# East Carolina

### Abstract

The Innocence Project is an agency that primarily uses DNA analysis to exonerate people wrongly convicted of crimes. Students in a non-majors biology lab have a hard time relating to and understanding the rather technical aspects of DNA analysis. A lab using the Innocence Project was designed to engage and motivate the students to learn about DNA structure, DNA function and DNA analysis techniques. The students work in groups of 4 as reviewers of a client application. The client is a person convicted of a crime who is proclaiming to be innocent. The client application includes information about the original conviction and a list of DNA evidence that was not previously analyzed. The students review the client's application and analyze the DNA evidence to make a recommendation to the Innocence Project review board (the other students in the class). The recommendation is made in the form of a group presentation. The lab activities are divided between 2 days. On the first day, the students use gel electrophoresis to separate fragments of DNA in a DNA sample. The students develop a DNA profile for that DNA sample and learn to calculate a DNA fingerprint frequency. Also on this first day, students determine their own genetic profile for 7 genetic traits and calculate their genetic frequencies for these traits in an effort to better help them understand DNA fingerprint frequencies. The second day students evaluate the DNA evidence from the case of their client. The students analyze one STR locus to make a recommendation about their client. Students have to explain the meaning of matches between DNA samples and relate it to the DNA fingerprint frequency of their client. The students also have to explain how the DNA evidence fits with the other evidence used in the original conviction. The students find this set of activities to be an enjoyable way to learn about DNA. Students in this class are engaged and consistently report this lab to be one of their favorites in all the semesters that we have used it.

### **Day One Activities**

The day one activities are a practice for using the equations and analysis that will be needed in day two. On day one, each student group loads and runs agarose gels with simulated DNA samples to create a DNA fingerprint of the nail color locus. The nail color locus is a simulation of a short tandem repeat (STR) site that has 3 alleles of different sizes. Students learn that STR sites are located in regions of DNA that do not code for proteins. The alleles of an STR site vary in size with different individuals having different sized alleles. STR sites are useful in forensics because they are highly variable between individuals due the varying number of short sequences repeated within the site, see Figure 1.



**Figure 1.** Short Tandem Repeat (STR) site This STR site has the sequence "TGT" repeated. In Suzie's DNA there are 4 copies of "TGT" while Mark's DNA has only 2 copies of "TGT". When this region is cut with a restriction enzyme Suzie's DNA fragment will be larger than Mark's DNA fragment for this STR site.

The students must identify which alleles are present in an individual and determine if the individual is homozygous or heterozygous for the locus. Over the years we have decided to use food coloring as the DNA samples to reduce the cost in our large multi-section lab course and to make the "DNA" bands easily identifiable (Figure 2a). Each student group gets a different combination of 3 of the 6 available samples to analyze on a gel as well as the color marker which contains all 3 possible alleles.

a)		<b>b</b> )
	Color MarkerSample 1Sample 2Sample 3Sample 4Sample 5Sample 6	Allele Color
		Blue
		Red
		Yellow

Figure 2. a) Nail Color DNA Gel with 6 DNA samples; samples 1, 2 and 3 have one allele while samples 4, 5 and 6 have two alleles. b) Nail Color Allele Frequencies

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The students learn to calculate the DNA fingerprint frequency (DF) of each sample for the nail color locus using the allele frequency (AF) of the alleles in the sample (Figure 2b). The DF is the frequency or likelihood of finding that particular DNA fingerprint for the nail color locus in an individual of the target population.

DNA samples with one allele use the following equation to calculate DF: DF = AF<sup>2</sup>

DNA samples with two alleles use the following equation to calculate DF: **DF= 2(AF<sub>1</sub>) (AF<sub>2</sub>)**; in which  $AF_1$  is the first allele frequency and  $AF_2$  is the second allele frequency.



### **Day Two Activities**

Each student group is given an Innocence Project case file for a person, the client, who was convicted of a crime. The client is claiming to be innocent of the crime and has asked the Innocence Project to investigate. The case file includes a description of the crime and the evidence used to convict the client. The case file is a modified version of the case questionnaire, the evidence questionnaire and the police questionnaire used by the North Carolina Center on Actual Innocence. In all cases, DNA evidence was not analyzed before the original trial. A simulated electrophoresis gel was prepared from the available DNA and is included in the case file for the students to analyze. See a sample gel in Figure 3.

λDNA marker	52-1	52-2	52-3	52-4
- 21,226 - 4973, 5148 - 4268 - 3530 - 2027 - 1904 - 1584 - 1375 - 947 - 831 - 564				
	Pepper (buccal) <mark>Victim</mark>	Allen (blood) <mark>Client</mark>	Blood on knife	Cells on knife handle

Allele Frequency				
0.65				
0.15				
0.20				
	-			

**Figure 3.** Sample Innocence Project Gel This gel includes a DNA marker, DNA from victim, client and from evidence collected at the crime scene.





 
 Allele Number
 Frequenc

 1
 0.009

 2
 0.308

 3
 0.078

 4
 0.201

 5
 0.104

 6
 0.518

 7
 0.162

 8
 0.405

 9
 0.215

**Figure 4.** THO1 DNA Locus Reference The THO1 DNA locus has 9 alleles. The allele frequencies and allele sizes in base pairs are listed.

The gels display the DNA fingerprints for the THO1 DNA locus, one of the STR loci included in CODIS, the FBI program that runs a criminal justice DNA database. The students are reminded that DNA is inherited from both parents; one copy from your mother and one copy from your father. This means that each person has 2 alleles for the THO1 DNA locus. A person could have 2 of the same allele and be homozygous for the DNA locus or have 2 different alleles and be heterozygous for the DNA locus. Students use the reference (see Figure 4) to determine which alleles are present in each DNA sample on their gel. If a DNA sample has one band then the individual is homozygous for the locus. If a DNA sample has 2 bands then the individual is heterozygous for the locus. If the DNA sample on the gel has more than 2 bands then it is a mixed sample with DNA from more than one person. The students must use the allele frequency information in the reference to calculate the DF for their client. If the DNA evidence does not match the DNA fingerprint of their client, then the student group can exclude their client from having left DNA at the crime scene. If the DNA evidence matches the DNA fingerprint of their client, then the student group will need to discuss the DF of their client. If the DF of their client is high then there are many others in the population with the same DNA fingerprint who could have left their DNA at the crime scene. If the DF of their client is low then there are few others in the population with the same DNA fingerprint that could have left DNA at the crime scene.

Each student group must analyze the gel as well as the previous evidence about their case to make a recommendation to the board. Students have time to prepare and give presentations during the class time. They are encouraged to summarize the case information in their own words and back up their claims with facts from the case. The students may recommend that their client be declared innocent and released from prison, that analysis of more STR sites be performed or that the case be dropped because the DNA does not exonerate the client. The rest of the class can ask questions and then vote whether or not to accept the group's recommendation. Student groups are graded on their presentations and interpretation of their gels. Students also evaluate each other's participation in the group.

These lab activities have real world applications even for the non-majors. The DNA analysis is relevant to any student who may enter law enforcement or serve on a jury. Student presentations help prepare students to communicate effectively and base conclusions on data and not conjecture.

#### References

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