Jumbosep™ Centrifugal Devices

 Convenient and reliable concentration, purification, and diafiltration of 15 to 60 mL biological samples.

Ordering Information				
Jumbosep Cent Part Number	trifugal Device Starter Kits Description	Pkg		
FD000K65	Generic starter kit, (no membrane inserts)	4/pkg		
FD003K65	3K Starter kit, gray	4/pkg		
FD010K65	10K Starter kit, blue	4/pkg		
FD030K65	30K Starter kit, red	4/pkg		
FD100K65	100K Starter kit, clear	4/pkg		
FD300K65	300K Starter kit orange	4/nka		

Jumbosep Cent	Jumbosep Centrifugal Device Mebrance Inserts		
Part Number	Description	Pkg	
OD003C65	3K membrane insert, gray	12/pkg	
OD010C65	10K membrane insert, blue	12/pkg	
OD030C65	30K membrane insert, red	12/pkg	
OD100C65	100K membrane insert, clear	12/pkg	
OD300C65	300K membrane insert, orange	12/pkg	

Accessory Products		
Part Number	Description	Pkg
FD001X65	Filtrate receiver and cap	12/pkg
FD002X65	Sample reservoir and cap	12/pkg
FD003X65	Insert release	24/pkg



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Note: The procedures herein are intended only as a guide. Users should always verify product performance with their specific applications under actual use conditions. If you have questions about the information presented in this guide, please contact our Technical Service department.



Introduction

Jumbosep™ centrifugal devices provide rapid and convenient concentration, diafiltration, and fractionation of 60 mL biological samples. A starting sample volume of 60 mL can be concentrated to 5 mL typically within 30 minutes. The Jumbosep device's ease of use saves valuable lab time.

Each Jumbosep centrifugal device contains a low protein-binding Omega™ membrane insert. This feature significantly reduces non-specific adsorption and enables the device to yield the highest recoveries. The Jumbosep centrifugal device is compatible with a number of solvents, giving the device the versatility to be used in a variety of different applications. Jumbosep centrifugal devices are ideal for buffer exchange or salt removal of chromatography eluates and gradient fractions; virus concentration or removal; concentration or desalting of enzymes, antibodies, growth factors, lymphokines, DNA, and RNA samples; and separation of antibodies and other biomolecules from cell culture.

Principle of Jumbosep™ Centrifugal Devices

Centrifugation up to 3,000 x g provides the driving force for filtration, moving sample toward the highly selective, low protein-binding Omega" membrane. Molecules larger than the membrane's nominal molecular weight cutoff (MWCO) are retained in the sample reservoir. Solutes and molecules smaller than the MWCO of the membrane pass through the membrane surface into the membrane insert and through the filtrate port into the filtrate receiver.

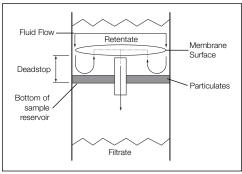
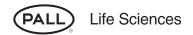


Figure 1

Molecular Weight Cutoffs Available

The Jumbosep centrifugal concentrator is available with five different molecular weight cutoffs: 3K, 10K, 30K, 100K, and 300K



Specifications

Materials of Construction

Filter Media: Omega™ Membrane (low protein-binding, modified polyether-sulfone)

Sample Reservoir and Filtrate Receiver: Polysulfone

Sample Reservoir Cap: Polyethylene Insert without Membrane: High density polyethylene Filtrate Receiver Cap and Insert Release: Polypropylene

Effective Filtration Area

15.2 cm²

Dimensions

Outside Diameter (maximum): 6 cm (2.4 in.)
Overall Height (fully assembled with cap): 11.3 cm (4.5 in.)

Capacities

Sample Volume (maximum): 60 mL Final Concentrate Volume: 3.5 to 4 mL Filtrate Receiver Volume (maximum): 60 mL Hold-up Volume (membrane/support): 0.2 mL

Operating Temperature Range

0 to 40 °C (32 - 104 °F)

pH Range

1 - 14

Maximum Centrifugal Force

 $3.000 \times a$

Centrifuge

Swinging-bucket rotor that accepts 250 mL bottles and is capable of spinning at 1,000 to $3,000 \times g$ (see pages 14-17).

Sanitization

Provided non-sterile; may be sanitized by filtering 70% ethanol through the device prior to use.

Applications

Jumbosep™ centrifugal concentrators replace dialysis, chemical precipitation, and lyophilization in the following applications:

- Concentrating and desalting enzymes, antibodies, growth factors, lymphokines, DNA, and RNA samples
- Buffer exchange or salt removal of chromatography eluates, and gradient fractions
- Harvesting antibodies and other biomolecules from cell culture
- Concentrating recombinant proteins in conditioned media
- Virus concentration or removal
- Crude fractionation of dilute protein mixtures
- Purification or clarification of tissue homogenates instead of ultracentrifugation
- Recovery of biomolecules from cell culture broths
- Removing debris and particulates from cell lysates
- Natural product screening



Choosing the Appropriate MWCO

For maximum retention, select a Jumbosep™ membrane insert with a MWCO that is 3 to 6 times smaller than the molecular weight of the molecule to be retained. For example, for a 100K protein, a 10K or 30K membrane insert would be the appropriate selection.

Components

Each Jumbosep device consists of the following components:

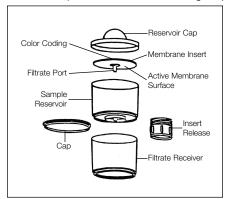


Figure 2
Generic Starter kit does not include membrane insert.

Jumbosep[™] Device Operation

Pre-Rinsing (Optional)

For the majority of applications, Jumbosep membrane inserts can be used without pre-rinsing. However, Omega" membrane inserts contain trace amounts of glycerine (a humectant) and sodium azide (a biocide). If these chemicals present possible assay interferences, they can be removed by the following procedure:

- 1. Assemble the device using the Instructions for Use (steps 1 through 8).
- 2. Filter 60 mL of deionized water or buffer through the Jumbosep device.
- 3. Empty the water or buffer from the filtrate receiver.
- 4. Repeat steps 2 and 3.

Note: If further flushing is required, filter 60 mL of 0.05N NaOH through the device. Empty and repeat the procedure. Wash the device clean by filtering 60 mL of deionized water. Use the membrane insert within 20 minutes or store it in buffer or deionized water to prevent irreversible membrane damage due to dehydration.



Non-Specific Adsorption

Omega™ membranes are made from polyethersulfone (PES) that has been specially modified to minimize protein binding. These membranes provide equivalent or higher recoveries than comparable regenerated cellulose membranes and offer exceptional biological and chemical resistance (see Chemical Compatibility, on pages 12-13).

Adsorption to device components is of particular concern when purifying microgram or nanogram levels of protein. Even with the advanced plastics used in Jumbosep™ devices, some adsorption may occur with particularly "sticky" proteins and biomolecules. Pre-treating Jumbosep centrifugal devices may further reduce non-specific adsorption to the device. To pre-treat, use the following procedure:

- 1. Fill the sample reservoir with 60 mL of 10% glycerin.
- 2. Soak overnight at room temperature.
- 3. Rinse the Jumbosep device with deionized water.
- Fill the sample reservoir with 60 mL of deionized water and spin. Empty water from the device.
- 5. Repeat step 4.

Caution: Use the device within 20 minutes to prevent irreversible membrane damage due to dehydration.

Instructions for Use

- 1. Separate the filtrate receiver from the sample reservoir.
- Hold the membrane insert by the edge with the filtrate port facing down and drop the insert into the sample reservoir.
- 3. Place the sample reservoir on a hard surface and, with both thumbs placed on the colored button in the middle of the membrane, press down firmly on the membrane insert. The membrane insert rests on the knobs at the bottom of the sample reservoir.

Note: The color of the button on the top of the membrane insert indicates the molecular weight cutoff of the membrane.

- 4. Attach the empty filtrate receiver to the bottom of the sample reservoir.
- Add 15 to 60 mL of sample to the sample reservoir. Place the cap on top of the reservoir to prevent evaporation during centrifugation.
- 6. Place the device in a swinging-bucket rotor that accepts standard 250 mL bottles. Remove any bottle adapters to ensure that the bottom of the bucket is flat. Presence of the adapters might cause deformation of the bottom of the Jumbosep device's filtrate receiver.

Note: Always counterbalance the rotor with another Jumbosep device containing an equivalent sample volume.

 Spin at 1,000 to 3,000 x g, typically for 15 to 40 minutes, to achieve the desired retentate volume. It is recommended that spin times and g-force be determined experimentally for each application.

Caution: Maximum g-force is 3,000 x g. Higher g-forces may cause retentate leakage into the filtrate.

- 8. At the end of spin time, stop the centrifuge and remove the Jumbosep devices. Separate the sample reservoir from the filtrate receiver in the following manner:
 - a) Hold the device so that both palms are placed on the filtrate receiver.
 - b) With both thumbs placed side by side on the sample reservoir, press upward (Figure 3).

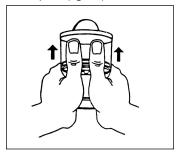


Figure 3

- 9. To recover retentate:
 - a) Pour off the retentate into a storage vessel. Some retentate will remain under the membrane insert.
 - b) To remove the remaining retentate, twist the insert release onto the sample reservoir (Figure 4).
 - c) Turn the sample reservoir sideways (taking care that the retentate remains in the sample reservoir).
 - d) Slide a pipette tip under the dislodged membrane insert and remove the remaining retentate.

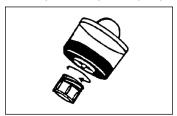


Figure 4

Polarization Control

Unlike many centrifugal devices, Jumbosep's filtration flow proceeds in the opposite direction from the centrifugal force. This unique design is responsible for Jumbosep's high flow rates due to two factors:

- Centrifugal force causes macromolecules and particulates which have specific gravities greater than the solute to be driven away from the membrane surface to the sample reservoir bottom.
- Macromolecule build-up (gel polarization phenomenon) on the membrane surface is minimized.

Diafiltration (Desalting and Buffer Exchange)

For salt removal or buffer exchange, use the following procedure:

- 1. Concentrate the sample at least ten-fold. For example, concentrate 60 mL to 6 ml
- 2. Reconstitute with exchange buffer and reconcentrate ten-fold.
- 3. Repeat the above steps 3-to-5 times to remove 95-99% of salt or buffer from the solution.



Chemical Compatibility

Reagent	Device Compatibility
Acetic Acid (10%)	•
Acetone (20%)	•
Acetonitrile (20%)	•
Ammonium Hydroxide (1N)	•
Ammonium Sulfate	•
Chloroform (1%)	•
Dimethyl Sulfoxide (20%)	•
Dimethyl Formamide (20%)	•
Ethanol (70%)	•
Ethyl Acetate	x
Formaldehyde (5%)	•
Formic Acid (1N)	•
Glycerol	•
Guanidine HCL (6M)	•
Hydrochloric Acid (1N)	•
Hydrogen Peroxide (10%)	•
Methanol (70%)	•
Methyl Ethyl Ketone (10%)	•
Phosphate Buffer	•
Phosphoric Acid (1N)	•
Polyethylene Glycol (0.1%)	•
Propanol (70%)	•
Saline Buffer (0.85%)	•
Sodium Dodecyl Sulfate (0.01M)	•
Sodium Hydroxide (1N)	•
Sodium Hypochlorite (0.05%)	•
Sulfuric Acid (1N)	•

• = Compatible \mathbf{x} = Not compatible

Note: Flow rates may be affected by the above reagents.

Chemical Compatibility (Cont)

Reagent	Device Compatibility
Terg-a-zyme* (1%)	•
Tris* Buffer (1M)	•
Ultrasil 11* (2%)	•
Urea (6M)	•

• = Compatible \mathbf{x} = Not compatible

Note: Flow rates may be affected by the above reagents.



Centrifuge Recommendations for Jumbosep™ Centrifugal Devices

Dimensions: 60 x 113 mm

0	D-t	Rotor	Di	Rotor
Centrifuge	Rotor	Angle	Places	Cavity (mL)
Beckman				
Avanti J25 & J2 series	JS-7.5	SW	4	250
J2 high capacity only	JS-4.3	SW	4	750
J6 series high capacity	JS-5.2	SW	4	1000
	JS-4.0	SW	4	1000
	JS-4.2	SW	6	1000
	JS-3.0	SW	6	1000
GS-6 series	GH-3.7	SW	4	1000
TJ-6 Bench top	TH-4	SW	4	250
IEC				
Centra* MP4 series	224	SW	4	250
Centra* GP8 series	52842	SW	4	250
	216	SW	4	750
	218	SW	4	750
	228	SW	4	750
Centra*-HN & HN-SII	245	SW	4	250
	268	SW	4	250
B-22M	963	SW	4	250
PR-7000/7000M	219	SW	4	750
	966	SW	6	1000
	949	SW	6	1000
	256	SW	4	1000

Accessories		etting for g-force 3,000 x g	Mfg listed rotor g-force maximum
7.000000.000	.,ooo x g	e,ccc x g	
none needed	2,326	4,028	10,400
349946	2,093	3,626	4,220
339108	1,988	3,443	6,935
339108	1,989	3,445	4,044
339108	1,876	3,250	5,010
339108	1,876		2,556
349849	2,096	3,631	3,200
339288	2,190		1,520
none needed	2,418	4,189	3,310
none needed	2,124	3,679	3,200
5780	2,129	3,688	2,550
5780	2,157	3,735	4,550
5780	2,150		2,500
none needed	2,245		1,500
none needed	2,422		2,000
none needed	2,315	4,009	10,500
5780	2,159	3,739	8,250
5780	1,912	3,311	7,400
5780	1,887		2,700
5780	1,971	3,413	3,525



Centrifuge Recommendations for Jumbosep™ Centrifugal Devices (Cont.)

Dimensions: 60 x 113 mm

Centrifuge	Rotor	Rotor Angle	Places	Rotor Cavity (mL)	
Jouan					
CR-CT & GR-GT series	T4	SW	4	250	
4.12-22	M4	SW	4	250	
C & G series 4.12-22	T4	SW	4	250	
	M4	SW	4	250	_
CR 3.12/CR 3.22	T4	SW	4	250	
	E4	SW	4	750	
C 3.12	T4	SW	4	250	_
	E4	SW	4	750	
KR.22	P6 w/o lid	SW	6	1000	
	P6 w/o lid	SW	6	1000	
	P6 w/lid	SW	6	1000	
	C6 w/o lid	SW	6	1000	
	C6 w/lid	SW	6	1000	
Sorvall*					
RC-5 series/ RC-28S&24	HS-4	SW	4	250	
	SH-3000	SW	4	750	_
6000 series table top models	H-1000B	SW	4	250	
RC-3 series	HS-4	SW	4	250	
	H-2000B	SW	8	250	
	HG-4L	SW	4	1000	
	H-4000B	SW	4	1000	
	H-6000A	SW	6	1000	

		tting for	Mfg listed
Accessories	desired 1,000 x g	g-force 3,000 x g	rotor g-force maximum
	.,	-, 3	
11175356	2,282		2,630
11174167	2,198	3,808	4,190
11175356	2,283		2,350
11174167	2,199	3,808	3,310
11175356	2,281		2,490
11174167	2,199		2,985
11175356	2,283		2,350
11174167	2,199		2,830
11178215 / 11175356	1,854	3,212	4,200
11178206 / 11178311	1,903	3,124	4,440
11178206 / 11178311	1,904	3,125	6,500
11178206 / 11178311	1,834	3,176	5,500
11178206 / 11178311	1,826	3,163	7,200
none needed	2,280	3,949	10,820
250 mL adapter	2,196	3,803	4,200
00186	2,162		2,190
none needed	2,280	3,949	6,925
00186	1,842		2,310
00443/00511	1,968	3,408	7,120
00443/00511	1,968	3,408	7,120
00443/00511	1,851	3,206	7,200



Troubleshooting		
Problem	Possible Cause	Possible Solution
Spin time too long	Centrifuge force too low	Increase g-force
	Biomolecule reached maximum concentration	Further concentration not possible
	Molecular weight cutoff of membrane insert too tight	Try membrane insert with the next highest MWCO
Loss of sample activity	Interference of trace amounts of glycerine and azide in the membrane	Follow pre-rinsing procedure on page 9
	Incompatibility of biomolecule and solvent	Adjust pH; change buffer type
	Sub-units of biomolecule may be passing through membrane into filtrate	Select lower molecular weight cutoff to retain sub-units
	Protein:protein interactions, gel layer formation	Decrease final concentration factor (increase final retentate volume)
Passage of biomolecule into filtrate	Poor retention of biomolecule by membrane	Select lower molecular weight cutoff membrane insert
	Membrane insert not sealed properly	Ensure that membrane insert is pushed all the way down
Smaller molecular weight molecule not passing completely through the	Retention of smaller molecule caused by larger retaining species inhibiting passage	Select higher molecular weight cutoff, allowing greater passage of smaller molecule
membrane		Use diafiltraton cycle to remove smaller molecule (see page 11)
Low recovery	Non-specific adsorption	Follow non-specific adsorption procedure (see page 10)
	Biomolecule not retained by membrane	Check for level of biomolecule in filtrate. If level is unacceptably high, choose a lower molecular weight cutoff (the sample is not lost – do not discard!)
	Membrane insert not seated properly	Ensure that membrane insert is pushed all the way down

Complementary Products

 Pall Life Sciences offers centrifugal devices for processing the following sample volumes:

<u>Device</u>	Sample Volume
Nanosep® Device	up to 0.5 mL
Microsep™ Device	0.5 to 3.5 mL
Macrosep® Device	3 to 15 mL
Jumbosep™ Device	15 to 60 ml

- BioTrace™, FluoroTrans® and Biodyne® Transfer Membranes offer precise performance and compatibility with nearly every detection system available.
- AcroWell™ 96-well Filter Plates with BioTrace NT and BioTrace PVDF Membranes exhibit high binding capacities for proteins and nucleic acids.
- Filtration Devices with Supor® Membrane are sterile, ready-to-use and maximize sample recoveries with low protein-binding membrane and low hold-up volumes.
- Enchant Protein Purification Kits deplete, fractionate, and purify abundant proteins.
- BioSepra® Chromatography Media used for purification of biomolecules and compounds.
- Minimate[™] Laboratory Tangential Flow Filtration Devices are typically used for the concentration of defiltration of 100 mL to 5 L samples.
- AcroPrep™ 96 and 384-well Filter Plates with a variety of membrane for high throughput protein assay applications.



WARNING

Employment of the products in applications not specified, or failure to follow all instructions contained in this product information insert, may result in improper functioning of the product, personal injury, or damage to property or the product. See Statement of Warranty in our most recent catalog.

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