



# **PROTEAN® II xi Multi-Gel Casting Chamber**

## **Instruction Manual**

**Catalog Number  
165-2025**



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# Section 1

## General Information

### 1.1 Introduction

The PROTEAN II xi multi-gel casting chamber allows as many as twelve 1.5 mm gels to be cast simultaneously, for use in the PROTEAN II xi cell or the PROTEAN II xi multi-cell. Monomer can be introduced from the top or the bottom to prepare identical gradient or uniform percentage gels. Cast gels can be stored for up to 1 week.

### 1.2 Specifications

#### Construction

Casting chamber	fabricated acrylic
Sealing plate	fabricated acrylic with captive, spring-loaded screws
Gasket	silicone tubing

#### Shipping weight

1.7 kg

#### Size

9.5 x 22.5 x 23.5 cm

#### Gel size

16 x 16 cm or 16 x 20 cm

#### Chemical compatibility

As the multi-gel casting chamber is acrylic, it is not compatible with many organic solvents. In particular, the unit should not be used with the following solvents as severe damage will result: acetic acid (glacial), acetone, carbon tetrachloride, chloroform, chromic acid (40%), diethyl ether, dimethyl formamide, ethyl acetate, toluene, xylene, and ethanol.

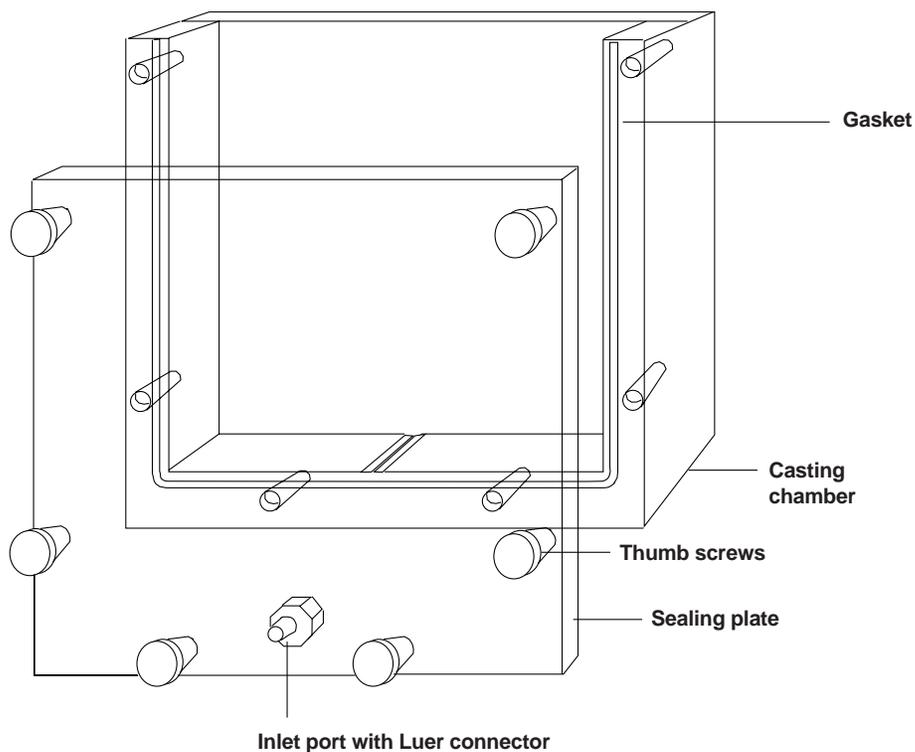
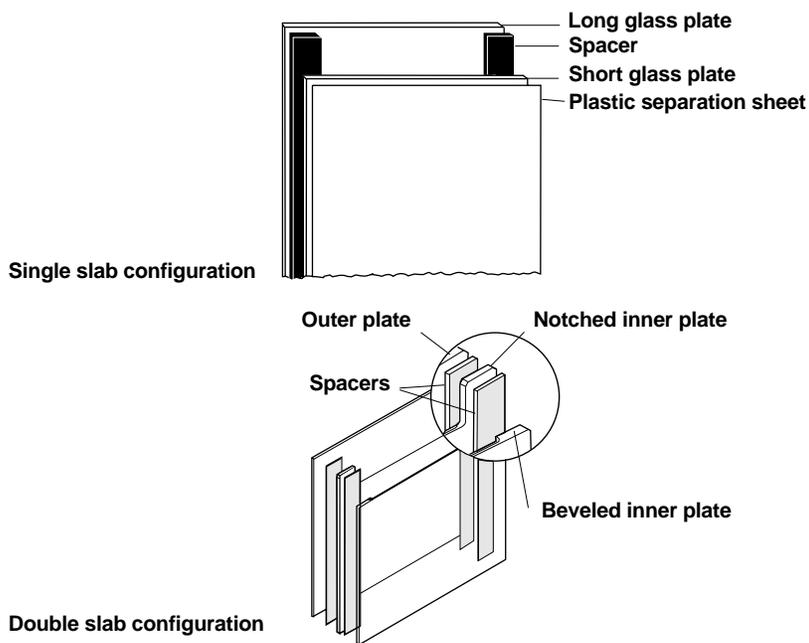


Fig. 1.1. PROTEAN II xi multi-gel casting chamber.

## Section 2 Preparation for Casting

In the PROTEAN II xi system, one pair of clamps will accommodate either one or two gel slabs. Each single slab requires one inner plate, one outer plate, and one pair of spacers. Running two slabs in one pair of clamps requires one inner plate, one outer plate, one notched inner plate, one beveled inner plate, and two pairs of spacers. Figure 2.1 shows the configuration required for casting single and double gels.



**Fig. 2.1. Diagram showing single and double slab configuration gel sandwiches as they would appear in the multi-gel casting stand.**

The PROTEAN II xi multi-gel casting chamber allows you to cast up to nine 1.5 mm slab gels in the single slab configuration or up to twelve 1.5 mm slab gels in the double slab configuration.

Since there are several choices of spacer thickness, the option of single or double slabs, and the choice of the number of gels, the total amount of space in the casting chamber taken up by plates and spacers can vary, sometimes leaving extra chamber space. When the chamber is not completely full, extra space must be taken up by acrylic blocks, and in some cases, by extra glass plates. The plastic separation sheets are used between the sandwiches to simplify separation of the plates after polymerization.

Before use, clean the casting chamber parts with laboratory detergent, rinse the parts thoroughly with distilled deionized water, and allow them to dry completely.

### 2.1 Loading the Chamber - Single Slab Configuration

1. Place the open casting chamber on a benchtop, open face up, and prop it up to about a 30° angle.
2. Place the first glass plate in the chamber. This plate should be an outer plate (tall plate).

3. Put one set of spacers on the first glass plate so that they are seated firmly against the side walls and lower corners of the chamber.

**Note:** To simplify the spacer positioning, the optional alignment card (165-2029) may be used. Insert the card between the spacers prior to the placement of the shorter glass plate. Remove the card prior to the addition of the monomer.

4. Carefully place a glass inner plate (short plate) on top of the spacers. Do not disturb the spacers.

**Note:** For 2-D applications the inner glass plate should be a beveled plate.

5. Place a plastic separation sheet on top of the inner glass plate. Be sure to remove the protective film from the separation sheet prior to use.
6. Repeat steps 2-5 until you have prepared the desired number of gel sandwiches.
7. Take up the remaining space in the chamber so that the plates and spacers will be held firmly in position when the sealing plate is in place. First, use the acrylic blocks, placing as many as will fit while still allowing the sealing plate to seal tightly. If some remaining space must be taken up to provide a tight seal, use an extra glass plate and/or a few plastic separation sheets. If you use more than one glass plate to take up space, put a plastic separation sheet between them to simplify separation after polymerization.
8. Seat the gasket firmly in the gasket notch on the perimeter of the open face.
9. Place the sealing plate on the chamber, aligning the six screws with the holes in the chamber walls. Gradually tighten the screws in a random fashion. When the screws are all moderately tight, hold the sealing plate against the casting chamber with one hand and tighten each screw further with the other hand.
10. Stand the casting chamber up and place it on a level surface. Do not tip the chamber upside down at this stage. Proceed to Section 3.

## 2.2 Loading the Chamber - Double Slab Configuration

1. Place the open casting chamber on a benchtop, open face up, and prop it up to about 30° angle.
2. Place the first glass plate in the chamber. This plate should be an outer plate (tall plate).
3. Put one set of spacers on the first plate so that they are seated firmly against the side walls and lower corners of the chamber.
4. Place a notched glass plate on top of the spacers carefully. Do not disturb the spacers.
5. Place a pair of spacers on the notched glass plate so that they are seated firmly against the side walls and lower corners of the chamber.
6. Place an inner plate (short plate) on top of the spacers carefully so as not to disturb the spacers.
7. Place a plastic separation sheet on top of the inner glass plate. Be sure to remove the plastic film from the separation sheet prior to use.
8. Repeat steps 2 through 8 until you have prepared the desired number of gel sandwiches.
9. Take up the remaining space in the chamber so that the plates and spacers will be held firmly in position when the sealing plate is in place. First, use acrylic blocks, glass plates, and separation sheets to fill this space. Place as many as will fill the casting chamber while not extending past its outer edge. If you use more than one glass plate to take up space, put a plastic separation sheet between them to simplify separation after polymerization.

10. Seat the gasket firmly in the gasket notch on the perimeter of the open face.
11. Place the sealing plate on the chamber, aligning the six screws with the holes in the chamber walls. Tighten the screws a little at a time in a random fashion. When the screws are all moderately tight, hold the sealing plate against the casting chamber with one hand and tighten each screw further with the other hand.
12. Stand the casting chamber up and place it on a level surface. Do not tip the chamber upside down at this stage.

### 2.3 The Inlet Port

The inlet port must be prepared so that the flow of monomer can be stopped and so that the port can be cleared of polymerized gel after use.

1. Cut a piece of Tygon® tubing (3 mm inner diameter), about 10 cm long.
2. Fit one end of the tubing onto a Luer connector or a stopcock. Connect the other end to the casting chamber port.
3. When monomer is introduced from the top, the casting chamber port should be kept closed by placing a pinch clamp on the tubing to prevent flow, or the stopcock should be in the "closed" position. Tubing from the reservoir or gradient maker is attached to the open end of a Luer connector or stopcock.

## Section 3 Casting Gels

Constant percentage gels can be prepared by introducing monomer from either the top or the bottom of the chamber. Gradients must be introduced from the bottom.

### 3.1 Estimating Monomer Volume

The volume of monomer required depends on several factors, including gel thickness, number of gels, and gel length, all of which can vary depending on your specific requirements. To make a precise determination of the volume of monomer required for your application, set up the casting chamber and measure the volume of deionized distilled water required to fill it to the desired level. Then disassemble the chamber, rinse and dry the parts, and repeat assembly and casting using monomer solution. Table 3.1 gives an example of volumes determined empirically for representative applications.

**Table 3.1. Monomer Volume Requirements for Casting Twelve 1.5 mm Gels Using the Double Gel Configuration**

Gel Type	PROTEAN II xi Cell Length Option	
	16 cm	20 cm
Lower gel-discontinuous system, leaving room for stacking gel with comb	410 ml	540 ml
Lower gel 2-D, leaving room for stacking gel, or continuous gel, no stacking gel with comb	460 ml	580 ml
2-D gel, no stacking gel	490 ml*	620 ml*

\* Maximum volume for any combination.

### 3.2 Casting from the Top (Non-Gradient Gels)

1. Be sure the pinch clamp is positioned on the inlet tubing so that the outward flow is blocked.
2. Mark a level on the chamber (with tape or a felt-tip pen) at the desired gel length, measuring from the bottom.
3. Introduce the monomer into one of the gel sandwiches (it is not necessary to pour monomer down all the plates). This is easily done using a funnel. Stop when you reach the desired level.
4. Overlay the monomer in each sandwich with 1.0 ml of water saturated sec-butanol. The amount of overlay solution and the rate of application must be the same on each gel to obtain gels of identical length. Optimally, the overlay solution should be applied to each gel simultaneously. This operation is best performed using a 1 ml syringe, with a pipet tip (yellow, type 35, catalog number 223-9035) attached, for each gel. It may be necessary to cut the top of the pipet tip to provide a snug fit onto the syringe. The syringes are filled with the overlay solution and are placed upright into the gel sandwiches. Using a flat plate, slowly overlay all the gels at the same time by pressing on all of the syringe plungers. Remove the syringes, and cover the chamber with a plate to prevent dust from settling on the gels.

### 3.3 Casting from the Bottom

The inlet on the PROTEAN II xi multi-gel casting chamber is used for casting linear and convex acrylamide gradient gels. Refer to your gradient maker instructions for preparation of the gradient solutions, etc.

1. Be sure the pinch clamp is positioned on the inlet tubing, or that the stopcock is closed, so that the flow to the chamber is blocked.
2. Mark a level on the chamber (with tape or felt-tip pen) at the desired gel length, measuring from the bottom.
3. Add monomer to the gradient maker and prepare it to begin pumping.
4. Slide the pinch clamp onto the Luer connector, or open the stopcock, to open the inlet port. Begin pumping in monomer.
5. Stop the pump when the monomer reaches the desired height, close off the pinch clamp or stopcock, disconnect the tubing upstream of the Luer connector or stopcock, and purge the gradient maker and tubing with water.
6. Overlay the monomer in each sandwich with 1.0 ml of water or water saturated sec-butanol. To obtain gels of identical length, the amount of overlay solution and the rate of application must be the same on each gel. Optimally, the overlay solution should be applied to each gel simultaneously. This operation is best performed using a 1 ml syringe, with a pipet tip attached, for each gel. It may be necessary to cut the top of the pipet tip to provide a snug fit onto the syringe. The syringes are filled with the overlay solution and are placed upright into the gel sandwiches. Using a flat plate, slowly overlay all the gels at the same time by pressing on all the syringe plungers. Remove the syringes, and cover the chamber with a plate to prevent dust from settling on the gels.

**Note:** Sec-butanol will chemically attack the acrylic plastic of the multi-casting chamber. This will cause crazing of the plastic. If alcohol is used to overlay the monomer solution, exercise caution in its disposal to minimize contact with the acrylic plastic.

## Section 4 Storage and Use of Gels

**Note:** Wear gloves for this operation.

1. After polymerization is complete, open the casting chamber and separate the gels by pushing a spatula down between the glass plates that are separated by plastic separation sheets.
2. Rinse off the sec-butanol overlay with distilled water. Wash off pieces of gel from the surface and remove rough edges of polyacrylamide extending out from the bottom with a razor blade.
3. Add 1 to 2 ml of 1x gel buffer (identical to the buffer in the gel) to the top of each gel. Store the gels in a tightly closed container or large zip-lock bag with a few milliliters of gel buffer for up to 1 week at 4 °C.

If stacking gels are to be used, they should be cast immediately prior to use.

4. On the day of use, remove gel(s) from storage and attach a pair of PROTEAN II xi clamps in the following manner:

Loosen the single screw of both the right and left sandwich clamps by turning counter-clockwise. Place each clamp by the appropriate side of the glass plate stack, with the locating arrows of the clamps facing up and toward the glass plates.

Grasp the glass plate sandwich firmly with your right hand. With your left hand guide the left clamp onto the sandwich so that the long and short plates fit the appropriate notches in the clamp. Tighten the single screw.

Place the right clamp on the right side of the plates and tighten the clamp screw.

5. Stand the sandwich upright on a level surface (a glass plate works well), loosen the clamp screws, and allow the gel sandwich to drop to the level surface. Re-tighten the clamp screws completely.
6. The gel sandwich is now ready for attachment to the PROTEAN II xi cooling core. For a complete description of cell setup and running the gels, including reagent preparation, refer to the PROTEAN II xi cell or the PROTEAN II xi multi-cell instruction manuals.

## Section 5 Equipment and Accessories

### 5.1 PROTEAN II xi Multi-Gel Casting Chamber

<b>Catalog Number</b>	<b>Product Description</b>
165-2025	<b>PROTEAN II xi Multi-Gel Casting Chamber</b> , includes casting chamber, sealing plate, Luer taper connector, silicone gasket, 15 plastic separation sheets, 4 acrylic blocks, and instructions
<b>Accessories</b>	
165-2026	<b>Silicone Sealing Gasket</b> , 3
165-1957	<b>Acrylic Blocks</b> , set of 4
165-1958	<b>Separation Sheets</b> , 15
165-2029	<b>Alignment Cards</b> , 2

#### **Other equipment required to cast gels:**

Glass plates, spacers, pump (for gradient gels)

## 5.2 PROTEAN II xi Cells

Catalog Number	Product Description
<b>PROTEAN II Slab Cells</b>	
165-1801	<b>PROTEAN II xi 16 cm Slab Cell</b> , no spacers or combs (order separately)
165-1802	<b>PROTEAN II xi 16 cm Slab Cell</b> , 1.5 mm spacers (4), 15 well combs (2)
165-1803	<b>PROTEAN II xi 16 cm Slab Cell</b> , 1.0 mm spacers (4), 15 well combs (2)
165-1804	<b>PROTEAN II xi 16 cm Slab Cell</b> , 0.75 mm spacers (4), 15 well combs (2)
165-1811	<b>PROTEAN II xi 20 cm Slab Cell</b> , no spacers or combs (order separately)
165-1812	<b>PROTEAN II xi 20 cm Slab Cell</b> , 1.5 mm spacers (4), 15 well combs (2)
165-1813	<b>PROTEAN II xi 20 cm Slab Cell</b> , 1.0 mm spacers (4), 15 well combs (2)
165-1814	<b>PROTEAN II xi 20 cm Slab Cell</b> , 0.75 mm spacers (4), 15 well combs (2)

All PROTEAN II xi cells include the central cooling core with gaskets and core caps, lower buffer chamber, lid with power cables, 2 sets of glass plates, 4 sandwich clamps, an upper buffer dam, a casting stand with gaskets, alignment cards, a leveling bubble, and instructions.

Catalog Number	Product Description
<b>PROTEAN II xi 2-D Cells</b>	
165-1931	<b>PROTEAN II xi 2-D Cell</b> , 1.0 mm, 16 cm
165-1932	<b>PROTEAN II xi 2-D Cell</b> , 1.5 mm, 16 cm
165-1933	<b>PROTEAN II xi 2-D Cell</b> , 1.0 mm, 20 cm
165-1934	<b>PROTEAN II xi 2-D Cell</b> , 1.5 mm, 20 cm

PROTEAN II xi 2-D cells include central cooling core, lower buffer chamber, lid with power cables, 2 sets of glass plates (with bevels), 4 sandwich clamps, 24 glass tubes, 2 tube cell adapters, 16 stoppers, 48 grommets (4-8 mm OD tubes), 2 2-D combs, 4 spacers, 1 upper buffer dam, slab casting stand, alignment cards, leveling bubble, and instructions.

Catalog Number	Product Description
<b>PROTEAN II Multi-Cells</b>	
165-1951	<b>PROTEAN II xi Multi-Cell</b> , includes 3 central cooling cores with gaskets, lid with power cables, tank, 1 upper buffer dam, PROTEAN II multi-gel casting chamber, leveling bubble, and instructions (order appropriate spacers, plates, clamps, combs, and accessories for your application).
165-1956	<b>PROTEAN II xi Multi-Cell 2-D Conversion Kit</b> , includes 2 cooling coils and manifold required for 2-D electrophoresis applications.

## 5.3 PROTEAN II xi Accessories

	Catalog Number	
	16 cm Cell	20 cm Cell
<b>Glass Plates</b>		
Inner Plate, 2	165-1821	165-1823
Outer Plate, 2	165-1822	165-1824
Frosted Inner Plate, agarose gels,* 2	165-1825	165-1826
Beveled Inner Plate, 2-D procedures,* 2	165-1827	165-1828
Notched Inner Plate	165-1832	165-1833

\*Used with regular outer plate.

### 5.3 PROTEAN II xi Accessories (continued)

	Catalog Number	
	16 cm	20 cm
<b>Spacers</b>		
0.5 mm	165-1841	165-1846
0.75 mm	165-1842	165-1847
1.0 mm	165-1843	165-1848
1.5 mm	165-1844	165-1849
3.0 mm	165-1845	165-1850
<b>Sandwich Clamps</b>		
Set, one left, one right	165-1901	165-1902

#### Miscellaneous Accessories

	Catalog Number
Alignment Cards, 2	165-2029
Upper Buffer Dam	165-1909
Slab Gel Casting Stand, with gaskets	165-1911
Replacement Gaskets, casting stand, 2	165-1912
Gradient Pouring Needle for bottom filing, 2	165-2007
Replacement Gaskets, central cooling core, 2	165-1913

#### Standards

Prestained Standards, high, 500 ml	161-0309
Prestained Standards, low, 500 ml	161-0305
Prestained Standards, broad, 500 ml	161-0318
Kaleidoscope Prestained Standards, 500 ml	161-0324
SDS-PAGE Standards, high, 200 ml	161-0303
SDS-PAGE Standards, low, 200 ml	161-0304
SDS-PAGE Standards, broad, 200 ml	161-0317
2-D SDS-PAGE Standards, 500 ml	161-0320
Silver Stain Standards, high 200 ml	161-0315
Silver Stain Standards, Low, 200 ml	161-0314

#### Stains and Dyes

Bromophenol Blue, 10 g	161-0404
Coomassie Blue G-250, 10 g	161-0406
Coomassie Blue R-250, 10 g	161-0400
Copper Stain & Destain Kit	161-0470
Ethidium Bromide Solution, 10 ml	161-0433
Ethidium Bromide Tablets, 10	161-0430
Silver Stain Kit	161-0443
Silver Stain Plus Kit	161-0449

**Buffers**

<b>Boric Acid, 500 g</b>	161-0750
<b>Boric Acid, 1 kg</b>	161-0751
<b>EDTA, 100 g</b>	161-0728
<b>EDTA, 500 g</b>	161-0729
<b>10x TBE Buffer, 1 L</b>	161-0733
<b>10x TBE Extended Range Buffer, 1 L</b>	161-0741
<b>10x Tris/Glycine Buffer, 1 L</b>	161-0734
<b>10x Tris/Gly/SDS Buffer, 1 L</b>	161-0732
<b>Tricine, 100 g</b>	161-0712
<b>Tris, 500 g</b>	161-0716
<b>Tris, 1 kg</b>	161-0719
<b>Urea, 250 kg</b>	161-0730
<b>Urea, 1 kg</b>	161-0731
<b>2-mercaptoethanol, 25 ml</b>	161-0710

**Detergents**

<b>CHAPS, 1 g</b>	161-0460
<b>CHAPSO, 1 g</b>	161-0465
<b>Cleaning Concentrate, 1 kg</b>	161-0722
<b>SDS, 25 g</b>	161-0300
<b>SDS, 100 g</b>	161-0301
<b>Triton-X100, 500 ml</b>	161-0407

**Gel Reagents**

<b>Acrylamide, 99.9%, 100 g</b>	161-0100
<b>Acrylamide, 99.9%, 500 g</b>	161-0101
<b>Acrylamide/Bis 19:1, 30 g</b>	161-0120
<b>Acrylamide/Bis 19:1, 150 g</b>	161-0123
<b>Acrylamide/Bis 29:1, 30 g</b>	161-0121
<b>Acrylamide/Bis 29:1, 150 g</b>	161-0124
<b>Acrylamide/Bis 37.5:1, 30 g</b>	161-0122
<b>Acrylamide/Bis 37.5:1, 150 g</b>	161-0125
<b>40% Acrylamide/Bis Solution, 19:1, 500 ml</b>	161-0144
<b>40% Acrylamide/Bis Solution, 29:1, 500 ml</b>	161-0146
<b>40% Acrylamide/Bis Solution, 37.5:1, 500 ml</b>	161-0148
<b>30% Acrylamide/Bis Solution, 19:1, 500 ml</b>	161-0154
<b>30% Acrylamide/Bis Solution, 29:1, 500 ml</b>	161-0156
<b>30% Acrylamide/ Bis Solution, 37.5:1, 500 ml</b>	161-0158
<b>Ammonium Persulfate, 10 g</b>	161-0700
<b>BAC, 5 g</b>	161-0204
<b>Bis, 5 g</b>	161-0200
<b>Bis, 50 g</b>	161-0201
<b>Dithiothreitol, 1 g</b>	161-0610
<b>Dithiothreitol, 5 g</b>	161-0611
<b>PAGE Reagent Starter Kit</b>	161-5100
<b>PDA, 10 g</b>	161-0202
<b>TEMED, 5 ml</b>	161-0800

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