

Tree Markers

Name _____ Date __/__/__

For this investigation you will be using *lambda* DNA to simulate tree DNA. You will run a gel electrophoresis on the DNA looking at a marker that signifies that the tree has the gene for frost resistance.

Materials

- Micropipette
- Electrophoresis gel box and power supply
- Micropipette Tips
- 18 micro liters of Tree one DNA (Lambda DNA/EcoR I digest)
- 18 micro liters of Tree two DNA (Lambda DNA/EcoR I, Hind III digest)
- 18 micro liters of Tree three DNA (Lambda DNA /BstE II digest)
- 18 Micro liters of Tree four DNA (Lambda DNA/ Hind III digest)
- 18 Micro liters of DNA Loading Dye
- 18 Micro liters of DNA Lane Marker
- Agargose
- Carolina Blu Stain for electrophoresis
- TBE
- Distilled water
- Vinyl gloves
- Safety glasses

Safety

- Carolina Blu will stain your hands and clothing.
- Wear safety glasses throughout the investigation.

Procedure

Loading the DNA into Gels

Use a micropipette to load the contents of each DNA tubes into separate wells in the gel. Be sure to use a new tip for each tube.

1. Pour liquid agargose into the gel chamber, allow to set.
2. Pour TBE buffer into gel box, just until it covers the solid gel.
3. Draw 18 μ L of the DNA Lane Marker into the pipette.
4. Using two hands, steady the pipette over the first well in the gel.
5. Carefully lower the pipette into the well, don't go too deep and puncture through the bottom.
6. Slowly expel the pipette contents into the well.
7. Repeat procedures 1-4 for the Tree one DNA, except use the second well.
8. Repeat the procedures 1-4 for the Tree two DNA, except use the third well.
9. Repeat the procedures 1-4 for the Tree three DNA, except use the fourth well.
10. Repeat the procedures 1-4 for the Tree four DNA, except use the fifth well.

Electrophoresis

1. Close the top of the chamber. Take the box to an electrical control station. Attach the black leads to the black cord and the red leads to the red cord.
2. After two chambers are connected to the station turn the power on and set the voltage to 165. If the system is connected correctly, the buffer solution will begin to bubble.
3. The loading dye will eventually resolve into two bands of color. The faster-moving, purplish band is the dye bromophenol blue. The slower-moving, aqua band is xylene cyanol. BP blue migrates through gel at the same rate as a DNA fragment of approximately 300 base pairs long. Xylene cyanol migrates at a rate equivalent to approximately 2000 base pairs.
4. Electrophoresis until the BP blue band is about 2cm from the end of the gel.
5. Turn the power off and disconnect the leads.
6. Carefully remove the casting tray and slide the gel onto a staining plate.
7. Stain the gel.

Name _____ Date __/__/__ Period __

1. Sketch what your gel looks like in the space below

2. Compare the lines for the four tree DNA types. The marker for frost resistance is found at the fourth row. Which tree(s) samples have the frost resistance gene marker? How confident are you with your results?

3. What information about your results should you share with your colleagues?

4. Do the results of your electrophoresis guarantee that the trees that have the frost resistant gene marker will pass this trait on to their offspring? Explain your answer.