PALL Laboratory

Application Note

AcroPrep™ 24-well Filter Plates with Various Pore Size for Ultrafiltration, Microfiltration and Macrofiltration

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Summary

Many techniques in Life Science research rely on efficient sample preparation methods. Concentration and purification of samples are key steps in the workflow that can greatly affect the outcome of the experimental results. Pall has increased the range of AcroPrep 24-well filter plates offered to help streamline membrane selection and provide effective solutions for multiple sample preparation needs. The new AcroPrep 24-well filter plates include membranes for ultrafiltration (molecular weight cut off ranging from 1 to 100 K), microfiltration (pore sizes ranging from 0.1 to 10 µm) and macrofiltration/particle filtration (30 – 40 µm non-woven media).

Introduction

Filtration is a standard process used in almost every lab for a wide variety of purposes. There are various types of filtration including ultrafiltration, microfiltration, and particle-macrofiltration (Table 1). Which type of filtration to use is dependent upon the goal to be achieved.

Table 1

Types of filtration media in the AcroPrep 24-well filter plate format.

Membrane Type	Pore Size (microns = µm)	Molecular Weight (Daltons = Da) Approximate	Material Removal
Ultrafiltration	0.002 – 0.1	10,000 – 100,000	Macro-molecules: colloids (proteins, carbohydrates polymers), some viruses,…
Microfiltration	0.1 – 10	> 100,000	Bacteria, …
Particle filtration (Macrofiltration)	10 – 1,000	N/A	Suspended particles, mammalian cells, bacteria, yeast …

The clarification, pre-filtration, and sterilization of samples remain an important function for a multitude of life sciences research applications. Microfiltration is a broad category of separation that ranges in pore size from 0.1 – 10 µm. There are two classic types of microfiltration processes that can be utilized in the sample preparation process depending on application requirements: depth filtration (remove particulates of varying sizes and high 'dirt' holding capacity), and membrane filtration (like sieves they retain all particles larger than the precisely controlled pore size on top of or within their structure). An advantage of membrane filtration is the membrane is bacteria and particle retentive (See Pall's 0.2 µm sterile filtration AcroPrep 24-well filter plate) with lower extractables than depth filtration media.

Depth and membrane filtration can be used in concert to generate a combination filter which is comprised of membranes or media of different pore sizes or varying materials to create an inline prefiltration/final filtration device (e.g. the Pall AcroPrep 24-well cell clarification and sterile filtration plate).

The concentration of dilute biomolecule solutions is common practice in research laboratories. The concentration of biomolecules is commonly performed via ultrafiltration through a size-exclusion mechanism typically rated by the molecular weight of the particles to remove, around 1,000 to 1,000,000 molecular weight cut-off (MWCO). Molecules larger than the membrane 'pores' will be retained by the membrane and concentrated during the ultrafiltration process. The biomolecules remain on the surface of the membrane and do not enter the polymer matrix. This attribute allows for > 90% recovery of target molecules which minimizes concern over non-specific binding of the target molecule. The use of ultrafiltration membranes does not shear nucleic acids, alter enzymatic activity, or cause up/down regulation of the protein. If the downstream application requires the removal of salts and /or detergents, ultrafiltration provides a convenient and efficient mechanism to change the ionic or pH environment.

Filter efficiency measures the percentage of particles that are removed from the fluid by the membrane. Large pore size filter materials are used to filter solutions prior to more detailed analysis. When selecting the best product for the application, several factors need to be considered such as sample volume, sample recovery, and the expected size of target to remove.

This document reports the data generated by ultrafiltration of proteins of known size with AcroPrep 24-well filter plates. Microfiltration and macrofiltration using the AcroPrep 24-well filter plates with relevant pore size were examined using calibrated latex beads mimicking relevant material diameter.

1. Material and Method

1.1 Consumables / Equipment

1.1.1 Pall Products

- AcroPrep 24-well filter plates with Omega™ membrane for ultrafiltration: 1, 3, 10, 30, 50, 100 K.
- AcroPrep 24-well filter plates with PES Supor® membrane for microfiltration: 0.1, 0.45, 0.8, 1.2, 5 µm.
- AcroPrep 24-well filter plates with PP/PE membrane for macrofiltration / Particles filtration: 30 – 40 µm.

Process		UltraFiltration					
Membrane		Omega					
Pore size		kDa					
		1	3	10	30	50	100
Pall Part No.	8PK	97049	97051	97053	97055	97057	97059
	2PK	97050	97052	97054	97056	97058	97060
Fisher Scientific Part No.	8PK	17399481	17319491	17339491	17359491	17379491	17399491
	2PK	17309491	17329491	17349491	17369491	17389491	17309501

** PES = Hydrophilic Polyethersulfone, **PP/PE = Polypropylene/Polyethylene.*

Process		MicroFiltration					Macro-Filtration
Membrane		Supor PES*					PP/PE**
Pore size		µm					µm
		0.1	0.45	0.8	1.2	5	30-40
Pall Part No.	8PK	97029	97031	97033	97035	97047	97061
	2PK	97030	97032	97034	97036	97048	97062
Fisher Scientific Part No.	8PK	97029	97031	97033	97035	97047	97061
	2PK	97030	97032	97034	97036	97048	97062

1.1.2 Consumables

• Latex beads solutions

The latex polystyrene beads and Opti-Bind◆ polystyrene sulfate particles were supplied by Sigma or Thermo Fisher Scientific, respectively, in 5 or 10 % (w/v) aqueous solution.

The polystyrene-based microparticles are colloidal particles, with calibrated diameters determined accurately by the supplier.

• Proteins used for testing solutions

Product	Brand	Product Number
PBS	Fisher Scientific	BP2944-100
Vitamin B12	Alfa Aesar	A14894
Blue Dextran (5kDa)	Sigma	90008-1G
Insulin	SAFC	91077C-1g
Cytochrome C	MP	101467
Lysozyme	VWR	0663-10G
Myoglobulin	Sigma	M06301G
Blue Dextran (20kDa)	Sigma	03714-1/G
Trypsin	Alfa Aesar	J63688
Ovalbumin	MP	950512
Bovine Serum Albumin	VWR	422381B
Transferrin	Sigma	90191 100MG
γ-globulins	Serva	22550.01
Catalase	Sigma	SRE0041-10G
Blue Dextran (2000 kDa)	Sigma	D5751-10G

• Various consumables

Product	Brand	Product Number
Disposable cuvettes for Spectrophotometer	VWR	612-5686
UV Microplates	Thermo Fisher Scientific	8404
NuPAGE◆ LDS Sample Buffer	Thermo Fisher Scientific	NP0007
SeeBlue◆ Plus2 Pre-stained protein standard	Thermo Fisher Scientific	LC5925
NuPAGE Novex 4-12% Bis-Tris	Thermo Fisher Scientific	NP0321BOX
Protein Stain Blue BANDit◆	VWR	K217-1L
NuPAGE MOPS SDS Running buffer	Thermo Fisher Scientific	NP0001

1.1.3 Equipment

- Centrifuge 5810, Eppendorf & A-2-DWP, A-2-DWP-AT rotors
- Multi-well Plate Vacuum Manifold, Pall PN: 5017
- Positive Pressure: 96, Waters◆
- Plate Reader Infinite◆ M200, Tecan
- Spectrophotometer, Biowave II, WPA
- pH/Conductivity meter, S470 SevenExcellence◆, Mettler Toledo

1.2 Methods

1.2.1 Filtration Condition

Table 1

Filtration condition used with the 24-well plates

	Ultrafiltration	Microfiltration	Macrofiltration
Membrane pore size	1, 3, 10, 30, 50, 100 K	0.1, 0.45, 0.8, 1.2, 5 µm	≈ 35 µm
Solution for retention study	Protein	Latex beads	Latex beads
Volume per well (for time-through & hold-up volume)	4 mL	3 mL	3 mL
Centrifugation	1,500 x g	1,500 x g	1,500 x g
Vacuum	15 in Hg	15 in Hg	< 5 in Hg
Positive pressure	50 psi	20 psi	20 psi

1.2.2 Water Pre-Rinse

pH, conductivity, and UV extractables were determined using 2 mL Reverse Osmosis (R.O.) water to rinse each plate, before and after filtration.

- UV extractables: 200 µL of the downstream/filtered water were loaded onto a UV microplate, in parallel with 6 x RO water (before filtration) as blanks. The UV microplate was scanned between 230-400 nm, to assess the presence of any UV extractables and compared to the UV profile of the RO water before filtration.
- pH / Conductivity: The downstream/filtered water was pooled into a centrifuge tube, which was used to measure the pH and conductivity of the downstream solution. The pH and conductivity of the initial RO water were also measured prior to filtration for comparison.

1.2.3 Hold-up Volume / Processing Time

The hold-up volume, or the volume of liquid retained in the filters, was determined by recording the volume (/weight) of the filtered samples in the collection plate (downstream) after filtration was compared to the volume (/weight) of the solution in the filter plate (upstream) before filtration. The difference represents the hold-up volume.

The time through, or filtration time, was determine by observing the time required for a solution of latex beads (3 mL) or protein solution (4 mL) to be filtered through the wells of the plate.

For centrifugation, protein solutions at 1 g/L were used while 0.1 g/L was used with vacuum and positive pressure.

1.2.4 Latex Beads Retention Test (Micro- / Macrofiltration)

Three mL aliquots of a latex bead solution (0.05 % in 0.01 % Tween20) were loaded into the wells of the AcroPrep 24-well filter plate. Each plate was used on top of a collection plate and processed either in centrifugation, vacuum, or positive pressure set up according to Table 1, until all the upstream solution had filtered through. The samples (200 µL) were loaded in a UV microplate and the absorbance was measured in a plate reader at 260 nm, which is the optimum wavelength for the latex bead solutions. The percentage of latex beads retained on the filters (retention) was obtained by comparing the corrected absorbances of the solution before (upstream) filtration and in the filtrate after filtration (downstream).

Because of the quick precipitation of the 100 µm diameter latex beads, the absorbances could not be recorded accurately. The retention for each filtrate was estimated by visual comparison to a standard curve for those beads.

1.2.5 Protein Retention Test (Ultrafiltration)

The protein solutions were prepared in 1X PBS buffer. The protein concentration was adjusted to 1 mg/mL (≈ 1 mAU) using the Biowave II spectrophotometer, after blanking the instrument with 1X PBS buffer. The 0.1 mg/mL protein solutions were prepared by a 1:10 dilution of the 1 mg/mL solution with PBS (1X).

Protein retention for the ultrafiltration plates was calculated by loading the 1 mg/mL protein solution onto the filter plate before processing by centrifugation, vacuum, or positive pressure according to the condition described in Table 1. The absorbance for each sample was recorded at 280 nm and corrected using 1X PBS. The percentage of protein retained on the filter plate (= retention) was obtained by comparing the corrected absorbances of the solution before (upstream) filtration and the filtrate in the collection plate after filtration (downstream).

1.2.6 SDS-PAGE

The filter plates containing 30 and 50 K Omega membranes were loaded with 0.3-1 mL solutions of protein. The initial concentration of the protein was 1 g/L and diluted accordingly to have clear bands on an SDS-page.

Individual wells of a 30 K 24-well filter plate were loaded with a solution of Myoglobulin (17.8 kDa), BSA (66 kDa), transferrin (79.5 kDa), IgG (150 kDa), and a mixture of 3 of them. The plates were centrifuged at 1,500 x g for 10 min. The concentrated protein (upstream) and the filtrate (downstream) were diluted and mixed with loading buffer before being heated up 10 min at 70 °C. Then, 20 µL of heated samples were loaded onto a 4 – 12 % Bis-Tris SDS-page with 1X NuPAGE MOPS SDS running buffer.

The gels were then stained with the Blue BANDit until the appearance of the bands and followed by water rinse.

2. Results and Discussion

2.1 Micro- / Macrofiltration plates: 0.1, 0.45, 0.8, 1.2, 5, 30 – 40 µm

The data generated for the AcroPrep 24-well filter plate in micro- and macro filtration were used to assess the integrity of the plates under three rounds of filtration.

Data in Table 2 shows the pH, conductivity, and UV extractables of RO water before and after filtration, for the various pore size plates.

Table 2

Average of the pH, conductivity, and UV extractables determined from a water pre-rinse of the 0.1, 0.45, 0.8, 1.2 and 5 µm AcroPrep 24-well filter plates

		Membrane Pore Size (µm)					
		0.1	**0.45**	**0.8**	**1.2**	**5**	**30 – 40**
pH	*Initial*	*6.2*	*6.2*	*6.3*	*6.3*	*6.2*	*6.3*
	Centrifuge	6.6	6.3	6.3	6.4	6.7	6.2
	Vacuum	6.3	6.2	6.3	6.3	6.5	6.4
	Positive Pressure	6.1	6.2	6.3	6.4	6.6	6.3
Conductivity (µS/cm)	*Initial*	*15.0*	*16.0*	*16.4*	*15.5*	*12.0*	*12.6*
	Centrifuge	15.9	16.5	16.4	14.1	13.2	13
	Vacuum	15.9	16.8	17.2	14.1	12.4	13.8
	Positive Pressure	15.8	16.3	16.3	14.3	12.6	12.5
UV Extractables	Centrifuge	No Significant differences in absorbance between water samples before and after filtration in UV range of 240 – 400 nm					
	Vacuum						
	Positive Pressure						

The data showed that the pH and conductivity did not vary from the initial values recorded before filtration, leading to the conclusion that no molecules influencing the pH and conductivity were being released from the plate.

This conclusion was confirmed by the absence of significant absorbance in the UV range for RO water after filtration through the AcroPrep 24-well filter plates when compared with the pre-filtration samples.

Data in Table 3 shows the average of the hold-up volume and time-through of 3 mL latex beads solution for the various plates.

Table 3

Average of the hold-up volume and time-through for 3 mL per well of latex beads solution (0.05 %) prepared in Tween 20 (0.01 %) for the 0.1, 0.45, 0.8, 1.2 and 5 µm 24-well filter plates

		Membrane Pore Size (µm)					
		0.1	**0.45**	**0.8**	**1.2**	**5**	**30 – 40**
Hold-up volume (µL)	Centrifuge	8.2	45.7	63.0	8.5	26.8	5.5
	Vacuum	12.0	29.3	77.9	11.6	44.2	13.7
	Positive pressure	15.4	35.1	30.3	6.8	24.3	4.3
Time through (mins)	Centrifuge	12	≤ 10	≤ 10	≤ 10	≤ 10	≤ 10
	Vacuum	2	< 1	< 1	< 1	< 1	< 1
	Positive pressure	3	5	2	< 1	< 1	< 1

The data showed that the hold-up volume varies in a non-linear way, according to the plate pore size and the filtration process used for the AcroPrep 24-well filter plates, with less than 63, 78 and 35 µL in centrifuge, vacuum and positive pressure, respectively.

In general, the time-through for all plates took less than 10 minutes to filter 3 mL of latex bead solution.

Data in Table 4 shows the average retention of various latex polystyrene beads in the AcroPrep 24-well filter plates. The polystyrene latex beads are calibrated and were used to mimic molecules of similar size.

Table 4

Latex bead retention from 3 mL per well of latex beads solution (0.05 %) prepared in Tween 20 (0.01 %) for the 0.1, 0.45, 0.8, 1.2 and 5 µm AcroPrep 24-well filter plates.

		Latex Bead Retention (%)		
Membrane Pore Size Plate	**Latex Bead Size (µm)**	**Centrifuge (1,500 x g)**	**Vacuum (15 inHg)**	**Positive Pressure (20 psi)**
0.1 µm	0.25	100.0 ± 0.1	100.1 ± 0.1	100.1 ± 0.1
	0.3	99.9 ± 0.2	100.0 ± 0.2	100.0 ± 0.2
	0.4	99.9 ± 0.1	100.0 ± 0.2	100.0 ± 0.1
0.45 µm	0.6	99.7 ± 0.1	99.5 ± 0.2	99.5 ± 0.3
	0.8	99.0 ± 0.1	99.9 ± 0.4	99.5 ± 0.3
	1.1	99.4 ± 0.3	99.2 ± 0.4	99.0 ± 0.7
0.8 µm	1.22	99.3 ± 0.3	99.1 ± 0.3	98.6 ± 0.3
	2	99.2 ± 0.4	98.6 ± 0.5	98.4 ± 0.8
	2.5	98.1 ± 0.9	98.7 ± 1.3	98.2 ± 0.9
	3	97.5 ± 0.7	99.8 ± 0.6	96.1 ± 0.8
1.2 µm	2	99.8 ± 0.3	99.5 ± 0.6	98.8 ± 0.9
	3	99.5 ± 0.8	98.7 ± 0.8	97.7 ± 1.2
	5	99.3 ± 1.1	98.9 ± 1.4	94.1 ± 3.2
5 µm	5	99.4 ± 1.0	96.7 ± 2.7	94.7 ± 3.3
	7	100.0 ± 0.1	97.6 ± 2.3	96.6 ± 3.2
30-40 µm	100	Visual assessment		
		Centrifuge (1,500 x g)	**Vacuum (< 15 inHg)**	**Positive Pressure (20 psi)**
		100.0	99.8 ± 0.2	99.9 ± 0.2

Latex beads solutions used to challenge the plates ranged from 1.5 to 5 times the membrane pore size. The data illustrates that all plates could retain more than 94% of latex beads with similar efficiency for centrifuge, vacuum, and positive pressure.

2.2. Ultrafiltration Plates: 1, 3, 10, 30, 50, 100 kDa

The data generated for the AcroPrep 24-well filter plate with an ultrafiltration (Omega) membrane were used to assess the integrity of the plates under centrifugation, vacuum, and positive pressure.

Table 5 shows the pH, conductivity, and UV extractables of RO water before and after filtration, for the various ultrafiltration plates.

Table 5

Average of the pH, conductivity and UV extractables determined from a RO water pre-rinse of the 1, 3, 10, 30, 50 & 100 K 24-well filter plates.

		Membrane Pore size (K)					
		1	3	10	30	50	100
pH	Initial	6.3	6.3	6.3	6.2	6.3	6.3
	Centrifuge	6.6	6.9	6.7	6.6	6.8	6.7
	Vacuum	6.7	6.7	6.7	6.7	6.7	6.7
	Positive pressure	6.9	6.8	6.7	6.6	6.7	6.6
Conductivity (µS/cm)	Initial	16.0	16.0	16.4	15.9	16.3	17.9
	Centrifuge	36.9	29.7	24.3	36.2	30.6	30.8
	Vacuum	55.2	42.7	28.4	41.7	30.0	33.8
	Positive pressure	37.0	37.4	28.2	37.2	31.3	32.4
UV extractables	Centrifuge	No Significant differences in absorbance for R.O. water before and after filtration, between 240 & 400nm					
	Vacuum						
	Positive pressure						

Table 5 shows a slight increase in pH and conductivity between the initial and filtered water. The absence of compound absorbing UV between 240 – 400 nm lead to the conclusion that this slight variability in pH and conductivity may not be related to molecules released by the AcroPrep 24-well filters plate.

Table 6 shows the time-through and hold-up volume of the various pore size plates for filtration of protein solution.

Table 6

Average of the hold-up volume and time-through for 4 mL per well of protein/dextran solution in PBS at 1 g/L or 0.1 g/L for centrifugation or vacuum/positive pressure, respectively.

		Membrane Pore Size (K)					
		1	3	10	30	50	100
Hold Up Volume (µL)	Centrifuge	19.3	9.8	26.5	24.6	12.8	7
	Vacuum	14	21.2	74.9	14.6	28.8	28.5
	Positive pressure	7.7	15.6	2.8	28.1	70.9	59.3
Time through (min)	Centrifuge	170	135	70	60	60	100
	Vacuum	165	135	85	60	60	30
	Positive pressure	95	70	45	50	55	25

The data showed that the hold-up volume varies in a non-linear way related to the membrane pore size of the plate. All plates showed a hold-up volume of less than 75 µL per well, when filled with 4 mL of protein solution per well.

The time taken for the plates to filter 4 mL of solution per well was recorded for each process. The proteins/dextran were around 3 to 10 times the size of the plates pore size. The data in Table 6 shows the smaller membrane cut-offs plate can take longer to filter 4 mL of solution per well than the larger cut-offs.

Table 7 shows the average retention of various molecules (protein, dextran, vitamin B12) based on their known molecular weight, for the AcroPrep 24-well filter plates with different membrane pore size/molecular weight cut-off (MWCO). Solutions used to challenge the ultrafiltration plates ranged from 1 to 20 times the plate MWCO.

Table 7

% Retention of 1g/L solution (protein, dextran, vitamin B12) for 1, 3, 10, 30, 50, 100 K MWCO 24-well filter plates. In centrifugation (A), vacuum (B), positive pressure (C).

A) Centrifugation (at 1,500 x g)

	Protein Size (kDa)	Membrane MWCO (K)					
		1	3	10	30	50	100
Vitamin B12	1.4	47.0 ± 1.5	49.2 ± 0.9				
Blue Dextran	5	96.2 ± 7.0	96.9 ± 1.6				
Cytochrome C	12.5	92.2 ± 1.2	93.7 ± 2.2	13.9 ± 7.1			
Blue Dextran	20			99.5 ± 0.5	98.3 ± 0.3		
Ovalbumin	45			96.3 ± 0.5	92.6 ± 0.6	44.9 ± 3.8	
BSA	66			98.0 ± 2.0	98.0 ± 0.3	91.7 ± 3.1	
Transferrin	79.5				92.6 ± 1.4	94.2 ± 2.4	27.3 ± 6.9
IgG	150						96.1 ± 1.1
γ-globulin	160				97.1 ± 1.7	96.5 ± 4.1	93.0 ± 1.4
Blue Dextran	2,000						97.1 ± 1.7

B) Vacuum (at 15 inHg)

	Protein Size (kDa)	Membrane MWCO (K)					
		1	3	10	30	50	100
Vitamin B12	1.4	53.1 ± 2.0	39.4 ± 5.3				
Blue Dextran	5	98.0 ± 2.4	93.6 ± 4.1				
Cytochrome C	12.5	93.6 ± 1.8	91.1 ± 1.7	17.0 ± 2.5			
Blue Dextran	20			99.9 ± 0.1	97.5 ± 0.8		
Ovalbumin	45			91.2 ± 2.1	78.1 ± 0.9	14.6 ± 3.2	
BSA	66			92.1 ± 6.6	96.2 ± 0.5	81.2 ± 4.9	< 4.1 %
Transferrin	79.5				92.6 ± 1.4	94.5 ± 1.5	< 14.8 %
γ-globulin	160				96.4 ± 0.9	98.1 ± 0.8	87.5 ± 6.5
Blue Dextran	2,000						96.4 ± 3.0

C) Positive pressure (at 50 Psi)

	Protein Size (kDa)	Membrane MWCO (K) 1	3	10	30	50	100
Vitamin B12	1.4	53.8 ± 0.6	37.5 ± 5.2				
Blue Dextran	5	98.6 ± 1.4	88.5 ± 8.7				
Cytochrome C	12.5	90.1 ± 0.7	90.8 ± 1.8	14.4 ± 3.9	7.5 ± 0.9		
Blue Dextran	20			98.4 ± 2.9	97.2 ± 2.1		
Ovalbumin	45			79.4 ± 15.3	71.2 ± 2.8	4.5 ± 1.8	
BSA	66			95.8 ± 3.2	94.9 ± 1.1	70.4 ± 11.7	< 8.3
Transferrin	79.5				86.7 ± 2.8	94.5 ± 1.5	< 15.2
γ-globulin	160				95.1 ± 1.7	96.1 ± 3.2	91.1 ± 3.2
Blue Dextran	2,000						96.1 ± 2.4

Table 7 shows that the molecules retained by the AcroPrep 24-well filter plates must be at least 1.5 to 2 times the plates MWCO for acceptable molecule retention under centrifugation, vacuum, or positive pressure. The nature of the molecule must be taken into account as a molecular weight given in Table 7 is for monomer protein. Oligomerisation may occur and affect the proteins retention.

Moreover, the data show that the smallest molecules/proteins tested were not retained as well as the biggest ones, making the plates advantageous for cleaning up small molecules/proteins from a mixture. This point was validated by using individual and mixtures of proteins before and after plate filtration and the retentate and filtrate by SDS-page (Figure 1) and assessing the retention (Table 8).

Table 8

Protein retention (%) from absorbances at 280 nm (A) and visualization into SDS-page (B) of protein solutions before and after filtration using centrifugation and 30 or 50 K 24-well plates.

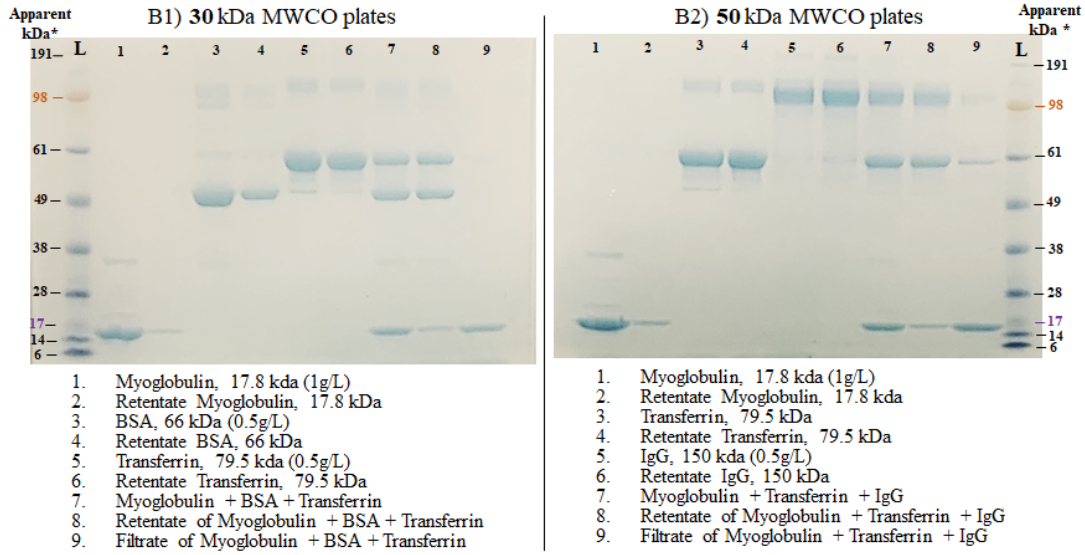
A) Protein retention

30 K MWCO			50 K MWCO		
Protein	**MW (kDa)**	**Retention (%)**	**Protein**	**MW (kDa)**	**Retention (%)**
Myoglobulin	17.8	≈ 0	Myoglobulin	17.8	≈ 0
BSA	66	98.0 ± 0.3	Transferrin	79.5	94.2 ± 2.4
Transferrin	79.5	92.6 ± 1.4	IgG	150	96.1 ± 1.1

B) SDS-page

Figure 1

SDS-PAGE of protein solutions before and after filtration through 30 K MWCO plates (B1) and 50 K MWCO (B2) plates. All retentates were resuspended in PBS buffer, accordingly.



1. Myoglobulin, 17.8 kda (1g/L)
2. Retentate Myoglobulin, 17.8 kDa
3. BSA, 66 kDa (0.5g/L)
4. Retentate BSA, 66 kDa
5. Transferrin, 79.5 kda (0.5g/L)
6. Retentate Transferrin, 79.5 kDa
7. Myoglobulin + BSA + Transferrin
8. Retentate of Myoglobulin + BSA + Transferrin
9. Filtrate of Myoglobulin + BSA + Transferrin

1. Myoglobulin, 17.8 kda (1g/L)
2. Retentate Myoglobulin, 17.8 kda
3. Transferrin, 79.5 kDa
4. Retentate Transferrin, 79.5 kDa
5. IgG, 150 kda (0.5g/L)
6. Retentate IgG, 150 kDa
7. Myoglobulin + Transferrin + IgG
8. Retentate of Myoglobulin + Transferrin + IgG
9. Filtrate of Myoglobulin + Transferrin + IgG

** Apparent molecular weight values vary depending upon electrophoresis gel/buffer system.*

3. Conclusion

Pall has extended the range of AcroPrep 24-well filter plates to allow customers to perform ultra-, micro- and macro filtration. The new range of AcroPrep 24-well filter plates have been characterized with solutions meant to assess the efficiency of the plates for screening and sample recovery. The broad range of membrane pore sizes allow concentration of samples as well as clean-up of solution by removal of small molecules in a 24-well filter plate format for sample screening.

The molecule of interest should be 3 – 6 times larger than the MWCO of the AcroPrep 24-well filter plates to ensure good retention/concentration of the molecule of interest. The purity and components of the solution should be taken into account as those can affect filtration and final sample quality.

The combination of several pore size plate can help streamline the sample preparation workflow and improve the purity of the molecule of interest.

4. References

- Media guide: Membranes and separation materials for diagnostics, research, and healthcare applications – Pall Life Sciences – www.pall.com/lab

10/21, PDF, GN21.1005