

# Quick Guide for DNA Fingerprinting Kit

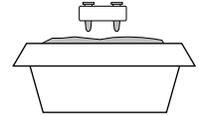
## Day 1 Preparing the DNA Samples

- Place the tube containing the restriction enzyme mix, labeled ENZ, on ice.
- Label one of each colored microtube as follows:
 

green	CS = crime scene
blue	S1 = suspect 1
orange	S2 = suspect 2
violet	S3 = suspect 3
red	S4 = suspect 4
yellow	S5 = suspect 5

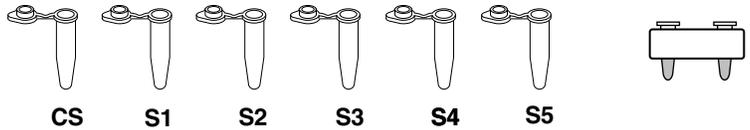


ENZ

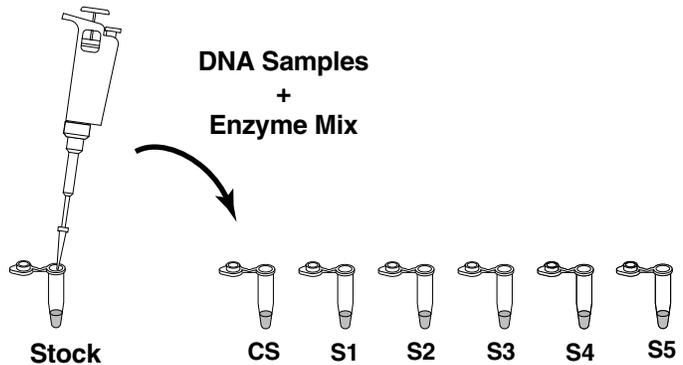


Ice

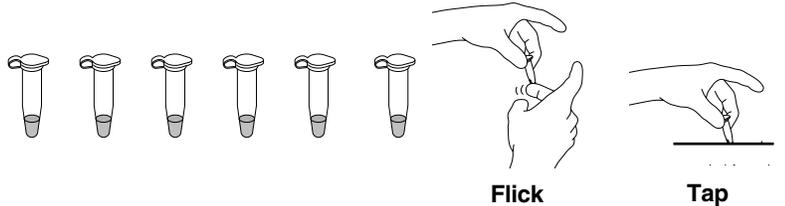
Label the tubes with your name, date, and lab period. Place the tubes in the foam microtube rack.



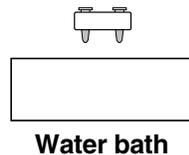
- Pipet 10  $\mu$ l of each DNA sample from the stock tubes and transfer to the corresponding colored microtubes. Use a separate tip for each DNA sample. Make sure the sample is transferred to the bottom of the tubes.
- Pipet 10  $\mu$ l of enzyme mix (ENZ) into the very bottom of each tube. Use a separate tip for each ENZ sample.



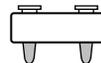
- Cap the tubes and mix the components by gently flicking the tubes with your finger. If a microcentrifuge is available, pulse spin in the centrifuge to collect all the liquid in the bottom of the tube. Otherwise, tap the tube on a table top.



- Place the tubes in the floating rack and incubate 45 min at 37 °C or overnight at room temperature in a large volume of water heated to 37 °C.

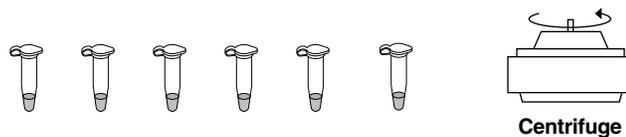


- After the incubation period, remove the tubes from the water bath and place in the refrigerator until the next laboratory period.

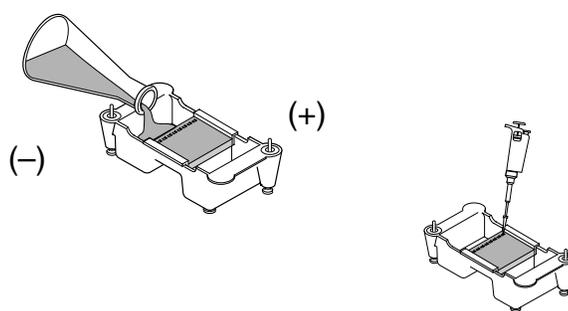
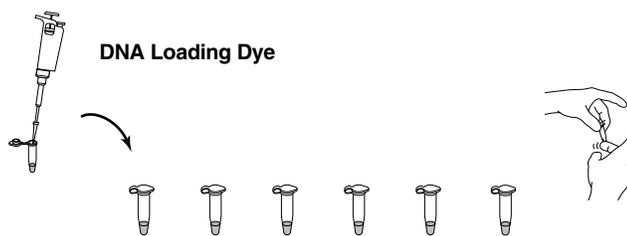


## Day 2 Gel Electrophoresis

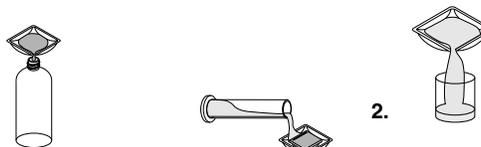
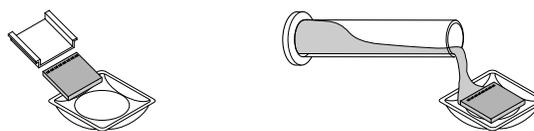
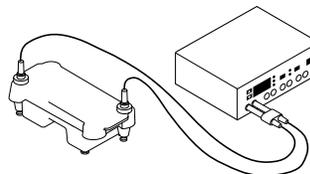
1. Remove your digested DNA samples from the refrigerator. If a centrifuge is available, pulse spin the tubes in the centrifuge to bring all of the liquid into the bottom of the tube.
2. Using a separate tip for each sample, add 5  $\mu$ l of loading dye "LD" into each tube. Cap the tubes and mix by gently flicking the tube with your finger.
3. Place an agarose gel in the electrophoresis apparatus. Fill the electrophoresis chamber with 1x TAE buffer to cover the gel, using approximately 275 ml of buffer.
4. Check that the wells of the agarose gels are near the black (-) electrode and the base of the gel is near the red (+) electrode.
5. Using a separate tip for each sample, load the indicated volume of each sample into 7 wells of the gel in the following order:
  - Lane 1: M, DNA size marker, 10  $\mu$ l
  - Lane 2: CS, green, 20  $\mu$ l
  - Lane 3: S1, blue, 20  $\mu$ l
  - Lane 4: S2, orange, 20  $\mu$ l
  - Lane 5: S3, violet, 20  $\mu$ l
  - Lane 6: S4, red, 20  $\mu$ l
  - Lane 7: S5, yellow, 20  $\mu$ l



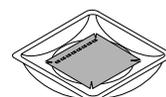
Centrifuge



6. Place the lid on the electrophoresis chamber. The lid will attach to the base in only one orientation. The red and black jacks on the lid will match with the red and black jacks on the base. Plug the electrodes into the power supply.
7. Turn on the power and electrophorese your samples at 100 V for 30 minutes.
8. When the electrophoresis is complete, turn off the power and remove the top of the gel box. Carefully remove the gel and tray from the gel box. Be careful—the gel is very slippery! Slide the gel into the staining tray.
9. Add 60 ml of DNA stain to the tray. Cover the tray with plastic wrap. Let the gel stain overnight, with shaking for best results.



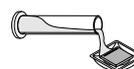
1.



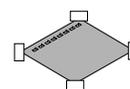
3.

## Day 3 Analysis of the Gel

1. Pour off the DNA stain into a bottle. Add 60 ml of water to the gel and let the gel destain 15 minutes.
2. Pour off the water into a waste beaker. Analyze the results with the help of your teacher.
3. Let the gel dry on gel support film or on your lab bench until completely dry. When the gel is dry, tape into your lab notebook for a permanent record.



2.



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