



## BIOTECH Project

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# Agarose Gel Electrophoresis with Food Color - Teacher Guide

What is electrophoresis? In this activity, students use agarose gel electrophoresis to determine the composition of various food color mixtures. Students learn how molecules can be separated and identified by electrophoresis. This activity is a good introduction to electrophoresis that requires a minimum of special reagents, and we like to do this lab before working with gel electrophoresis of DNA.

## Classroom time needed for this lab

- 50 minutes (if agarose gels are poured before class)
- 90 minutes (if students pour the agarose gels)

## You will need to prepare the following materials (recipes to follow):

- 1 x TBE
- Agarose
- Various food colorings: Green, Red, Yellow, Blue (We use Schilling brand food color)
- Practice Loading dye

This activity is similar to another electrophoresis activity that uses biological dyes.

## You will need the following equipment and supplies:

- Micropipets and tips to load dye samples
- Small microcentrifuge tubes (0.5 ml or 0.65 ml size)
- Electrophoresis units and power supplies
- 1X TBE for electrophoresis units
- Microwave oven
- Hot water bath for keeping agarose liquified

## Setting up the classroom for this lab:

Distribute at each lab station (we use four students per group):

- 12 ul of each food color sample in small, labeled microcentrifuge tubes
- 4 small microcentrifuge tubes each containing 12 ul practice dye
- Micropipet and tips
- Electrophoresis unit and power supply (Label each electrophoresis unit with a number written on a piece of tape)

We use a central station for agarose and 1X TBE. We use a hot water bath to hold four small bottles, each containing 125 ml of 0.8% agarose. The high temperature keeps the agarose liquified. We have four large bottles at room temperature, each containing 1000 ml of 1X TBE.

### **During the lab:**

If necessary, demonstrate to students how to use the micropipet. While the agarose is solidifying in the gel tray, the students can practice using the micropipet with the practice dye. After the gel is solidified and 1X TBE is added to the electrophoresis unit, students can practice loading the gels with the practice samples. Important: Practice samples are loaded at the positive (red) end of the gel, and the dye samples are loaded in the middle set of wells.

### **After the lab:**

The 1X TBE in the electrophoresis units can be poured back into the large bottles for reuse, and the used gels can be disposed in the regular trash or taken home by the students. If the students want the gels, wrap them in plastic wrap. Unfortunately, the dyes in the gel diffuse rapidly, so the banding patterns will disappear overnight.

### **Other notes:**

1. We use the Graduate Micropipets available from Life Technologies, and we use regular yellow tips (1-200 ul). The Graduate micropipets are much less expensive than micropipets commonly found in research laboratories, and they are more likely to survive in student hands.
2. Be sure to calculate how much agarose you will need for the lab. You can add a defined volume of water into the gel tray to determine the volume of agarose that would be needed for each tray.
3. Be sure to calculate how much 1X TBE you will need

for the lab. Using a graduated cylinder, you can pour a defined volume of water into the electrophoresis unit to determine the volume of 1X TBE that would be needed for each unit.

4. If there is not enough time during the class for students to pour the gels and run the electrophoresis units, you can pour the gels before class. After the gel solidify, they can be stored in the refrigerator in a ziplock plastic with some 1X TBE buffer.

## Recipes

### 1X TBE Buffer

TBE is the buffer used in gel electrophoresis

#### Equipment

Balance  
1000 ml beaker  
1000 ml graduated cylinder  
Magnet stirrer and stir bar  
Large container for TBE solution

#### Chemicals

Tris base  
Boric acid  
EDTA  
Deionized or distilled water

1. Put 10.8 g Tris base, 5.5 g boric acid, and 0.74 g EDTA into a 1000 ml beaker.
2. Fill a graduated cylinder with 1000 ml of deionized or distilled water. Add the water to the 1000 ml beaker containing the dry chemicals.
3. Put beaker on magnetic stirrer, and add stir bar to solution. Keep stirring until chemicals are dissolved. This solution is 1X TBE.
4. Store 1X TBE buffer solution in a large container. The 1X TBE can be kept indefinitely at room temperature. This buffer can be reused several times before disposing down the sink.

### 0.8% Agarose

Agarose is the gel matrix used to separate molecules, such as DNA and dyes, during electrophoresis.

**Equipment**

Microwave oven  
250ml bottle or flask  
Plastic wrap (if using flask)  
Balance and weighing paper  
Hot water bath (or hot plate with pot of water)

**Chemicals**

Agarose powder  
1X TBE buffer solution

1. Weigh 1 gram of agarose on a folded piece of weighing paper and add to empty 250 ml bottle or flask.
2. Add 125 ml of 1X TBE buffer solution to agarose.  
Note: The container should never be filled more than half-way in order to prevent the solution from boiling over.
3. If a bottle is used, cap the bottle loosely to release air during boiling. If you using a flask, cover the opening and neck of flask with plastic wrap.
4. Mix solution by swirling. Microwave the agarose solution at high heat until powder is completely dissolved. The length of time required will vary depending on the microwave oven. The molten agarose solution should look clear (no floaties).
5. To keep the agarose liquified (for example, during several biology classes), store the bottle or flask of agarose in a hot water bath between 600 and 700C. Be sure that the bottle or flask is covered to prevent evaporation. A hot plate with a pot of water can substitute for a laboratory water bath.
6. If there is agarose left over in the container, you can let the agarose solidify and store it at room temperature until next use. Be sure that the container is well covered.

**Food color samples for electrophoresis**

The food color solutions are diluted into sugar water for loading into an agarose gel. The sugar increases the density of the sample so that it sinks to the bottom of the well.

**Equipment**

5 or 10 ml pipet  
4 small tubes  
Balance  
Weigh paper  
10 or 25 ml graduated  
cylinder

**Chemicals**

Table sugar  
Red food coloring (Schilling)  
Blue food color (Schilling)  
Yellow food color  
(Schilling)  
Green food color (Schilling)

1. Prepare a 10% sugar solution. Weigh 1 gram of sugar and add to 10 ml water. Mix to dissolve sugar in water.
2. Put 2 ml 10% sugar solution into an empty tube. Add two drops (~0.1 ml) from one of the food color bottles to the sugar solution. Mix to evenly distribute food color in sugar solution.
3. Repeat steps 1 and 2 for each food color.
4. The food color solutions can be stored indefinitely in the refrigerator.

*Practice dye sample*

You can use the blue food color sample as a practice dye sample. Prepare individual sample tubes for each student by adding 12 ul of the practice dye to a small microcentrifuge tube (0.5 or 0.65 ml size). We usually prepare one tube for each student.

*Preparing individual samples*

Before the lab, label small microtubes (0.5 or 0.65 ml size) with R, Y, B, or G. Prepare enough tubes for all the lab stations. In our labs, the students work in groups of four, and each group needs two of each of the labeled samples.

**Tube Biological Dye**

R Red  
Y Yellow  
B Blue  
G Green

Before classes start, distribute the food color samples into the proper microtubes. Each tube gets 12 ul of the dye sample, enough for a single well. Make sure each dye is added to the correct set of numbered tubes. Store samples in refrigerator

until needed.

### Sources of laboratory materials

Item	Supplier and catalog no.	
TBE buffer, dry powder, 2/pk	Intermountain C-5556-2	
Agarose	Life Technologies 15510-019	
	Genemate yellow 1-200 ul pipet tips, 1000/bag	ISC P-3200-1
Graduate micropipets, 5/pack	Life Technologies 10245-066	
Horizon 58 horizontal gel electrophoresis apparatus	Life Technologies 41060-013	
Life Technologies Electrophoresis Power Supply	Carolina P7-21-3700	
Microfuge tubes, 0.5 ml, 1000 per box	VWR 20170-310	
*Precision utility water bath	VWR 13470-030	
Corning hot plate/stirrer	VWR 33920-219	
Ohaus balance, CT series 200g	VWR 11378-059	
Tris base, 1kg	VWR JT4099-2	
Boric acid. 500 g	VWR JT4035-1	
EDTA, disodium salt, dihydrate, 100 g	Life Technologies 15576-010	

\*Hot plates can be substituted

### DATASHEET

Link to the Worksheet/Datasheet for the Electrophoresis Dyes lab

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