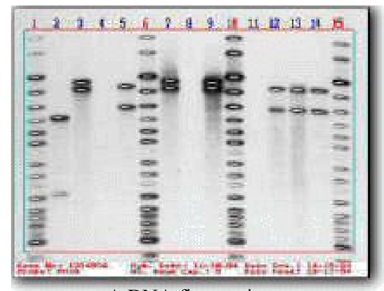
DNA Fingerprinting-What it is and how it works


A fingerprint

You probably know that a fingerprint is a mark left by a person's fingers. Everyone has a unique fingerprint, so fingerprints can be used to identify a person. If a criminal leaves a fingerprint at a crime scene, that fingerprint could be the evidence that convicts the criminal of the crime.


A DNA fingerprint

In some ways, a DNA fingerprint is like a regular fingerprint. Remember that the DNA sequence of everyone is a little different (except for identical twins.) If some blood, hair, semen, or skin cell evidence is left at the scene of a crime, a DNA fingerprint can be used to determine who left the evidence. DNA fingerprinting can also be used to determine relationships between people. For example, a DNA fingerprint can help determine who is the father of a particular child.

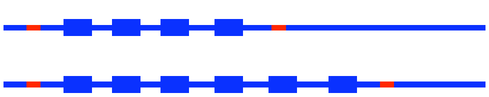
There are some important differences between DNA fingerprinting and regular fingerprinting. With regular fingerprints, just one good print is needed to match to a person. DNA fingerprinting is more complicated. When you studied the basic structure of DNA, you learned that all DNA molecules are the same. All DNA has deoxyribose sugar, phosphate groups and the four bases A, T, C and G. What makes everyone different is the order and number of each base pair in their DNA. If we were to look at every single base pair in a person's DNA, we would find that no two people have exactly the same sequence (except for---whom?). The problem is that the number of base pairs in our DNA is so huge—many billions of letters-that it would take too long to look at all of the DNA.

VNTR Fingerprinting

Instead of looking at all the DNA when a DNA fingerprint is made, certain special areas of DNA are used. These areas are believed to be parts of the DNA that do not code for any genes, but can be very different in different people. These areas of DNA have repeats of the same basic sequence, for example GATCGG repeated over and over again. These repeats are next to each other on the DNA molecule. This is called being "in tandem" because it is like two people riding on a tandem bicycle (a bicycle built for two.) Just like people on a tandem bicycle ride next to each other, tandem DNA sequences are found next to each other in the DNA, like this: GATCGG GATCGG GATCGG. How many repeats of the sequence are there? You should have counted three repeats of the GATCGG sequence.


A Tandem Bike

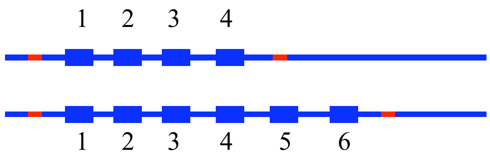
The number of tandem repeats can vary a lot from one person to another. One person could have eighteen repeats of the same DNA sequence, while another person could have fifty repeats. These special segments of repeating tandem DNA that can vary from one person to another are called **V**ariable **N**umber **T**andem **R**epeats (or **VNTR**s for short.) It is these VNTRs that are most often used in DNA fingerprinting. Because there are many different VNTRs on the different chromosomes, it is extremely unlikely that any two people will have exactly the same VNTRs. In fact, the chance that two people will have the same VNTRs is usually less than 1 in a billion.


VNTRs on chromosomes
Person A

Remember that everyone gets one chromosome from their father, and one from their mother. This also means that every person has VNTRs that they got from their father and their mother. Usually, people get different length VNTRs from each parent, but it is also possible to have two VNTRs that are exactly the same.

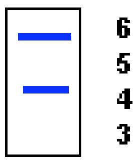
The picture above shows a small piece of the chromosomes of a Person A. Each line is a chromosome. One came from the person's father and one came from the person's mother. Each rectangle on the chromosomes is a VNTR. How many VNTRs are on each of this person's chromosomes? Count them and check your answer on the next page.

You should have counted 4 VNTRs on the first chromosome and 6 VNTRs on the second:



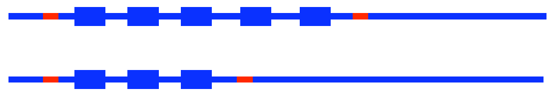
When you learned about gel electrophoresis, you learned that smaller sized pieces of DNA will move through the gel faster than larger pieces. A piece of DNA with a large number of VNTRS is larger than a piece with a small number of VNTRS. The larger piece will move more slowly through the gel. In the case of the person whose chromosomes we see above, the piece with the 4 VNTRS will move more quickly through the gel than the piece with 6 VNTRS. This will make a pattern on the gel that looks like this:

Gel Electroporesis
Person A



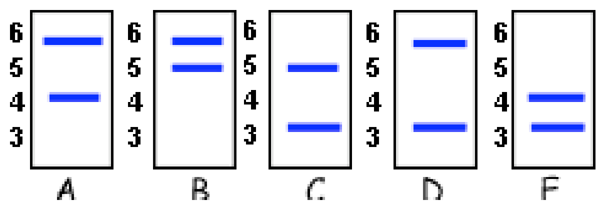
The numbers to the right of the gel are the sizes of the VNTRs.

Here is a picture of another person's chromosomes and their VNTRs. Count the number of VNTRS on each chromosome:



VNTRs on chromosomes
Person B

Now look at the different pictures from gel electrophoresis below and pick the one you think matches the number of VNTRs in person B above:



Which is the right answer? If you picked 'C', you're right, because person B has 3 VNTRs on one chromosome and 5 VNTRs on the other. Gel C shows a band at 5 and 3.

One of the problems with DNA fingerprinting is that the amount of DNA found at a crime scene is often very small. Small amounts of DNA are very hard to see with gel electrophoresis. VNTRs are very useful for DNA fingerprinting because only a very small amount of DNA is necessary. When a sample is being prepared for this type of DNA fingerprinting, it can be multiplied many times through a process called Polymerase Chain Reaction or PCR. PCR makes many copies of an area of a chromosome. Scientists can choose areas of the chromosome that contain the VNTRs, put them through the PCR reaction, and accurately make many billion copies of that DNA segment. With this much DNA, it is easy to see the bands on the gel. You will learn more about PCR later.

VNTR fingerprinting takes three main steps. First the DNA is extracted (taken out) from the cells or sample. Then, the DNA is put through PCR so that billions of copies of the DNA are made. Then the amplified (copied) DNA is run through gel electrophoresis and analyzed. Typically, many different VNTR sites are used so that there are many bands on the gel to analyze. This makes VNTR DNA fingerprinting more accurate because the greater the amount of VNTRS used, the lower the chance that two unrelated people could have the same gel pattern.

When scientists use VNTR fingerprinting, they must be extremely careful not to contaminate the samples they are using. Samples must be collected and handled with gloves at all times, and carefully stored. If any other person's DNA gets into the sample, the accuracy of the test is ruined because while the PCR reaction is making billions of copies of the sample DNA, it will also make billions of copies of the contaminate DNA and ruin the results.

On the next page you will find some practice problems using VNTR DNA fingerprinting.

VNTR DNA Fingerprinting Practice Problems and Questions

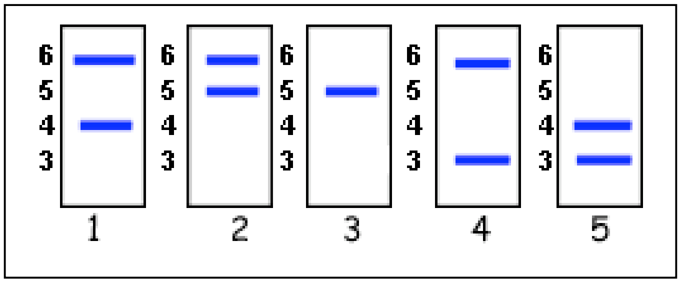
<u>Questions:</u>

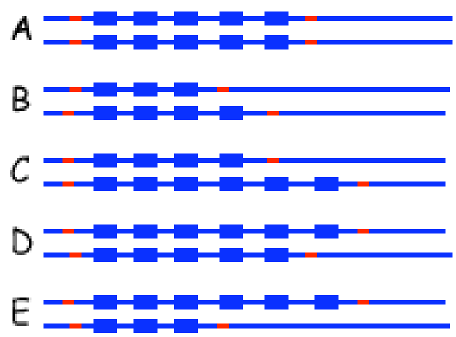
<u>Please answer in complete sentences, on binder paper. DO NOT WRITE ON THIS PAGE.</u>

1. What does VNTR stand for? What are VNTRs?
2. Why do most people show two different bands on gel electrophoresis for each VNTR?
3. Why is VNTR DNA fingerprinting such a valuable technique in solving crimes?
4. Why must scientists be extra careful when using VNTR fingerprinting?
5. What does PCR do?
6. Why can't we look at the entire DNA sequence of a person when doing DNA fingerprinting?
7. What are two different types of samples collected at crime scenes that are used in DNA fingerprinting?

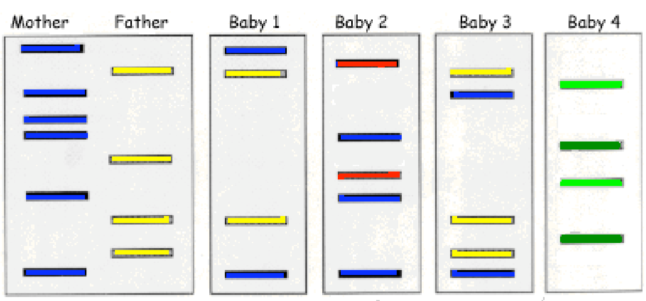
<u>Problems:</u>

1. Below are pictures of DNA fingerprints after gel electrophoresis (labeled 1-5). Match each one to the pair of chromosomes (labeled A-E) that would give each result.

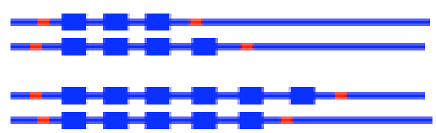




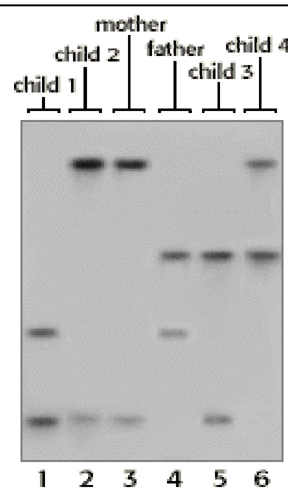
2. A mother and father have twins that are not identical. Which two babies below could be theirs? Remember that the babies each receive only one chromosome of each pair from each parent.



3. Below are the chromosomes from a person showing two sets of VNTRS on two different pairs of chromosomes. Draw a picture of the DNA fingerprint that this person would have.



4. Look at the gel to the right. Compare the banding patterns of the four children to the patterns of the mother and father. Is this father the father of all the children, some of them or none of them? Explain your answer.



RFLP Fingerprinting

Another form of DNA fingerprinting is called **R**estriction **F**ragment **L**ength **P**olymorphism, or **RFLP** fingerprinting for short. In this type of fingerprinting, special enzymes called *restriction enzymes* are used to cut DNA into pieces. These enzymes come from bacteria and they only cut DNA in specific places. For example, the restriction enzyme called EcoR1 cuts DNA between the G and A in the sequence GAATTC. Here is how it would look:

Original DNA sequence	EcoR1 cute DNA between the G and A on both sides of the DNA	DNA cut into two pieces
G-A-A-T-T-C	**G-A-A-T-T-C**	**G A-A-T-T-C**
C-T-T-A-A-G	C-T-T-A-A-G	C-T-T-A-A G

The EcoR1 enzyme moves along the DNA and cuts wherever there is the GAATTC sequence. Look at the DNA segment below. How many places are there for EcoR1 to cut?

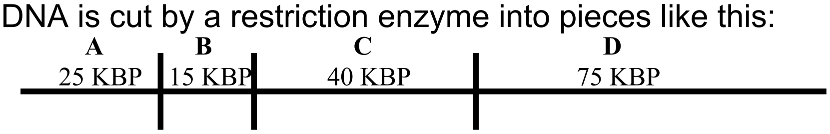
T-A-T-G-A-A-T-T-C-G-G-T-A-C-G-T-G-A-A-T-T-C-A-A-T-C-C-G-A-A-T-T-C-T-A-T-G-C
A-T-A-C-T-T-A-A-G-C-C-A-T-G-C-A-C-T-T-A-A-G-T-T-A-G-G-C-T-T-A-A-G-A-T-A-C-G

You should have found three sites for EcoR1 to cut:

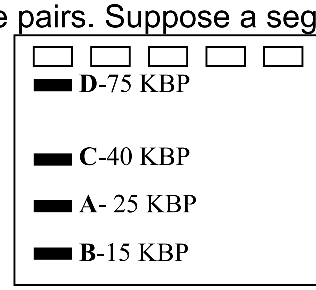
T-A-T-G-A-A-T-T-C-G-G-T-A-C-G-T-G-A-A-T-T-C-A-A-T-C-C-G-A-A-T-T-C-T-A-T-G-C
A-T-A-C-T-T-A-A-G-C-C-A-T-G-C-A-C-T-T-A-A-G-T-T-A-G-G-C-T-T-A-A-G-A-T-A-C-G

When EcoR1 cuts this DNA, it cuts it at three places into four different segments. EcoR1 is only one of many different restriction enzymes. Each different enzyme cuts DNA at a different site. By using different enzymes, a scientist can cut DNA into many smaller pieces that can be run out on a gel during electrophoresis. Remember that in gel electrophoresis, DNA fragments separate by size. Because these segments have different sizes, they will separate onto a gel at different rates.

To make it easier to understand, we will now draw the DNA as a single line and ignore the actual bases. Instead, we will label each segment with a number that represents the number of base pairs in that piece. The unit KBP means Kilo Base Pairs, or thousands of base pairs. Suppose a segment of DNA is cut by a restriction enzyme into pieces like this:



If these pieces are then run on a gel, the gel would look like this one



If different people's DNA is cut by restriction enzymes and then run out on a gel, each person's DNA will leave a different pattern.

RFLP Fingerprinting Practice Problems-put answers on your own paper! Do not write on this page!

1. What are restriction enzymes?
2. What makes each restriction enzyme different?
3. What will happen if the DNA from different people is cut by restriction enzymes and then run out on a gel?
4. What does KBP stand for?
5. The DNA below is cut by restriction enzymes as shown. Draw what the gel would look like..