Name \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Period \_\_\_\_\_\_\_

**Accelerated Biology**

**"Seeking Justice"**

Watch the video clip, and read the following information below in order to answer the Pre–Lab questions on the following page. http://www.youtube.com/watch?v=RLpalVirlTk

**Introduction**

A DNA profile or DNA ‘fingerprint’ is different for every single person, except identical twins. DNA profiles can be produced from biological samples of hair, skin, or blood. They can be used to identify whom the sample came from by comparing it to a number of different people’s profiles and matching it. Police use DNA profiling to determine who was present at a crime scene.

To produce a DNA profile, one must look to areas of the DNA sequences that contain differences among individuals. These areas are called [polymorphisms](http://www.biotechnologyonline.gov.au/topitems/glossary.html#polymorphism) (poly = many, morph = form). These are often introns or other non–coding regions of DNA. Two different methods are often used to make the profile.

**Method 1 – RFLP Analysis**

RFLP (Restriction Fragment Length Polymorphism) was what you used to make the gel in the paper DNA lab to identify the parents of Baby X. What this means is that *Restriction* enzymes made different *Length Fragments* for each individual.

As you should recall, restriction enzymes cut very specifically between bases in a sequence. For example, the enzyme EcoRI cuts between the guanine (G) and the adenine (A) in the sequence GAATTC. Because no two people have exactly the same sequence of bases in their DNA (except identical twins), the cuts will produce DNA pieces of different lengths. When the DNA pieces are separated on an electrophoresis gel, the resulting pattern is a bit like a strip of bands of different thicknesses at different distances from each other. This pattern is called a DNA profile. Probes are used to identify where specific sequences are on the DNA fragments formed.

If only a small amount of DNA is available, PCR (Polymerase Chain Reaction) can be used to amplify, or copy, the sequence before the restriction enzymes are added. PCR mimics DNA replication that occurs naturally within cells, but at a much faster pace.

**Method 2 – STR Analysis**

Another technique was developed that would allow smaller samples of DNA to be analyzed without the use of probes. Just one nanogram of DNA is usually a sufficient quantity to provide good data.

There are sections in our DNA where a sequence of bases is repeated a number of times. For example: GTAC GTAC GTAC GTAC GTAC GTAC. These are called [short tandem repeats](http://www.biotechnologyonline.gov.au/topitems/glossary.html#STR) (STRs). The number of repeats within an STR varies between individuals in a population. To produce a DNA profile, several known STRs are selected and only those sequences are copied using [polymerase chain reaction](http://www.biotechnologyonline.gov.au/topitems/glossary.html#polymerasechainreaction) (PCR). Millions of copies of the selected STRs are produced. Since they have different lengths in different people, when run on a gel with electrophoresis, they separate forming a unique banding pattern.

The Federal Bureau of Investigation (FBI) has identified 13 core STR loci (places in DNA) that are now routinely used in the identification of individuals in the United States, and Interpol has identified 10 standard loci for the United Kingdom and Europe. These loci are used commonly in criminal investigations to match crime scene data to suspect data. The current DNA database maintained by the FBI, known as the Combined DNA Index System (CODIS), contains case samples (DNA samples from crime scenes or "rape kits") and individuals' samples (collected from convicted felons or arrestees) that are compared automatically by the system's software as new samples are entered. Contrary to how DNA analysis is portrayed on popular television shows, DNA samples are not analyzed within the course of an hour. Rather, the U.S. currently has an enormous backlog of samples waiting to be typed and entered into the database.

**Pre–Lab Questions**

After having read the information above and watching the video clip, answer the following questions.

1. Why are regions that are non–coding used for profiling rather than segments of DNA that code for needed proteins?
2. Why is a DNA profile called a “fingerprint?”
3. Other than helping to convict criminals, what are 4 other uses of DNA profiling?
4. What is the function of restriction enzymes?
5. Why is RFLP analysis less useful than using STRs?
6. Can contamination of a sample be used as an argument against identification of a criminal? Explain. (this one is strictly answered in the video)
7. What is the function of PCR?
8. How many loci (or markers) are used by the FBI in a DNA profile?
9. What does CODIS stand for? What is it?
10. Now read the case below and the procedure so that you know what you will be doing in lab! To get good results, it is important to work efficiently. KNOW WHAT YOU ARE DOING!

**The Case**

A horrible crime has been committed. A 33 year–old woman was found stabbed to death in her home in an affluent suburb outside of Chicago. Time of death was determined to be 2am. She had just finalized a divorce from her husband the previous day. Her 2 young children were at their grandparents’ house and the woman had spent a night out with friends. In addition to the large amounts of blood found near the victim’s body, smaller amounts of blood and tissue were found under her finger nails and on the knife found to be the murder weapon, left on the floor. Preliminary typing identified the blood under the fingernails as Type A. The victim’s blood was Type B.

**The Suspects**

The investigators were very interested in the whereabouts of her ex–husband. He claims to have been at home all night alone. He had a prior record after having been arrested for assault 3 years ago when he initiated a bar fight with a man he found flirting with his wife. He was convicted and is currently serving a suspended sentence.

In addition, a traffic stop was made after a neighbor called police upon seeing a suspicious looking 1995 Dodge Neon stopped in front of the victim’s house around midnight. Most vehicles in the neighborhood were newer models and the neighbor claimed she did not like the look of the scruffy young man driving the car. When police questioned the man, they found a knife in the trunk of the car along with other tools and $1500 in cash. The young man claimed that he had been doing some free–lance work in the area and had gotten lost on his way home and had stopped to look at a map. The money, he claimed, was from the clients he had done work for. The clients he claims to have worked for, he says, left for vacation and are not reachable. Upon looking up his record, they found he was released from prison 10 months prior after serving time for armed robbery.

Not convinced that either suspect would be the murderer, the investigators also looked to the registered offenders in the area. They found a newly released felon who lived in the next town but frequented the bar the victim was at earlier that night. Though there was no reason to arrest the man, they could match the DNA to that on file due to his previous crimes. His DNA is not yet in the database, but is available for testing.

The case caused a media frenzy. Friends of the victim swear her ex–husband murdered her, while friends of the husband claim to have seen the young man near the house on several occasions. The court of public opinion already has the ex–husband convicted. You must run the DNA samples and compare them to find out which person’s DNA was found at the crime scene.

**Procedure**

1. Ms. Lubecke has already prepared the agarose gel for you! You’re welcome ☺ CARFULLY, place the gel into the chamber. If you do not handle it with care, it could rip and you will not get good results.
2. You have 5 microtubes with DNA samples in them. The colors and where the DNA was obtained from are listed in the table below. Load the DNA into the well exactly as depicted in the diagram below. NOTICE: there will be 3 extra wells. Also label the positive and negative ends of the gel.

Well 1 – Orange

Well 2 – Green

Well 3 – Purple

Well 4 – Yellow

Well 5 – Blue

The last three wells will remain empty!

|  |  |  |
| --- | --- | --- |
| **Well #** | **Color** | **DNA Sample Taken From** |
| 1 | Orange | Standard DNA |
| 2 | Green | Crime scene |
| 3 | Purple | Ex–husband |
| 4 | Yellow | Registered Felon |
| 5 | Blue | Scruffy Man in Dodge Neon |

\_\_\_ Charge

\_\_\_ Charge

1. When you are finished loading the wells’ raise your hand and Ms. Lubecke will pour in the buffer. Attach your chamber to the power source. The power supply should be set at 180 volts.
2. Label 2 staining trays with label tape. Write your names and class period on them.

**While the gel is running, answer the following questions.**

1. Based on what you have read above, if there were no DNA evidence, who do you think would most likely be convicted of the crime? Why?
2. When you get the results back tomorrow, how will you know who’s DNA matches the crime scene?
3. Look at the gel. It should be foggy, but you should see a blue substance in each lane moving down the gel. What is the function of this dye? (Hint, it is not to see the DNA.)
4. What causes the DNA move down the gel? Be specific!
5. What can you infer about the DNA fragments based on how far they move in the gel. Explain.
6. Turn off your power supply when instructed. Carefully remove the gel from the chamber and place it into the staining tray. Cover the tray with Saran wrap® and place it where Ms. Lubecke instructs you to. Ms. Lubecke will apply the stain for you. You will see the results tomorrow in class.

**Analysis Questions**

The gel has been stained and probes have been added. Draw the results that you see on the picture of the gel on provided below. TYPE your answers to the following questions for h.w.

1. Whose DNA was found under the fingernails of the victim and on the knife?
2. What does this evidence tell you about that suspect?
3. What is some other evidence that will be necessary to convict the person of the murder?
4. What can you use the standard to determine?
5. Imagine a crime where there were no suspects. Why is CODIS important?
6. There are many false convictions – people convicted of crimes they did not commit. What do you think these convictions are based upon?