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Amicon Ultra-4 Centrifugal Filter Devices

for volumes up to 4 mL

Amicon® Ultra-4 3K, 10K, 30K, 50K, and 100K devices for research use only; not for use in diagnostic procedures

Introduction

Amicon® Ultra-4 centrifugal filter devices provide fast ultrafiltration, with the capability for high concentration factors and easy concentrate recovery from dilute and complex sample matrices. The vertical design and available membrane surface area provide fast sample processing, high sample recovery (typically greater than 90% of dilute starting solution), and the capability for 80-fold concentration. Typical processing time is 10 to 40 minutes depending on Molecular Weight Cutoff (MWCO). Solute polarization and subsequent fouling of the membrane are minimized by the vertical design, and a physical deadstop in the filter device prevents spinning to dryness and potential sample loss. The concentrate is collected from the filter device sample reservoir using a pipettor, while the ultrafiltrate is collected in the provided centrifuge tube. The device can be spun in a swinging-bucket (for optimal performance) or fixed-angle rotor. Amicon® Ultra-4 devices are supplied nonsterile and are for single use only.

The Amicon® Ultra-4 product line includes 5 different cutoffs (MWCO):

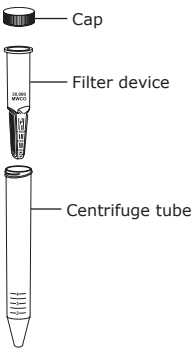
- Amicon® Ultra 3K device — 3,000 MWCO
- Amicon® Ultra 10K device — 10,000 MWCO
- Amicon® Ultra 30K device — 30,000 MWCO
- Amicon® Ultra 50K device — 50,000 MWCO
- Amicon® Ultra 100K device — 100,000 MWCO

Applications

- Concentration of biological samples containing antigens, antibodies, enzymes, nucleic acids (DNA/RNA samples, either single- or double-stranded), microorganisms, column eluates, and purified samples
- Purification of macromolecular components found in tissue culture extracts and cell lysates, removal of primer, linkers, or molecular labels from a reaction mix, and protein removal prior to HPLC
- Desalting, buffer exchange, or diafiltration

Materials Supplied

The Amicon® Ultra-4 device is supplied with a cap, a filter device, and a centrifuge tube.



Required Equipment

- Centrifuge with swinging-bucket (preferred) or fixed-angle rotor with wells/carriers that can accommodate 17 mm × 124 mm 15 mL conical-bottomed tubes

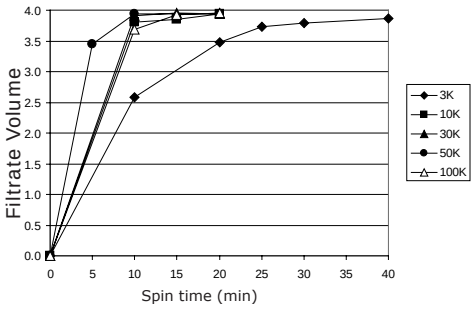
  CAUTION: To avoid damage to the device during centrifugation, check clearance before spinning.
- Pipettor with 200 microliter (µL) tip for concentrate recovery

Performance - Protein Concentration

Flow Rate

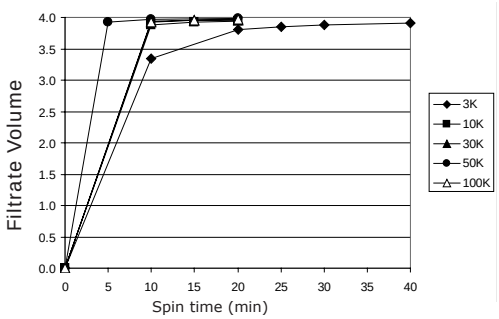
Factors affecting flow rate include sample concentration, starting volume, chemical nature of solute, relative centrifugal force, centrifuge rotor angle, membrane type, and temperature. Figures 1 and 2, and Tables 2 and 3 can be used to estimate the time required to achieve a given volume of filtrate or concentrate for a variety of protein markers. A typical spin time for a 4 mL sample is approximately 10 to 40 minutes (depending on device molecular weight cutoff). While most of the sample is filtered in the first 10 to 20 minutes of centrifugation, the lowest concentrate volume (30–75 µL) is reached after spinning for 20 to 40 minutes.

Figure 1. Typical Filtrate Volume vs. Spin Time (Swinging-bucket rotor)



Spin conditions: 4,000 × g, room temperature, 4 mL starting volume. Protein markers used: Cytochrome c for 3K and 10K, BSA for 30K and 50K, and IgG for 100K, n=6.

Figure 2. Typical Filtrate Volume vs. Spin Time (Fixed-angle rotor)



Spin conditions: 7,500 × g for 3K, 10K, 30K, and 50K, 5,000 × g for 100K, room temperature, 4 mL starting volume. Protein markers used: Cytochrome c for 3K and 10K, BSA for 30K and 50K, and IgG for 100K, n=6.

Table 2. Typical Concentrate Volume vs. Spin Time (Swinging-bucket rotor)

	Concentrate volume (µL)				
Spin time (min)	**3K device**	**10K device**	**30K device**	**50K device**	**100K device**
10	1,369	176	73	32	264
15	–	76	46	–	36
20	478	58	37	30	33
25	228	–	–	–	–
30	159	–	–	–	–
40	94	–	–	–	–

Spin conditions: 4,000 × g, room temperature, 4 mL starting volume. Protein markers used: Cytochrome c for 3K and 10K, BSA for 30K and 50K, and IgG for 100K, n=6 (mean value of 3 device lots). Shaded volumes were used for the calculation of protein recovery in Table 5.

Table 3. Typical Concentrate Volume vs. Spin Time (35° Fixed-angle rotor)

	Concentrate volume (µL)				
Spin time (min)	**3K device**	**10K device**	**30K device**	**50K device**	**100K device**
10	613	97	42	23	53
15	–	54	30	–	30
20	170	35	22	15	26
25	118	–	–	–	–
30	92	–	–	–	–
40	62	–	–	–	–

Spin conditions: 7,500 × g for 3K 10K, 30K, and 50K, 5,000 × g for 100K, room temperature, 4 mL starting volume. Protein markers used: Cytochrome c for 3K and 10K, BSA for 30K and 50K, and IgG for 100K, n=6 (mean value of 3 device lots). Shaded volumes were used for the calculation of protein recovery in Table 5.

Protein Retention and Concentrate Recovery

The membranes used in Amicon® Ultra devices are characterized by a molecular weight cutoff (MWCO); that is, their ability to retain molecules above a specified molecular weight. Solutes with molecular weights close to the MWCO may be only partially retained. Membrane retention depends on the solute's molecular size and shape. For most applications, molecular weight is a convenient parameter to use in assessing retention characteristics. For best results, use a membrane with a MWCO at least two times smaller than the molecular weight of the protein solute that one intends to concentrate. Refer to Table 4.

Chemical Compatibility

Amicon® Ultra centrifugal devices are intended for use with biological fluids and aqueous solutions. Before use, check the sample for chemical compatibility with the device.

Table 6. Chemical Compatibility of Amicon® Ultra Filter Devices

Acids	**Concentration**		**Concentration**
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%*
Alkalis			
Ammonium hydroxide	≤ 10%	Sodium hydroxide	≤ 0.5 M
Alcohols			
n-Butanol	≤ 70%	Isopropanol	≤ 70%
Ethanol	≤ 70%	Methanol	≤ 60%
Detergents			
Alconox® detergent	≤ 1%	Sodium dodecyl sulfate (SDS)	≤ 0.1%
CHAPS detergent	≤ 0.1%	Tergazyme™ detergent	≤ 1%
Lubrol® PX detergent	≤ 0.1%	Triton® X-100 surfactant	≤ 0.1%
Nonidet® P-40 surfactant	≤ 2%	Tween® 20 surfactant	≤ 0.1%
Sodium deoxycholate	≤ 5%		
Organic solvents			
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%*		
Miscellaneous			
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 Information

This section lists the catalogue numbers for Amicon® Ultra Centrifugal Filter Devices.

MWCO	Qty/ pk	Amicon® Ultra-0.5 device	Amicon® Ultra-2 device	Amicon® Ultra-4 device	Amicon® Ultra-15 device
3K	8	UFC500308	UFC200324	UFC800308	UFC900308
	24	UFC500324		UFC800324	UFC900324
	96	UFC500396		UFC800396	UFC900396
	500	UFC5003BK			
10K	8	UFC501008	UFC201024	UFC801008	UFC901008
	24	UFC501024		UFC801024	UFC901024
	96	UFC501096		UFC801096	UFC901096
	500	UFC5010BK			
10K IVD*	8			UFC801008D	UFC901008D
	24			UFC801024D	UFC901024D
	96			UFC801096D	UFC901096D
30K	8	UFC503008	UFC203024	UFC803008	UFC903008
	24	UFC503024		UFC803024	UFC903024
	96	UFC503096		UFC803096	UFC903096
	500	UFC5030BK			
50K	8	UFC505008	UFC205024	UFC805008	UFC905008
	24	UFC505024		UFC805024	UFC905024
	96	UFC505096		UFC805096	UFC905096
	500	UFC5050BK			
100K	8	UFC510008	UFC210024	UFC810008	UFC910008
	24	UFC510024		UFC810024	UFC910024
	96	UFC510096		UFC810096	UFC910096
	500	UFC5100BK			

* Amicon® Ultra-4 and -15 10K devices are for in vitro diagnostic (IVD) use. All other devices are for research use only.

Notice

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