

Name: _____

Date: _____

A Process to Dye For

Introduction: How can a mixture of molecules, too small to be seen with even a high-powered microscope, be separated from one another? Such was the dilemma facing scientists until the development of a process that has now become standard in many laboratories world-wide- **gel electrophoresis**. Laboratories rely heavily on this proven and reliable technique for separating a wide variety of samples, from DNA used in forensics and for mapping genes, to proteins useful in determining evolutionary relationships.

Purpose: The purpose of this activity is to demonstrate the separation technique known as gel electrophoresis. This process will be used to identify dye samples by charge, molecular mass, and shape. The information will also be used to identify the composition of an “unknown” mixture of dyes.

Materials:

Electrophoresis Buffer

Agarose Gel

Dye Samples

Electrophoresis Chamber & Power Supply

Pipette and tips

Ruler

Safety Precautions

Be sure all connecting wires, terminals and work surfaces are dry before using the electrophoresis units. Electrical Hazard: Treat these units like any other electrical source- very carefully! Do not try to open the lid of the unit while the power is on. Exercise extreme caution in handling the dyes: they will readily stain clothing and skin.

Procedure:

1. Assemble the gel electrophoresis unit according to the instructions given by your teacher.
2. Place the electrophoresis unit on a flat surface of the lab table.
3. Place a sheet of white paper on the counter horizontally next to or below the chamber. Draw a rectangle representing your gel on the sheet of paper. Number your wells in your rectangle.
4. Make sure you have a gel in your chamber along with buffer solution that covers the gel.
5. Label your piece of paper according to which dye sample you are going to put in each well.

6. Withdraw 10ul of dye from each micro centrifuge tube by filling only the needle tip of the pipette. (Remember it is a very small amount) **Use a clean tip for each sample to avoid contamination.** Dispense a sample of each dye into a different well in the gel. Record the name and well number of each dye in your data table.
7. Discard all of the used pipette tips when you are finished loading all of the wells.
8. Place the lid on the chamber and connect it to the power supply. Set the power supply to 100V for about 15-20 minutes. Check on the gels frequently to make sure you do not over run your gels. When the gels are done turn off the power supply.
9. When the power is off, remove the cover and carefully remove the gel from the chamber and put it onto a white piece of paper/or a light box.
10. Measure the distance each dye migrated in millimeters (mm). This distance should be measured from the side of the well closest to the direction the dye traveled to the leading edge of each colored band. Record this number in the data table as Migration distance (mm).
11. Use the gel drawing worksheet to make an accurate drawing of the bands in the gel. Colored pencils may be helpful in identifying each band in the drawing.
12. Record the charge for each group of molecules by noting to which pole (+/-) each dye sample was traveling when the electricity was turned off.
13. Use the migration distance to complete the last column of the data table ranking the five unknown dye samples from fastest (#1) to slowest (#5). You DO NOT need to rank the unknown!!
14. Throw the paper with the gel into the trash can! Save the buffer in the chamber. Leave these at your lab station!!

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Data Table

Dye Name	Dye Well #	Migration Distance (mm)	Migration Direction (+/-)	Dye Molecules Speed Ranking

Post Lab Questions

1. Which dye(s) traveled the farthest?
2. Why didn't all the dyes travel the same distance of the same direction from the wells?
Explain your answer.
3. List the dyes that were used in your "unknown" dye sample.
4. Write one reasonable explanation to support the answer written for Question #3.

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Gel Drawing Worksheet



Sample
Wells

