Ordering Information for Mini-PROTEAN II Accessories

Catalog Number	Product Description
165-2941	**Mini-PROTEAN II Cell,** without combs or spacers
165-2943	**Casting Stand,** with gaskets, to cast 1 or 2 gels
165-2908	**Outer Glass Plates,** 8.3 x 10.2 cm, 10
165-2909	**Frosted Inner Glass Plates,** 7.3 x 10.2 cm, 10
165-2946	**Clamp Assembly,** to cast 1 gel
165-2933	**1.5 mm Spacers,** 4
165-2918	**1.5 mm Comb,** 5 well
165-2922	**1.5 mm Comb,** 10 well
161-0722	**Bio-Rad Cleaning Concentrate**

Stock Solutions (per liter)

A. Tris-acetate (TAE), 50x
242 g Tris base
57.1 ml glacial acetic acid
100 ml 0.5 M EDTA (pH 8.0)

B. Tris-borate (TBE), 10x
108 g Tris base
55 g boric acid
40 ml 0.5 M EDTA (pH 8.0)

C. Gel Loading Buffer, 6x
0.25% bromophenol blue
0.25% xylene cyanol FF
30% glycerol in water

References

Sambrook, J., Fritsch, E. F. and Maniatis, T., Molecular Cloning, A Laboratory Manual, 2nd edition, Cold Spring Harbor Laboratory Press, 1989.

Instructions for Using the Frosted Inner Glass Plates with the Mini-PROTEAN® II Dual Electrophoresis Cell

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9204273 Rev B

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Completely read the Mini-PROTEAN II dual slab cell instruction manual prior to preparation of the agarose gels. The frosted inner glass plate supports the vertical agarose gels used to separate high molecular weight DNA fragments. Vertical agarose gels are fragile, and require the use of 1.5 mm spacers and a 5 or 10 well comb. The Mini-PROTEAN II cell can run two agarose gels simultaneously (up to 20 samples) in as little as 45 minutes.

Procedure

Prior to assembling the frosted glass plate sandwich, be sure both the frosted inner plate and the standard outer plate are completely free of foreign particles. This can be done by cleaning the plates with Bio-Rad's cleaning concentrate or a similar product. Rinse the plates with deionized, distilled water, then follow the rinse with a

reagent grade ethanol wash. Allow the plates to dry completely.

Assemble the plates and spacers as described in the Mini-PROTEAN II instruction manual. Make sure the frosted side of the small plate is facing inward toward the outer plate and that the spacers are 1.5 mm thick.

Prepare a 1% or higher agarose solution in 1x TBE or 1x TAE buffer (see appendix for buffer recipe). The agarose concentration must be at or greater than 1%, otherwise the gel may be damaged during casting and handling.

With a 10 ml disposable pipette, pipette approximately 10 ml of hot agarose between the glass plates, to a height of approximately 0.5 cm from the top.

Slide the comb carefully between the glass plates, without forming air bubbles below or adjacent to the wells. This can be done by sliding

the comb into the agarose at an angle. The comb must be in position within 20 seconds of pouring, because the agarose will solidify quickly.

When the comb is set, the plates will begin to expand. This will cause the agarose level to drop. To correct this problem fill a pasteur pipette with hot agarose and slowly add the agarose to the gel as needed, until the agarose level stops dropping.

Allow the gel to solidify for approximately 15 minutes.

Carefully begin removing the comb from the gel by gently pushing the comb outward against the frosted plate to allow air to enter the wells. Slowly pull the comb out of the gel while gently pushing the comb outward, allowing air to enter the wells.

When the comb has been removed, remove excess agarose above the gel by scraping the outer plate with a razor blade.

Assemble the the upper buffer chamber as described in the Mini-PROTEAN II instruction manual.

Fill the upper buffer chamber with the 1x buffer used in the gel, and flush out each well with a pasteur pipette prior to sampling loading. To temporarily store an agarose gel, cover the top with the 1x buffer used in the gel. The gels should be used the same day they are prepared.

Recommended Running Conditions

DNA Fragment Size	Volts	Time
1-10 kb	75 V	45 minutes
11-23 kb	50 V	90 minutes