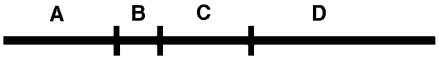
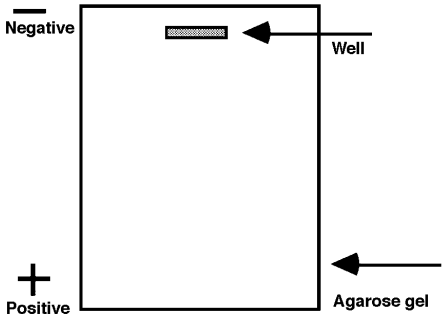
Prelab Activity 2 A Review of Electrophoresis

Agarose gel electrophoresis is a procedure used to separate DNA fragments based on their sizes. DNA is an acid and has many negative electrical charges. Scientists have used this fact to design a method that can be used to separate pieces of DNA. A solution containing a mixture of DNA fragments of variable sizes is placed into a small well formed in an agarose gel that has a texture similar to gelatin. An electric current causes the negatively-charged DNA molecules to move towards the positive electrode.

Imagine the gel as a strainer with tiny pores that allow small particles to move through it very quickly. The larger the size of the particles, however, the slower they are strained through the gel. After a period of exposure to the electrical current, the DNA fragments will sort themselves out by size. Fragments that are the same size will tend to move together through the gel and form bands.

A piece of DNA is cut into four fragments as shown in the diagram. A solution containing the four fragments is placed in a well in an agarose gel. Using the information given above, draw (to the right) how you think the fragments might be separated. Label each fragment with its corresponding letter.





Have your teacher check your diagram before you proceed.

1. Where would the larger fragments, those with the greater number of base pairs, be located, toward the top of the gel or the bottom? Why?

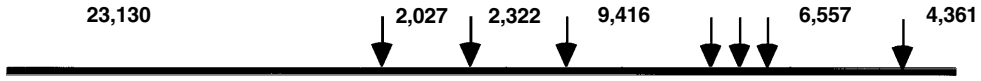
2. Suppose you had 500 pieces of each of the four fragments, how would the gel appear?

3. If it were possible to weigh each of the fragments, which one would be the heaviest? Why?

4. Complete this rule for the movement of DNA fragments through an agarose gel.

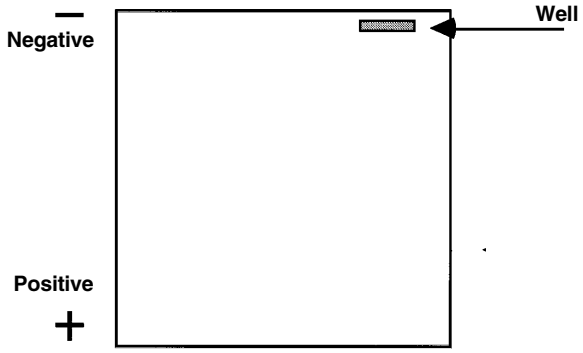
   The larger the DNA fragment, the …

This diagram represents a piece of DNA cut with *Hin*dIII at each of the restriction sites pointed to by the arrows. The numbers represent the number of base pairs in each fragment.



5. How many fragments were produced by the restriction enzyme *Hin*dIII?

6. On the gel diagram, show how you believe these fragments will sort out during electrophoresis. The two fragments with no length indicated will be too small to be visualized on the gel.



7. Label each fragment with its correct number of base pairs.