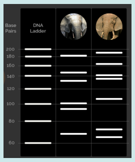


Click and Learn
CSI Wildlife: Using genetics to hunt elephant poachers

hhmi | BioInteractive

Student Worksheet

ABOUT THIS WORKSHEET
Forensic scientists collect and analyze scientific evidence to solve crimes. One type of evidence they use is genetic data. In this activity, you will use DNA analysis to solve several crimes related to elephant conservation, a field of science known as wildlife forensics. This worksheet complements the Click and Learn "CSI Wildlife." (http://www.hhmi.org/biointeractive/csi-wildlife).

MATERIALS

- Access to HHMI BioInteractive's Click and Learn "CSI Wildlife"
- 1 copy of the data handout
- Four-function calculator

PROCEDURE

Congratulations! You have just been hired by an international police force to work as a forensic scientist dedicated to protecting wildlife and investigating illegal activity where protected animals are involved. In particular, you will be focused on the protection of African elephants. You are excited to put your scientific skills to work. To help you learn more about what you will do in your new position, your boss asks you to complete HHMI BioInteractive's Click and Learn "CSI Wildlife." But before you begin, she asks you to answer the following questions.

1. Elephants are a keystone species and play a pivotal role in shaping the forests and savannas in which they live. Knowing the importance of elephants, an international group of scientists conducted a census to estimate the number of African elephants. The Great Elephant Census data was released in August 2016, and unfortunately, the results reveal that African elephants have decreased by 30% in just the past seven years. What ideas do you have for why the number of elephants is declining?

2. You have likely heard of investigators using DNA fingerprinting to identify individuals in crime scene investigations. Briefly describe your current understanding of the process of DNA fingerprinting.

Part 1: Now that you have shared with your boss what you already know, she is ready to send you for training. Complete the activities that are in **Case One** in HHMI BioInteractive's Click and Learn "CSI Wildlife."

3. Your colleagues in the legal department are preparing to take a case to trial. To help them, rewrite your description of the process of DNA fingerprinting so a jury could understand it.

Now you are ready for your first major assignment. Your colleagues are investigating a newly seized tusk and wonder if the tusk is from a recently discovered crime where 10 elephants from the same population were found dead with their tusks removed. The genotype of the seized tusk for five short tandem repeat (STR) loci is shown in Table 1. Allele names for each of the STR loci correspond to their size in base pairs (bp). Table 2 lists the genotypes of the same five STR loci for 10 recently slaughtered elephants.

	FH19	FH60	FH67	FH71	FH129
Seized tusk	193 193	147 147	97 103	62 62	152 160

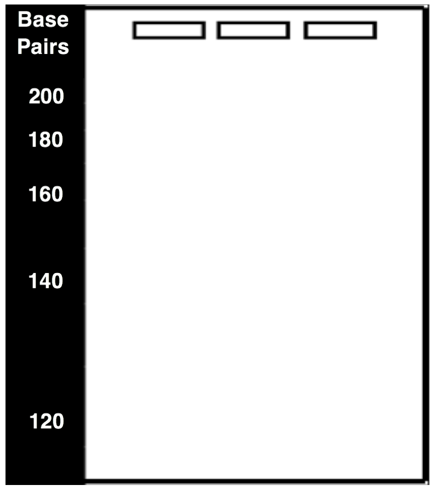
Table 1: Genotype for five STR loci recovered from a seized elephant tusk.

	FH19	FH60	FH67	FH71	FH129
Elephant 1	189 193	147 147	97 97	62 64	152 154
Elephant 2	185 193	147 147	95 103	62 62	152 152
Elephant 3	183 185	147 147	95 97	62 64	152 162
Elephant 4	193 193	147 147	91 105	62 64	152 152
Elephant 5	187 187	145 147	95 95	62 64	152 160
Elephant 6	187 193	145 147	91 97	64 64	152 160
Elephant 7	193 193	149 151	97 97	60 62	156 160
Elephant 8	193 193	147 147	97 103	62 62	152 160
Elephant 9	193 193	147 147	97 97	62 62	160 160
Elephant 10	189 193	147 147	95 105	62 62	152 156

Table 2: Genotypes using five STR loci for 10 slaughtered elephants.

4. Did the seized tusk come from one of the recently slaughtered elephants? _________ What evidence supports your finding?

5. Draw the expected pattern of bands on a gel that shows the alleles for the seized tusk and the elephant you identified as a potential match, for STR loci FH60 and FH129. Include a DNA ladder that has DNA fragments that are 125, 150, and 175 base pairs (bp) in size.



6. Your next challenge is to calculate the probability that another elephant from the same population would have the same genotype as the one from the seized ivory sample.

a. Calculate the frequency of each of the alleles found in the ivory sample using data from the 10 slaughtered elephants. Start by examining the data in Table 2 for STR locus FH19. The ivory sample was homozygous for the 193-bp allele. In the sample of 10 elephants, 12 alleles were also 193 bp in size, out of a total of 20 alleles. Look at how these data are used to determine the frequency of the allele in Table 3. Use the same approach to calculate the frequency of the other four STR loci in the table on the following page. Some of the data is provided for you.

STR locus	FH19	FH60	FH67	FH71	FH129
# of copies of Allele 1 in the group of 10 elephants studied	12	16	9		
Total number of alleles in the group of 10 elephants studied	20			20	
Frequency of Allele 1	12/20 = **0.60**				
Number of copies of Allele 2 in the group of 10 elephants studied	12		2		
Total number of alleles in the group of 10 elephants studied	20		20		
Frequency of Allele 2	12/20 = **0.60**				

Table 3. Frequency of alleles found in the seized tusk in the slaughtered elephants' population for five different STR loci.

b. Use the formulas below to calculate the probability of another individual having the same genotype for each individual locus. Enter the formulas and probability in Table 4.

- If the ivory sample is **homozygous** for an STR locus, the probability ($P_{(genotype\ for\ locus\ X)}$) that another individual has the same genotype for the STR locus is:

$$P_{(genotype\ for\ locus\ X)} = \text{(frequency of Allele 1)}^2$$

- If the ivory sample is **heterozygous** for an STR locus, the probability ($P_{(genotype\ for\ locus\ X)}$) that another individual has the same genotype for the STR locus is:

$$P_{(genotype\ for\ locus\ X)} = 2 \times \text{(frequency of Allele 1)} \times \text{(frequency of Allele 2)}$$

STR locus	FH19	FH60	FH67	FH71	FH129
Formula with filled-in values for the probability	$(0.60)^2$				
Calculated probability for each locus	0.36				

Table 4. The probability that another elephant in the same population has the same genotype at each individual STR locus.

c. To determine the probability that another individual has the same genotype for each of the five STR loci (in other words, the same genetic fingerprint), multiply all the individual probabilities together.

What is the probability that all five match? _____________

7. Impressed with your work, your boss now asks you to apply what you have learned to the genotype for the ivory sample for all 16 STR loci, listed in Table 5 below. The first five STR loci are the same as the ones listed in Table 4. In addition, your colleague has been able to collect additional samples of elephants from the same population as the ones shown in Table 2. He now has allele frequency data based on 80 alleles, shown in the table below.

STR locus	FH19	FH60	FH67	FH71	FH129	FH39	FH40	FH48
Allele 1 size (bp)	193	147	97	62	152	239	242	170
Allele 1 frequency	0.28	0.66	0.36	0.51	0.39	0.31	0.68	0.05
Allele 2 size	193	147	103	62	160	243	242	172
Allele 2 frequency	0.28	0.66	0.10	0.51	0.25	0.41	0.68	0.38
STR locus	**FH94**	**FH102**	**FH103**	**FH126**	**FH127**	**FH153**	**S03**	**S04**
Allele 1 size (bp)	225	177	149	94	247	169	141	154
Allele 1 frequency	0.65	0.64	0.13	0.18	0.04	0.011	0.55	0.8
Allele 2 size	225	181	151	110	247	171	143	154
Allele 2 frequency	0.65	0.14	0.61	0.11	0.04	0.08	0.39	0.8

Table 5. The ivory sample allele size and their associated allele frequency in the population for all 16 STR loci from a sample of 40 elephants (80 alleles).

a. Compare the frequency of the alleles for STR loci FH19, FH60, FH67, FH71, and FH129 that you completed in Table 3 with the values in Table 5. Why do you think the values differ?

b. Calculate the probability that another individual has the same genotype at all 16 STR loci by using the same procedure as Steps 6b and 6c and then multiply all the individual probabilities together.

Probability of an identical match with another elephant at all 16 STR ___________________

Part 2: Your colleagues can tell that you have the necessary skills to help stop the killing of elephants for ivory and to bring poachers to justice. Your boss promotes you to lead a new case. To solve this case, you first need to complete **Case Two** of HHMI BioInteractive's Click and Learn "CSI Wildlife."

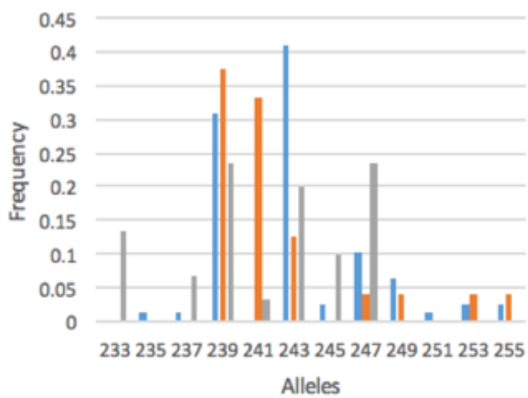
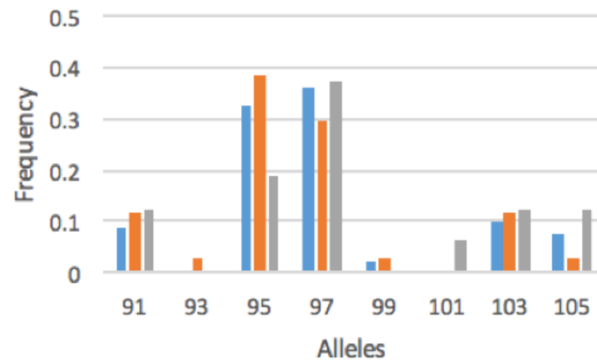
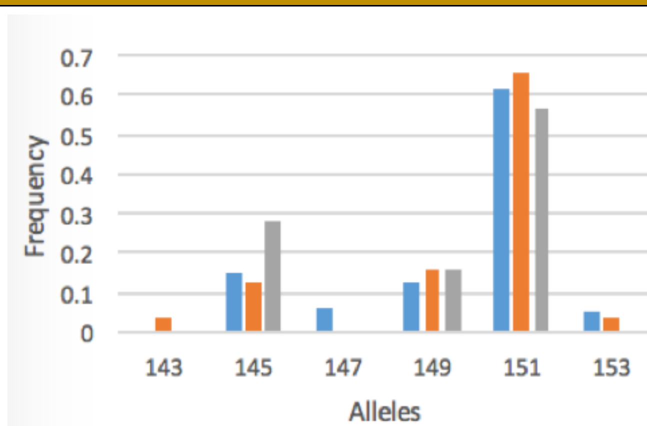
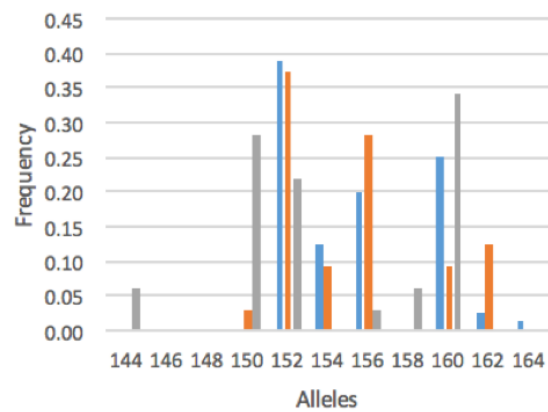
The details of the case: Another shipment of ivory has been seized. Law-enforcement officials captured one of the people shipping the ivory. Through interviews with this individual and his contacts, the police determine the timing of the poaching event, but they still do not know the location. At the time the ivory was collected, three suspects were each caught poaching elephants in parks from three different parts of Africa. Suspect one was poaching in the country of Tanzania, Suspect 2 in Cameroon, and Suspect 3 in Zambia.

Your boss would like you to link the genotype of one of the seized tusks to one of the three reference populations taken from each country. You determine the genotype of the seized tusk for four STR loci, shown in Table 6.

FH39	FH67	FH103	FH129
239	97	151	154
241	97	153	160

Table 6. The genotype for four STR loci of a seized tusk.

Your lab assistant summarizes the allele frequency data for the three possible source populations in Figure 2 below.

<table>
<tr><th>STR Loci</th><th>FH39</th><th>FH67</th></tr>
<tr><td>Allele frequencies from three elephant populations</td><td>
</td><td>
</td></tr>
<tr><td>Alleles in seized tusk</td><td>239, 241</td><td>97, 97</td></tr>
<tr><th>STR Loci</th><th>FH103</th><th>FH129</th></tr>
<tr><td>Allele frequencies from three elephant populations</td><td>
</td><td>
</td></tr>
<tr><td>Alleles in seized tusk</td><td>151, 153</td><td>154, 160</td></tr>
</table>

■ Tanzania ■ Zambia ■ Cameroon

Table 7. Comparison of the alleles from the ivory tusk to the reference populations in Tanzania, Zambia, and Cameroon.

8. Make a claim about which country in Africa is most likely to be near the source population of the seized tusks. Your claim will be used in court, so make sure to highlight the evidence you used to make your claim.

Part 3: Late one night, rangers in a park hear a gunshot and quickly move to investigate. One elephant has been slain, but the quick actions of the rangers make the poacher flee, which saves the lives of many elephants. Eventually, a suspect is captured with one pair of elephant tusks. Your boss gives you the tusk and a blood sample from the killed elephant. She asks you to determine whether the tusks came from the elephant that was killed.

You analyze the samples and the results are shown in Figure 1. Each STR locus is shown in a different color.

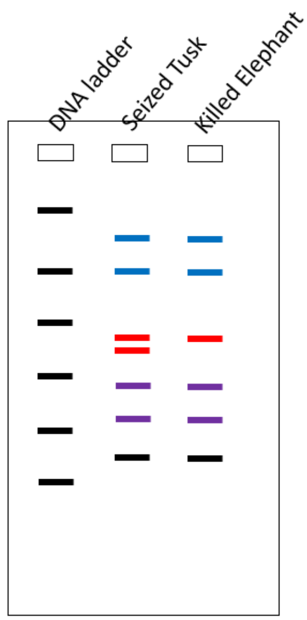


Figure 1. The gel with the alleles for four different STR loci for the elephant tusk and the killed elephant.

9. Use the evidence in Figure 1 to determine whether the tusk came from the slain elephant. Write a brief summary of your findings for the case file.

10. It is very important for scientists to consider sources of error in an experiment and alternative explanations. What sources of error or alternative explanations might change the answer you gave in Step 9?

11. After completing this activity, do you believe that DNA fingerprinting is useful as evidence in criminal cases and can help in reducing elephant poaching? Provide three pieces of evidence from this experience to support your answer.

REFERENCE

This activity is based on the annotated research paper Wasser, S. K., Brown, L., Mailand, C., Mondol, S., Clark, W., Laurie, C., & Weir, B. S. (2015). Genetic assignment of large seizures of elephant ivory reveals Africa's major poaching hotspots. *Science*, **349**(6243), 84-87. The underlying data are available on the Dryad Digital Repository: http://dx.doi.org/10.5061/dryad.435p4.