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# LESSON PLAN:

## Left at the Scene of the Crime!

An Introduction to Forensic Science

Grade level: 6-12, College  
Subject area: Forensic Science, Biotechnology

## General Information

### LEARNING OBJECTIVES:

In this Forensic Science Lesson, students explore many different aspects of a crime scene investigation. First, students utilize critical reading skills through the interpretation of a "Police Report". Next, students will analyze simulated crime scene and suspect samples using the Kastle-Meyer test, Blood Group Typing, and Agarose Gel Electrophoresis. The results will be presented as a Forensic Science report in the "court of law". After performing this investigation, your students will have experience with data collection, critical analysis of results, and scientific inquiry. This will help them to transform advanced topics in genetics and biotechnology into a concrete scientific understanding.

### ESSENTIAL KEY QUESTIONS:

- What is Forensic Science?
- What are some advantages and disadvantages of different forensic techniques?
- How can biotechnology be used in Forensic Science?
- How does Forensic Science work within the context of a criminal investigation?

### CONTENT STANDARDS:

#### NGSS Alignment:

HS-LS1: From Molecules to Organisms: Structures and Processes

HS-LS3: Heredity: Inheritance and Variation of Traits

LS3.A: Inheritance of Traits

LS3.B: Variation of Traits

Science and Engineering Practices:

Asking Questions

Developing and Using Models

Analyzing and Interpreting Data

Crosscutting Concepts:

Cause and Effect

Structure and Function

#### Common Core State Standards:

ELA/Literacy:

RST.11-12.1

RST.11-12.9

WHST.9-12.9

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## Lesson Introduction

Forensic science (or forensics) is the application of scientific knowledge to answer questions of interest within the legal system. This interdisciplinary field uses scientific techniques from diverse fields like Biotechnology, Toxicology, Chemistry, and Physics to characterize physical evidence found at the scene of a crime. Forensics professionals perform all stages of evidence collection and analysis. Throughout the process, they keep careful notes documenting chain of custody and tests performed. After analyzing the evidence, forensic scientists act as witnesses at trials by providing detailed reports and expert testimony.

Forensic science has its roots in antiquity. For hundreds of years, researchers devised forensic strategies to distinguish between guilt and innocence. In the early 1800's, the chemist James Marsh devised a test to determine whether samples contained the common poison arsenic. By the end of the 19th century, the Scottish physician Henry Faulds recognized the potential of using human fingerprints as a method of identification. Modern forensics labs analyze crime scene evidence using cutting-edge techniques such as DNA fingerprinting, X-ray fluorescence spectroscopy, and trace chemical analysis. While many forensic labs use state-of-the-art technology, forensic scientists still use classical techniques to examine the physical evidence found at a crime scene.

Before being analyzed in the forensic science laboratory, samples must be carefully collected to prevent contamination or sample degradation. Each sample is labeled and sealed in a tamper-proof evidence bag before being removed from the crime scene. A careful record is kept of all people who have handled the evidence as it is transferred from the crime scene to the forensic laboratory to make sure it remains unadulterated. Similarly, while the samples are being analyzed, forensic scientists keep careful records of all tests performed, as their results become evidence in the court of law.

It is important to point out that a forensic match provides strong evidence that the suspect was present at the crime scene. However, the evidence does not prove that the suspect committed a crime. It cannot prove when a suspect was at the crime scene or intent. Many other lines of evidence need to come together to build a case.

### **PRESUMPTIVE VS. CONFIRMATORY TESTS**

Since the amount of biological evidence collected at a crime scene is often vanishingly small, it is important that forensic scientists select the correct tests for analysis. To preserve as much of the evidence as possible, initial testing involves simple, cost-effective chemical tests that require trace amounts of sample. These "presumptive" tests strongly indicate the identity of the tested substance. However, other common substances may produce a positive result (a "false positive") with presumptive tests. In these situations, further testing is required to verify the identification.

To identify the composition of a piece of evidence beyond a doubt, forensic scientists will use confirmatory tests. Like presumptive tests, confirmatory tests are highly specific and sensitive. Unlike presumptive tests, these specialized tests rarely produce false positives. However, confirmatory tests can be expensive and time-intensive. This is why confirmatory tests are performed after the presumptive tests have identified which samples will yield the best results. The results from a confirmatory test allow the forensic scientists to make a positive identification of the sample.

### **FORENSIC ANALYSIS OF BLOOD SAMPLES**

To identify biological samples as blood, two common tests may be performed. The first test is the Kastle-Meyer test, which is a presumptive test for blood. This test uses the chemical phenolphthalein to detect the presence of

blood in crime scene samples. Phenolphthalein is best known as an acid-base indicator. At pH values between 0 and 8, the chemical is colorless. Above a pH of 8, phenolphthalein changes to a bright pink color. However, phenolphthalein also changes color in the presence of an oxidizing agent. This property allows it to be used as a blood indicator. At the molecular level, the blood protein hemoglobin reacts with hydrogen peroxide to create water and free oxygen molecules. The free oxygen molecules catalyze the oxidation of phenolphthalein, resulting in the color change from clear to bright pink.

**Table 1 - Blood Type Distribution in the USA**

Group	%	Actual # in millions
A	42	127.7
B	10	30.3
AB	4	12.2
O	44	133.8
<b>Total</b>	<b>100</b>	<b>304.0</b>

The Kastle-Meyer test is an ideal presumptive test for blood because it is fast, it is highly specific for blood, and it can reliably detect blood in dilute samples. However, there are a few chemicals that react with phenolphthalein to produce a false positive (e.g. iron and copper oxides). An experienced examiner can distinguish between these results and true positive results by studying the color and timing of the reaction. For example, a true positive sample undergoes a color change to pink within 10 seconds of the addition of the hydrogen peroxide, and only once the hydrogen peroxide has been added. If the pink color change occurs before the addition of the hydrogen peroxide, then it is a false positive. Once a biological sample has been identified as blood, further testing can be performed.

The second test used to identify blood samples in the forensic science lab is blood group testing. This test relies on the interaction of blood cell surface antigens (A or B) with their corresponding antibodies (Figure 1). If the antibody in solution recognizes the blood cell surface antigens, large complexes are formed. These complexes precipitate out of solution, which is visible to the naked eye as a granular solid. In contrast, when the antibody does not recognize the blood cell surface antigens, the appearance of the sample does not change. This precipitation reaction (agglutination) is specific to blood, which is why it is classified as a confirmatory test.

**Figure 1 – Blood Group Typing**

	Group A	Group B	Group AB	Group O
Red blood cell type				
Antibodies in Plasma	Anti-B	Anti-A	None	Anti-A & Anti-B
Antibodies in Red Blood Cell	A antigen	B antigen	A & B antigens	None

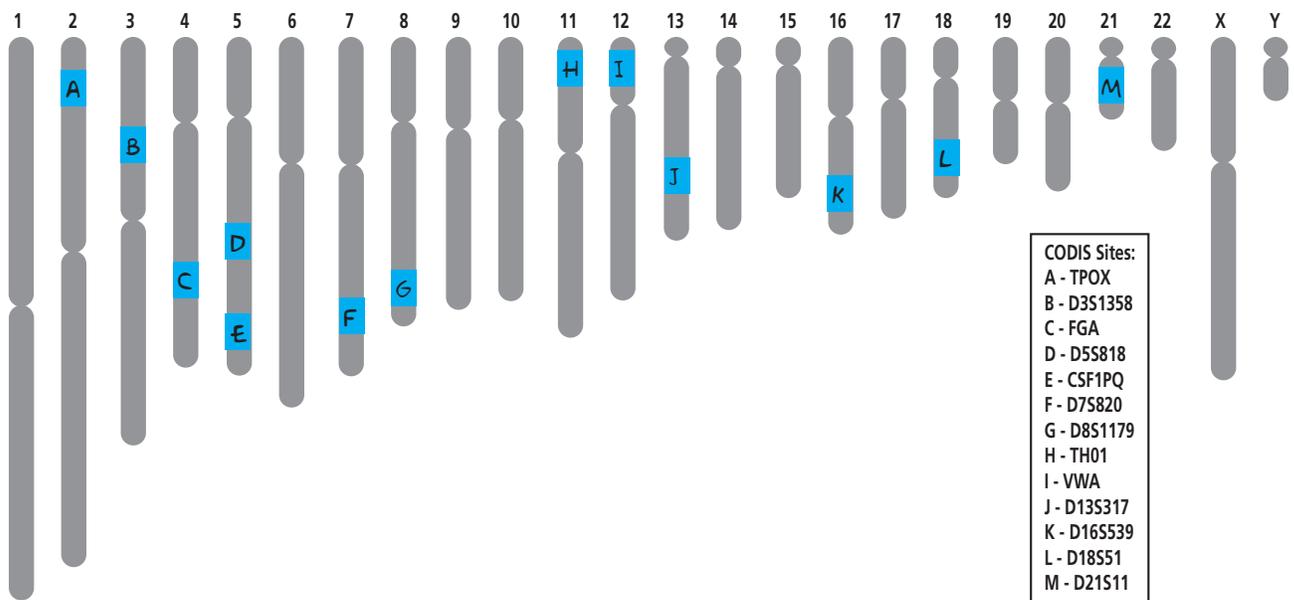
Blood group typing is an easy and cost-effective forensic test to rule out potential suspects. For example, imagine that a crime scene blood sample is type O. Using the blood group information, we can't say for sure that the suspect with Type O is also the one who left blood at the crime scene, since many individuals can share a blood type (Table 1). However, we can definitively state that the person with Type AB did not leave the blood at the crime scene. Using serological evidence in conjunction with physical evidence and eyewitness accounts will always help narrow down the possibilities in a case.

**DNA FINGERPRINTING ANALYSIS**

After a crime scene sample has been confirmed to be blood, forensic scientists will extract and analyze DNA from the cells. In humans, DNA is packaged into 23 pairs of chromosomes. Although most of this DNA is identical between individuals, small sequence differences, or "polymorphisms", occur at specific locations throughout the genome. These polymorphisms include single base pair changes and repetitive DNA elements. The polymerase chain reaction (PCR) can be used to analyze polymorphisms at several loci within the human genome. Since an individual's genome contains a different combination of polymorphisms, we can generate a unique "DNA fingerprint" for that person.

Forensic scientists use DNA fingerprinting to analyze biological evidence collected at crime scenes. After the crime scene sample is analyzed, it is compared to DNA fingerprints from the suspects or those stored in CODIS (COmbined DNA Index System), a database of DNA fingerprints from convicted offenders, other crime scenes, and missing persons (Figure 2). Each DNA fingerprint analyzes thirteen separate loci, making the odds of an exact match less than one in a trillion. This evidence is then used in court to link a suspect to a crime scene.

**Figure 2 – Human DNA Sequences Analyzed by CODIS**



## Pre-Lab Assessment

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**Before performing the exercise, be sure that students are comfortable with the following topics:**

1. Forensic science is not a separate field of science. Instead, it is the application of scientific knowledge to answer questions of interest within the legal system.
  - a. Proper evidence collection, documentation, and handling
  - b. Classical and modern techniques are used to analyze evidence
  - c. Forensic scientists prepare reports detailing their experiments and act as expert witnesses in court
  - d. Results from forensic science experiments provide evidence to support a case, but do not alone prove guilt.
2. Presumptive versus confirmatory tests
  - a. A presumptive test strongly indicates the identity of a tested substance.
    - i. Advantages – fast, sensitive, low-cost, good for screening samples
    - ii. Disadvantages – high false positive rate
  - b. A confirmatory test makes a positive identification of a crime scene sample.
    - i. Advantages – sensitive, definitive test, low false positive test
    - ii. Disadvantages – expensive, time-consuming
3. Testing crime scene samples for blood
  - a. The Kastle-Meyer Test is a presumptive test for blood
    - i. Uses phenolphthalein to detect the presence of blood in crime scene samples. Positive samples change pink.
      1. Advantages – fast, sensitive, inexpensive
      2. Disadvantages – high false positive rate
  - b. Blood group typing is a confirmatory test for blood
    - i. Uses antibodies to detect the presence of blood antigens in crime scene samples. Positive samples will agglutinate.
      1. Advantages – sensitive, confirms the presence of blood, assigns blood group type which can be used as evidence
      2. Disadvantages – more expensive, time-consuming
4. DNA Fingerprinting analyzes polymorphisms at several loci within the human genome.
  - a. DNA is extracted from biological samples, amplified using the Polymerase Chain Reaction, and analyzed using agarose gel electrophoresis
  - b. After the crime scene sample is analyzed, it is compared to DNA fingerprints from the suspects or those stored in CODIS.

## Laboratory Guidelines

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### LABORATORY SAFETY:

1. Gloves and goggles should be worn routinely as good laboratory practice.
2. Exercise extreme caution when working with equipment that is used in conjunction with the heating and/or melting of reagents.
3. DO NOT MOUTH PIPET REAGENTS - USE PIPET PUMPS.
4. Exercise caution when using any electrical equipment in the laboratory.
5. Always wash hands thoroughly with soap and water after handling reagents or biological materials in the laboratory.

### LABORATORY NOTEBOOKS:

#### Before starting the experiment, students should:

- Carefully read the introduction and the protocol. Use this information to form a hypothesis for this experiment.
- Predict the results of the experiment.

#### During the experiment, students should:

- Record any observations in a lab notebook.

#### After the experiment, students should ask the following questions:

- Interpret the results – does the data support or contradict the hypothesis?
- If the experiment was repeated, what should change? Revise the hypothesis to reflect this change.

Be sure to have your students reflect upon these points in their lab notebooks. Students should also submit a formal lab report in the form of a crime scene report upon completion of the exercise.

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## Activity #1: Reviewing Witness, Suspect, and Alibi Statements

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*Students should work in groups of 2-4.*

### REQUIRED MATERIALS:

- Print outs of the Police Report (page 8)

Before performing the experiments, students will critically read and analyze the police report. The statements in this document will be used in conjunction with the forensic evidence to complete the expert statement at the end of this exercise.



## EDVOTEK POLICE REPORT

**RESPONDING OFFICER:** Evie Dence

**DETAILS OF EVENT:**

Late one night Dr. Elektra Phoresis worked on an important biotechnology experiment in the laboratory. She was very close to creating a groundbreaking vaccine that could save many lives. After working in the lab all day, Elektra decided to go home to eat dinner and get a good night's rest. The next morning the lab was in shambles. The scientist found that many important pages were ripped from her lab notebook. Furthermore, security footage showed that someone had stolen some critical reagents from the laboratory.

Upon investigating the crime scene, Officer Dence identified a broken window in the laboratory as a potential entry point by the suspect. The forensic scientists believe the perpetrator may have been cut on the broken glass, as several blood-like samples were found around the crime scene. The potential biological samples were collected as evidence to be analyzed.

**SUSPECTS:**

**Suspect 1: Mr. Cy Ence**

Mr. Ence owns the pharmaceutical company that was trying to buy the vaccine. The negotiations had stalled because Ence wanted to wait until the Stage One Clinical Trials had been performed. Phoresis had started talking to other pharmaceutical companies, which angered him. Ence stated that he was home with his wife at the time of the lab break-in, but their accounts of the evening do not match up entirely. Ence also had some minor lacerations on his hands that he said were from a recent fall.

**Suspect 2: Dr. Gene Ticode**

Dr. Ticode is a colleague of Dr. Phoresis. They have always been highly competitive with one another. The suspect was working on the same vaccine, but the preliminary results from the Phoresis lab were more promising. Ticode says he was at home watching television when the break-in occurred, but there were no other witnesses to confirm his alibi.

**Suspect 3: Ms. Vi Russ**

Ms. Russ is a graduate student in the Phoresis lab. She had been working with Phoresis on the vaccine at the time it was stolen. Russ had been bragging about her ideas to improve the vaccine. She said she would sell the information to Mr. Ence, negating the deal with Dr. Phoresis. Ms. Russ said she was at a movie with her boyfriend at the time of the break-in. Corroborating evidence includes movie stubs and credit card receipts for concessions.

**SAMPLES PENDING FORENSIC ANALYSIS:**

Potential blood evidence found at the crime scene. Tests to be performed include Kastle-Meyer testing, Blood Group Typing and DNA fingerprinting.



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## Activity #2: Identifying Blood Samples

Students should work in groups of 2-4.

### REQUIRED MATERIALS:

- Edvotek Kit 191 – reagents and components for Module I
- 95-100% Ethanol

### OBJECTIVE:

Several fluids were found at the crime scene. In this experiment, students will use the Kastle-Meyer test to determine which crime scene samples contain blood.

### PRE-LAB PREPARATION:

Before performing this experiment, the students should read the background information for kit 191 and the procedure for Module I.

Prepare the evidence as outlined by kit 191 on pages 15 and 16. Take care to prevent cross contamination of the crime scene samples, as the Kastle-Meyer test is very sensitive. Results will vary depending upon instructor's preparation. At least one "crime scene" sample should test positive for blood.

When performing the experiment, be sure to emphasize good laboratory practices (proper personal protective equipment, exercise caution when using equipment, hand washing, etc.). Be sure to emphasize the fact that cross contamination can lead to false positive results.

### Cat. #191

### Forensics Blood Typing

**For 10 Lab Groups.** Introduce students to some of the techniques used by forensics scientists for analyzing blood. First, check for the presence of blood typing using the phenolphthalein test. Then, apply the concept of blood type-based screening for potential suspect(s) present at a crime scene.

[www.edvotek.com/191](http://www.edvotek.com/191)



## Activity #2: Discussion Questions

### 1. Why would we perform the Kastle-Meyer test before other forensic tests?

The Kastle-Meyer test is a presumptive test for blood. It is fast, easy, and cost-effective, so it is performed before other tests. However, since it has a high rate of false positives, the results must be confirmed with further testing.

### 2. Would the Kastle-Meyer test detect blood that is not human in origin?

Yes, the Kastle-Meyer test would react with any hemoglobin-based blood (i.e., animal blood) in the same way. Therefore, further testing is required to determine from which species the blood originated.

## Activity #3: Forensic Blood Typing

*Students should work in groups of 2-4.*

### REQUIRED MATERIALS:

- Edvotek Kit 191 – reagents and components for Module II
- Markers

### OBJECTIVE:

The Kastle-Meyer test identified that a sample collected at the crime scene was potentially blood. In this experiment, students will use the blood group typing to confirm that the crime scene sample was blood. In addition, they will determine the blood types of crime scene and suspect samples.

### PRE-LAB PREPARATION:

Before performing this experiment, the students should read the background information for kit 191 and the procedure for Module II.

Prepare the evidence as outlined by kit 191 on page 16. These samples should be prepared no more than 24 hours before starting the experiment. Mr. Ence is represented by S1, Dr. Ticode is represented by S2, and Ms. Russ is represented by S3. Results will be found in the instructors guide for kit 191.

## Activity #3: Discussion Questions

### 1. Why is blood group testing a confirmatory test for blood?

This test relies on the interaction of blood cell surface antigens (A or B) with their corresponding antibodies. If the antibody in solution recognizes the blood cell surface antigens, large complexes are formed. These complexes precipitate out of solution, which is visible to the naked eye as a granular solid. This precipitation reaction (agglutination) is specific to blood, which is why it is classified as a confirmatory test.

### 2. How can forensic scientists use blood group evidence?

Although blood groups cannot be used to identify a particular suspect, this test can be used to narrow down the list of potential suspects. Suspects who match the blood group will be examined more closely, and suspects that do not match the blood group can be eliminated from the inquiry.



## Activity #4: DNA Fingerprinting

*Students should work in groups of 2-4.*

### REQUIRED MATERIALS:

- Kit – DNA Fingerprinting by PCR Amplification <http://www.edvotek.com/130>
- Fixed Volume MiniPipets® 35 µl [http://www.edvotek.com/587-2\\_0](http://www.edvotek.com/587-2_0)
- Disposable micropipet tips <http://www.edvotek.com/636>
- Electrophoresis Apparatus <http://www.edvotek.com/504>
- Power Supply <http://www.edvotek.com/509>
- White Light Box (optional) <http://www.edvotek.com/552>
- Microwave or Hot Plate

### OBJECTIVE:

Previous testing has identified which crime scene samples were blood. In this DNA fingerprinting exercise, students will analyze crime scene and suspect samples using agarose gel electrophoresis.

### PRE-LAB PREPARATION:

Before performing this experiment, the students should read the background information and the procedure for kit 130. Perform the experiment as outlined in the literature for kit 130. Be sure to review the Instructor's Guide for best practices. Mr. Ence is represented by S1, Dr. Ticode is represented by S2, and Ms. Russ is represented by S3. Results of the experiment will be found in the instructors guide for kit 130.

#### Cat. #130

#### DNA Fingerprinting by PCR Amplification

**For 8 Gels.** Forensic DNA fingerprinting has become a universally accepted crime-fighting tool. Recent advances use the polymerase chain reaction (PCR) to amplify human DNA obtained from crime scenes. This experiment, based on a crime scene scenario, has an inquiry-based component.



[www.edvotek.com/130](http://www.edvotek.com/130)

## Activity #4: Discussion Questions

### 1. Does DNA evidence at a crime scene prove that the suspect committed a crime?

A match does not prove when the person was at the crime scene, or that they committed the crime, or intent. Many other lines of evidence need to come together to prove that a person committed a crime.

### 2. Why does CODIS look at 13 different loci when making DNA matches?

The probability of two unrelated samples matching at one position in the genome is fairly high. However, as more points of comparison are added, the probability of two samples being identical gets smaller and smaller. In fact, the chances of two individuals having exactly the same DNA profile is ~30,000 million to one.

#### Cat. 5062

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- 1 Cat. #636  
Yellow Micropipet Tips  
(1 - 200 µl / 2 Racks of 96)
- 1 Cat. #130  
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# EDVOTEK DEPARTMENT OF FORENSIC SCIENCES

Final Report Prepared by: \_\_\_\_\_ in cooperation with \_\_\_\_\_, \_\_\_\_\_, and \_\_\_\_\_.

**Information gathered from Police Report:**

Suspect 1: \_\_\_\_\_

Motive: \_\_\_\_\_

Alibi: \_\_\_\_\_

Suspect 2: \_\_\_\_\_

Motive: \_\_\_\_\_

Alibi: \_\_\_\_\_

Suspect 3: \_\_\_\_\_

Motive: \_\_\_\_\_

Alibi: \_\_\_\_\_

**Forensic Analysis of Crime Scene Samples:**

The results of the Kastle-Meyer test are as follows:

Sample ID	Sample Description	Kastle-Meyer Test
Positive Control		
Negative Control		
Crime Scene Sample 1		
Crime Scene Sample 2		
Crime Scene Sample 3		
Crime Scene Sample 4		
Crime Scene Sample 5		
Crime Scene Sample 6		
Crime Scene Sample 7		
Crime Scene Sample 8		

In my own words, the results show the following:

\_\_\_\_\_  
\_\_\_\_\_  
\_\_\_\_\_



# EDVOTEK DEPARTMENT OF FORENSIC SCIENCES

PAGE TWO, CONTINUED

The results of the blood group typing are as follows:

	A	B	AB	O	CS	S1	S2	S3
Anti A	<input type="radio"/>							
Anti B	<input type="radio"/>							

In my own words, the experimental samples show the following:

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The results of the DNA Fingerprinting  
experiment are as follows:

In my own words, the results show the following.:

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**Conclusions** (include information gathered from Police Report):

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I affirm that these results are true and correct to the best of my knowledge.

Dated this \_\_\_\_ day of \_\_\_\_\_, 20\_\_\_\_

\_\_\_\_\_  
Signature of Forensic Scientist