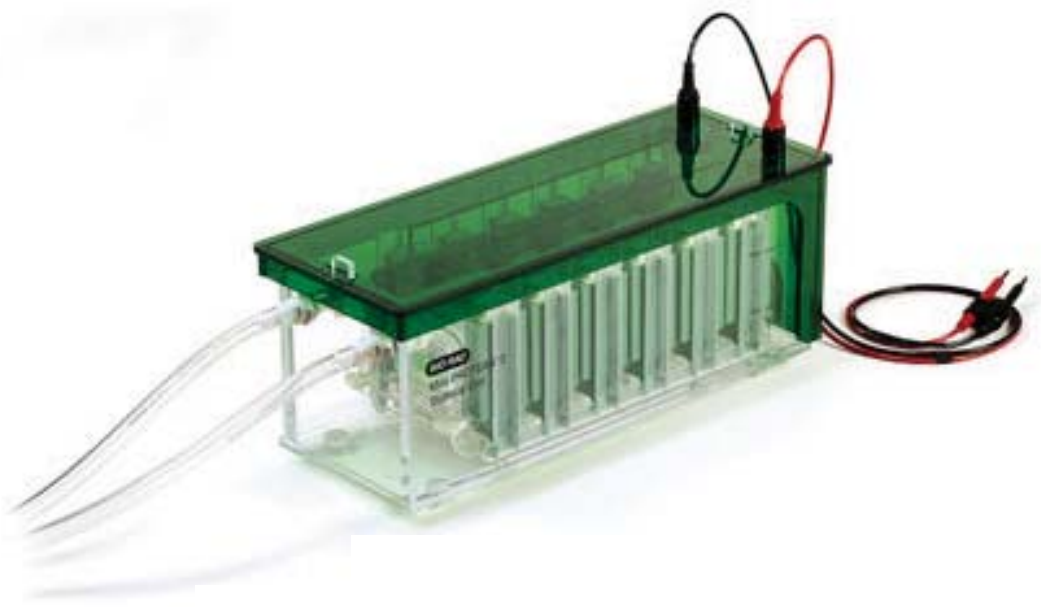


Mini-PROTEAN® 3 Dodeca™ Cell

Instruction Manual

Catalog Number
165-4100

BIO-RAD

For Technical Service Call Your Local Bio-Rad Office or in the U.S. Call **1-800-4BIORAD** (1-800-424-6723)

Table of Contents

Section 1
General Information

1.1 Introduction

The Mini-PROTEAN 3 Dodeca Cell is a multi-cell for mini-vertical gel electrophoresis. It can run four to twelve identical polyacrylamide gels simultaneously. The Dodeca cell includes clamping frames, buffer dams, a drain line, and gel releasers. Ready Gel precast gels are compatible with the Mini-PROTEAN 3 Dodeca Cell. Handcasting electrophoresis gels is also an option; see Section 5 for recommended handcast accessories.

Specifications

Tank and lid	Acrylic
Clamping frame	Polycarbonate and liquid crystal polymer
Upper electrode holder	Polycarbonate with 109 mm (43") platinum wire
Lower electrode assembly	Polycarbonate with 89 cm (35") platinum wire
Drain line	Tygon tubing
Drain line connectors	Delrin
Cooling coil	Acrylic
Cooling coil connector tubing	Tygon
Maximum buffer volume	4.4 liters
Minimum buffer volume	3.4 liters
Overall size	41.5 x 15 x 16.2 cm (L x W x H)
Precast gel compatibility	Ready Gel precast gels
Handcast gel compatibility	Mini-PROTEAN 3 glass plates and spacers
Safety limits	500V, 500W
Weight	5 kg (11 lb)

Note: Dodeca cell components are not compatible with acetone or ethanol. Use of organic solvents voids all warranties. The Dodeca cell is compatible only with aqueous reagents.

1.2 Safety

Power to the Mini-PROTEAN 3 Dodeca cell is supplied by an external DC voltage power supply (not included). The output of the power supply must be isolated from external ground to insure that the DC voltage output floats with respect to ground. All Bio-Rad power supplies meet this important safety requirement. Regardless of the power supply used, the maximum specified operating parameters for the Mini-PROTEAN 3 Dodeca cell are as follows:

500 VDC	maximum voltage limit
500 W	maximum power limit
40 °C	maximum ambient temperature limit

The current to the cell enters the unit through the lid assembly and provides a safety interlock to the user. The circuit between the cell and the lid's upper electrodes is broken when the lid is removed. Always turn off the power supply before removing the lid. **Do not attempt to use the cell without the safety lid.**

Important: This Bio-Rad product is designed and certified to meet *IEC61010-1 and EN61010-1 safety standards. Certified products are safe to use when operated in accordance with the instruction manual. This instrument should not be modified or altered in any way. Alteration of this instrument will

- Void the warranty
- Void the IEC61010-1 and EN61010-1 certifications, and
- Create a potential safety hazard

Bio-Rad is not responsible for any injury or damage caused by use of this instrument for purposes other than those for which it is intended or by modifications of the instrument not performed by Bio-Rad or an authorized agent.

*IEC61010-1 and EN61010-1 are internationally accepted electrical safety standards for laboratory instruments.

1.3 Components

To get the best performance from your Mini-PROTEAN 3 Dodeca cell, familiarize yourself with the components by assembling and disassembling the cell before using it.

Buffer tank and lid	The buffer tank and lid combine to fully enclose the inner chamber during electrophoresis. The lid cannot be removed without disrupting the electrical circuit.
Cooling coil	The integrated cooling coil chills the lower buffer chamber. The cooling core must be connected to a refrigerated circulator.
Electrode assembly	The lower buffer chamber contains the anode. The lid of the Dodeca cell houses the cathode.
Drain port	The drain port allows buffer to be removed from the tank. The drain line attaches via a quick connect fitting; this assembly is provided with the cell.
Clamping frame[1]	The clamping frame holds two gel sandwiches in place. It houses the notched gasket.
Buffer dam	The molded, one piece buffer dam is used when running only one gel in a clamping frame.
Gel releaser	The gel releaser is a small green tool that loosens the two glass plates following electrophoresis.
Gel sandwich	A spacer plate and a short plate with a polymerized gel form a gel sandwich.
Gel cassette	A Ready Gel precast gel or a handcast gel in an empty Ready Gel cassette is referred to as a gel cassette.

[1] U.S. Patent 6,436,262.

Section 2
Setting up the Dodeca Cell for Electrophoresis

2.1 Buffer preparation

Prepare 3.4–4.4 liters of running buffer (minimum/maximum) for electrophoresis. Buffer should be equilibrated to room temperature prior to electrophoresis. Premixed buffers are available (see Section 5).

2.2 Gel Preparation

Ready Gel precast gels or handcast gels using Mini-PROTEAN 3 glass plates can be run in the Mini-PROTEAN 3 Dodeca cell. Note: All gels in a single electrophoresis experiment must be of the same gel type, with the same buffer and the same % acrylamide.

2.2.1 Ready Gel Precast Gel Cassette Preparation

Note: The Mini-PROTEAN 3 Dodeca cell is guaranteed only for use with Ready Gel precast gels.

For complete Ready Gel precast gel instructions, order catalog number 161-0993, Ready Gel Applications Guide. The Applications Guide is also available on the internet at www.bio-rad.com in the literature section.

a. Take the Ready Gel precast gel out of its storage pouch.

b. With a razor blade, cut the tape along the black line at the bottom of the gel.

c. Pull the clear tape at the bottom of the Ready Gel Cassette to expose the bottom edge of the gel.

Repeat for additional Ready Gel precast gels up to a maximum of 12 gels.

Note: If an odd number of gels is to be run, use the buffer dam on the opposite side of the clamping frame with only one gel to create the clamp assembly.

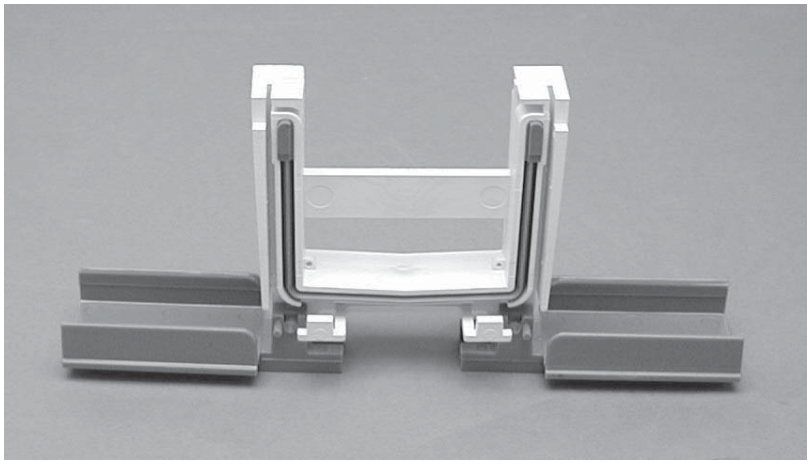
2.2.2 Handcast Gel Preparation Using Mini-PROTEAN 3 Glass Plates

a. Cast gels per the instructions with the Mini-PROTEAN 3 multi-casting chamber.

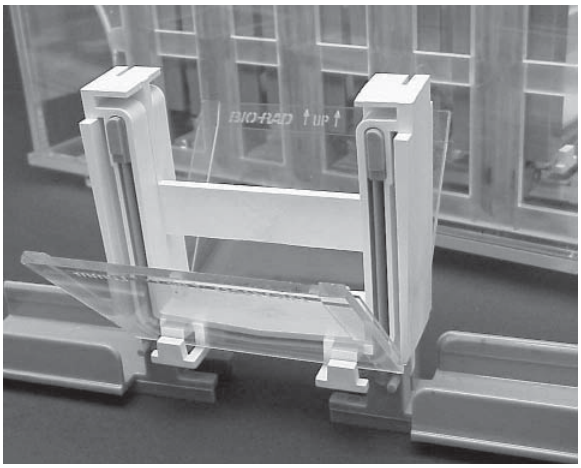
b. Remove the gels from the casting chamber or from the storage container.

2.3 Assembly

a. Set the clamping frame to the open position.

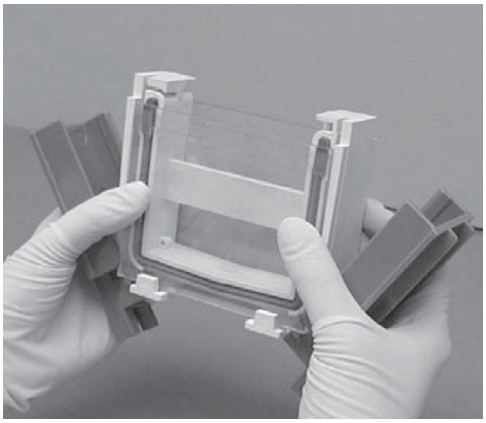


b. Place gel sandwich or gel cassette into the clamping frame with the Mini-PROTEAN 3 short plates facing inward. Note: the clamping frame requires 2 gels to create the clamp assembly. If an odd number of gels are being run, use the buffer dam.

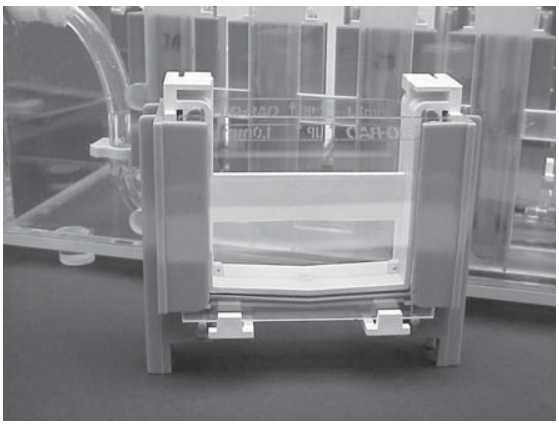


c. Lift the gel sandwiches or cassettes into place against the green gaskets. Make sure the Mini-PROTEAN 3 short plates sit just below the notch in the gasket.

d. While gently squeezing the gel sandwiches or cassettes against the green gaskets, slide the arms of the clamping frame over the gels, locking them into place. This forms the clamp assembly.



e. The arms of the clamping frame push the short plates up against the notch in the green gasket, creating a leak-proof seal. At this point, the sample wells can be washed out with running buffer, and sample can be loaded.



f. Place the Dodeca cell on a stir plate.

g. Lower each clamp assembly into one of the six slots in the tank.

h. Fill the tank with at least 2.5 liters of buffer (3.5 liters recommended) and drop a 38 mm (1.5") stir bar into the buffer tank. Do not exceed the maximum fill line. Fill each clamp assembly with 150 ml of running buffer (900 ml total for 6 clamp assemblies.)

i. Turn on the stir plate and make sure the stir bar is mixing effectively.

2.4 Required Accessories for Cooling

The Mini-PROTEAN 3 Dodeca cell has a cooling coil with quick connect fittings that must be connected to a refrigerated circulator. The refrigerated circulator must be able maintain the buffer temperature of 18–20 ºC during electrophoresis[2]. Tubing with 3/8" ID connects the Dodeca cell to the refrigerated circulator. To connect the Dodeca cell to the refrigerated circulator,

a. Insert the male fittings (supplied in a separate bag) into the tubing on the refrigerated circulator inlet and outlet tubing.

b. Connect the male fittings from the refrigerated circulator to the female fittings on the Mini-PROTEAN 3 Dodeca cell. The cooling coil in the Dodeca cell has no specific orientation, so it is not important which direction the flow goes.

   In addition to the refrigerated circulator, the Dodeca cell should be used with a stir bar and stir plate.

   By using the maximum volume of buffer, the refrigerated circulator, and the stir plate, the effects of heat on electrophoresis results are minimized.

[2] The refrigerated circulator used during development of the Dodeca cell was set to 15 ºC and had a flow rate of 3.8 liters per minute. Under these conditions and using the maximum volume of buffer, the buffer temperature during electrophoresis was maintained at 18–20 ºC.

2.5 Running the Gels

Insert the red and black color-coded power cables into the corresponding red and black jacks of the power supply. Set the power supply to run at 200 V constant for 35 minutes. For best results, use the PowerPac 200 power supply.

Application note:

The initial current draw for one 1.0 mm Ready Gel precast gel in a Laemmli formulation is about 50 mA; a Dodeca cell filled with 12 precast gels will draw 600 mA at the start of a run. The current will drop approximately 50% during the run.

Section 3
Maintenance

Mini-PROTEAN 3 Dodeca cell tank and lid, clamping frames, buffer dam, gel releaser	Rinse thoroughly with distilled water after every use.
Glass plates and combs	Wash with a laboratory detergent, then rinse thoroughly with distilled water.
Glass plates and combs (when more stringent cleaning is required)	Soak in 10 N KOH for 30 minutes (max.) and then rinse thoroughly with distilled water.

The buffer in the lower tank may be reused up to 10 times. Sodium azide (final concentration of 0.02%) is recommended to help minimize contamination.

Ultimately the number of times the buffer is used depends on the number of gels run each time and the conditions at which the runs are performed. (The primary concern is ion depletion.) Prepare fresh buffer if the following problems appear:

- changes in protein mobility
- decrease in band sharpness
- longer run times
- a band at the bottom of the gel which is difficult or impossible to destain

Section 4
Troubleshooting Guide

Problem	Cause	Solution
1. Smile effect – band pattern curves upward at both sides of the gel	a. Center of the gel running hotter than either end	a. Improper cooling. Use cooling coil and refrigerated circulator to maintain buffer temperature of 20 ºC.
	b. Power conditions are excessive	b. Decrease voltage from 200 V to 150 V or fill buffer to maximum volume.
2. Run takes unusually long time.	a. Ion depletion in running buffer (lower tank)	a. Prepare and use fresh buffer.
	b. Running buffer too concentrated in the clamp assembly	b. Check buffer protocol and dilute buffer if necessary.
3. Changes in protein mobility or band sharpness.	a. Ion depletion in running buffer (lower tank)	a. Prepare and use fresh buffer.
4. Clamp assembly leaks	a. Improper assembly	a. Be sure the green gasket is clean, free of cuts, and is inserted completely into the core.
		b. Be sure the gel sandwich or gel cassette is seated at the bottom of the clamping frame before closing the clamps.
5. SDS is precipitating.	a. Running buffer is too cold.	a. Set the refrigerated circulator to maintain a buffer temperature of 18–20 ºC.
6. Vertical streaking of protein	a. Sample overload or precipitation	a. Dilute sample, selectively remove predominant protein in the sample, or reduce voltage by about 25% to minimize streaking.
		b. The ratio of SDS to protein should be enough to coat each protein molecule, generally 1.4:1. It may require more SDS for some membrane protein samples.
7. Lateral band spreading	a. Diffusion out of the wells prior to turning on the current	a. Minimize the time between sample application and power start up.

Problem	Cause	Solution
	b. Ionic strength of sample lower than that of gel	b. Use same buffer in sample as in gel or stacking gel.
8. Skewed or distorted bands	a. Poor polymerization around the sample wells	a. Degas stacking gel solution thoroughly prior to casting; increase ammonium persulfate and TEMED concentration by 25%, wipe comb with TEMED just before casting the stacking gel, insert comb carefully to prevent trapping of air bubbles in sample well.
	b. Salts in sample.	b. Remove salts by dialysis, desalting column, etc.
	c. Uneven gel interface	c. Decrease the polymerization rate. Overlay gels very carefully.
9. Lanes constricted at the bottom of the gel.	a. Ionic strength of sample higher than that of surrounding gel.	a. Desalt sample and neighboring samples.

Section 5
Product Information and Accessories

Catalog Number	Description
Mini-PROTEAN 3 Dodeca Cell and Accessories	
165-4100	**Mini-PROTEAN 3 Dodeca Cell,** includes 6 clamping frames, 2 buffer dams, drain line, and 2 gel releasers
165-4101	**Mini-PROTEAN 3 Dodeca Cell,** with multi-casting chamber, (includes 165-4100 and 165-4110, order glass plates and combs separately)
165-4102	**Replacement clamping frame**, 1
165-4103	**Lower electrode assembly with platinum wire**
165-4104	**Replacement drain line**
165-4105	**Replacement cooling coil**, includes connector tubing
165-2948	**Replacement power cables**
165-3130	**Buffer dam**

Catalog Number	Description

Mini-PROTEAN 3 Multi-Casting Chamber

165-4110	**Mini-PROTEAN 3 multi-casting chamber**[*], includes acrylic blocks and separation sheets, tapered luer fitting, and stopcock valve
165-4114	**Acrylic blocks**, 6 mm thickness, 8
165-4115	**Separation sheets**, 15
165-4116	**Mini-PROTEAN 3 multi-casting chamber,** with glass plates[**], 0.5 mm, includes 15 sets of glass plates
165-4111	**Mini-PROTEAN 3 multi-casting chamber,** with glass plates[**], 0.75 mm, includes 15 sets of glass plates
165-4112	**Mini-PROTEAN 3 multi-casting chamber**, with glass plates[**], 1.0 mm, includes 15 sets of glass plates
165-4113	**Mini-PROTEAN 3 multi-casting chamber**, with glass plates[**], 1.5 mm, includes 15 sets of glass plates

[*] Note: Mini-PROTEAN 3 glass plates and combs must be ordered separately

[**] Note: Mini-PROTEAN 3 combs must be ordered separately

Mini-PROTEAN 3 Glass Plates and Combs

165-3308	**Short plates**, 5
165-3309	**Spacer plates with 0.5 mm spacers**, 5
165-3310	**Spacer plates with 0.75 mm spacers**, 5
165-3311	**Spacer plates with 1.0 mm spacers**, 5
165-3312	**Spacer plates with 1.5 mm spacers**, 5

Combs (2/Pack)

	0.5 mm	0.75 mm	1.0 mm	1.5 mm
5-well Comb	–	165-3352	165-3357	165-3363
9-well Comb	–	165-3353	165-3358	165-3364
10-well Comb	165-3350	165-3354	165-3359	165-3365
15-well Comb	165-3351	165-3355	165-3360	165-3366
Prep/2-D Comb	–	165-3356	165-3361	165-3367
IPG Comb	–	–	165-3362	165-3368

Catalog Number	Description
Model 485 Gradient Former	
165-4120	**Model 485 Gradient Former**, includes body with valve stem and tubing connection kit
Power Supply	
164-5052	**PowerPac™ HC power supply**, 100–120/220–240 V
Premixed electrophoresis buffers	
161-0772	**10x Tris/Glycine/SDS**, 5 L
161-0757	**10x Tris/Glycine**, 5 L
161-0760	**10x Tris/Tricine/SDS**, 6 x 1 L
161-0770	**10x Tris/Boric Acid/EDTA**, 5 L
161-0773	**50x Tris/Acetic Acid/EDTA**, 5 L

Section 6
Warranty Information

The Mini-PROTEAN 3 Dodeca cell is warranted for 1 year against defects in materials and workmanship. If any defects should occur during this warranty period, Bio-Rad Laboratories will replace the defective parts without charge. However, the following defects are specifically excluded:

- Defects caused by improper operation
- Repairs or modifications performed by anyone other than Bio-Rad Laboratories or their authorized agent.
- Damage caused by accidental misuse
- Damage caused by disaster
- Common replacement parts including platinum wire and power cables
- Damage caused by the use of organic solvents

For inquiries or to request repair service, contact your local Bio-Rad office

Warranty Information

Model _____

Catalog Number _____

Date of Delivery _____

Serial Number _____

Invoice Number _____

Purchase Order Number _____