platte stellen.



Dialyse

Die Dialyse kann dann in einem Schritt erfolgen. Der Vorgang wird beschleunigt, wenn man den Puffer zwischendurch mehrmals wechselt. Dazu wird der Dialyzer einfach in ein neues Röhrchen mit Dialyselösung gesetzt bzw. innerhalb der Deep-wellplatte umgesetzt.

Dialysezeit

Die Dialysezeit hängt von der zu dialysierenden Substanz ab und der Ausschlussgröße der Membran. Die typische Dialysezeit für den Mini-Dialyzer liegt zwischen 30 min und 4 h mit wieder-holtem Wechsel des Dialyse-puffers (alle 15 bis 60 min). Richtwert für Salze: Wechsel alle 30 min.

Probe entnehmen

Pipette 2-200 µl auf 140 µl einstellen. Kolben bis zum ersten Druckpunkt herab-drücken und dort halten. Pipettenspitze in die runde Öffnung einführen und leicht andrücken. Dann Probe vorsichtig heraus-pipettieren.

Aufbewahrung

Probe zur weiteren Verwendung in ein vorbereitetes Gefäß geben.

Materialien

Einzeldialyzer oder Dialyzerstreifen (8 verbundene Einzeldialyzer)

Wahlweise 96-Deepwellplatte (2 ml) oder Mikrozentrifugen-röhrchen (2 ml).

Pipette 2-200 µl (für Probe), Pipette 1-2 ml (für Dialyse-lösung).

Mikrozentrifugen-röhrchen oder Mikrotiterplatten zur Aufnahme der Probe nach der Dialyse.

Spitze Pinzette zum Entnehmen der Einzeldialyzer aus Mikrozentri-fugenröhrchen.

Geeignete Schutzhand-schuhe und Schutzbrille

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Chemical resistance

ZelluTrans/Roth Mini Dialyzer

Materials and Methods

ZelluTrans/Roth Mini Dialyzer: MWCO 3,5, 6-8, 12-14 kDa

Test Sample: 100 µl of Congo Red color solution in water dd

Dialysis solution: 1,4 ml of tested chemical

Incubation: 18 h

Determination Method: Optical integrity and leak-tightness to air pressure

Acetonitrile	Good
Acetone	Good
Chloroform	Good
Dimethyl sulfoxide	Good
Ethanol 70%	Good
Ethanol 98%	Good
Ethylacetate	Good
Ethylene glycol	Good
Glycerol	Good
n-Hexane	Good
iso-Propanol	Good
Methanol 98%	Good
Methylene chloride	Good
1-Propanol	Good
Tetrahydrofuran	Good
Toluene	Good
Hydrogen peroxide 30%	Good

Acetic acid 25%	Good
Acetic acid 96%	Good
Formic acid 25%	Good
Formic acid 100%	No
Hydrochloric acid 10%	Limited
Hydrochloric acid 25%	No
Hydrochloric acid 37%	No
Hydrofluoric acid 50%	No
Nitric acid 25%	No
Nitric acid 65%	No
Phosphoric acid 25%	Limited
Phosphoric acid 85%	No
Sulfuric acid 98%	No
Ammonium hydroxide 1N	Limited
Ammonium hydroxide 25%	Limited
Potassium hydroxide 1N	Limited
Potassium hydroxide 32%	No
Sodium hydroxide 1N	Limited
Sodium hydroxide 32%	No

Good chemical resistance	Good
Limited chemical resistance, e.g. pore size cannot be guaranteed	Limited
No chemical resistance, use not recommended	No

Sample Volume Recovery

ZelluTrans/Roth Mini Dialyzer

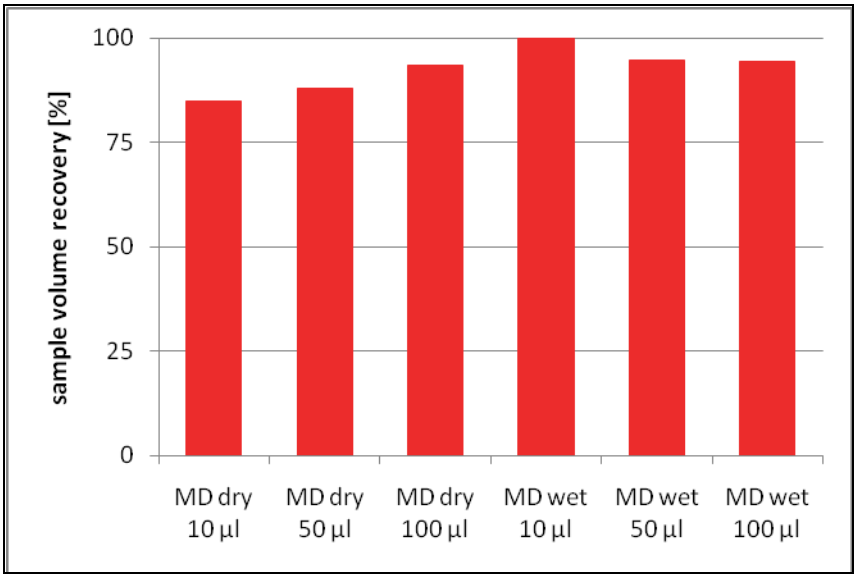
Materials and Methods

ZelluTrans/Roth Mini Dialyzer: MWCO 6-8 kDa (Art. No. 4769.1 ff)

Sample: 10 µl, 50 µl, or 100 µl respectively of Aq. dest.

Determination Method: Gravimetry

Sample	Recovery [%]
MD dry 10 µl	84.86
MD dry 50 µl	88.01
MD dry 100 µl	93.60
MD wet 10 µl	100.00
MD wet 50 µl	94.53
MD wet 100 µl	94.37
MD: Mini Dialyzer	
Dry: dry membrane	
Wet: prewetted membrane	



Application Note - Protein Binding (BSA)

ZelluTrans/Roth Mini Dialyzer

Materials and Methods

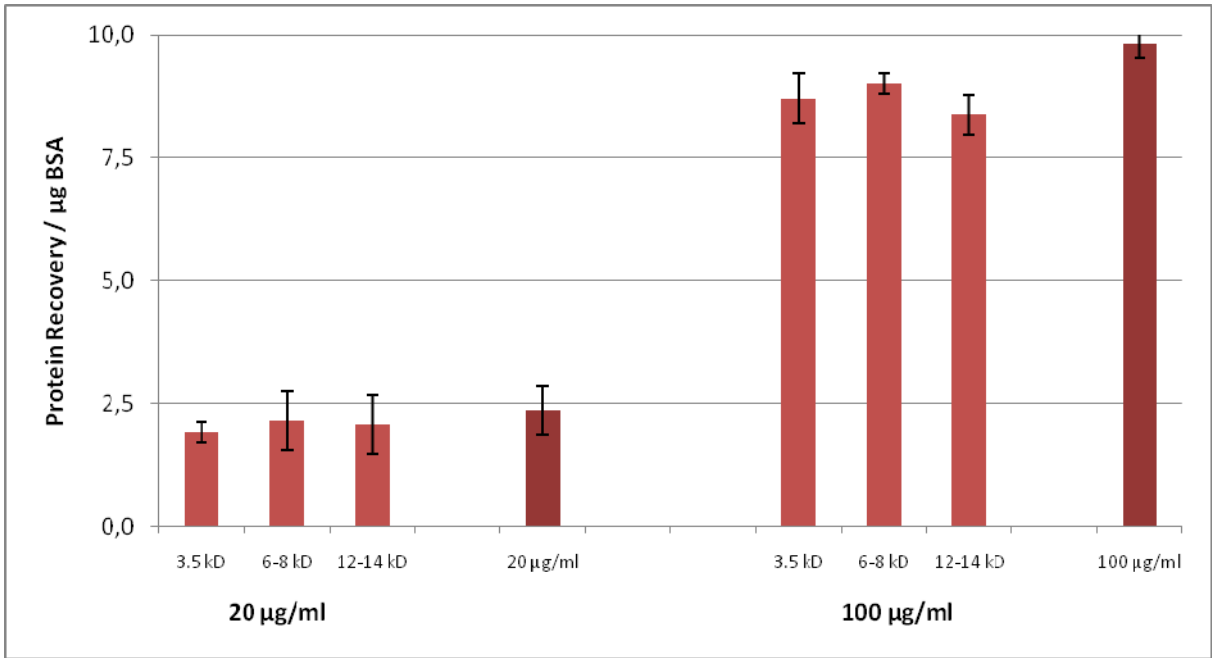
Sample: 100µl of BSA solution in PBS (20 and 100 µg/ml), n= 5 - 6

Dialysis Buffer: 1,8 ml of Aq. dest., exchange after 1 h, Incubation time: 4h at RT

Determination Method: Protein determination according to Bradford,

Tecan Sunrise photometer

Protein recovery	µg Protein	SD
3.5 kD (20 µg/ml)	1.92	0.2
6-8 kD (20 µg/ml)	2.14	0.6
12-14 kD (20 µg/ml)	2.08	0.6
Initial value (20 µg/ml)	2.35	0.5
3.5 kD (100 µg/ml)	8.71	0.5
6-8 kD (100 µg/ml)	9.02	0.2
12-14 kD (100 µg/ml)	8.38	0.4
Initial value (100 µg/ml)	9.83	0.3



Application Note - Urea Removal

ZelluTrans/Roth Mini Dialyzer

Materials and Methods

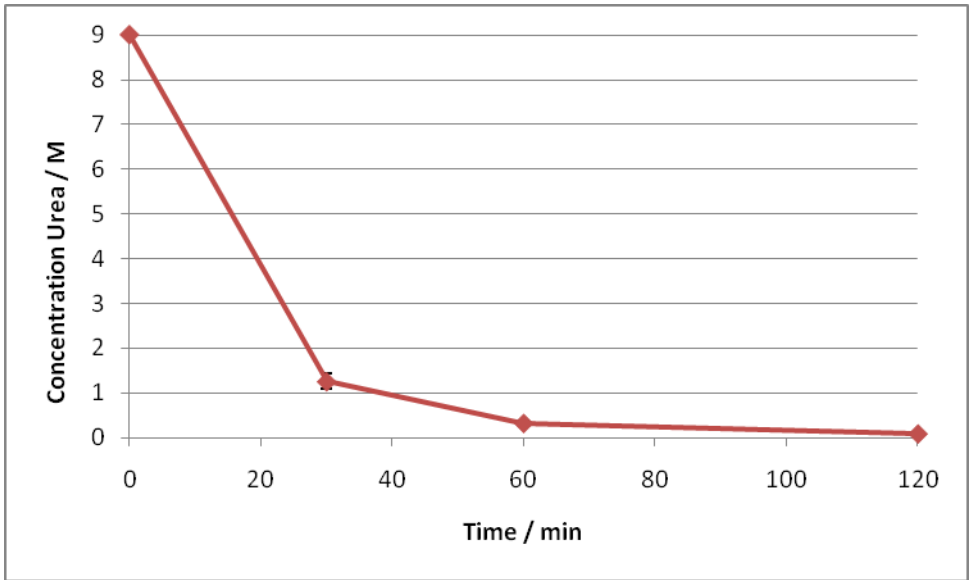
ZelluTrans/Roth Mini Dialyzer: MWCO 3,5 kDa (Art. No. 4765.1 ff)

Sample: 100 µl of 1 mg/ml BSA in 9 M Urea

Dialysis Buffer: 1.8 ml 10 mM Tris, pH 8.3, Buffer exchange interval 30 min, RT

Determination Method: Wescor VAPRO 5520 Osmometer

t in min	Residual Urea in M	SD
0	9	0,00
30	1,25	0,17
60	0,32	0,06
120	0,09	0,02



Remark: Due to the high mobility of urea in solutions and the short diffusion distances in ZelluTrans/Roth Mini Dialyzer, there is only a minor osmotic effect, resulting in a slightly higher volume recovery of about 110 µl.

Application Note - Salt Removal

ZelluTrans/Roth Mini Dialyzer

Materials and Methods

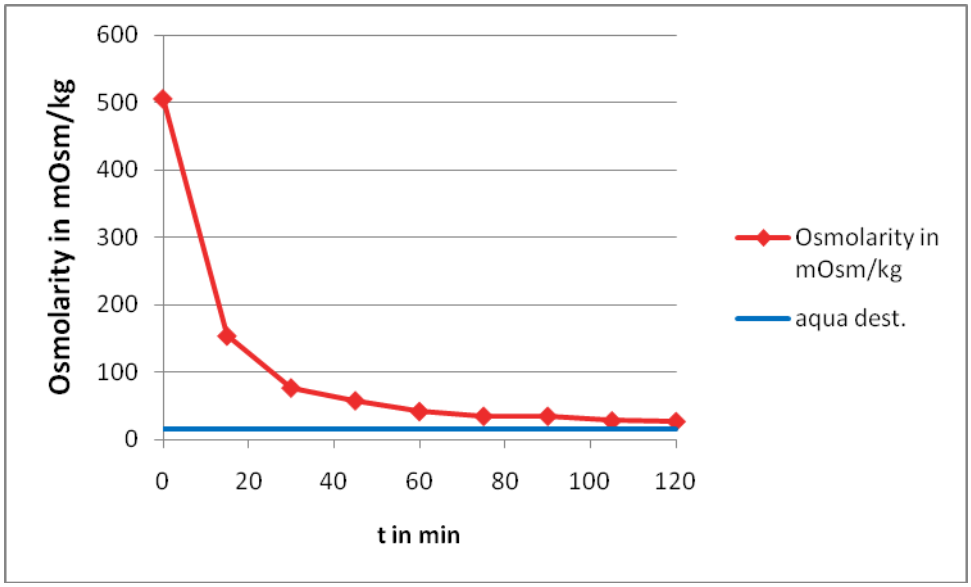
ZelluTrans/Roth Mini Dialyzer: MWCO 6-8 kDa (Art. No. 4769.1 ff)

Sample: 100µl of 50mM H_2NaPO_4/ HNa_2PO_4, 150mM NaCl, pH 7.4

Dialysis Buffer: 1.8 ml Aq. dest., Buffer exchange interval 15 min, 20 °C

Determination Method: Wescor Osmometer Vapor 5520

t in min	Osmolarity in mOsm/kg
0	506
15	154
30	77
45	58
60	42
75	35
90	35
105	28
120	27
aqua	15



Application Note - Dialysis Speed of different MWCO

ZelluTrans/Roth Mini Dialyzer

Materials and Methods

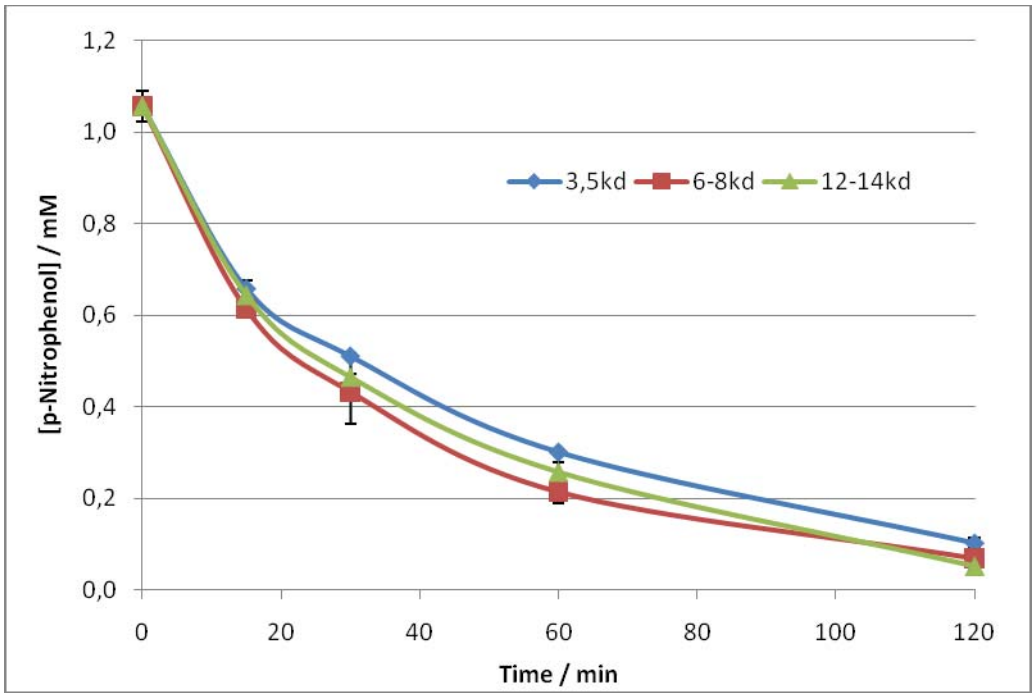
ZelluTrans/Roth Mini Dialyzer: MWCO 3.5 kD, 6-8 kD, 12-14 kD

Sample: 100µl of 1mM *p*-Nitrophenol in PBS, pH 7.4, n=3

Dialysis buffer: 1.8 ml of PBS, pH7.4, exchange interval 30 min.

Determination Method: Tecan Sunrise Photometer, 420 nm

	mM *p*-NP		
Time / min	**3.5 kD**	**6-8 kD**	**12-14 kD**
0	1.06	1.06	1.06
15	0.66	0.61	0.64
30	0.51	0.43	0.47
60	0.30	0.21	0.26
120	0.10	0.07	0.05



Application Note - Dialysis of Sucrose

ZelluTrans/Roth Mini Dialyzer

Materials and Methods

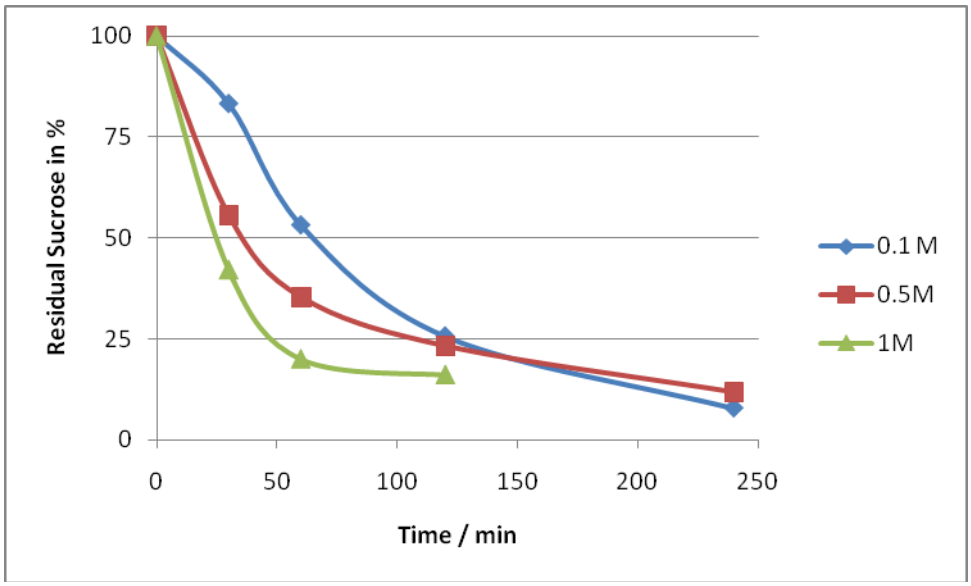
ZelluTrans/Roth Mini Dialyzer: MWCO 6-8 kDa (Art. No. 4769.1 ff)

Sample: 100µl of 1M, 0,5 M or 0,1 M Sucrose, respectively, in Aq. dest

Dialysis Buffer: 1.8 ml Aq. dest., Buffer exchange interval 30 min, 20 °C

Determination Method: Wescor Osmometer Vapor 5520

Time	Residual Sucrose in % of original concentration		
min	0.1 M	0.5M	1M
0	100.0	100.0	100.0
30	83.2	55.5	42.1
60	53.1	35.3	20.0
120	25.6	23.2	16.1
240	7.8	11.8	



Remark: With higher molarities of sucrose (e.g. 0,5 M and 1 M), an increasing osmotic effect appears resulting in dilution of the sample, which is simulating a higher speed of dialysis. To prevent this, it is best to keep the concentration difference around 100 mM. If this is not possible or desired, reduce the sample volume to 50 µl and close the ZelluTrans/Roth Mini Dialyzer with sealing for MD. For salt, this osmotic effect was not as pronounced as for sugars.

Application Note - CHAPS Removal

ZelluTrans/Roth Mini Dialyzer

Materials and Methods

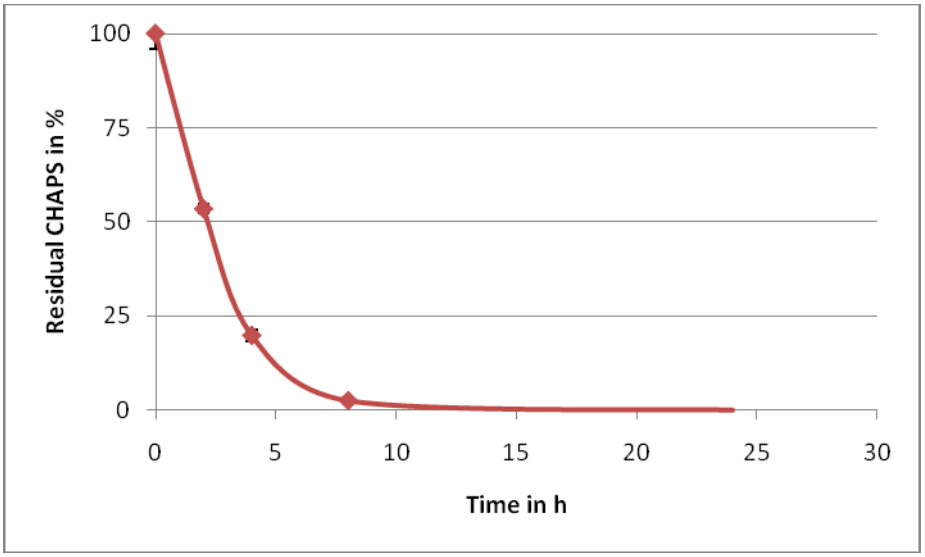
ZelluTrans/Roth Mini Dialyzer: MWCO 6-8 kDa (Art. No. 4769.1 ff)

Sample: 100µl of 1 mg/ml BSA in Aq. dest., 1% (w/v) CHAPS

Dialysis Buffer: 1.8 ml Aq. dest., Buffer exchange interval 2 h for 8 h, 20 °C

Determination Method: Agilent HP1100/MSD

t in h	Residual CHAPS in %
0	100
2	53,4
4	19,8
8	2,5
24	0,0



Application Note - SDS Removal

ZelluTrans/Roth Mini Dialyzer

Materials and Methods

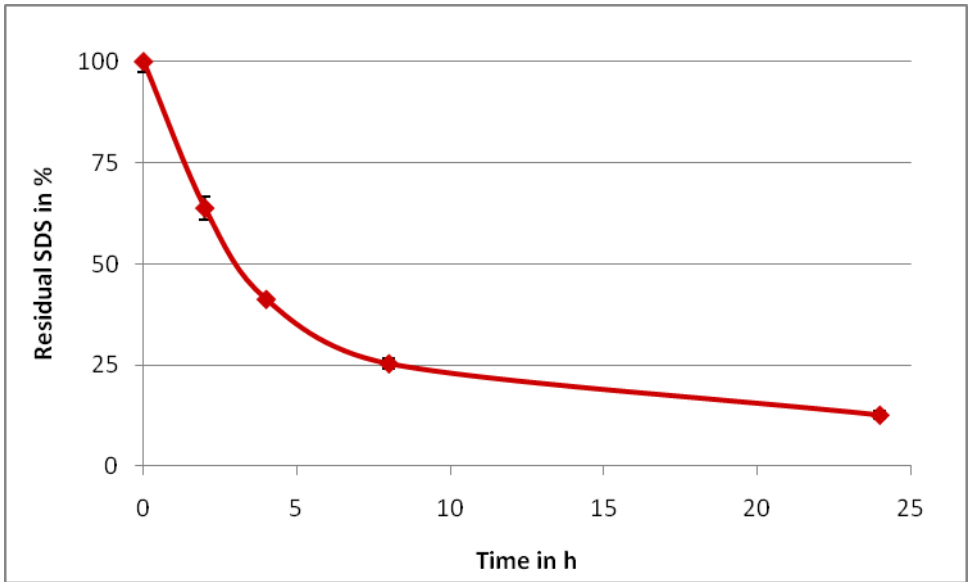
ZelluTrans/Roth Mini Dialyzer: MWCO 6-8 kDa (Art. No. 4769.1 ff)

Sample: 100µl of 1 mg/ml BSA and 0.1 % (w/v) SDS in PBS, pH 7.4

Dialysis Buffer: 1.8 ml Aq. dest., Buffer exchange interval 2 h for 8 h, 20 °C

Determination Method: Agilent HP1100/MSD, ESI negative mode

t in h	Residual SDS in %
0	100
2	63,8
4	41,2
8	25,3
24	12,6



Application Note - Volume reduction by evaporation

ZelluTrans/Roth Mini Dialyzer

Materials and Methods

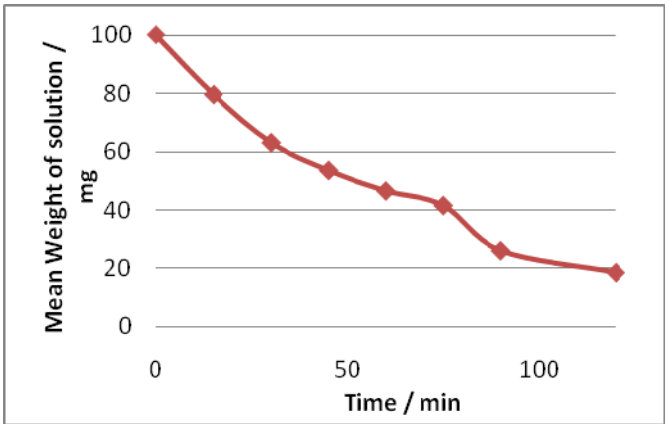
ZelluTrans/Roth Mini Dialyzer: MWCO 3,5 kDa (Art. No. 4765.1 ff)

Sample: 100 µl of PBS (Phosphate buffered saline: 10mM H_2NaPO_4/ HNa_2PO_4, 150mM NaCl, pH 7.4)

Determination Method: Gravimetric analysis of the weight of the residual solution in the Mini Dialyzer

Experimental: MD were filled with 100 µl PBS and placed in an “Egg Crate“ (from Corning) which was attached to a stand to allow air to pass the Mini Dialyzer. Ambient temperature, n=2.

Time / min	Weight of residual solution in Mini Dialyzer/ mg		
	Sample 1	Sample 2	**Mean**
0	100	100	**100**
15	81	78	**79.5**
30	64	62	**63**
45	55	52	**53.5**
60	46	47	**46.5**
75	38	45	**41.5**
90	30	22	**26**
120	26	11	**18.5**



Remark: Results may vary with temperature and air flow