20 Questions to master inquiry

DNA Fingerprinting Kit

Big Idea 3

Mastering Inquiry can be easy with Bio-Rad

Use the following 20 Questions for student based inquiry about the processes contained in this kit. Whenever possible let your students come up with protocols and let them choose the variables to test.

Level 1 questions are simple to adapt and do not add extra days onto running this lab. An example of how to organize and execute a Level 1 question is given below.

Level 2 questions may add a few days onto the lab and may require a few additional materials to complete.

Level 3 questions are for students seeking a real challenge and will require additional days, techniques, and materials to answer.

Example

Level 1, Question #9: How much DNA is needed to be able to stain a gel with Fast Blast or other “safe” stains?

Have students do dilutions on the DNA to determine at what level the DNA is no longer visible. One way to set this experiment up is:

- Have 2 student groups load the normal 20 ul of DNA sample into their gels (loading dye already added).
- Have 2 student groups remove 15 ul of sample + loading dye into new tubes. Dilute by adding 5 ul of 1x loading dye.
- Have 2 student groups remove 10 ul of sample + loading dye into new tubes. Dilute by adding 10 ul of 1x loading dye.
- Have 2 student groups remove 5 ul of sample + loading dye into new tubes. Dilute by adding 15 ul of 1x loading dye.

Compare band intensity between gels. Left over samples can be combined to refine experiment.

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Level 1 Questions

1. How important is enzyme concentration for a DNA digest?
2. How important is DNA concentration (substrate) for DNA digest?
3. How important is digest time?
4. How important is digest temperature?
5. How important is thoroughly mixing the sample prior to digest?
6. How important is agarose concentration in the gel?
7. How important is buffer concentration in the chamber?
8. How important is voltage at which the gel is run?
9. How much DNA is needed to be able to stain a gel with FAST Blast or other 'safe' stains?

Level 2 Questions

11. How important is restriction enzyme concentration when adding more than one enzyme to the same tube?
12. How important is overall reaction volume?
13. How important is restriction buffer concentration when doing a restriction digest?
14. Are enzymes as effective after exposure to UV light?
15. Can I mutate DNA using UV light? Does this change restriction sites?

Level 3 Questions

16. If I cut the DNA sample and then ligate together followed by another restriction digest will I get the same restriction pattern?
17. Can I ligate precut samples together to make a plasmid?
18. Can I cut a band out of the gel and ligate it into a plasmid?
19. Can I make a restriction map of a known plasmid using multiple restriction enzymes?
20. Some enzymes exhibit star activity when reaction conditions are not optimal. How can I determine if or when an enzyme exhibits star activity?