Standard Agarose Gel Electrophoresis

Maxi-gel Preparation

1. Seal the ends of the gel tray securely with strips of colored lab tape. Press the tape firmly to the edges of the gel tray to form a fluid-tight seal.

2. Level the gel tray on a leveling table or workbench.

3. Position the comb over the gel tray. For adjustable combs, adjust the height with the aid of the thumb screws so that it remains 1 to 2 mm above the base of the tray.

4. Mix agarose and melt in microwave oven.

5. When agarose is completely dissolved, allow to cool slowly.

6. After agarose has cooled to about 60°C, pour the agarose into the gel tray.

7. Allow the gel to solidify at room temperature for about 1 hour.

8. Carefully remove the comb from the solidified gel. For very low percent gels, try refrigerating the gel first to prevent damage when removing the comb.

9. Remove the tape from the edges of the gel tray.

10. Place the tray into the gel box containing buffer.

11. Submerge the gel under 2 to 6 mm buffer. Use greater depth overlay with increasing voltages to avoid pH and heat effects.

Sub-Mini Agarose Gel Electrophoresis

Micro-Gel Preparation

1. Make 400 ml of 0.7% agarose:

To make:	400 ml
0.7% agarose	2.8 g agarose
1X TBE	40 ml 10X TBE
	360 ml H2O

2. Bring agarose to a boil in a 1000 ml covered erlenmeyer flask. Boil until all clear particles are dissolved.

3. Remove from heat and allow to cool briefly.

4. Dispense in 10 ml amounts into culture tubes (40), cap the tubes with "Kim-Kaps" and seal with a strip of parafilm. Refrigerate in radioactive/flammable storage refrigerator until use.

Pouring Gels

1. To actually pour a microgel, take desired number of tubes from the refrigerator and place them in a beaker of water containing a few boiling chips. Bring water to a boil.

2. Continue boiling until all particles of agarose have dissolved.

3. Remove from heat and allow to cool to a point where they can be held by hand for enough time to pour the gel.

4. Position the gel comb over the plate(s).

5. Use the 10 ml pipet labeled "for mini gels" to transfer the solution to a small glass plate. Empty the pipet slowly so as not to cause the solution to overflow the plate.

6. Allow the gel to cool to room temperature. Store any unused gels on 2 or 3 moistened micro wipes in a large covered petri plate. They can then be stored for several days.

Separation of DNA in agarose

Agarose in gel (percent)	Efficient range of separation of linear DNA molecules (kilo-bases)
0.3	60-5.0
0.6	20-1.0
0.7	10-0.8
0.9	7-0.5
1.2	6-0.4
1.5	4-0.2
2.0	3-0.1

Gel Recipes

BRL H4 Boxes

Agarose:	0.7%	0.8%	1.0%	1.2%
250 ml gel	1.75 g	2.0 g	2.5 g	3.0 g
300 ml gel	2.10 g	2.4 g	3.0 g	3.6 g

Buffer tray volume = 1800 ml

Loading capacities

300 ml gel volume = 0.032 ml/well for 1 mm comb, 20-lane

250 ml gel volume = 0.025 ml/well for 1 mm comb, 20-lane

BioRad Slab Gel Box

Agarose:	0.7%	0.8%	1.0%	1.5%
150 ml gel	1.05 g	1.20 g	1.50 g	2.25 g

Buffer tray volume = 1300 ml

Minigel Box

Agarose:	0.7%	0.8%	1.0%	1.5%
30 ml gel	0.21 g	0.24 g	0.30 g	0.45 g

Buffer tray volume = 200 ml