

## User Guide

# MultiScreen® 96-well Ultrafiltration Plates with Ultracel®-10K Membrane

For use in the separation, concentration, desalting, preparation of proteins and other biomolecules.

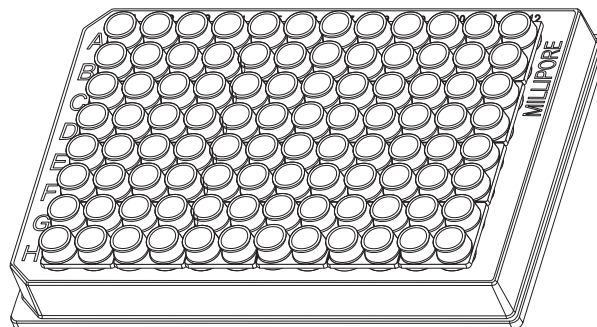
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## Introduction

The MultiScreen® 96-well Ultrafiltration Plate with Ultracel® membranes (MultiScreen® Ultrafiltration Plates) are used in the processing of aqueous biological solutions in volume ranges from 0.1 to 0.5 milliliters (mL). The ultrafiltration plate is used in a centrifugal pressure mode only and is compatible with standard centrifuge microtiter plate swinging bucket rotors. It is designed to fit a 96-well microtiter receiver plate for use in ultrafiltrate collections.

These plates have been developed and QC- tested specifically for protein removal prior to sample analysis and for sample purification, concentration and desalting of biological solutions. The low binding ultrafiltration membrane used in this product has 10000 Daltons (Da) nominal molecular weight limit (NMWL).

The MultiScreen® Ultrafiltration Plates can be used with conical 100 microliters ( $\mu\text{L}$ ), standard 300  $\mu\text{L}$  or 700  $\mu\text{L}$  well microtiter deep well receiver plates.



## Materials Required

The MultiScreen® Ultrafiltration Plates include microtiter plate storage lids. The user must supply the following items:

- Centrifuge capable of a minimum of 2000  $\times$  g-force with a swinging bucket rotor and 96-well plate carrier. Do not use this plate with vacuum filtration.
- Pipettors or robotic liquid handlers for transferring 500  $\mu\text{L}$  and 25  $\mu\text{L}$  and 10  $\mu\text{L}$  volumes.
- 96 well microtiter plate receiver tray. Some of the receiver plates that this MultiScreen® filter plate is compatible with are as follows:
  - Corning® 96-well EIA/RIA Clear Flat Bottom Polystyrene Not Treated Microplate (Cat. No. CLS9017)
  - Greiner 96-well plates, polypropylene, 300  $\mu\text{L}$ /well, V bottom clear well (Cat. No. M8185)
  - Greiner 96-well plates, polypropylene, 0.5 mL/well, clear (Cat. No. Z667234)
  - Greiner 96 well plates, polypropylene, with full skirt (Cat. No. Z711063)

For other receiver plates, compatibility should be confirmed prior to use.

## Specifications

Filter Plate Well Capacity	500 $\mu$ L
Maximum centrifugal force	4000 $\times$ g

### Filter Plate Dimensions

Length	127.8 $\pm$ 0.2 mm
Width	85.5 $\pm$ 0.2 mm
Height (Without Lid)	20.1 mm
Membrane Surface Area	32.4 mm <sup>2</sup>

### Material of Construction

Plate	Polypropylene with Silicone O-ring
Lid	Polystyrene

### Membrane

Ultracel <sup>®</sup> 10K Membrane	10000 NMWL regenerated cellulose
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## Limitations and Precautions

- Operate centrifuge within temperature range of 4 to 26 °C.
- The MultiScreen<sup>®</sup> Ultrafiltration Plates may exhibit a variance in flow properties with viscous, particle-laden samples such as serum. Depending on the centrifuge, wells on the outside edges (i.e., rows A, B, G, and H) of the filter plate will tend to filter slightly faster than those on the inside of the plate. This will NOT interfere with data analysis or assay results (see Table 1).
- This product is recommended for use in initial starting volumes of 100–500  $\mu$ L but is limited to the volume capacity of the 96-well receiver plate. For samples exceeding 300  $\mu$ L (up to 500  $\mu$ L), use of a deep well plate will ensure that the receiver plate will contain all the ultrafiltrate. For samples generating an ultrafiltrate less than 50  $\mu$ L the use of a small volume conical well plate is recommended.
- The ultrafiltrate volume generated can be increased by using a larger starting volume of sample or more centrifuge spin time. Use of a 500  $\mu$ L sample of serum or plasma will generate a larger ultrafiltrate volume (See Table 1).
- MultiScreen<sup>®</sup> Ultrafiltration Plates are designed for centrifugal pressure mode only. Do not use this plate with vacuum filtration.
- Centrifuge microtiter plate carriers (plate bucket) must be flat to provide uniform support during centrifugation. If the carrier base is not flat, performance may be compromised.

## Operation and Performance

The MultiScreen<sup>®</sup> Ultrafiltration Plate is operated in a centrifugal pressure mode at a recommended temperature of 4 to 26 °C and g force of 2000 to 4000  $\times$  g. When mounted on top of a microtiter receiver plate, the MultiScreen<sup>®</sup> Ultrafiltration Plate will fit into all standard microtiter plate swinging bucket rotors.

To minimize evaporation of sample prior to analysis and during centrifugation, the top of the MultiScreen<sup>®</sup> Ultrafiltration Plate should be covered with the lid.

## General Operating Procedure

1. Mount the MultiScreen<sup>®</sup> Ultrafiltration Plate on top of a standard microtiter plate (for sample volumes up to 300  $\mu$ L) or on top of a deep well plate (for sample volumes up to 500  $\mu$ L).
2. Load samples into the filter plate using a standard multichannel pipettor or liquid handling instrument.
3. Place the loaded filter plate assembly into the centrifugal plate carrier and centrifuge up to 4000  $\times$  g for desired spin time. Refer to Table 1 for additional information regarding centrifugation guidelines for different spin times and sample types. The wells of the MultiScreen<sup>®</sup> filter plate can handle up to 500  $\mu$ L of sample (requires deep well receiver plate).
4. To recover the sample ultrafiltrate, remove the MultiScreen<sup>®</sup> 96-well Ultrafiltration Plate with Ultracel<sup>®</sup> membranes from the microtiter receiver plate. The ultrafiltrate is ready for analysis or storage.
5. If applicable recover the retentate after ensuring it is well mixed (concentrated protein layer may form on the membrane during filtration). Avoid touching the membrane with the pipette tip as this will clog the pipettor or damage the membrane.

**NOTE:** Not all cases will result in retentate to be recovered.

**Table 1:**

Final Ultrafiltration Volume by receiver plate row for various sample type and times at 25 °C and 3000 x g with 300 µL sample.

Spin Time	Sample Type	Average Ultrafiltrate Volume by Row (µL)							
		A	B	C	D	E	F	G	H
15 min	BSA	267	269	273	271	275	273	271	270
	Buffer	288	290	291	292	295	291	290	288
	CmT	272	269	265	261	262	264	266	271
	FBS	80	64	47	32	33	48	66	83
30 min	BSA	256	260	264	262	266	264	263	261
	Buffer	280	285	286	287	290	288	286	285
	CmT	272	274	277	276	279	274	272	274
	FBS	86	72	55	41	43	57	74	90
45 min	BSA	248	253	257	255	259	257	255	254
	Buffer	273	280	282	282	286	284	283	281
	CmT	264	268	272	272	284	270	268	270
	FBS	101	86	68	53	55	71	90	107
60 min	BSA	241	247	252	250	254	252	251	250
	Buffer	267	275	278	279	283	281	280	278
	CmT	257	263	266	268	270	266	264	265
	FBS	105	91	73	59	61	76	94	111
90 min	BSA	230	238	243	242	247	245	243	242
	Buffer	255	266	270	272	277	275	273	272
	CmT	245	254	259	261	264	260	258	259
	FBS	110	97	80	67	69	83	100	116

BSA = Bovine Serum Albumin, Buffer = Phosphate Buffered Saline, CmT = Chymotrypsinogen, FBS = Fetal Bovine Serum

**Table 2:**

Typical Cross talk in MultiScreen® Ultrafiltration Plates

Location	1	2	3	4	5	6	7	8	9	10	11	12
<b>A</b>	0.000%		0.000%		0.000%		0.000%		0.001%		0.000%	
<b>B</b>		0.000%		0.000%		0.000%		0.000%		0.000%		0.000%
<b>C</b>	0.000%		0.000%		0.000%		0.000%		0.000%		0.000%	
<b>D</b>		0.000%		0.001%		0.000%		0.000%		0.000%		0.000%
<b>E</b>	0.000%		0.000%		0.000%		0.000%		0.000%		0.000%	
<b>F</b>		0.000%		0.000%		0.000%		0.000%		0.000%		0.000%
<b>G</b>	0.000%		0.000%		0.000%		0.000%		0.000%		0.000%	
<b>H</b>		0.000%		0.000%		0.000%		0.000%		0.000%		0.000%

Water, Yellow: 0.05% Fluorescein in water

**Table 3:**

Typical protein recovery at 3,000 x g for MultiScreen® Ultrafiltration Plate with Ultracel®-10K membrane

Solute / Concentration	Molecular Weight	Typical % Recovery from concentrate
Cytochrome C (0.25 mg/mL)	12.4 K	93.3 ± 5.8 %
A-Chymotrypsinogen (1 mg/mL)	25 K	97.9 ± 4.2 %
Bovine serum albumin (1 mg/mL)	67 K	97.9 ± 4.2 %
Bovine IgG Fraction II (1 mg/mL)	156 K	98.4 ± 0.8 %

## Chemical Compatibility

The solutions listed in the table below have been evaluated for chemical compatibility in the MultiScreen® 96-well Ultrafiltration Plate with Ultracel® membranes. Contact with some organic chemicals may cause leaching from component parts. If leaching is suspected, run solvent blanks before performing assays.

### CAUTION: Please note the following

1. These recommendations assume pure solutions at room temperature and pressure without applied stresses. Time of exposure is not considered. These are critical assumptions as polymer properties are strongly affected by environmental conditions, time, the presence of external stress and the presence of additives. It is not safe to assume that property changes are linearly related to changing temperature. A 10 °C increase in temperature, for example, may place the test conditions closer to the glass transition of the polymer, thus allowing greater penetration of solvent molecules. This has a plasticizing effect, further lowering the glass transition and resulting in a modulus drop of up to three orders of magnitude.
2. These recommendations assume that each polymer category has a uniform chemistry, molecular weight distribution and thermomechanical history. This assumption will never be true and, in some cases, variation has a distinct influence on compatibility. Crystalline morphology and degree of crystallinity influences compatibility of semicrystalline polymers and can vary significantly. Such specific information concerning polymers evaluated does not accompany published compatibility tables.
3. The definition of solvent compatibility for our products differs from that used in determining the ratings given in published compatibility tables. Such tables are generally concerned with chemical attack and significant losses in strength and / or dimensional changes. A top designation, for example, might be designated for solvent polymer combinations with <10% swelling, which is high. Other compatibility tables may make recommendations based upon dimensional change as a function of time. This is difficult to relate to a membrane that may respond almost immediately to immersion in solvent. In addition, solvent-membrane compatibility requires additional consideration of filtration-specific factors. None of these published compatibility guides, for example, monitors the solvent's ability to wet a membrane or increase extractables.
4. This table does not consider solvent safety issues.

**Table 4:**

Chemical compatibility of the MultiScreen® 96-well Ultrafiltration Plate with Ultracel® membrane.

<b>Acids</b>	<b>Concentration</b>		<b>Concentration</b>
Acetic acid	≤ 50%*	Phosphoric acid	≤ 30%
Formic acid	≤ 5%*	Sulfamic acid	≤ 3%
Hydrochloric acid	≤ 1.0 M	Sulfuric acid	≤ 3%
Lactic acid	≤ 50%	Trichloroacetic acid (TCA)	≤ 10%*
Nitric acid	≤ 10%	Trifluoroacetic acid (TFA)	≤ 30%*
<b>Alkalis</b>			
Ammonium hydroxide	≤ 10%	Sodium hydroxide	≤ 0.5 M
<b>Alcohols</b>			
n-Butanol	≤ 70%	Isopropanol	≤ 70%
Ethanol	≤ 70%	Methanol	≤ 60%
<b>Detergents</b>			
Alconox® detergent	≤ 1%	Lubrol® PX detergent	≤ 0.1%
CHAPS detergent	≤ 0.1%	Nonidet™ P-40 surfactant	≤ 2%
Sodium deoxycholate	≤ 5%	Triton® X-100 surfactant	≤ 0.1%
Sodium dodecyl sulfate (SDS)	≤ 0.1%	Tween® 20 surfactant	≤ 0.1%
Tergazyme® detergent	≤ 1%		
<b>Organic solvents</b>			
Acetone	not recommended	Ethyl acetate	not recommended
Acetonitrile	≤ 20%	Formaldehyde	≤ 5%
Benzene	not recommended	Pyridine	not recommended
Carbon tetrachloride	not recommended	Tetrahydrofuran	not recommended
Chloroform	not recommended	Toluene	not recommended
Dimethyl sulfoxide (DMSO)	≤ 5%*		
<b>Miscellaneous</b>			
Ammonium sulfate	Saturated	Phenol	≤ 1%
Diethyl pyrocarbonate	≤ 0.2%	Phosphate buffer (pH 8.2)	≤ 1 M
Dithiothreitol (DTT)	≤ 0.1 M	Polyethylene glycol	≤ 10%
Glycerine	≤ 70%	Sodium carbonate	≤ 20%
Guanidine HCl	≤ 6 M	Tris buffer (pH 8.2)	≤ 1 M
Imidazole	≤ 100 mM	Urea	≤ 8 M
Mercaptoethanol	≤ 0.1 M		

\* Contact with this chemical may cause materials to leach out of the component parts. Solvent blanks are recommended to determine whether leachables represent potential assay interferences.

## Product Ordering

These products can be purchased on-line at [SigmaAldrich.com/products](https://SigmaAldrich.com/products).

Description	Qty / Pk	Catalogue Number
MultiScreen® 96-well Ultrafiltration Plate with Ultracel® 10 K membrane	5/Pk	MAUF01005

### Accessories

Corning® 96-well EIA/RIA Clear Flat Bottom Polystyrene Not Treated Microplate	25/pk	CLS9017
Greiner 96-well plates, polypropylene, 300 µL / well, V bottom clear well	100/pk	M8185
Greiner 96-well plates, polypropylene, 0.5 mL/well, clear	80/pk	Z667234
Greiner 96-well plates, polypropylene, with full skirt	100 pk	Z711063

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