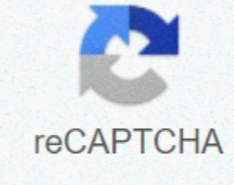




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1.3.1 student response sheet answers

Introduction Bones can provide a snapshot of the identity of a person- they can predict height, growth, gender, ethnicity and even age. However, that is what lies inside these hard calcified tissues, DNA placed inside the body's cells, that holds the key to true genetic identity. Tissue consists of many cells, the building blocks of life. Hidden inside the cells of the body, you will find chromosomes. These structures house your genes and contain the DNA code necessary for the production of all the proteins that keep your body functioning. Your DNA provides a unique code of over three billion base pairs. Unless you are an identical twin, there is no other person on the planet with the same code. And although only a tenth of a percent of this DNA differs from person to person (there are still 3 million base pairs!), the regions that vary provide a true genetic blueprint of a person. This wonderful molecule is small - invisible to the naked eye - but it is often the only key that can link murderers to a crime, parents of their children or a person to their own bones. In PBS, you learned about the molecular biology techniques that allow scientists to explore our DNA. PCR, Polymerase Chain Reaction, is the copier; the revolutionary process that allows scientists to recreate even the smallest stain of DNA. Limiting endonucleases (enzymes) are molecular scissors that can cut DNA in specific locations. Your specific code determines how many times this set of scissors will cut and the number and size of DNA pieces that will be left behind. These pieces can then be separated and compared using the process of gel electrophoresis. As these fragments move, their varying lengths drive them through the gel at different speeds. Scientists can use these RFLPs, Restriction Fragment Length Polymorphisms, a set of DNA puzzle pieces unique to you, to create a pattern called a DNA fingerprint. Like the unique fingerprint from your hands, this DNA fingerprint provides important information about human identity, giving you at the slightest level a clue as to what makes you. Using the DNA of the ID skeleton in lesson 2, you used basic forensic anthropology to analyze bones and to provide a preliminary snapshot of the two individuals found in the park. Using the tracks you have uncovered, the local police have run these descriptions through their missing personal files. Two people matching the description for each skeleton have been reported missing in the past year. You will now work as a forensic DNA analyst to evaluate DNA samples found in the bones of the skeletons and compare each unique DNA fingerprint with the genetic material of those who have disappeared. In this activity, you will continue working with the skeleton your team analyzed in 2. You will compare the DNA found inside the bones of the skeleton with with people who match the profile you entered. DNA samples provided by the family of those who have disappeared will each be cut, or digested, with two restriction enzymes in separate reactions and will be compared to DNA isolated from the humerus of the uncovered skeleton. DNA extracted from the skeleton has already been digested with the same two enzymes. DNA work takes care and precision. Work carefully to identify these individuals and ultimately give their families some peace. Equipment Computer with Internet Access Laboratory journal Edvotek DNA Detectives Kit Predigested DNA from skeletal DNA samples from missing persons #1 and #2 Restriction enzymes (EcoRI and HindIII) Reaction buffer Microcentrifuge tubes 37 ° C water bath An agarose gel (0.8% agarose; 8 Wells) Tris-Acetate-EDTA (TAE) Gel Electrophoresis Buffer Gel Electrophoresis Micropipettor (20 µl) Disposable Micropipette Tips Light Box Safety Glasses Lab Apron Activity 1.3.1 Student Resource Sheet Procedure PART I: DNA digestion with limitation enzymes 1. Analyze the DNA of the two people who have been linked to your skeleton: Missing Person 1 and Missing Person 2. 2. Get four clean microcentrifuge tubes. All other reagents should be located at the laboratory station. 3. Note microcentrifuge tubes 1-4 for four limitation enzyme digestive reactions. Make sure you mark each tube with the name of your group. Also be sure to mark the contents of each pipe in the laboratory journal. You will process four DNA samples: Tube 1- Missing Person 1 DNA cut with Enzyme 1 Tube 2- Missing Person 1 DNA cut with Enzyme 2 Tube 3- Missing Person 2 DNA cut with Enzyme 1 Tube 4- Missing Person 2 DNA cut with Enzyme 2 PLTW LABORATORY JOURNAL Record the each contents of the tube in your laboratory journal. 4. Use the micropipettor to dispense 10µl enzyme reaction buffer in each of the four marked tubes. 5. Add the DNA and enzymes to tubes 1-4 according to the following information. Each tube will have a final volume of 40µl. NOTE: Remember to use a fresh pipette tip for each transfer. You dont want to contaminate your samples. Tube 1- 15µl Missing Person 1 DNA + 15µl Enzyme 1 Tube 2- 15µl Missing Person 1 DNA + 15µl Enzyme 2 Tube 3- 15µl Missing Person 2 DNA + 15µl Enzyme 1 Tube 4- 15µl Missing Person 2 DNA + 15µl Enzyme 2 6. Cap each tube and gently knock on the side of each tube to mix. Gently tap the tube on your desk to make sure all the contents are at the bottom of the tube. 7. Incubate your pipes in a 37°C water bath for 45-60 minutes. Place the pipes in the bath as directed by your teacher. At the end of the incubation period, you need to add 5µl 10X gel load solution to the tubes 1-4 to stop the reactions. Depending on time, your instructor can complete this step for you. While your samples are digesting, continue with steps 8 -10. 8. Get a Resource sheet from your teacher. 9. With your partner, review the science behind the limitations. Use the following website to complete the questions and activities in Part A of the Student Response Sheet. DolanDNALearningCenter: Limitation enzymes 10. Review the process of gel electrophoresis by viewing the animations listed below. Use information from the websites to complete the activity described in part B of the student response sheet. PART II: Gel electrophoresis of restriction fragments Now that you have digested your DNA, you need to analyze smaples using gel electrophoresis. 11. If you make the gel, follow the teacher's instructions to melt and pour agarose. 12. Carefully remove the comb from the agarose gel and place the gel in the electrophoresis chamber. 13. Fill the chamber with TAE buffer and make sure the gel is completely submerged. 14. Get the test tubes 1-4. Gently flick on the side of each tube to mix the contents. NOTE: Digested DNA samples from the skeleton and standard DNA marker will be placed at each laboratory station or table. 15. Warm samples, including standard DNA marker and DNA extracted from the skeleton, for two minutes at 65°C. Your teacher will refer you to the heating block or water bath. NOTE: Allow the samples to cool down for a few minutes before placing them on the gel. 16. While the samples are cooling, you can practice loading samples on the training gels. 17. When ready, put 40µl of each sample into the gel. Be sure to use a new tip for each sample. Lane 1: Standard DNA Lane 2: DNA from skeleton digested with Enzyme 1 Lane 3: DNA from skeleton digested with Enzyme 2 Lane 4: Tube 1- Missing Person 1 / Enzyme 1 Lane 5: Tube 2- Missing Person 1 / Enzyme 2 Lane 6: Tube 3- Missing Person 2 / Enzyme 1 Lane 7: Tube 4- Missing Person 2 / Enzyme 2 2.18. Draw a diagram of the gel in the portable lab. Be sure to clearly specify which sample is in which good. 19. Follow the teacher's instructions to install the gel electrophoresis unit and connect the power supply. Be sure to check the direction of your gel. REMEMBER: The DNA containing wells should be closer to the negative pole and further away from the positive pole. 20. Follow the teacher's instructions to turn on the power supply. 21. Check the DNA samples 5 minutes after turning on the power supply. Make sure that the load color migrates out of the well and moves towards the positive rod. 22. Check the gel every 10 minutes. NOTE: The gel should take 30-45 minutes to run. During this time, continue work on the Students Resource List. 23. When the gel has finished running, follow the teacher's instructions to stain and visualize the DNA fragments. 24. Examine your results and analyze the pattern of fragments you see. PLTW LABORATORY JOURNAL MAKES sketch of the gel. Do the DNA fingerprints from the bone sample match up with one of the missing? Explain your findings. REFLECTION QUESTION: Suggest reasons why it was useful to digest each of your samples with two different restriction enzymes? How does the results of the gel reinforce this point? 26. Return to the case report created by the forensic anthropologist. Add a new headline: DNA analysis: Referring to the results of gel electrophoresis, you need to clearly describe the findings in the DNA analysis. Make sure that your explanation is clear (and well supported by what you see in the gel) and give a clear conclusion about the identity of the skeleton in the park. If possible, attach a marked photo of your gel to the finished case report. Final conclusion: Use information from the whole case to summarize the identification experience. Conclusion 1. Write a paragraph explaining what you want to say to the other family to convince them that the science techniques used prove that the bones do not belong to their loved ones. 2. Explain how the code in your DNA relates to your physical appearance, as well as your body's function. How can a change in this code affect the body? 3. Other than information from bones and from DNA analysis, what other characteristics/identifiers can be used to identify this skeleton? 1571457599 10/18/2019 23:59 23:59