

Title:
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Appropriate Life Science, High School, Honors, or Advanced Placement Biology
Level:
Abstract: In this paper simulation, students will "cut" DNA samples from a mother, a baby, a husband, and a rape suspect using a restriction endonuclease. They will then "run" the DNA fragments on a "gel" to simulate the process of electrophoresis. A fluorescent probe is then washed over the gel. Finally, students will analyze the gel to identify the father of the baby.

Time Required: Teacher Preparation: Minimal.
In Class: One standard class period to perform the simulation and one standard period to discuss the results.

Living
Environment

1-Analysis, Inquiry and Design: 1- Purpose of Scientific Inquiry: 1.1c, 1.2b, 1.3a;
3- Analyzing Observations: 3.3; 4- Content: 2-In heritance: 2.1b,e,f; 2.2c; 4-
Continuity of life: 4.1b.

## Additional Teacher Information

## Information With Which Students Should Be Familiar

- individuals differ at the DNA level
- chromosome and DNA structure
- Mendelian inheritance
- restriction endonucleases are enzymes typically found in bacteria that cut DNA in very specific locations
- electrophoresis separates charged molecules on the basis of size (of the molecule and of the charge)


## Materials Required (per team of students)

- large sheet of paper or poster board for gel (at least $60 \mathrm{~cm} \times 80 \mathrm{~cm}$ )
- scissors and tape (or glue)
- set of base sequences representing the "Standards," "Mother DNA," "Child DNA," "Suspect DNA," and "Husband DNA"
- set of "Probe" sequences copied onto brightly colored paper
- envelopes to act as wells to hold restriction fragments


## Preparation Instructions

No special preparation is required other than standard preparation. Before class, minimal time is required to gather supplies needed for the actual procedure. This is intended as a group activity with two to four students per group. Each group needs a copy of each of the DNA samples, the standard samples, and the probe samples. To emphasize the "fluorescence" of the complimentary probe sequences, you might want to copy them onto brightly colored paper. In addition, each group will need enough scissors and glue or tape. At least one or two class periods will be required to introduce the topics of electrophoresis, DNA structure, variable number tandem repeats, and the action of endonucleases.

During class, one class period is required for the actual simulation. One additional class period could be used to discuss the results and situations in which this procedure could be used.

## Helpful Hints

- Copy the DNA probes onto brightly colored paper to emphasize their fluorescence.
- Copy the "mother," "child," "husband," and "suspect" DNA onto different colored paper or in some other way mark them so they can be distinguished after they have been cut out.
- The "scenarios" in the Analysis and Discussion section of the student lab can be used as a class discussion, or have the students write the answers out onto paper.
- Technical background may be used as a supplemental reading for enrichment activities.


## Answers to Questions from Technical Background:

1. What is "junk" DNA?

Junk DNA is DNA that, for what we know now, is not transcribed into mRNA.
2. Sketch a flow diagram that shows your understanding of the steps involved in DNA profiling.

| Isolate DNA from sample. | ----------> | "Cut" DNA with endonuclease. | ---------> | Electrophorese fragments. I I V |
| :---: | :---: | :---: | :---: | :---: |
| Analyze results. | <--------- | Wash with fluorescent probe. | <------- | Transfer to nitrocellulose. |

3. Why does any one individual have a maximum of only two copies of a VNTRs at a specific locus? Mendelian inheritance regulates the passage of genes from parent to offspring. One allele will come from the mother, the second allele will come from the father. Therefore, a person can have at most two different copies of a VNTR.
4. How would restriction endonucleases assist a bacterial cell fight infection from a virus? Restriction endonucleases are produced by bacteria to help fight viral infections. When a virus infects its host, it "shoots" its nucleic acid into the host. The bacterial endonucleases would then be able to "cut" this viral genome and make it nonfunctional.
5. A short nucleotide sequence is determined to be a VNTR. The sequence is composed of the bases AGTCTTGA. Write an imaginary gene sequence that would be found on the chromosomes from an individual heterozygous at that locus with one allele containing 3 copies of the VNTR and the second locus containing 6 copies of the VNTR.
chromosome A
5'..AGCTAGCTAGGAGTCTTGAAGTCTTGAAGTCTTGAAGCTCGCTA... $3^{\prime}$
homolog
5…AGCTAGCTAGGAGTCTTGAAGTCTTGAAGTCTTGAAGTCTTGAAGTCTTGA
AGTCTTGAAGCTCGCTA…3'
6. The following is the nucleotide sequence from an individual. Only one strand of the chromosome is shown:

5'...AACGTGACGTCGATCGAGCTACGTAAAGCTTAGCTACGATTAGCTAGC...3'
a. Write the nucleotide base sequence of the complimentary strand of DNA. $3^{\prime} . . . T T G C A C T G C A G C T A G C T C G A T G C A T T T C G A A T C G A T G C T A A T C G A T C G . . .5 ' ~$
b. Show the results of a digest using the restriction endonuclease HindIII.

$$
\begin{aligned}
& 5^{\prime} \cdots A^{\prime} \text { AGCGTGACGTCGATCGAGCTACGTAAGCTACGATTAGCTAGC...3' } \\
& 3^{\prime} \cdots T T G C A C T G C A G C T A G C T C G A T G C A T T T C G A ~ A T C G A T G C T A A T C G A T C G \ldots 5 ' ~
\end{aligned}
$$

c. Identify the nucleotide sequence that would be labeled if the probe $3^{\prime}$ GCAGC $_{5}$ ' was used.

$$
\begin{aligned}
& 5^{\prime} \ldots \text { AACGTGACGTCGATCGAGCTACGTAA AGCTTAGCTACGATTAGCTAGC...3' } \\
& 3^{\prime} \ldots \text {...TTGCACTGCAGCTAGCTCGATGCATTTCGA ATCGATGCTAATCGATCG...5' }
\end{aligned}
$$

## Answers to Questions from Student Lab:

1. Sketch the electrophoresis gel before and after the addition of the radioactive probes. In the "before" diagram, use pencil to sketch the fragments. In the "after" sketch, use colored pencil to show the bands that showed up with the probe.
(Numbers represent fragment length after digest. Bold numbers identify probed sequences.)

| Stand | $\underline{\mathrm{M}}$ | $\underline{\mathrm{H}}$ | $\underline{\mathrm{S}}$ | $\underline{\mathrm{C}}$ |
| :--- | :--- | :--- | :--- | :--- |
|  |  |  |  |  |
| $\mathbf{2 9}$ |  | $\mathbf{2 9}$ |  | $\mathbf{2 9}$ |
|  |  |  |  |  |
| $\mathbf{2 6}$ |  |  | $\mathbf{2 6}$ |  |
|  |  |  |  |  |
| $\mathbf{2 3}$ | $\mathbf{2 3}$ |  |  |  |
|  |  |  |  |  |
| $\mathbf{2 0}$ |  |  | $\mathbf{2 0}$ |  |
|  | 19 |  |  | 19 |
| $\mathbf{1 7}$ |  | $\mathbf{1 7}$ |  |  |
|  |  | 16 |  |  |
| $\mathbf{1 4}$ | $\mathbf{1 4}$ |  | 14 | $\mathbf{1 4}$ |
|  |  |  |  |  |
| $\mathbf{1 1}$ |  |  |  |  |
|  | 10 |  |  |  |
| $\mathbf{8}$ | $2 @ 8$ | $2 @ 8$ | $2 @ 8$ | $2 @ 8$ |
|  | $2 @ 7$ | $2 @ 7$ | $3 @ 7$ | $2 @ 7$ |
|  | $2 @ 5$ | $3 @ 5$ | $2 @ 5$ | $3 @ 5$ |

2. Discuss the results of the DNA gel you made in class. Analyze the banding patterns according to Mendelian principles: The child inherited one allele from each parent. In our example, the mother donated one of the child's labeled alleles. Which man is more likely to have donated the other allele? Which of the two men was most likely the father of the child. Explain the reason for this answer. How sure can you be? What other procedures could you have done to be more certain?

The husband most likely fathered the child. He had fragment lengths of 29 and 17 base pairs that combined with the fluorescent probe; the suspect had fragment lengths of 26 and 29 that combined with the probe. The child had fragment lengths of 29 and 14 that reacted with the probe. The restriction fragment of 14 came from the mother, while the one of length 29 had to come from the husband. Several other digests could be performed to the isolated DNA to be more sure of the results.
3. DNA profiles from two suspects (SA and SB ) are shown below along with evidence (E) found at a murder scene (a blood drop). The marker ( M ) is shown for reference of sizes. Based on the results shown, which of the two will be most likely exonerated in this case? Explain your conclusion.


Based on this gel, it is safe to assume that suspect A (SA) did not commit the crime. None of the bands from his profile match up with the evidence. It would also be a safe assumption that suspect $B$ might have been at the scene of the crime since his profile matches the evidence profile. More probes and/or restriction endonuclease digests would need to be performed to determine probable cause.
4. For each of the following scenarios, discuss the implications of DNA analysis on the outcome(s). Also, think and discuss the bioethics involved in some of these cases. There are no right or wrong answers in this section.

These can be used as class discussions focusing on biomedical ethics and validity of technology. Answers will vary from students. There are no right or wrong answers in these cases.
5. Find an article from the popular press in which DNA profiling was used for whatever reason. Attach the article to your answer paper. Read the article and then assess the validity of the report. Try to pick out errors as well as correct points. In criminal cases, do you believe that a "jury of your peers" would be intelligent enough to understand this forensic evidence?

Answers will vary.

## Annotated References

Alberts, Bruce, et al. (1994) Molecular Biology of the Cell, 3rd ed. Garland Publishing Company, New York

An intensive college text focusing on cell molecular biology with great discussions on current biotechnology including Northern blots, Southern blots, and Western blots, all of which would be helpful for this activity on DNA profiling.

Campbell, Neil. (1996) Biology, 4th ed. Benjamin Cummings Publishing Company, New York.

A college-level general biology text good for understanding DNA structure and function as well as other biological concepts.

Elwell, Lynn. (1995) DNA Goes to Court. Carolina Tips, October, 58(4).
A nice supplemental reading on VNTRs, RFLP technology, and PCR technology. It includes a short lesson on using playing cards to demonstrate the mathematics behind DNA profiling.

## How Many Cats? <br> New York State Learning Standards

## Standard 1: Inquiry Analysis and Design

Key Idea 1: Key Idea 1: The purpose of scientific inquiry is to develop explanations of natural phenomena in a continuing and creative process.
1.1- Elaborate on basic scientific and personal explanations of natural phenomena and develop extended visual models and mathematical formulations to represent one's thinking.
c. Science provides knowledge, but values are also essential to making effective and ethical decisions about the application of knowledge.
1.2- Hone ideas through reasoning, library research, and discussion with others, including experts.
b. Inquiry involves making judgments about the reliability of the source and relevance of information
1.3- Work towards reconciling competing explanations; clarify points of agreement and disagreement.
a. Scientific explanations are accepted when they are consistent with experimental and observational evidence and they lead to accurate predictions.

Key Idea 3: The observations made while testing proposed explanations, when analyzed using conventional and invented methods, provide new insights into natural phenomena.
3.3- Assess correspondence between the predicted result contained in the hypothesis and actual result, and reach a conclusion as to whether the explanation on which the prediction was based is supported.

## Standard 4: Living Environment

Key Idea 2: Organisms inherit information in a variety of ways that result in continuity of structure and function between parent and offspring
2.1 - Explain how the structure and replication of genetic material result in offspring that resemble their parents
c. Every organism requires a set of coded traits....Heredity is the passage of these instructions from one generation to another.
e. In sexually reproducing organisms, the new individual receives half of the genetic information from it's mother and half from it's father.
f. In all organisms the coded instructions for specifying the characteristics of the organisms are carried in DNA, a large molecule formed from subunits....
2.2- Explain how the technology of genetic engineering allows humans to alter genetic makeup of organisms.
c. Different enzymes can be used to cut, copy and move segments of DNA.

Key Idea 4: The continuity of life is sustained through reproduction and development.
4.1 - Explain how organisms, including humans, reproduce their own kind.
b. ...Other organisms reproduce sexually with half the genetic information typically contributed by each parent.

## Technical Background: DNA Profiling

DNA profiling (also called DNA fingerprinting) is now being used in some criminal and legal cases where DNA samples are available to determine identity or parentage. DNA may be extracted from relatively small samples of cells, such as a blood stain the size of a nickel (about two drops) or a semen stain the size of a dime. When performed under properly controlled conditions and interpreted by an experienced forensic scientist, such profiling can link a suspect to a particular incident with compelling accuracy or completely exonerate a suspect.

At the DNA level individual people are about $99.9 \%$ identical; they differ on average in 1 out of 1000 base pairs. Some of these differences are in genes which lead to the visible differences between us. Some of the differences, however, are in "junk" DNA (DNA which as far as is known is not transcribed into RNA). In 1984, Sir Alec Jeffreys discovered these short nucleotide sequences ( 3 to 30 base pairs in length) that were repeated multiple times ( 10 to 100 times) in the non-coding region of DNA. These are known as Variable Number Tandem Repeats, or VNTRs. The most variable sequences known are tandemly repeated sequences; the basic unit of a repeat is usually a sequence of 2 to 300 base pairs. In tandem repeats, each unit has the same orientation (e.g., CATCAT). Different repeated sequences appear in different places in the genome. In each case, what is variable is the number of copies of the sequence in an allele. So for example, if the repeated sequence were CAT, one allele might have 3 copies [CATCATCAT] whereas, another allele might have 7 copies [CATCATCATCATCATCATCAT]. In a given population there may be dozens or even hundreds of different alleles. Of course, any individual has only two alleles, one on each of the homologous chromosomes; each of which was inherited from one parent. Since there are so many alleles in a population, most people are heterozygous for alleles of any given VNTR. Because of the great variability in alleles of VNTRs, if one examines enough different VNTRs (6 to 12) in a given person, one can put together a molecular picture or "DNA profile" or "DNA fingerprint" of that person.

These DNA profiles can be used for identification of tissue left at the scene of a crime (e.g., semen from a rape victim) or for paternity testing, in which case the VNTR alleles in the child which are not present in the mother must have come from the biological father. DNA profiling was introduced into the courtroom in 1988. The O.J. Simpson case brought this technique into the limelight. DNA profiling can be used to completely exonerate a suspect if the profile of the suspect does not match the profile of the evidence. However, the converse is not always true! Additional DNA profiling work must be done and population statistics will assist in the "match probability" determination. As of yet, there is no way to determine that an individual committed a crime with $100 \%$ probability, based solely on DNA profiling. Population studies must be done to determine the frequency of each allele in the population. Using that information, calculations can be made to determine the chance that a random person in the population would have the same alleles as the suspect or alleged father.

The following is a description of the techniques necessary to distinguish between different VNTR alleles.

1. DNA is isolated from the tissue sample (a procedure not included in this simulation).
2. DNA is then cut with a restriction endonuclease to produce specific fragments. Restriction endonucleases are produced in bacteria, primarily as a mechanism to protect them from virus infections. The word endonuclease can be interpreted as "an enzyme that 'works' on the interior of a nucleic acid strand"; the word restriction can be interpreted as "specific." Restriction endonucleases work by recognizing a specific nucleotide sequence (from 5 to 10 base pairs long) and then "cutting" the DNA at that specific site. More than 100 restriction endonucleases have been identified and commercially produced. Restriction endonucleases are named from the bacteria from which they were first isolated. For example, the restriction endonuclease EcoRI was the first endonuclease (I) isolated from the bacteria Escherichia coli, strain RY 13 (EcoR); HindIII was the third (III) endonuclease isolated from Hemophilous influenza, strain d. For performing DNA profiling, it is necessary to use a restriction enzyme which does not cut within the variable sequence, but does cut in flanking regions which are the same in everyone. In our example, we will use the enzyme Hind III, which recognizes the sequence:

$$
\begin{aligned}
& 5^{\mathrm{AAGCTT}_{3}} \\
& 3^{\mathrm{TTTCGAA}_{5}}
\end{aligned}
$$

and cuts the DNA between the two As, leaving one fragment that ends with
$\cdots \mathrm{A}_{3}$
and another fragment that begins with
5'AGCTT. . .
. . TTCGA ${ }_{5}$
$3^{\prime}$ A. .

Within the human genome, this particular sequence is found frequently; therefore, Hind III will cut the human genome into very many pieces, perhaps as many as a million different fragments! (Remember that the DNA has been isolated from millions of cells and the enzyme will cut each chromosome in the same place so that the result will be millions of copies of each different fragment.)
3. The fragments need to be separated according to size. Electrophoresis is used to separate molecules based on charge and/or size of the molecules. A "gel" made of agarose (a polysaccharide isolated from seaweeds) is cast and the "wells" are filled with DNA samples. The wells are located at the anode (negative) end of the apparatus and the power is turned on. The DNA fragments, having a negative charge due to the many phosphate groups, begin to migrate toward the cathode (positive) end of the gel. After an appropriate period of time, the power is turned off. The agarose acts as a "filter" that impedes the migration of the fragments. The longer fragments are found closer to the anode end; the smaller fragments are found closer to the cathode end. A "ladder" composed of fragments of known sizes is run along side the unknowns to provide size standards.
4. For technical reasons, the DNA smear is transferred to a nylon filter to prevent further movement of the DNA fragments, but the bands remain in the same relative places on the filter as they were in the gel. In the process of transferring the DNA from the gel to the nylon filter, the double strands of the DNA are separated, and the filter tightly binds the now single-stranded DNA. This will permit a complementary sequence of DNA to bind to the sample DNA which is stuck on the filter. At this point, if all the DNA were stained there would be so many fragments from each sample that the DNA would appear to be an undifferentiated smear down each lane in the gel. In order to distinguish between the DNA of one individual and that of another, we need to see only the particular fragments of interest - in this case those containing the VNTR sequences. Fluorescently labeled sequences of single-stranded DNA, called probes, have been produced which recognize (i.e. are complementary to) a particular DNA sequence. Under the proper conditions, the probe will bind only to sequences which are complementary to it; e.g., a probe for the CAT sequence will contain tandem repeats of GTA. Labeled probes are usually at least 18 base pairs long, but for simplification in our simulation, we are using smaller sizes. Additionally, complementary strands of DNA are always antiparallel, so to be accurate we should specify the ends as follows:
the target is:

$$
5^{\prime} \text { CATCAT } 3^{\prime},
$$

whereas the probe is: $\quad 3^{\prime}$ GTAGTA $5^{\prime}$.
Probe which doesn't bind is washed away, and the fluorescence is "visualized" by exposing to specific wavelength light. This process may be repeated by removing the first probe and adding different probes in sequence. This allows a single filter to be tested with multiple probes, each of which visualizes a different VNTR. Optimally, the VNTRs chosen for examination will come from many different chromosomes.

## Study Questions:

1. What is "junk" DNA?
2. Sketch a flow diagram that shows your understanding of the steps involved in DNA profiling.
3. Why does any one individual have a maximum of only two copies of a VNTRs at a specific locus?
4. How would restriction endonucleases assist a bacterial cell fight infection from a virus?
5. A short nucleotide sequence is determined to be a VNTR. The sequence is composed of the bases AGTCTTGA. Write an imaginary gene sequence that would be found on the chromosomes from an individual heterozygous at that locus with one allele containing 3 copies of the VNTR and the second locus containing 6 copies of the VNTR.
6. The following is the nucleotide sequence from an individual. Only one strand of the chromosome is shown: 5…AACGTGACGTCGATCGAGCTACGTAAAGCTTAGCTACGATTAGCTAGC... $3^{\prime}$
a. Write the nucleotide base sequence of the complimentary strand of DNA.
b. Show the results of a digest using the restriction endonuclease HindIII.
c. Identify the nucleotide sequence that would be labeled if the probe ${ }_{3}$, GCAGC $_{5}$, was used.

# How Many CATs? A DNA Profiling Simulation 

## Objectives

In this lab, you will investigate the technology behind DNA profiling (DNA fingerprinting). During this simulation you will work through the theory of DNA profiling and grapple with some analytical and ethical questions. It will help to reinforce basic concepts such as base pairing in DNA as well as the principles of restriction enzyme digestion, gel electrophoresis, and probe hybridization. From the results of your simulated gel, you will be able to determine the paternity of a child.

## Introduction

DNA profiling (also called DNA fingerprinting) is now being used in some criminal and legal cases where DNA samples are available to determine identity or parentage. DNA may be extracted from relatively small samples of cells, such as a blood stain the size of a nickel (about two drops) or a semen stain the size of a dime. When performed under properly controlled conditions and interpreted by an experienced forensic scientist, such profiling can link a suspect to a particular incident with compelling accuracy or completely exonerate a suspect.

At the DNA level individual people are about 99.9\% identical; they differ on average in 1 out of 1000 base pairs. Some of these differences are in genes which lead to the visible differences between us. Some of the differences, however, are in "junk" DNA (DNA which as far as is known is not transcribed into RNA). In 1984, Sir Alec Jeffreys discovered short nucleotide sequences (3 to 30 base pairs in length) that were repeated multiple times (10 to 100 times) in non-coding regions of DNA. These are known as Variable Number Tandem Repeats, or VNTRs. In each case, what is variable is the number of copies of the sequence in an allele. So for example, if the repeated sequence were CAT, in one allele there might be 3 copies [CATCATCAT] whereas another allele might have 7 copies [CATCATCATCATCATCATCAT].

In a given population there may be dozens or even hundreds of different VNTR alleles. Of course, any individual has only two alleles, one on each of the homologous chromosomes (one each of which was inherited from one parent). Since there are so many alleles in a population, most people are heterozygous for alleles of any given VNTR. If one examines enough different VNTRs (6 to 12) in a given person, one can put together a molecular picture or "DNA fingerprint" of that person. This can be used for identification of tissue left at the scene of a crime (semen from a rape victim) or for paternity testing, in which case the VNTR alleles in the child that are not present in the mother must have come from the biological father.

## The Simulation

The activity simulates the following situation. A married woman has been raped. Her attacker has been identified, tried, and found guilty and is now serving a prison sentence. During the legal proceedings, the woman discovered that she was pregnant. If the child was fathered by her husband, she wanted to have this child. However, if the child was fathered by the rapist, the woman wished to abort the fetus. The woman and her husband agreed to have DNA samples collected from the fetus by amniocentesis, and they both gave samples to the lab. The rapist did not have to voluntarily give a DNA sample because it was obtained from the semen stains on the bed linens which were held as evidence by the police.

Materials (per group of two students):

- large sheet of paper or poster board for gel (at least $60 \mathrm{~cm} x 80 \mathrm{~cm}$ )
- scissors and tape (or glue)
- set of base sequences representing the "Standards," "Mother DNA," "Child DNA," "Suspect DNA," and "Husband DNA"
- set of "Probe" sequences copied onto brightly colored paper


## Procedure

1. Cut out the strips of DNA sequences for each of the individuals and the standards. Be sure to keep each individual's DNA strands separate from each others.
2. We will be using the restriction endonuclease Hind III. Mark the sample strip at the recognition sites for the restriction enzyme (AAGCTT) and cut the strip all the way across between the two A's of each restriction site.
3. Use the desktop or a large sheet of newsprint to simulate the gel electrophoresis apparatus. The standards should be placed first. Use the top of the desk or paper to represent the wells of the gel. Exact distances from the origin in the "well" are not important, as long as all fragments of the same length are placed the same distance from the well. The larger fragments are placed closest to the well with the smaller ones being placed further away in descending order beneath the well. The standards should span almost the whole distance, leaving perhaps 5 cm at the bottom.
4. Place the fragments of the mother's sample to the right of the standard sample well. Note that the mother's 12-base fragment should be the same distance from its well as the standard 12-base fragment is from its well.
5. Continue placement of the remaining samples in the same manner, moving to the right across the paper or desk top in the following order: husband, suspect and child. When complete, each sample should contain five different fragments.
6. On a separate sheet of paper, sketch the results of this electrophoresis event. Remember that in "real life" these fragments would be invisible to the naked eye.
7. These fragments must next be differentiated from one another by use of the "probe." Construct DNA probes by cutting the simulated fluorescent probes from the brightly colored paper containing the probe sequences. These DNA probes will be used to see ("visualize" as the scientists call it) the VNTR sequences. (Remember that the probe sequence, ${ }_{3}{ }^{\prime}$ GTAGTA $_{5}$, is complementary to the VNTR sequences, $5^{\prime}$ CATCAT ${ }_{3}$ ' With a probe in hand, scan the "gel" and position a probe on each complementary sequence. Each labeled fragment represents a part of one chromosome of a homologous pair.
8. On your previous sketch of the unmarked gel, identify the fluorescently marked fragments by lightly coloring over them with a colored pencil.

Analysis and Discussion (Answer the following on a separate sheet of paper.)

1. Sketch the electrophoresis gel before and after the addition of the radioactive probes. In the "before" diagram, use a regular pencil to sketch the fragments. In the "after" sketch, use a colored pencil to show the bands that showed up with the probe.
2. Discuss the results of the DNA gel you made in class. Analyze the banding patterns according to Mendelian principles: the child inherited one allele from each parent. In our example, the mother donated one of the child's labeled alleles. Which man is more likely to have donated the other allele? Which of the two men was most likely the father of the child. Explain the reason for this answer. How sure can you be? What other procedures could you have done to be more certain?
3. DNA profiles from two suspects (SA and SB) are shown below along with evidence (E) found at a murder scene (a blood drop). The marker (M) is shown for reference of sizes. Based on the results shown, which of the two will be most likely exonerated in this case? Explain your conclusion.

4. For each of the following scenarios, discuss the implications of DNA analysis on the outcome(s). Also, think and discuss the bioethics involved in some of these cases. There are no right or wrong answers in this section.

- Suppose a pregnant 14-year-old claimed the pregnancy resulted from her father raping her and he claimed her boyfriend was the father. Could this procedure be used?
- What if the samples represented two suspects in the rape instead of the husband and suspect, and a stain from the woman's clothing instead of a fetus? Would this be sufficient evidence to convict (or exonerate) a suspect of the crime?
- Could blood on the clothes of a murder suspect be used to implicate him in the crime?
- On occasion, pediatricians have been asked by a husband to test his baby to determine if he is really the father, but without informing the mother. What should the doctor do?
- The U.S. military has just instituted a policy of taking and freezing a blood sample from each recruit. If necessary the frozen sample will be subjected to DNA profiling (e.g. to identify remains). Do you agree with this policy?
- The Tomb of the Unknown Soldier in Washington, DC contains the remains of soldiers from different branches of the armed services. The parents of one of the Marines assumed missing in action during the Vietnam War had evidence to believe that their son was one of the Unknowns buried. There was also evidence from the government. The remains were exhumed and tested using DNA profiling techniques;
it turns out that it was indeed their son. The parents are planning to remove the remains and bury them in their hometown. Do you think this test should have been done?
- Some states like California mandate that all men convicted of sex felonies give a blood sample for DNA profiling before they are released from prison. The information is stored on a computer. When a sex crime occurs in the state, a DNA profile will be run on the evidence and run on computer to check whether a match exists with the DNA of a previous convict. New York State has proposed do likewise. What do you think?
- What limitations can be seen in these procedures?

5. Find an article from the popular press in which DNA profiling was used for whatever reason. Attach the article to your answer paper. Read the article and then assess the validity of the report. Try to pick out errors as well as correct points. In criminal cases, do you believe that a "jury of your peers" would be intelligent enough to understand this forensic evidence?

- $5^{\text {AGGCTITTH }}{ }^{\prime}$

- $5^{\text {A'AGCTTCATCATITA3 }}{ }^{\prime}$
. $\mathbf{5}^{\text {'月GCTTCATCATCATTR }}{ }_{3}$
- $5^{\text {AGGCTTCATCATCATCATITA }}{ }_{3}$
- $5^{\text {'AGCTTCATCATCATCATCATTTA }}{ }_{3}$
- $5^{\text {AGGCTTCATCATCATCATCATCATITA }}{ }_{3}$
- $5^{\text {'月GCTICATCATCATCATCATCATCATTR }}{ }_{3}$


## TEMPLATE FOR DNA PROBES

(SHOULD BE COPIED ON BRIGHTLY COLORED PAPER, OR COLOR MAY BE ADDED BY STUDENTS AS THEY "MAKE" THE PROBES)



|  | $\mathbf{3}^{\prime \text { GTAGTA }_{5 '}}$ | $\cdot \mathbf{3}^{\text {GTAGTA }_{5 '}}$ | $\cdot \mathbf{3}^{\text {GTAGTA }_{5 '}}$ |
| :--- | :--- | :--- | :--- |






| . $3^{\prime}$ GTAGTA $_{5}{ }^{\prime}$ | . $3^{\prime \text { GTAGGTA }_{5}}$ | . $3^{\prime}$ GTAGTH $_{5}{ }^{\prime}$ | . $3^{\text {GTAGTA }}{ }_{5}$ |
| :---: | :---: | :---: | :---: |







| . $3^{\text {GTAGGTH }}{ }^{\text {' }}$ | . $3^{\text {GTAGTA }}{ }^{\prime}$ | . $3^{\text {GTAGTH }}{ }_{5}$ | - $3^{\text {GTAGTA }}{ }^{\text {' }}$ |
| :---: | :---: | :---: | :---: |

## DNA SAMPLES FROM VARIOUS PLAYERS

Each box represents a small piece of DNA from one chromosome;
each person has two alleles to represent the pair of homologous chromosomes
mother

5'CTAGAAGCTTAAAGCTTCATCATTTAAGCTTCAAAGCTTTCGACCTAAATTGC $\mathbf{3}^{\prime}$

5'CTAGAAGCTTAAAGCTTCATCATCATCATCATTTAAGCTTCAAAGCTITCGAC $\mathbf{3}^{\prime}$

## husband

5'CTAGAAGCTTAAAGCTTCATCATCATTTAAGCTTCAAAGCTITCGACCTAAAT $\mathbf{3}^{\prime}$

5'CTAGAAGCTTAAAGCTTCATCATCATCATCATCATCATTTAAGCTTCAAAGCTT3'
suspect

5'CTAGAAGCTTAAAGCTTCATCATCATCATCATCATTTAAGCTTCAAAGCTTTCG3'

$$
\text { 5'CTAGAAGCTTAAAGCTTCATCATCATCATTTAAGCTTCAAAGCTTTCGACCTA }{ }_{3}
$$

## child

5'CTAGAAGCTTAAAGCTTCATCATTTAAGCTTCAAAGCTITCGACCTAAATTGC $\mathbf{3}^{\prime}$

5'CTAGAAGCTTAAAGCTTCATCATCATCATCATCATCATTTAAGCTTCAAAGCTT ${ }_{3}{ }^{\prime}$

