

# DNA Gel Electrophoresis

<b>Rationale:</b>	
<b>Special Observations:</b>	
<b>Results:</b>	
<b>Interpretation :</b>	

<b>Experiment Date:</b>	Source: Rafael, Nathan
<b>Experiment Time:</b>	Kruer-Zerhusen, Dujduan Waraho
<b>Primary Experimenter (contact):</b>	Assembled: 6/27/2012
<b>Other Experimenters:</b>	
<b>Image Filename:</b>	

Reagent	Details	Quantity
<b>Agarose</b>		1%
<b>1X TAE</b>	Used for making gel, running buffer, and dilutions of samples	50 mL
<b>EtBr</b>	For post-staining	n/a
<b>Ladder* (premixed with dye)</b>	(brand; size)→	6 μL
<b>6X Loading Dye</b>	1 ul dye for 5 ul DNA	** XμL
<b>DNA samples</b>	See below chart for details	* μL

\*For fragments ≤1kb, use NEB 2-log ladder for clearer visualization of fragment size.

$$(X\mu L) = \frac{\mu L \text{ sample}}{4}$$

<b>Lane 1</b>	<b>Lane 2</b>	<b>Lane 3</b>	<b>Lane 4</b>	<b>Lane 5</b>	<b>Lane 6</b>	<b>Lane 7</b>	<b>Lane 8</b>	<b>Lane 9</b>	<b>Lane 10</b>
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## **Procedure:**

### **Critical Steps:**

- Swirl agarose to make sure that all is melted (but not too vigorously to avoid bubbles)

### **The following is for a 1% Gel**

**Measure 0.5 g agarose and add to flask**

**Add 50 mL 1X TAE to the flask**

**Cover flask with saran wrap (should be on top of the microwave)**

**Microwave the agarose for 1:00, or until all agarose is dissolved and the solution is completely clear and bubbling**

- o If the agarose is still suspended/undissolved, microwave for additional 10-20 seconds
- o CAREFUL, bottle will be extremely hot, watch out for steam coming from the bottle, hot gloves should be by the microwave

**Obtain gel tray, box, and comb, then rinse with dH<sub>2</sub>O**

- o Comb size depends on experiment

**Place gel tray in gel box sideways – the tray's rubber gasket should make a watertight seal against the gel box**

- o Make sure the gasket hasn't derailed

**Insert gel comb**

**Pour hot agarose solution carefully into mold**

**Use a sterile pipette tip to sweep bubbles away from the important parts of the gel, then cover the gel while it cools to prevent debris from falling in**

**Prepare DNA samples with dye & appropriate volume (at least 200 ng for visualization, avoid more than 1 ug if you need to distinguish between similarly sized fragments)**

**When agarose has completely solidified, gently pull tray out of box, rotate tray, then re-insert tray so the wells are on the black side**

**Pour 1X TAE into the box until it just covers the lanes**

**Record locations of samples, load accordingly, and run at 100 V for 50 mins (alternatively, 80V for 60-75 mins)**

**Soak in EtBr bath for 20-30 mins then image according to posted instructions**