INSTRUCTIONS FOR "mini-gel" ELECTROPHORESIS USING THE IDEA SCIENTIFIC **MINI-SLAB**

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INTRODUCTION

The Idea Scientific MINI-SLAB complements the other "mini" and "micro" equipment found in most laboratories and has been found to increase productivity dramatically. The utility of the MINI-SLAB derives from the following features which also make it markedly different from larger format electrophoresis.

1. Electrophoresis with the MINI-SLAB is rapid (normally 20 to 90 minutes).
2. Staining and destaining times are short.
3. The system has extreme sensitivity.
4. Several gels can be polymerized at once with economic storage for later use.
5. MINI-SLAB gels can be easily dried and stored.

ITEMS YOU MAY WISH TO PURCHASE

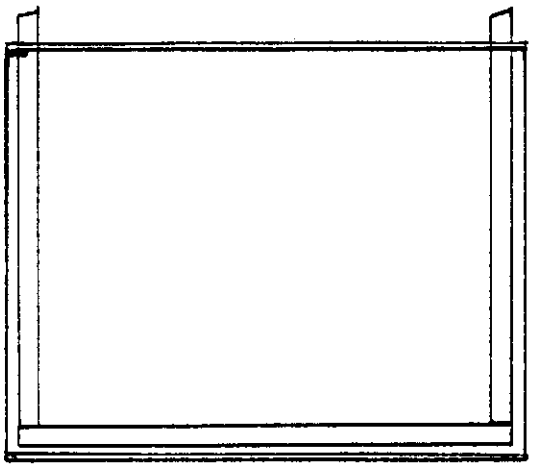
1. A Hamilton™ 700 series 10 ul syringe with a Chaney adapter works well for loading samples. Gels which are 0.8 and 1.0 mm thick can be loaded with Gel Loading Pipet Tips (Idea Scientific Product #7210).
2. Extra 10x10 cm and 10x15 cm glass plates are available from Idea Scientific. Extra 8x10 glass plates can be purchased from Arthur H. Thomas Co. (Thomas Catalog #6686-M20 is a pkg. of 12, #6686-M23 is a pkg. of 144 plates). Thomas phone # is 609-467 2000.
3. Rolls of cellophane membrane for the gel dryer are available from Idea Scientific. (Product #1080).
4. The small, rust-free binder clips are available from Idea Scientific. (Product # 1062).

CASTING OF INDIVIDUAL GELS

1. Be sure to use a comb and spacer set from the same matched set in which it was received. Idea Scientific comb and spacer sets are hand-matched so that all parts are within 0.01 mm of thickness. Using comb and spacer sets that differ by more than 0.02 mm in thickness can lead to the formation of a "skin" around a thinner comb while a comb which is too thick is difficult to force between the glass plates. Combs are made of either Teflon™ or of vinyl. Teflon™, a white pliable material, must be used if the gel is to be stored for more than a few days with the comb in it. Teflon™ combs are also easier to remove from high percentage gels. Vinyl combs which come in various colors and have been permanently non-stick coated by a special process, are easier to use than Teflon™ and can be used in all other cases. The vinyl spacers are provided in 20-cm lengths so that they can be customized for use with equipment made by other manufacturers. (To customize spacers, chop them to the desired length with a single-edged razor blade. Never use scissors, which may leave a burr.) (Note: If a comb is to be used with an agarose gel containing Ethidium Bromide, mark the comb and do not use it with non-Ethidium Bromide formulations. Ethidium Bromide inhibits the polymerization of acrylamide).

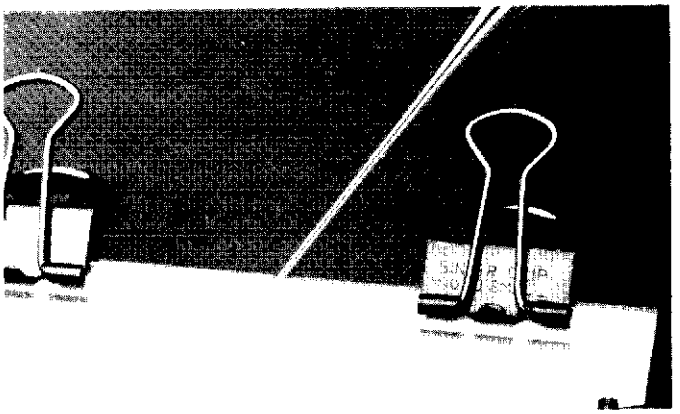
2. Trim spacers to appropriate sizes (9.6 cm long spacers work well as both the side and bottom spacers for the 10.2 cm square plates). Make a glass sandwich as shown below. Positioning the glass plates with one plate approximately 2 mm higher than the other will make sample application easier.

Position plates with rear plate 2 mm higher



Bottom spacer should be 9.6 mm long

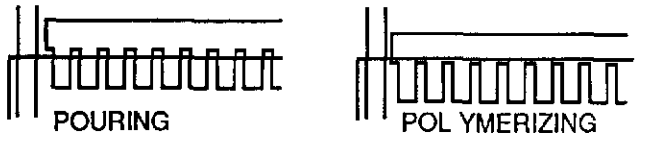
Indent all spacers 3 mm from the edges of the glass plates. Clip glass plates together with the small clips provided. Position the clips so the pressure points are directly on the spacer. (Improper positioning will bend the glass). Use at least three clips on each side, and four on the bottom. The glass sandwich will rest on the benchtop on the bottom clips. Using a pasteur pipet, put molten 1.5% agarose (dissolved in water rather than buffer) around the sandwich. (Hint: It is very handy to make a stock of 1.5% agarose and dispense 5 ml aliquots of it into 13-ml culture tubes. When agarose is needed, just loosen the top of the tube, pop the tube into the microwave, and in 15 seconds it is ready for use.) When sealing, do not use agarose which is too hot, as it will shrink away from the spacers during cooling. If necessary, allow it to cool in the pipet.



SEALING GLASS PLATES WITH AGAROSE

3. Prepare the gel formulation and pour the gel. If the gel does not have a stacking gel, the comb should be positioned between the glass plates with the top of the tooth (well-former) slightly above the glass edge. This allows air to escape between the teeth of the comb.

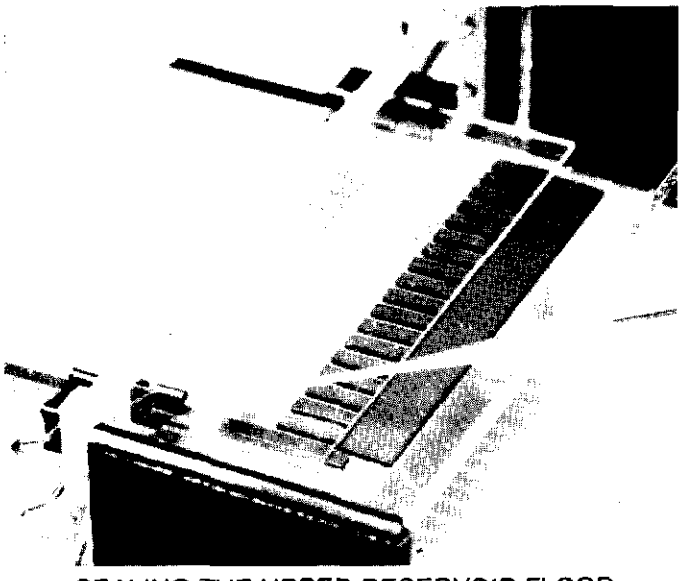
When the gel sandwich is filled with the gel solution, gently push the comb down until the teeth are completely surrounded by gel.



Note: Some scientists then position a larger clamp over the comb teeth to squeeze the liquid from between the teeth and the glass.

4. Prepare the gel for electrophoresis by removing the clips from the bottom of the gel and then removing the bottom spacer. Remove the side clips near the top of the gel and slide the gel into the slot of the upper reservoir. Position the gel with the bottom of the sample wells slightly above the slot. Clip the gel to the holder (which extends downward from the upper reservoir floor) using a clip on each side. Be sure the pressure point of each clip is directly on the spacer to avoid bending the glass. Be careful not to dislodge the side spacers when positioning the gel.

5. Hold the upper reservoir so that the gel is in a horizontal position and seal the top of the slot with molten 1.5% agarose. To do this, flow the agarose into the large front gap with a pasteur pipet and hold the gel horizontal for 10 seconds. After this, flip the gel over and seal the small rear gap. Allow the agarose to cool an additional 10 seconds and then hold the gel in its vertical running position. Check to see that the gaps at the sides of the gel are sealed and add a few drops of agarose to close any possible gaps. This process of sealing with agarose eliminates the need for gaskets.



SEALING THE UPPER RESERVOIR FLOOR

6. Put enough buffer in the lower reservoir to immerse the lower 3 mm of the gel. Do not put the upper reservoir in place yet. Clear the bubbles away from the area where the gel will be immersed. Now tip the lower reservoir so that the end is about 3 cm above the benchtop. Place the upper reservoir on the lower reservoir while it is tipped and slowly lower the apparatus to the benchtop. The bubbles at the bottom of the gel should be swept out from between the glass plates by this procedure. If bubbles remain, lift the upper reservoir, dry the gap between the glass plates with a paper towel and repeat. This immersion method eliminates the need for bubble removal with a cumbersome bent-end pasteur pipet. Pour enough buffer into the upper reservoir to be sure electrical contact with the electrode on the lid can be made. Do not add excess buffer to the lower reservoir as the gels are air cooled.

7. If necessary, clear the wells of unpolymerized gel. Load the gel. Loading the gel is easier if the plates were originally positioned with one plate higher then the other. After loading, position the cover on the apparatus, connect the cables and turn on power.

COMMON RUNNING CONDITIONS

		CONSTANT	
GEL TYPE	VOLTS	mAMPS	WATTS
SDS-PAGE	50 to 300	2 to 15	2 to 10
Native Gels	20 to 300	1 to 10	1 to 3
Nucleic Acids	40 to 300	2 to 20	1 to 5

Note: This table demonstates the wide range of running conditions possible with the MINI-SLAB. Monitor the temperature of each run to determine proper running conditions for each gel formulation. Excessive power levels may break the glass.

HINTS

1. Stacking gels can be cast in the apparatus. Just rinse off the n-Butanol overlay, seal the running gel in the apparatus, and then cast the stacking gel in place. This ensures that the side-spacers are sealed and will not leak. Use a stacking gel of 4% or more acrylamide.

2. Agarose mini vertical gels are adaptable for many uses. Warm the glass plates before pouring them. and allow them to solidify with the gel in a horizontal position. The 14-well combs have a large gap between the wells for easy removal from agarose gels. Agarose gels occasionally slide out from between the plates. Eliminate this problem by filling the area occupied by the bottom spacer with agarose and supporting the gel on a Scotch-Brite™ pad. Frosting the glass by sanding it with emery cloth will make the agarose adhere to the glass.

PUBLICATIONS

Refer to the first microslab electrophoresis paper: **Matsudaira, P.T. and Burgess, D.R.**, Anal. Biochem. **87**, (1978), pages 386 to 396., when publishing data obtained with the MINI-SLAB. A more complete bibliography can be obtained from Idea Scientific.

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