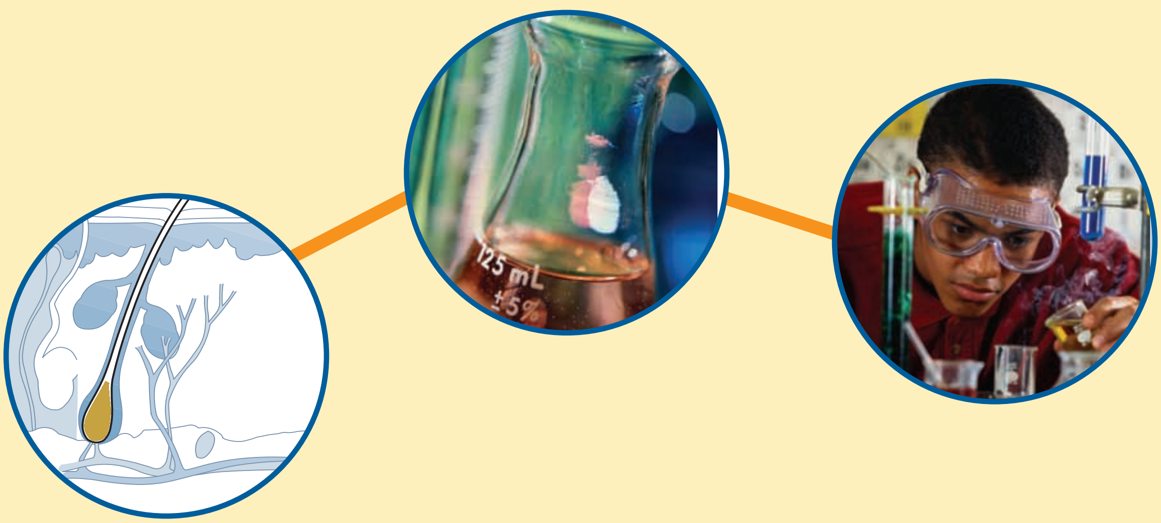
truTV

Forensics in the Classroom

Developed as part of a continuing educational partnership with the **American Academy of Forensic Sciences** *and in collaboration with the* **National Science Teachers Association.**

www.trutv.com/forensics_curriculum



truTV
NOT REALITY. ACTUALITY.™

WARNING and SAFETY PRECAUTIONS

Working with your students on these projects gives you an excellent opportunity to instill in them good work practices, in particular the practice of dealing safely and efficiently with chemicals and other materials that may be potentially hazardous and/or expensive. It is important to comply with the warnings and safety instructions concerning the use of your materials, as well as any laws regarding the disposal of the materials. For safety and for avoiding waste, you should inform your students of these warnings, instructions and laws, and the reasons behind them. The goal is to educate the students without implanting or enhancing any unreasonable fears.

Safety is an important part of any laboratory exercise. Plastic safety products, as illustrated below, can help protect your students as they conduct chemistry experiments such as those in the FIC units. To find out more about plastics log on to the American Plastics Council at http://www.americanplasticscouncil.org.

Plastic safety devices, if used properly, can help save kids from injuries.

¥ Safety eye goggles
¥ Splatter-proof face shields
¥ Plastic apron to keep dangerous spills away
¥ Heat-resistant plastic gloves, chemical-resistant vinyl gloves
¥ Shatter-resistant plastic containers for acids
¥ Child-resistant plastic safety caps for storing dangerous substances
¥ Plastic film for spill-resistant laboratory beaker sealing
¥ Plastic tubs immune to harsh solvents for appropriate disposal
¥ Emergency plastic face mask and eyewash bowl ¥ Flame retardant countertops made with plastic composite

MS Unit 1 Teacher Overview: It’s Magic!

Level of Difficulty: Intermediate

UNIT DESCRIPTION

A crime has been committed in the school involving vandalism and a cut gas line in the cafeteria. The school resource officer (a teacher or principal) will enlist the help of biology students to lead the investigation and help him/her solve the crime. The unit begins with the resource officer introducing the crime and leading the students on a tour of the crime scene. Students will make notes and take pictures (Polaroid® or digital) of the crime scene for later review. With the help of a tipster, investigators discover a link between the crime and a recent initiation rite for an underground club. Using the information garnered from the club s Web site, students begin to analyze the evidence. In Lesson 2, students will use chemical indicators to analyze what appears to be stomach contents found at the crime scene. Positive tests for raw liver and sugar (from soda) establish a link to the club s inductees, because eating and drinking large quantities of these was a requirement of initiation. Next, students will use wet-mount and scale-cast slides of the evidence hair found in the stocking to compare and contrast with the club inductees hair samples. Students discover that three of the inductees could be the source of the evidence hair. With this information, students proceed to blood analysis of the sus-pects. By using blood typing and DNA profile analysis, students are able to pinpoint the perpetrator who cut the gas line. Students complete the Investigative Report and then submit their findings to the school resource officer to close the case.

IN ADDITION TO THIS TEACHER OVERVIEW, UNIT 4 INCLUDES:

Lesson 1: Introduction to Forensics and the Mystery

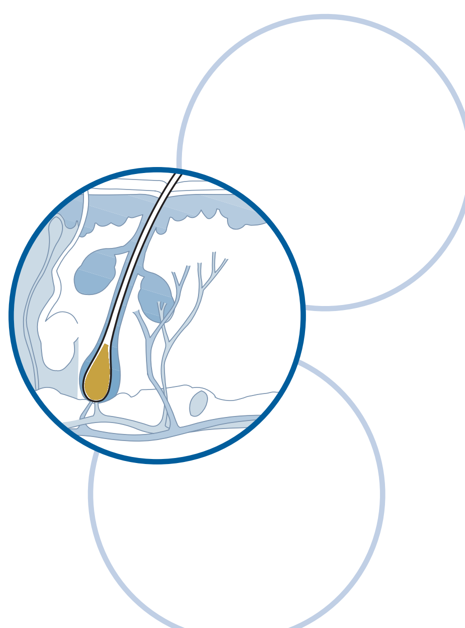
¥ *Forensics Terms and FAQ Sheet handout*
¥ *Mystery Synopsis handout*
¥ *Forensic Tests handout*
¥ *Part II: The Cafeteria (optional)*

Lesson 2: Indicators and Enzymes Test

¥ *Indicators and Enzymes Activity Sheet*
¥ *Persons of Interest handout*

Lesson 3: Hair Analysis

¥ *Hair Identification Activity Sheet*



Lesson 4: Blood, Chromosomes, and DNA Analysis

¥ *Kastle-Meyer Color Test Activity Sheet (Optional: see Appendix)*
¥ *Blood Analysis Activity Sheet*
Part I: Blood Typing
Part II: Karyotype Analysis
Part III: DNA Profile Analysis
¥ *Investigative Report*

Unit Epilogue

DESIRED OUTCOMES:

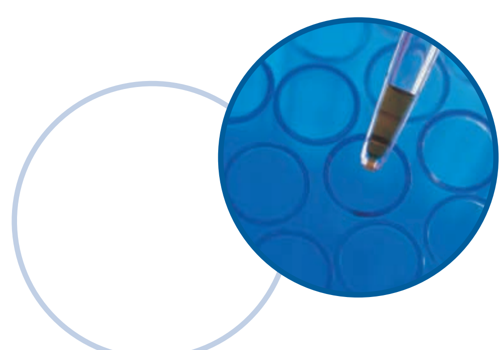
After completing the unit, students should:

¥ *Develop basic skills in observation, data collection, and data analysis*
¥ *Understand organic molecule properties, including enzyme function*
¥ *Identify uses for chemical indicators*
¥ *Understand the structure of hair and how it is analyzed in forensic investigations*
¥ *Identify and use various tests using blood components*
¥ *Differentiate between presumptive and conclusive tests*
¥ *Analyze and synthesize several pieces of data to reach a conclusion*
¥ *Understand and perform scientific inquiry*

NATIONAL STANDARDS ADDRESSED:

After completing this unit, students should:

¥ *Develop an understanding of the cell (including enzyme function, organic molecules, and indicators)*
¥ *Develop an understanding of the molecular basis of heredity (including chromosomes, DNA, and genetic inheritance)*
¥ *Understand and perform scientific inquiry*
¥ *Analyze and synthesize several pieces of data to draw a conclusion*
¥ *Develop an understanding of science and technology*



IMPORTANT TERMS:

Active Site: *The region of an enzyme s surface to which a specific set of substrates bind.*

Alleles: *Alternate forms of the same gene; T is the allele coding for tall pea plants, while t is the allele coding for short pea plants.*

Aneuploidy: *The condition of having an abnormal number of chromosomes; for example, a female with Turner s syndrome has 45 chromosomes because she inherits only one X for the sex chromosome pair.*

Autosome: *Any eukaryotic chromosome that is not a sex chromosome; autosomes are present in the same number and kind in both males and females of the species.*

Codominant Inheritance: *When two alleles act in unison to create a trait or characteristic, neither is dominant over the othe; both alleles are expressed. AB blood type is caused by the inheritance of IAIB.*

Disaccharide: *A type of carbohydrate formed by two simple sugars bonded covalently; an example is sucrose (formed by two glucose molecules).*

Down Syndrome: *A condition in which an individual has three of chromosome number 21; symptoms include mental retardation, short stature, larger than normal tongue and forehead.*

Enzyme: *A type of protein known as a biological catalyst because it speeds up chemical reactions by reducing the amount of activation energy needed for the reaction; examples include lactase, amylase, lipase, and peroxidase.*

Genotype: *The genetic makeup of an organism; the pea plant has the TT genotype for the height gene.*

Hair Cortex: *The layer of the hair shaft containing pigment-storing cells.*

Hair Cuticle: *The outermost layer of the hair shaft.*

Hair Root: *The part of the hair below the epidermis.*

Hair Shaft: *The part of the hair above the epidermis.*

Karyotype: *A picture of an individual s chromosomes taken with the help of a microscope when a cell is actively dividing.*

Lock-and-Key Theory of Enzyme Function: *The theory that an enzyme fits into a substrate site like a lock into a key as the result of the respective shape of the enzyme and the specific substrate site at which it is to operate.*

Monosaccharide: *A type of carbohydrate called a simple sugar; the most common are glucose and ribose.*

Multiple Allele Inheritance: *When there are more than two alleles inherited for the same gene, such as IA, IB, and i for the blood type gene.*

Organic Molecules: *Carbon-based molecules found in living things (e.g., carbohydrates, proteins, lipids, and nucleic acids).*

Peroxidase: *An enzyme that speeds up the breakdown of hydrogen peroxide into water and oxygen.*

Phenotype: *The physical appearance of an individual organism as opposed to the general characteristics of the species to which the organism belongs.*

Polysaccharide: *A type of carbohydrate formed by linking many monosaccharides to form a long chain; glycogen, starch, and cellulose are examples.*

Protein: *A chain of amino acids joined by peptide bonds; examples include structural proteins such as keratin and collagen, as well as functional proteins such as enzymes.*

Substrate: *In the present context, a molecule broken down or otherwise operated on by an enzyme.*

Turner's Syndrome: *A condition in which a female has only one X for the sex chromosomes; symptoms include short stature, webbed neck, and reproductive sterility.*

XYY "Super" Male: *A male with an extra Y chromosome; symptoms include tall stature and higher-than-average testosterone production.*

Control Test: *Generally, a test of a system that includes all the same features as the experiment system except for the feature being tested/studied; a test run that acts as a comparison.*

MYSTERY SYNOPSIS:

Early one Monday morning, employees at a high school discover that someone broke into the school over the weekend. Most of the damage seems to be cosmetic vandalism, but a cafeteria worker smells gas upon entering the kitchen. As a result, the administration cancels classes that day, fearing a potential explosion or fire. The custodian finds that a gas line in the cafeteria has been cut. Because no valuables were stolen, the school resource officer and investigators suspect that students are the culprits. Investigators find what might be drops of blood on and next to the cut gas line. They also discover what appears to be stomach contents on the cafeteria floor. Lastly, they find a discarded knee-high stocking that contains strands of hair. School officials receive a tip from a student who tells them that an underground club inducted five new members over the weekend. This information comes courtesy of a web site maintained by the club. According to the site, the five inductees had an initiation ceremony that included eating raw liver and drinking large amounts of soda. Initial questioning of the five persons of interest reveals little information, but investigators are able to get hair samples and physical descriptions.

MATERIALS NEEDED:

Reproducibles

1. *Forensics Terms and FAQ Sheet*
2. *Mystery Synopsis/Part II: The Cafeteria (Part II is optional)*
3. *Indicators and Enzymes Activity Sheet*
4. *Persons of Interest handout*
5. *Hair Identification Activity Sheet*
6. *Blood Analysis Activity Sheet Part I: Blood Typing Part II: DNA Profile Analysis*
7. *Kastle-Meyer Blood Identification Test (see Appendix)*
8. *Investigative Report handout, detailing what information needs to be provided to conclude the investigation*
9. *Unit Epilogue*

Lab Materials

Indicators and Enzymes Activity — Lesson 2:

- *Raw calf liver*
- *Soda (preferably a soda one that is not dark and is high in sugar content)*
- *Blender*
- *4 test tubes and test tube rack (or spot plates)*
- *Droppers*
- *Hot plate and beaker*
- *Tongs*
- *Wax pencil for marking test tubes*
- *Distilled water*
- *Hydrogen peroxide*
- *Biuret reagent*
- *Iodine*
- *Benedict s indicator*
- *Monosaccharide solution (e.g., glucose, dextrose)*
- *Disaccharide solution (sucrose)*
- *Polysaccharide solution (starch)*
- *Protein solution (gelatin)*

Hair Analysis Activity — Lesson 3 (per group):

¥ Microscope
¥ 10 slides
¥ 10 coverslips
¥ Dropper bottles (for water)
¥ Forceps (tweezers)
¥ Hair samples from three different individuals (e.g., black hair, red hair, and bleached blonde hair)
¥ Clear fingernail polish (Sally Hansen® HARD AS NAILS® works well)
¥ Permanent marker

Blood Analysis Activity — Lesson 4:

¥ Glue sticks or tape
¥ Scissors
¥ Copy paper
¥ Kastle-Meyer ingredients (Optional: see Appendix):
  - ethanol (95%) or distilled water
  - Kastle-Meyer reagent*
    phenolphthalein**
    potassium hydroxide
    absolute ethanol
    (100%) zinc dust
  - hydrogen peroxide (3%)
  - sample of animal blood (can be obtained from a butcher) OR beef or calf liver (both contain high blood content) homogenized in a blender
  - filter paper (cotton swabs may be used instead, if needed)
  - latex/vinyl gloves
  - chemical splash goggles
  - droppers
  - biohazard or other disposable bag

* Premixed Kastle-Meyer reagent can be purchased from Antec, Inc.:
1-800-448-2954 or 1-502-636-5176

** Phenolphthalein can be purchased online from the following vendor, Sigma:
http://www.sigmaaldrich.com or 1-800-325-3010; Product number P8903

ORDER OF ACTIVITIES:

1. *Introduction to the Crime Scene and the Mystery*
2. *Introduction to Forensics*
3. *Indicators and Enzymes Activity*
4. *Hair Analysis Activity*
5. *Kastle-Meyer Test (Optional: see Appendix)*
6. *Blood Analysis Activities:*
   - ¥ *Blood Typing*
   - ¥ *Karyotype Analysis*
   - ¥ *DNA Profiles*
7. *Investigative Report*
8. *Epilogue*

You can present students with the story s epilogue when all activities have been successfully completed, or hold a classroom discussion to share various theories and the mystery conclusion.

Note: *Any time you use chemicals in your classroom or lab, be sure to read and follow the safety instructions for use and disposal that are provided in the MSDS information sheet that came with your lab materials.*

ADDITIONAL RESOURCES FOR TEACHERS:

http://www.thegateway.org
Performing a search for forensics or criminology will produce articles of interest.

http://www.aafs.org
The Resource/Forensics section provides links to forensics publications and organizations.

LESSON 1: INTRODUCTION TO FORENSICS AND THE MYSTERY

OBJECTIVE:

Students will learn that a crime has been committed at their school. After viewing the crime scene and learning about the mystery, students will review the Forensics Terms and FAQ Sheet as an introduction to forensic science. (The extent of the forensics review should be based on students familiarity with forensic science.) Based on the information they have at hand, students will determine how to proceed in conducting the investigation to solve the crime.

MATERIALS NEEDED:

Reproducibles

- *Forensics Terms and FAQ Sheet*
- *Part I: Mystery Synopsis*
- *Part II: The Cafeteria (Note: This handout should be used only for students who don t get to visit the "crime scene."]*

Materials

- *Polaroid® or digital cameras (one for each team or enough for the class to share)*
- *Crime Scene elements:*
  - *a. cafeteria setting (if using the cafeteria isn t possible, work with your custodial staff to find a safe, suitable area)*
  - *b. stocking or knee-high hose with hair strands in it*
  - *c. cut tubing or garden hose to simulate gas line*
  - *d. simulated blood drops*
  - *e. simulated stomach contents (see Lesson 2 for recipe)*
  - *f. crime scene or caution ribbon*
  - *g. access to the Internet or the reproducible showing a web site s home page*

TIME REQUIRED:

- *Teacher Prep Time: 30 minutes*
- *Class Time: 60 — 90 minutes*

LESSON DESCRIPTION:

In this lesson, students will learn about the mystery from the school s resource officer, who will come to class to announce that a crime has been committed. Students will then be instructed to gather note-taking materials and Polaroid® or digital cameras to begin their crime-scene investigation. (Teachers will have set up the crime scene in the school s cafeteria before the first les son.) Students will be allowed to view the crime scene and take notes, but will not collect evidence at that time.

TEACHER

After visiting the crime scene and returning to class, students will be provided with the Mystery Synopsis, followed by the Forensics Terms and FAQ Sheets. They will be introduced to forensic science and will learn about various investigative techniques and evidence examination methods used by investigators. Then, after discussing what they see and know about this particular crime, students will begin their investigation. (Note: You may wish to begin the Forensics unit with the Introduction to Forensics and the FAQs without mentioning the crime. Then time the arrival of the police or resource officer to coincide with finishing the introduction. Students may then proceed directly to the crime scene and mystery synopsis.)

BACKGROUND INFORMATION:

Before beginning this lesson, you should be acquainted with the history of forensics and types of tests that can be performed. Be prepared for lots of questions from the students about various forensics-related topics, because they will be curious about certain techniques that they have seen on TV. They may have many misconceptions that need to be corrected.

You should also obtain permission from the principal and cafeteria manager to set up a crime scene. Explain the scope of the unit and the times you will be visiting the crime scene. You may want to choose a remote area of the cafeteria that doesn t see a lot of foot traffic in case the crime scene has to be used during lunchtime. If your school will not allow you to set up the crime in the cafeteria, you may choose an alternate location but will need to amend the directions and descriptions slightly.

Next, discuss the crime with the school resource officer or community police officer. School resource officers are usually local policemen or security personnel who are assigned to specific schools and are present on campus during the day. Most police officers are more than willing to participate in school activities. Discuss the crime by sharing the mystery synopsis and the crime scene setup. Gauge how interested the officer is by asking if he/she would like to simply introduce the crime, lead a tour of the crime scene, or be involved in both the introduction and the conclusion. Officers may also want to discuss their department s local forensic lab with students, as well as describe how not to contaminate a crime scene. This will be important as you begin this activity. It is exciting to have students submit their crime reports to the officer, even if the officer simply collects them and returns them to you. Police officers enjoy meeting students who are not in trouble!

If you are unable to enlist the help of a police or school resource officer, you may want to ask the principal, the assistant principal, or another person from within the administration to help introduce the crime. Keep in mind, you may certainly introduce the crime yourself, although it creates a more dynamic lesson to have another individual do it. You may also decide to offer extra credit to students who solve the crime first.

LESSON STEPS:

Teacher Preparation

1. *Establish and coordinate the participation of your school resource officer or local police officer to arrive and announce the crime to the class. (Optional)*

2. *Obtain permission from your principal and cafeteria manager to set up the crime scene.*

3. *Section off an area of the cafeteria with crime scene or caution tape. (This can be obtained from a police officer.)*

4. *Place a piece of cut piping or tubing at the crime scene (preferably sticking out from behind a stove or oven) with drops of ketchup or simulated blood on the end of the tube. Also place some drops on the floor around the tubing.*

5. *In one area of the crime scene, place the knee-high stocking with pieces of hair (refer to Lesson 3 for hair color) on and around the stocking.*

6. *In the same general area of the knee-high stocking, make a small pool of the simulated stomach contents, as if the individual had torn the stocking off of his/her head to regurgitate his/her food.*

7. *If the crime scene remains during a lunch period, make sure all elements are intact before you bring your next class to the area.*

8. *Obtain digital cameras from the library or computer teachers, or write a note to parents asking permission for students to bring in personal Polaroid® or digital cameras on the crime scene day.*

Note: *Any time you use chemicals in your classroom or lab, be sure to read and follow the safety instructions for use and disposal that are provided in the MSDS information that came with your lab materials.*

Lab Execution

1. Announce the crime. (Either yourself or by having the school resource or local police officer do it.) Inform students that they will visit the crime scene and are responsible for taking meticulous notes about what they see, being careful to include descriptions of anything they might consider to be evidence. Divide the class into teams. Establish rules for what students may and may not touch and explain that they will not be collecting evidence at this time but merely viewing the crime scene and taking pictures of what they deem to be important. (If students are not able to take pictures, direct them to sketch the crime scene in their notes.)

2. *After coming back to class, introduce the Mystery Synopsis. (It is written in a way that you can use it as the first step after visiting the crime scene.) Briefly discuss the mystery to ensure that students understand its basic components. Many questions are left unresolved, so you may have to remind students that they should try to arrive at those answers. Instruct students to write down their notes and questions that need to be answered. They can add to these notes each day.*

   (**Note:** *If you are unable to create a mock crime scene for students to see, you can use the optional Part II: The Cafeteria handout and associated diagram to help them visualize the crime scene.)*

3. *When you reach the part in the Mystery Synopsis that discusses the club s web site, you can stop and have students access the mock site at http://www.philophiles.com.*

   If you do not have access to the Internet in class, you can encourage students to access the site at home, or use the web page handouts provided in this lesson.

4. *Hand out the Forensics Terms and FAQ Sheet as an introduction or a review. Begin the discussion by stimulating students prior knowledge. If you have already completed a forensics unit in class, your students will likely recall their experiences with it. Even if this is their first unit, many students will be familiar with current TV shows that use forensics as the central theme. If this is your first unit, ask students to write a definition of forensics in their notebooks before you start the discussion, so that everyone gets a chance to answer. The discussion will naturally revolve around the specific mystery of this unit, but try to make sure that it is not limited to the specific mystery.*

   (**Note:** *You may wish to begin the unit with the Introduction to Forensics and the FAQs. Then time the arrival of the police or resource officer to coincide with the end of the introduction. Once students learn of the crime, they may proceed directly to the crime scene and mystery synopsis.)*

5. *Lead a discussion about the possible evidence that students observed and what investigative techniques might be of use in solving the crime. (If they do get to observe the mock crime scene, be sure that all the potential pieces of evidence are mentioned in your discussion.) Lead students to conclude that analysis of the liquid on the floor may prove to be useful in light of the clues provided by the secret club s web site.*

   (**Note:** *It is important to emphasize to students that in this investigation, they will perform the investigative labs in sequence. In a typical criminal investigation, however, examination of evidence can be simultaneous and is based on the time-sensitive nature of any evidence present.)*

ACADEMIC EXTENSIONS/MODIFICATIONS:

1. If space for a crime scene setup is not available or feasible during the school day, teachers can set up the scene after school hours and use a Polaroid® camera to take pictures of it. Instead of actually visiting the "scene," students will rely on the pictures provided to them and the Mystery Synopsis to visualize the setting. Additionally, there is an optional handout, Part II: The Cafeteria, in the Synopsis that describes the crime scene for students who do not get to see the actual site.

2. To abridge the first lesson, distribute the Forensic Terms and FAQ Sheets as homework, and begin the class with the introduction to the crime scene and mystery.

3. Ask the truTV Mobile Crime Unit for your city to come visit the school and allow the students to tour the van/bus. If one isn t available, ask a local police detective or sheriff to visit your school.

4. If circumstances allow, you can conduct an interview of a law official (after he/she has been briefed about the scope of this forensics unit) and have it videotaped for all your classes.

FORENSICS FAQ SHEET

Q: What is forensic science, and how can it aid in criminal investigations?

A: *Forensic science is not limited to just criminal investigations. It is essentially the application of science to law in events subject to criminal or civil litigation. More commonly, though, it is applied to the investigation of criminal activity. The term "forensic science" includes many different technical fields, including (but not limited to) physics, chemistry, biology, engineering, psychology, and medicine. Forensic scientists might study a Questioned Document (QD), DNA evidence found at a crime scene, or the mental and emotional state of a suspect. Investigators turn to forensic scientists to discover additional evidence that requires specialized training to analyze and interpret.*

Q: How long have investigators been using forensic science?

A: *Forensic science has been around for nearly 900 years. The first recorded application of medical knowledge to the solution of a crime was in the year 1248. The first known use of a forensic chemical analysis was in 1836, when James Marsh, a Scottish chemist, detected arsenic poisoning in connection with a criminal investigation. Techniques involving blood typing have been used since 1900, when Karl Landsteiner discovered human blood types. Developed only within the past 20 years, DNA tests are now commonplace and are revolutionizing the field.*

Q: What are some types of evidence that investigators look for?

A: *Investigators look for clues such as:*

- ¥ *fingerprints, palm prints, and footprints*
- ¥ *shoeprints*
- ¥ *fibers from clothes*
- ¥ *handwriting on a ransom note*
- ¥ *presence of chemicals*
- ¥ *blood spatters*
- ¥ *DNA samples (can be obtained from hair, skin cells, blood, semen, or saliva)*
- ¥ *residue from accelerants (e.g., compounds used to speed up fires set by an arsonist)*
- ¥ *gunshot residue on hands and clothing*
- ¥ *bullet casings*
- ¥ *tool marks (e.g., marks left on a bullet by a gun when fired)*
- ¥ *insect and mold growth in a body, as well as body temperature (to determine time of death)*
- ¥ *bullet residue around bullet holes*
- ¥ *patterns of gunshot residue spray (can help determine the distance the shooter was from the victim)*
- ¥ *gunpowder burns*

STUDENT HANDOUT

Q: Why are fingerprints important?

A: *If you look at the palms of your hands and soles of your feet, you will see a maze of lines in your skin that curve, break apart, and join back together. The places where skin ridges break apart and join together are unique for every person. This unique pattern allows forensic investigators to trace a print found at the scene of the crime back to a specific person. Even identical twins have different fingerprints!*

Though one of the older forms of investigative techniques, fingerprint identification is not without some controversy. One recent court ruling declared that fingerprint examination and identification did not qualify as a science, in part because an examiner subjectively decides if two sets of prints match. There is no uniform set of requirements used by all analysts to determine a positive match, so critics argue that fingerprint identification should not be considered scientific evidence. It is important to note, however, that other court challenges to the science of fingerprint identification have been rejected.

Q: How long after a crime can DNA evidence be collected?

A: *DNA is a wonderfully stable molecule. Researchers have been able to recover usable DNA from Egyptian mummies thousands of years old. Each individual strand of DNA is made with strong, unreactive bonds. The strands of DNA twist around each other to form the well-known double helix, concealing weaker hydrogen bonds in the middle of the molecule. There are so many billions of hydrogen bonds that even though one is not strong by itself, the cumulative effect is strong enough to keep DNA intact.*

Q: Are some forensic tests, by their nature, NOT conclusive?

A: *Yes, not all tests performed by forensic investigators are conclusive. Some tests, such as morphological hair analysis (microscopic comparison between the appearance of two or more hairs for points of similarity), are presumptive, meaning they do not provide absolute proof for what the investigator is testing. When investigators use presumptive tests, which are often quick, easy, and sensitive ways to initially screen evidence from a crime scene, they must then follow up with conclusive tests of the issues of interest.*

Q: What is microscopic hair analysis, and how is it useful in a criminal investigation?

A: *When a crime is committed, physical evidence in the form of hair is often left behind by the perpetrator and/or the victim. Police collect these hair samples and forensic scientists examine the structure (morphology) of the individual hairs microscopically in an attempt to identify potential suspects or victims. Microscopic hair analysis is useful because it can narrow the field of suspects. It can also determine whether or not the evidence is human or animal hair.*

STUDENT HANDOUT

Q: Can microscopic hair analysis provide positive identification of a suspect?

A: A positive ID based on hair morphology alone is rare. Hair samples are used, however, to obtain DNA samples from suspects and victims. DNA evidence is more conclusive in nature.

Q: What is the difference between a "suspect" and a "person of interest"?

A: Sometimes investigators designate people as "suspects," and sometimes they refer to them as "persons of interest"; however, no published definition distinguishes the difference between the two. Generally speaking, investigators consider someone a suspect once he/she becomes an official focus of an investigation as the result of initial evidence or circumstances making it likely that the person in question was a perpetrator of the crime under investigation. Once someone is deemed a suspect, police must follow certain rules for interrogation. For example, police must advise a suspect of his/her Miranda rights, and if a suspect requests a lawyer, the police must stop their questioning until a lawyer is present. If someone is simply a person of interest, however, police can do some initial probing for information without such restrictions in place. If the investigation is to probe more deeply into someone s background and possible connection to a crime, the judicial system then insists that the police treat that person as a suspect.

Q: What changes are occurring in the field of forensic science?

A: Experts believe forensic science will continue to evolve so as to provide faster and more accurate tests and techniques for the solution of crimes. One current focus of the field is to scrutinize closely its many analytic techniques in order to strengthen their use in investigations, mainly by eliminating as many potential errors as possible. For example, by comparing cases from all over the world that involve similar uses of handwriting analysis or ballistics tests, investigators can establish improved practices. Many in the community of forensic science hope to improve on the techniques already in place by establishing standards and using careful error analysis.

STUDENT HANDOUT

FORENSICS TERMS

The following are just a few of the many terms related to the field of forensic science.

Acid: *A corrosive substance that has a sour taste. (Students SHOULD NOT taste any acids in the lab!) When mixed with water, an acid yields H+ ions.*

Autopsy: *the internal and external examination of a body after death. An autopsy (the literal meaning can be expressed as "see for oneself") is performed to confirm or determine the cause of death and establish predeath conditions, such as the type of food last consumed and the time it was consumed.*

Ballistics: *the study of the motion of bullets and their examination for distinctive characteristics after being fired. Examiners can use this evidence to match bullets or bullet fragments to specific weapons.*

Base: *a slippery substance that has a bitter taste. When mixed with water, a base yields OH- ions.*

Bloodstain (or Blood Spatter) Interpretation: *the interpretation of size, shape, orientation, and distribution of blood pooled or spattered on various surfaces at a crime scene. Information about the event can be derived from proper interpretation of the stains.*

Bullet Track: *the path of a bullet as it passes through matter, such as a body or a wall.*

Catalyst: *a substance that accelerates a chemical reaction but is not itself permanently changed by the reaction.*

Chromatogram: *the recording of the results of a chromatography procedure.*

Chromatography: *the process of separating a chemical mixture such as ink into its individual substances.*

Composite Drawing: *a sketch of a suspect produced from descriptions by one or more eyewitnesses.*

Criminology: *the study of criminal activity and how it is dealt with by the law.*

Diacritics: *marks such as an accent or a tilde that indicate the correct pronunciation of a letter or combination of letters which, that without the mark, would be pronounced differently.*

DNA: *deoxyribonucleic acid. Occurring in the form of double-helix strands, DNA contains genetic code. In each individual, identical DNA occurs in the nucleus of every cell and serves to define that individual's characteristics. In addition to the portions of the DNA that encode the proteins making up all the individuals of a species, there are portions of "junk" DNA unique to each individual within the species. Often an individual's DNA appears in the blood and other bodily fluids. This provides a powerful technique for uniquely identifying the person or animal that left traces of such fluids at a crime scene. Indeed, this is the best method presently known for such identification.*

STUDENT HANDOUT

DNA Profiling: *the process of testing to identify DNA patterns or types. In forensic science this testing is used to indicate parentage or to exclude or include individuals as possible sources of bodily fluid stains (e.g., blood, saliva, or semen) and other biological evidence (e.g., bones, hair, or teeth.)*

Evidence: *anything that has been used, left, removed, altered, or contaminated during the commission of a crime or other event under investigation.*

Fingerprint: *the unique patterns created by skin ridges found on the palm sides of fingers and thumbs.*

Forensic Science: *the application of science to technical questions relating to events that may lead to civil litigation or criminal prosecution.*

Gene: *a unit of inheritance consisting of a sequence of DNA. Individually or collectively, genes determine a particular characteristic in an organism.*

Indicator: *a chemical used to determine the presence of an acid or a base.*

Latent Fingerprint: *a full or partial fingerprint made by deposits of oils and/or perspiration, not usually visible to the human eye. Various technologies, including lasers, can be used to visualize latent prints so that they can be recorded (usually by photography) for comparison with previously recorded fingerprints.*

Lie Detector: *also known as a "polygraph." A machine that charts how respiration and other bodily functions change as questions are asked of the person being tested. An attempt to knowingly provide false answers can cause changes in bodily functions. Lie detector tests are usually not admissible in court. Essentially all scientists not directly engaged in the use of polygraphs consider polygraph results to be unreliable.*

Luminol: *a chemical that is capable of presumptively detecting bloodstains diluted up to 10,000 times. Luminol is used to identify blood that has been removed from a given area. It is an invaluable tool for investigators at altered crime scenes. It is luminous (gives off light) and thus at a darkened scene highlights the distribution of what may be blood.*

Microscopic Hair Analysis: *An investigative procedure of examining hair shafts for identifiable characteristics.*

Morphology: *The branch of biology that deals with the form and structure of organisms without consideration of function.*

pH: *"power of hydrogen" — a measure of the concentration of H+ ions found in a solution; the lower the pH, the higher the concentration.*

STUDENT HANDOUT

Physical Evidence: *any object, as distinguished from witness statements, that can help explain an event under investigation. For example, physical evidence can establish that a crime has been committed, and sometimes it can provide a link between a crime and its victim or between a crime and its perpetrator.*

Point-by-Point Analysis: *when comparing a known object to one that needs to be identified, analysts will break down photos of each into small portions and compare the respective similarities within those portions.*

Questioned Document (QD) Analysis: *the procedure of examining handwriting, watermarks, and other qualities of a document that may indicate it s origin and author.*

QD Examiner: *one who analyzes documents professionally, often for investigative purposes.*

Ridge Characteristics: *ridge endings, bifurcations, enclosures, and other details, which, if present in both of two fingerprints being compared, must match for their common origin to be established.*

Serology: *the science dealing with the properties and actions of serums in blood; also known as "blood analysis."*

Solute: *a dissolved substance.*

Solvent: *a liquid substance capable of dissolving or dispersing one or more substances.*

Standard: *in handwriting analysis, material whose source and origin are known and used for comparative purposes.*

Superglue Fuming: *a technique used to visualize latent fingerprints on nonporous surfaces. A chemical in the glue reacts with and adheres to the finger oils, producing a visible substitute for the underlying prints.*

Toxicology: *the study of poisons and drugs and their effects on humans and animals.*

Trace Evidence: *material deposited at a crime or accident scene that can be detected only through a deliberate-processing procedure. An individual entering any environment will deposit traces of his or her presence, and this material can be used as evidence. Common types of trace evidence are hairs and clothing fibers.*

Trajectory: *the path of an object as it moves through space, usually referring in the forensic context to a bullet or other projectile.*

STUDENT HANDOUT

Unit 4 Mystery Synopsis: The Cafeteria Caper

THE SETUP:

"Attention students. May I have your attention, please? First, let me state that no one has been hurt." The booming voice of Mr. Holden, the principal of Park Haven High School, echoed throughout the campus. He usually commanded attention, but on that particular Monday morning any voice on the PA system would have worked.

Those who arrived early could not enter the school, and at least four police cars were parked outside. Although no one knew yet what had happened, rumors of serious foul play were jumping from one student to the next like honeybees in a patch of wildflowers. Everyone waited for Mr. Holden to clear things up.

"Students," the principal continued, "I know this will come as a shock, but we have to send all of you home today." Now he definitely had everyone s attention! All over campus, people whistled and whooped in approval of the news. Some students groaned. But everyone wondered if any of the rumors were actually true. "This weekend, someone broke into the school and caused some serious damage in the cafeteria. Because this threatens the safety of everyone in our school, we ve been forced to cancel school today."

"Hey, Mr. Holden," yelled a voice from the crowd. "What happened? Did somebody run off with this week s mystery meat lunch special?" The students broke into laughter and everyone turned to look at the wise guy. Of course it was Jesse Martin, the school s funny man. Mr. Holden was not pleased and told Jesse to report to his office tomorrow at 8:00 am sharp instead of going to homeroom.

The laughter subsided. Everyone waiting outside Park Haven High School turned their attention back to Mr. Holden and the serious matter at the heart of the rumors. "Now," continued Mr. Holden, "I know that some of you have heard that this situation involves a corpse. That is absolutely not true. In fact, as I stated earlier, no one s been hurt at all."

(If one were to survey the students at that exact moment, half would have expressed relief, while the other half would have been a little disappointed that the situation was turning out to be not quite as interesting as the rumors had indicated.)

The principal went on. "I m sure you won t be surprised to hear that my father used to have something to say about rumors and gossip." A collective sigh rose up from the crowd. Mr. Holden always found a way to work in one of his father s sayings. "My father said that rumors and gossip are like weeds. Sometimes weeds grow pretty flowers, but they re still just weeds trying to choke the life out of other plants." As always, he paused for a moment to give the students time to absorb the meaning of his father s sage advice. By the blank looks on their faces, Mr. Holden knew that he would have to wait a very long time for them to grasp the implication of that one.

STUDENT HANDOUT

Mr. Holden cleared his throat and pushed on. "Our assistant principals and I have already begun to hear some rumors about what went on this past weekend. We can t tell yet if what we ve heard is good information or bad. But if anybody has any information about what happened, come and tell me about it in the office. We ll decide if it s a plant or a weed. Get it?"

Spencer Rhodes thought about that for a moment. A plant or a weed? He was certain that the information he had was definitely a plant.

Spencer was a student who normally kept to himself. He doubted that Mr. Holden even knew who he was. He was a very good student, but he wasn t an "active" student. Good grades alone didn t get you noticed by the big shots at Park Haven High. For that matter, Spencer had a hard time getting noticed by anyone, even other students. Maybe the information he had would change all of that.

After waiting until most of the students had cleared out, Spencer convinced one of the assistant principals, Mr. Warden, that he had some helpful information and he was escorted to the main office. As they walked to the office, Mr. Warden kept shaking his head, saying "Senseless vandalism senseless." And as they walked by the trophy case, Spencer saw some of the evidence of it. The case had been smashed open, and many of the trophies were broken into pieces on the floor. Seeing the demolished trophies, Spencer felt even more confident about the information he had for Mr. Holden. It also confirmed his earlier decision to stop associating with the guys most likely responsible for trashing the trophy case. If he hadn t followed his instincts, there was a good chance that he, too, might have been caught up in the past weekend s activities. Whew, thought Spencer, as he made a mental note to keep that little piece of information to himself for now. He couldn t help wondering, though, why a vandalized trophy case would create an evacuation situation. He quickly concluded that there had to be more pieces to this puzzle.

"Mr. Holden, this is, uh What did you say your name was again?" Mr. Warden looked to Spencer for a little help.

"Spen—" Spencer cleared his throat and looked around. There were also a couple of other people in the room. Spencer thought they might be detectives. "Spencer Rhodes, sir."

"Rhodes, that s right." Spencer doubted that Mr. Warden really remembered. "Anyway, he says he has an idea about who committed these senseless acts of vandalism."

"Is that true, son?" Mr. Holden asked.

Spencer looked at Mr. Holden and at the people he assumed were detectives. They all seemed to be patiently waiting to hear what he had to say. "Well, sir, I don t have any direct proof, but I think some kids did this to the school to get initiated into an underground club."

"Underground club?" For the first time in Spencer s presence, one of the other people in the room a woman spoke up. "Spencer, I m, uh, Detective Wulff. Bingo, thought Spencer. I should have introduced myself right away." She offered her hand for him to shake. "Principal Holden called us in early this morning to let us know that someone had broken into the building over the weekend. Please go on."

STUDENT HANDOUT

"Well, the club has a web site. I, um know about the club and have seen their site. I thought that something was going on this weekend, so I sort of hack ah, found a way to get on to their site." Spencer nervously glanced around the room to some very bewildered looks. "The people in the club call themselves Philophiles."

As if they had planned it, Detective Wulff, Mr. Holden, and Mr. Warden all said "Philophiles?" in unison.

"Yes, Philophiles. Their name was inspired by Philo Farnsworth.

Mr. Warden jumped into the conversation, asking, "We don t have a student by that name, do we?"

"Uh, no, sir. Well, I don t think so, but if there is, the club is not related to him," Spencer answered. "Philo Farnsworth invented television." Spencer got some polite nods, but no one seemed familiar with the name. He could tell that everyone wondered why a club would name itself after Philo Farnsworth. "Only recently did people begin to give him full credit for the invention. He was 14 years old when he first conceived the TV. It s all pretty amazing."

"Yeah, yeah, yeah. Whatever. Tell us about "

"Spencer," Said detective Wulff cutting off Mr. Warden, shooting the assistant principal a look as if to say, "I m lead detective. I ll handle it." She returned her gaze to Spencer. "What makes you think that the club members are responsible for what went on here?"

"It s the web site. It talks about the initiation ceremony from this past weekend." Spencer paused, trying to figure out the best way to say what he wanted to say. "It doesn t say anything specifically about breaking into the school. But it lists the five inductees. I know them. I m familiar with their club motto, and I know how they feel about this school. They all feel like their genius isn t recognized. That our school places too much importance on popularity and sports."

"And Philo Farnsworth was also one whose genius was underappreciated?" added Detective Wulff, seeing the connection.

Spencer smiled and nodded. "As soon as I saw the trophy case, I realized that that was something they might do as part of their initiation. You know, destroy one of the symbols of what bums them out the most the way the school tends to worship jocks, not brains." He could tell that everyone was trying to understand what he was telling them. The silence seemed uncomfortably long. Spencer knew he was doing the right thing by sharing the information. Still, he couldn t help feeling like he was betraying everyone in the club. After all, they had once asked him to consider becoming a member. But he hadn t joined because he felt like the members were moving in a direction that betrayed their original mission. And that was even before this latest incident.

Detective Wulff finally broke the silence. "Is there a way we can see that web site? Because what you re telling me still doesn t seem like enough to bring those students in for questioning."

"Not a problem at all. We can go into my office and take a look," Mr. Holden offered.

STUDENT HANDOUT

Spencer felt a little funny sitting down in Mr. Holden s big leather chair. As he was typing in the URL, he nervously bragged a little. "It requires a password to get to the site, but that was easy enough to figure out. Televisionman and then the date of Philo Farnsworth s birth, 08 19 06." Spencer quickly typed it in as he recited the number, and up popped the Philophiles page.

The room was quiet for a moment as everyone looked at the site. When they got to the part about what the inductees had to eat during the initiation process, they all had the same reaction: "Yuck!" Raw liver and soda seemed like an awful combination to everyone.

After viewing the web site, Detective Wulff thanked Spencer for his help and left to go to the cafeteria the source of danger and the cause of the evacuation. Detective Wulff also hoped that it would give her the evidence she needed to bring in the five inductees for questioning.

THE CAFETERIA

Very early that Monday morning, Ms. Wright, one of the cafeteria workers, arrived to find a strong stench in the kitchen, quickly realizing the stench s source. The smell came from what looked like someone s stomach contents on the floor. She had no idea how long it had been there. All she knew was that it wasn t there on Friday afternoon and that it reeked!

Curiously, right next to the smelly mess was what looked like a woman s knee-high stocking with hairs in it. That puzzled Ms. Wright. She debated whether to call someone about it or simply clean it up, but then she noticed another smell. At first the stench of the stomach contents was so strong that she hadn t noticed the other smell. But there was no doubt about it. Gas was escaping from somewhere.

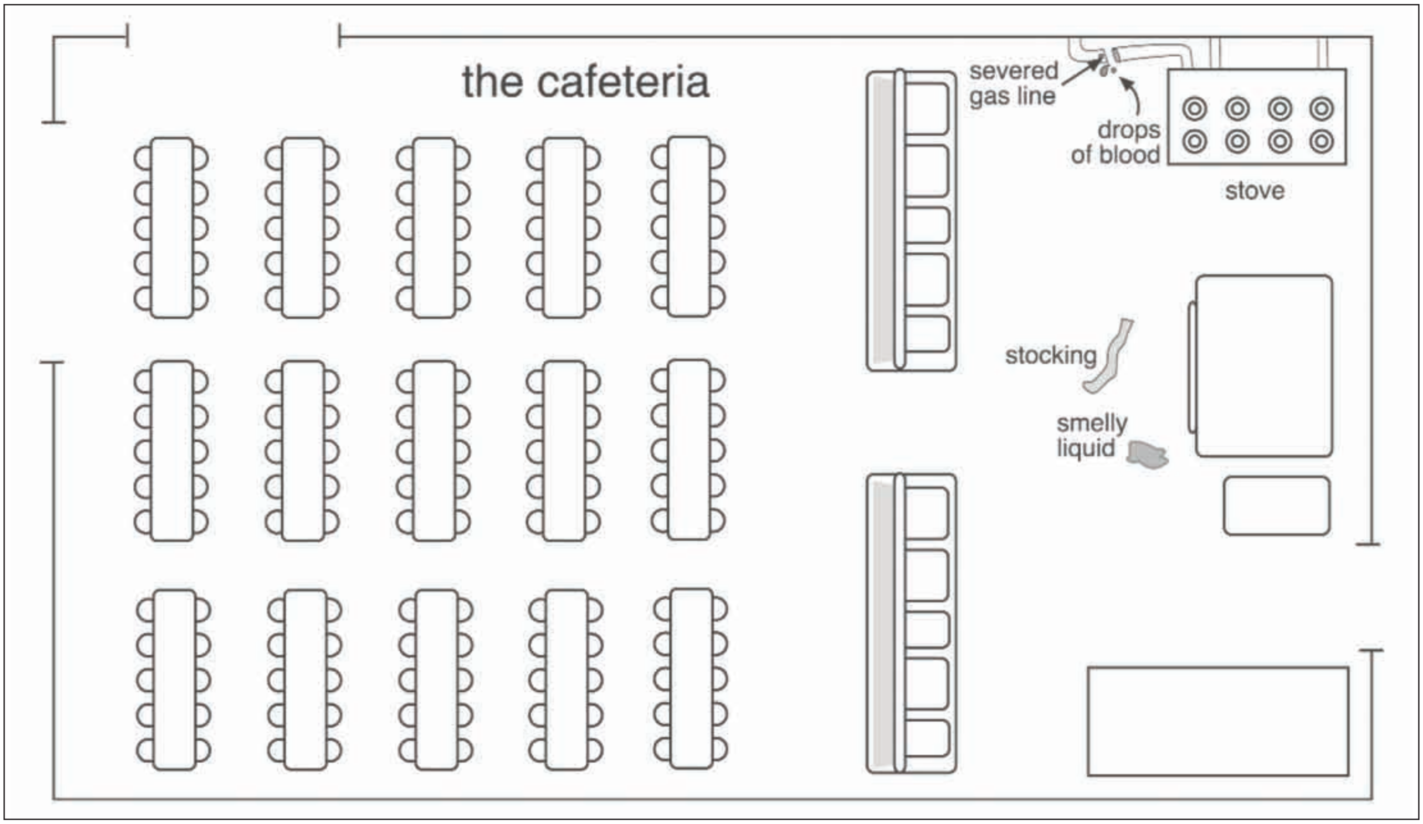
That made Ms. Wright s decision easy. She immediately called the main office. Then she was told that other parts of the school had been damaged, too.

One of the custodians quickly isolated the source a gas line had been cut behind one of the big ovens. While they waited for the gas company and the police to arrive, Principal Holden and his assistants debated whether they should cancel school that day.

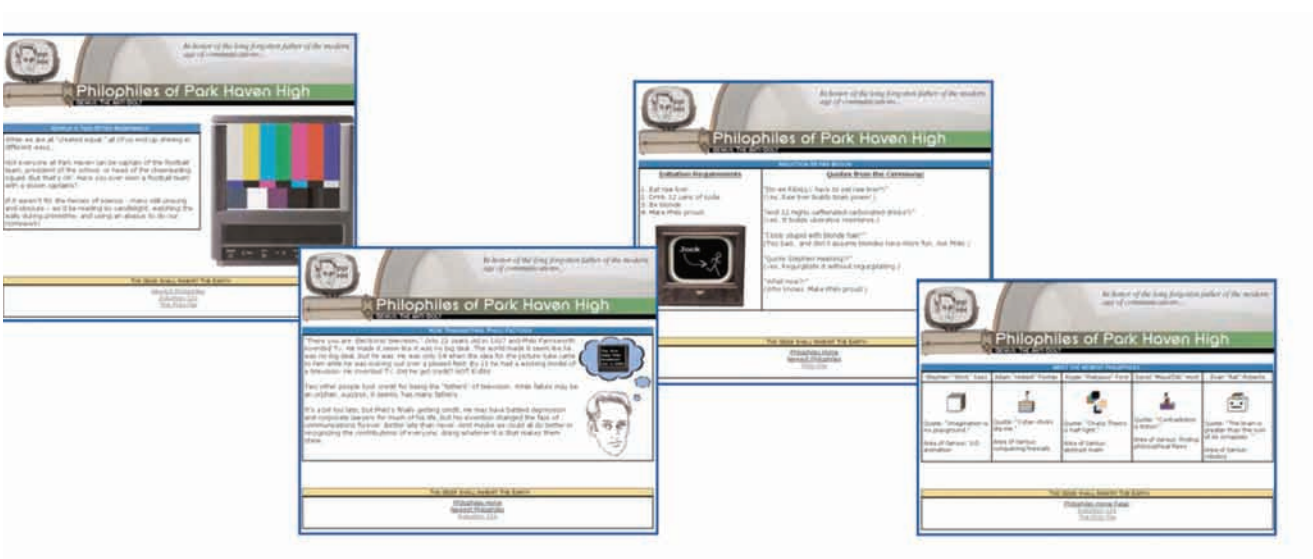
By the time Detective Wulff entered the kitchen, the scene was no different from the one Ms. Wright had seen (and smelled!) earlier. The smells weren t quite as strong because the gas had been turned off. Detective Wulff felt confident that there was some possible evidence that they could test, so she asked that nothing be touched or cleaned up. In addition to the stomach contents, the knee-high stocking, and the cut gas line, she saw what looked like a few drops of blood on the floor near the gas line. She figured that whoever cut the line must have also cut himself or herself. The detective had to remind herself that she should not presume that the boys from the Philophile club were involved. Maybe the evidence, though, would point in that direction.

STUDENT HANDOUT

CRIME SCENE DIAGRAM



PHILOPHILES WEB SITE



STUDENT HANDOUT

LESSON 2: INDICATORS AND ENZYMES TEST

OBJECTIVE:

After examining the crime scene and evidence at hand, students will begin their first activity by examining the contents of the liquid found at the crime scene. They will learn to use various indicators to test for the presence of organic molecules such as proteins and sugars. They will also use deductive reasoning to determine the contents of the stomach fluid found at the crime scene, and then establish a link between the perpetrator(s) and the underground club.

MATERIALS NEEDED:

Reproducibles

- *Indicators and Enzymes activity sheet*
- *Persons of Interest information sheet (This handout should be presented only after students confirm that liver was present in the stomach contents.)*

Note: *Any time you use chemicals in your classroom or lab, be sure to read and follow the safety instructions for use and disposal that are provided in the MSDS information that came with your lab materials.*

Equipment and Chemicals

- *Safety goggles*
- *Lab aprons*
- *Heat-protective gloves or a hot mitt (**Note:** You might not need this if you are using tongs, but it is smart to have some protective gloves around just in case.)*
- *Raw calf liver (whole and homogenate)*
- *High-sugar content clear soda (color change of the indicators may be masked by darker sodas)*
- *Hydrogen peroxide*
- *Biuret reagent*
- *Iodine*
- *Benedict s indicator*
- *Blender*
- *Droppers or pipettes*
- *4 test tubes and test tube rack (or spot plates)*
- *Hot plate and beaker (for hot water bath)*
- *Tongs*
- *Wax pencil for marking test tubes*
- *Distilled water*
- *Monosaccharide solution (glucose)*
- *Disaccharide solution (sucrose)*
- *Polysaccharide solution (starch)*
- *Protein solution (gelatin)*

STUDENT HANDOUT

Indicators and carbohydrates can be purchased online from the following vendors:
The Lab Depot: http://www.labdepotinc.com
Carolina Biological Supply: http://www.carolina.com

TIME REQUIRED:

- *Teacher Prep Time: 60—90 minutes*
- *Class Time: 90 minutes (two classes)*

LESSON DESCRIPTION:

Forensic investigators often analyze stomach contents during an autopsy. During a forensics investigation, stomach contents are usually weighed and tested for the presence of toxins (e.g., alcohol or poison). In this mystery, students find a thick, smelly fluid at the scene of the crime that appears to be stomach contents. An underground club s web site mentions that inductees have to eat raw liver and drink large quantities of soda as part of their initiation. Students will test the stomach contents for the presence of peroxidase, which is plentiful in liver, by exposing the contents to hydrogen peroxide. They will observe the production of oxygen bubbles and will conclude that peroxidase is present.

Because peroxidase is found in large amounts in liver, students may deduce that members of the underground club MAY have been involved. At the least, given Spencer s report and the information from the web site, they will understand that these club members will be of interest to the police.

This lesson also gives students the opportunity to test the stomach contents for the presence of protein (also in liver), monosaccharides (in soda), and polysaccharides (not present). They will use these tests to establish a link between the underground club and the perpetrators. When they have concluded their analysis of the results, present them with the Persons of Interest list.

BACKGROUND INFORMATION:

Organic chemistry is an integral part of the biology curriculum. This lab gives you the chance to teach organic chemistry in an interesting and purposeful format. It is important to teach the basic properties of carbohydrates and proteins, including enzymes, before completing this lab. Students should be familiar with the differences between monosaccharides, disaccharides, and polysaccharides as well as foods in which each of these are found. They should also be familiar with the role of enzymes in digestion and the lock-and-key theory of enzyme function. The following web sites review basic properties of enzymes.

- *http://www.tvdsb.on.ca/westmin/science/sbi3a1/digest/enzymes.htm*

STUDENT HANDOUT

It is also important to emphasize lab safety skills before students begin the lab. These safety reminders are found in the student version of the lab. Finally, peroxidase (also known as catalase) is one of the most common enzymes in organisms. While it is found in many plants and animals, it is found in very high concentrations in the liver. This is why liver was chosen to demonstrate the properties of this enzyme.

LESSON STEPS:

Teacher Preparation

Each student or lab station will need the following:

1. *To create the "stomach contents," blend calf liver and soda in a blender until it is liquefied. Calf liver can be found in the freezer section at your local grocery store.*
2. *Set up the lab equipment with the materials list needed for groups of two.*
3. *The control tests will allow students to view the color change of the indicators so they can recognize these changes during the actual test. Create your glucose solution by dissolving 10 grams of glucose or dextrose into 700 ml of distilled water. Create a sucrose solution by dissolving 10 grams of table sugar into 700 ml of distilled water. Create the starch solution by slowly dissolving 10 grams of starch into 700 ml of distilled water while heating the solution. Glucose (or any monosaccharide) and starch can be purchased at any biological or chemical supply company.*
4. *To create the protein solution, dissolve 1 envelope (about 7 grams) of clear gelatin into 700 ml of distilled water by heating the solution.*
5. *Place the beaker solutions in a central location accessible to students.*

Lab Execution

1. *To begin the first lab activity, thoroughly review the background information on the student activity sheet by emphasizing the differences between the three types of carbohydrates and explaining which indicators are used for each type. You may wish to supplement the lab with illustrations of monosaccharides, disaccharides, polysaccharides, enzymes, and substrates.*
2. *Discuss the function of peroxidase, where it is found, and how enzymes are substrate-specific. The introduction on the activity sheet gives a detailed review of the reaction you will observe between the liver and hydrogen peroxide.*
3. *Place a piece of raw liver in a test tube to demonstrate how hydrogen peroxide is broken down when it comes into contact with peroxidase. Point out the oxygen bubbles in the test tube and ask students to identify the source. Refer to the equation for assistance.*

STUDENT HANDOUT

4. Remind students to use safety precautions when completing the tests. Reiterate rules for using chemicals in the lab (e.g., those involving inhaling, skin contact). Material Safety Data Sheets are provided with all ordered chemicals. Refer to the MSDS sheets for proper handling and disposal of any and all chemicals. Biuret reagent is caustic and should be thoroughly rinsed
off if it comes into contact with skin or clothing. Because the Benedict s test requires a hot water bath, make sure students wear goggles during this test.

5. During the control tests, make sure students do not cross-contaminate the solutions by using the same dropper for two different solutions.

6. Emphasize why students are performing the control tests. Because there is no test for disaccharides, they should realize that if the carbohydrate doesn t test positive during the Benedict s test or the iodine test, it must be a disaccharide.

7. At the end of the lesson, review or check the answers to the analysis questions and discuss the results with the class. Guide the class to the conclusion that the group of club inductees should be further investigated due to the lab results. Once that has been established, distribute the Persons of Interest handout for discussion and review.

ACADEMIC EXTENSIONS/MODIFICATIONS:

¥ Lipid testing can also be done using the "stomach contents." Simply use a brown paper bag and place a few drops of the sample on the bag. Allow the bag to dry. Hold the bag up to the light; if the bag is transparent, the sample contains lipids.

¥ Students can research the methods and procedures for forensic toxicology tests.

¥ If lack of time is an issue, students can work cooperatively to complete the tests. One group can test for monosaccharides, one for disaccharides, and one for polysaccharides. Each group should complete the enzyme test.

STUDENT HANDOUT

INDICATORS AND ENZYMES ACTIVITY SHEET

INTRODUCTION:

Forensic scientists analyze stomach contents during an autopsy by weighing the contents of the stomach and testing for poisons. During many investigations, toxicology tests will also include drug and alcohol testing. The type and amount of a drug present can be critical information for a forensic investigator. The focus of this lab is to analyze the components of the "stomach contents" found at the scene of the crime to determine if raw liver and sugar are present. A positive test for these foods would provide enough supporting evidence to investigate the club inductees, because the police have learned that eating raw liver and drinking large quantities of soda are initiation requirements for the club.

Once the stomach contents have been tested for sugars and proteins, the next step is to test specifically for the presence of liver. This is possibe because of the presence of an enzyme, peroxidase, that is found in the liver. Peroxidase speeds up the breakdown of hydrogen peroxide, a waste product of cell metabolism that can be toxic in high concentrations. The breakdown that occurs is as follows:

$$\underset{\text{Hydrogen Peroxide}}{2H_2O_2} \longrightarrow \underset{\text{Water}}{2H_2O} + \underset{\text{Oxygen}}{O_2}$$

Peroxidase, or catalase, is an enzyme found in animals and plants and in high concentration in the cells of the liver. Peroxidase has an active site in which it fits perfectly, like a key in a lock, with hydrogen peroxide. Other examples of enzymes are lipase, which breaks down only lipids; amylase, which only breaks down amylose; and lactase, which only breaks down lactose. When peroxidase, found in the liver, is exposed to hydrogen peroxide, large numbers of oxygen bubbles and water are produced, as well as water; therefore, a positive test for liver peroxidase will produce many oxygen bubbles when the liver is exposed to hydrogen peroxide.

PROCEDURE:

Part I: Control Test for Monosaccharides

Control tests allow scientists to have a basis for comparison during an experiment. These control tests will allow you to view the color change of indicators used to test for the presence of organic molecules. The following is a list of indicators and their expected results: positive for GSR.

- ¥ *Biuret reagent is a blue liquid that turns lavender to violet in the presence of a protein.*
- ¥ *Benedict s solution is a blue liquid that turns green in a small amount of monosaccharide, yellow in a larger amount of monosaccharide, and orange or red in a great amount of monosaccharide.*
- ¥ *Iodine is an orange liquid that turns blue or black in the presence of a polysaccharide.*

OBJECTIVE:

Your teacher will give you directions for setting up your hot water bath.

1. *Put on your goggles.*
2. *Using a wax pencil, label four clean test tubes 1, 2, 3, and 4.*
3. *Add 20 drops of distilled water to test tube 1.*
4. *Add 20 drops of monosaccharide (glucose) solution to test tube 2.*
5. *Add 20 drops of disaccharide (sucrose) solution to test tube 3.*
6. *Add 20 drops of polysaccharide (starch) solution to test tube 4.*
7. *Now add 10 drops of Benedict s solution to each test tube.*
8. *Place the four test tubes into the hot water bath (it does not have to be boiling) for 2—3 minutes.*
9. *Carefully remove the test tubes from the water bath and observe any color changes.*
10. *Complete the data table and analysis questions for the Monosaccharide Control Test.*
11. *Follow instructions from your teacher for correct waste disposal, then wash your test tubes and proceed to Part II.*

DATA TABLE I: MONOSACCHARIDE CONTROL TEST

Test Tube	Beginning Color	Ending Color	Positive or Negative for Monosaccharides
Distilled Water			
Monosaccharide Solution			
Disaccharide Solution			
Polysaccharide Solution			

ANALYSIS

1. *Identify the control group in this experiment.*

2. *What color does Benedict s solution turn in the presence of a monosaccharide?*

3. *Give three examples of foods that contain monosaccharides and give the specific monosaccharide found in those foods.*

Part II: Control Test for Polysaccharides

1. *Label four clean test tubes 1, 2, 3, and 4.*
2. *Repeat steps 2—5 in Part I.*
3. *Now add 10 drops of iodine solution to each test tube and observe any color changes.*
4. *Complete the data table and analysis questions for the Polysaccharide Control Test.*
5. *Follow instructions from your teacher for correct waste disposal, then wash your test tubes and proceed to Part III.*

DATA TABLE II: POLYSACCHARIDE CONTROL TEST

Test Tube	Beginning Color	Ending Color	Positive or Negative for Monosaccharides
Distilled Water			
Monosaccharide Solution			
Disaccharide Solution			
Polysaccharide Solution			

ANALYSIS:

1. *Identify the control group in this test.*

2. *What color did iodine turn in the presence of a polysaccharide?*

3. *Name five foods that contain polysaccharides.*

4. *If iodine is dropped on your paper, it turns black. How can you explain this color change?*

Part III: Control Test for Proteins

Note: *Biuret reagent is caustic to skin and clothing. Rinse immediately upon contact and notify your instructor about any spills.*

1. *Put on your goggles. Label two clean test tubes 1 and 2.*
2. *Add 20 drops of distilled water to test tube 1.*
3. *Add 20 drops of protein (gelatin) solution to test tube 2.*
4. *Now add 10 drops of Biuret reagent to each test tube and observe any color changes.*
5. *Complete the data table and analysis questions for the Protein Control Test.*
6. *Follow instructions from your teacher for correct waste disposal, then wash your test tubes and proceed to Part IV.*

DATA TABLE III: PROTEIN CONTROL TEST

Test Tube	Beginning Color	Ending Color	Positive or Negative for Monosaccharides
Distilled Water			
Gelatin			

ANALYSIS:

1. *Identify the control group for this test.*
2. *What color did Biuret turn in the presence of a protein?*
3. *Name five foods that contain protein.*
4. *Biuret reagent turns the skin brownish-purple. Why does this happen?*

STUDENT HANDOUT

Part IV: Stomach Content Analysis

Note: *When the indicators are added to the stomach contents, DO NOT MIX! Simply view the color of the indicator, not the stomach contents. During the protein test, you may get a very dark blue, lavender, or almost black color. This is a positive result.*

Remember to use goggles for these tests!

1. *Put on your goggles.*
2. *Label four clean test tubes 1, 2, 3, and 4.*
3. *Add 20 drops of the stomach contents to each test tube.*
4. *Now use the procedures for monosaccharide, polysaccharide, and protein testing to test the stomach contents for these organic molecules.*
5. *Complete the data table and analysis questions for Stomach Content Analysis.*
6. *Follow instructions from your teacher for correct waste disposal, then wash your test tubes and proceed to Part V.*

DATA TABLE IV: STOMACH CONTENT ANALYSIS

For each organic molecule, place a check mark if the stomach contents tested positive and a minus sign if the stomach contents tested negative.

Organic Molecule	Positive (+) or Negative (-)
Monosaccharides	
Polysaccharides	
Proteins	

ANALYSIS:

1. *Which organic molecules were present in the stomach contents?*

2. *If the stomach contents belonged to a club inductee, could you identify the food source for these organic molecules? Check the nutritional labels on these foods to verify your answer.*

STUDENT HANDOUT

Part IV: Stomach Content Analysis

Part V: Peroxidase Activity Test

1. *Put on your goggles.*
2. *Label four clean test tubes 1, 2, 3, and 4.*
3. *Add 20 drops of distilled water to test tube 1.*
4. *Add 20 drops of raw liver homogenate to test tube 2.*
5. *Add 20 drops of the soft drink to test tube 3. Allow the fizz to settle.*
6. *Add 20 drops of the stomach contents to test tube 4.*
7. *Now add 20 drops of hydrogen peroxide to each test tube and record the results in the data table for the Peroxidase Activity Test.*
8. *Follow instructions from your teacher for correct waste disposal, and then wash your test tubes.*

DATA TABLE V: PEROXIDASE ACTIVITY TEST

For each organic molecule, place a check mark if the stomach contents tested positive and a minus sign if the stomach contents tested negative.

Test Tube	Positive (+) or Negative (-)
Distilled Water	
Raw Liver	
Soft Drink	
Stomach Contents	

ANALYSIS:

1. *Identify the control group(s) for this experiment.*

2. *What evidence do you have that the raw liver contains peroxidase?*

3. *Did the soda test positive for peroxidase? Why or why not?*

STUDENT HANDOUT

4. *Did the stomach contents test positive for peroxidase?*

5. *Is there enough evidence to support further questioning of the club inductees?*

CONCLUSION:

Write two to three sentences summarizing the results of this activity as they relate to your investigation. You will refer to this summary later in the investigation.

STUDENT HANDOUT

PERSONS OF INTEREST INFORMATION SHEET

THE PHILOPHILE INDUCTEES

Based on the information provided on the club s web site and the results of the liquid analysis, Detective Wulff felt that there was enough suspicion to bring the five inductees and their parents in for questioning. A search warrant was issued, allowing the detective to collect hair and DNA samples from each of them. What follows are partial excerpts from police notes on the questioning of five boys, all of whom are suspected of associating with the underground club known as the Philophiles. All five are considered persons of interest.

Roger Ford (Person of Interest 1) — *Roger, 16, was small with black curly hair. Although he seemed nervous, he denied any knowledge of the events that occurred at Park Haven High School (PHHS) or any knowledge of the Philophiles. When presented with the fact that his name was listed on a Philophiles web site, he said that it wasn t him and that he had a very common name.*

Stephen Sasz (Person of Interest 2) — *Stephen, 15, was tall and skinny and had bleached blonde hair. He said he recently bleached it as a fashion statement. He denied any knowledge of the Philophiles or the events that occurred at PHHS. When presented with the fact that his name was listed on a Philophiles web site, he said anyone can put anything on the Web and that he didn t know why someone would associate him with a group he s never heard of.*

Evan Roberts (Person of Interest 3) — *Evan, 15, was short with bleached blonde hair. He seemed extremely nervous. He said that the first he knew about the events at PHHS was Monday morning. When questioned, he also said that he had heard of the Philophiles club, but didn t know anything about it. When presented with the fact that his name was listed on a Philophiles web site, he shrugged his shoulders and said someone was playing a prank on him.*

David Host (Person of Interest 4) — *David, 16, was of average build and had bleached blonde hair. He also claimed to have recently bleached it. He denied any knowledge of the events that occurred at PHHS or the Philophiles. When presented with the fact that his name was listed on a Philophiles web site, he said, "I don t know what to say. I have no knowledge of that organization."*

Adam Fischer (Person of Interest 5) — *Adam, 16, was rotund with red hair and freckles. He acted as though everything was a big joke. He denied any knowledge of the events that occurred at PHHS or the Philophiles. He laughed at the name of the club. When presented with the fact that his name was listed on a Philophiles web site, he stated that he was a popular guy and that his name was probably listed on numerous web sites.*

STUDENT HANDOUT

LESSON 3: HAIR ANALYSIS

OBJECTIVE:

Students will use various microscopy techniques for observation and data collection to analyze hair samples found at the crime scene.

MATERIALS NEEDED:

Reproducibles

- *Hair Identification Activity Sheet, including a hair follicle and shaft diagram and a data collection table*
- *Persons of Interest information sheet (if not already covered at the end of Lesson 2)*

Lab Supplies (For groups of 2)

- *Microscope*
- *10 slides*
- *10 coverslips*
- *Dropper bottles (water)*
- *Forceps*
- *Hair samples from 3 different individuals (black hair, red hair, bleached blond hair)*
- *Clear fingernail polish (Sally Hansen® HARD AS NAILS® works well)*
- *Permanent marker*

Note: *Any time you use chemicals in your classroom or lab, be sure to read and follow the safety instructions for use and disposal that are provided in the MSDS information that came with your lab materials.*

TIME REQUIRED:

- *Teacher Prep Time: 90 minutes*
- *Class Time: 90 minutes*

LESSON DESCRIPTION:

The discovery of the club s web site (which describes the initiation rites involving eating liver and consuming soft drinks) led students in Lesson 2 to analyze the stomach contents. Because the stomach contents appear to match the requirements of the initiation rites, the five club inductees mentioned on the web site were brought in for questioning. Based on the evidence, the five students are considered Persons of Interest. (A sheet that includes information about the Persons of Interest is provided at the end of Lesson 2.) With a search warrant, investigators were able to obtain hair samples from each of them. Students will now create a wet-mount slides and a scale-cast slides of the five suspects hair samples. They will sketch and collect data on the suspect hair and then repeat the procedure for the hair found on the stocking at the crime scene. They will then compare and contrast the suspect hair and the evidence hair and determine which suspect(s) could have been at the scene of the crime.

STUDENT HANDOUT

BACKGROUND INFORMATION:

Before beginning this activity, it is important to adequately prepare for questions regarding hair anatomy and physiology. To research additional information on this topic, refer to the following web sites:

- ¥ http://www.trutv.com/forensics/lab
- ¥ http://www.thegentletouch.com/hairbiol/h-morph1.htm
- ¥ http://www.natural-hair.com/structure.html

You may choose any three types of hair to use for this investigation, making sure that hair from three of the suspects is the same as the evidence hair. The differences between black hair, red hair, and bleached blonde hair are striking and work well for this lab. If you choose to teach your students how to do permanent mounts, you will need to order mounting media from a biological supply company. Most mounting media contains xylene. You will also want to review basic microscope techniques with your students, and be sure to follow all instructions for safety and disposal provided on the MSDS sheet. If you use xylene, be especially aware of any chemical sensitivities your students may have and provide protective face gear.

LESSON STEPS:

Teacher Preparation

1. *Obtain hair samples from individuals with natural black hair, natural red hair, and bleached blonde hair.*
2. *Cut the hair into 1-inch sections and separate the hair samples into three jars or envelopes.*
3. *Create 5 envelopes or zippered plastic bags for each group. Using forceps, place 15 to 20 1-inch pieces of black hair in the first envelope and label it "Suspect 1." Now place the same amount of bleached blonde hair into the second, third, and fourth envelopes and label them "Suspect 2," "Suspect 3," and "Suspect 4." Now using the red hair, create an envelope labeled "Suspect 5." Create one last envelope for yourself. It should contain the same bleached blonde hair used to create envelopes for Suspects 2, 3, and 4. Label this envelope "Evidence."*

(**Note:** *You may want to enlist the help of a hair stylist to get your hair samples. The lab requires that much hair because you need 4—5 strands for each slide to get good results, and that only leaves only 5—10 strands more. Sometimes the hair falls out of the envelope or gets lost.)*

4. *Gather necessary materials for each group according to the materials list.*

5. Using the hair from your Evidence envelope, create one scale cast for each group. (Be sure to label these slides as such.) You will distribute these slides to the groups for their examination only AFTER they have completed examining all the suspects hair. Then, as the groups near completion of their wet-mount slide examinations, create a wet-mount slide of the evidence hair for them to examine. Try to prepare the wet-mount slides in a timely manner so that they do not dry out before students can examine them.

6. Practice making the scale-cast slides before the lab so you can help students who may be having trouble. Also practice identifying the differences between the scales on damaged hair and on healthy hair.

Lab Execution

1. It is best to begin the lesson by reviewing what students have learned about the case so far. Then, introduce the lab by asking them to read the Introduction on the activity sheet aloud. Discuss the anatomy and physiology of hair while labeling the parts of the hair follicle and shaft on the handout. Students will label their diagrams and fill in the functions for the structures as you review. Emphasize the differences between chemically treated hair and natural hair. Point out the differences in the scale patterns of the two hair types.

2. Before beginning the hair analysis activity, ask students why it is important to analyze the hair samples from the suspects and the hair found at the crime scene.

3. Divide your students into groups of two. You want the groups to be small so that each group member can analyze each hair sample.

4. Emphasize the importance of keeping each sample separate from the other samples so that the evidence is not contaminated.

5. Remind students to label each slide as it is created so that data collection is accurate. It is easy to mix up hair samples, and this could affect whether a person is considered innocent or guilty.

6. Reiterate rules for using the chemicals in the lab (e.g., those involving inhaling, skin contact). Material Safety Data Sheets are provided with all ordered chemicals. Refer to the MSDS sheets for proper handling and disposal of any and all chemicals.

7. While students are waiting for the scale casts to harden, encourage them to create their wet mount slides and begin their data collection. As they begin to make and examine their wet-mount slides, this is a good time for you to begin preparation of a wet-mount slide of the evidence hair for each group. DO NOT distribute the evidence slides yet.

8. As the groups completes their suspect slide examinations and analyses, distribute the evidence hair slides. You should provide the evidence slides only after the suspects hair has been analyzed. This will ensure objectivity.

9. As a class, summarize the data collected by reviewing the drawings, and ask students to conclude which suspects could have been at the scene of the crime. Be sure to emphasize that the findings indicate that three of the suspects could be the source of the evidence hair but do not necessarily prove a positive identification.

ACADEMIC EXTENSIONS/MODIFICATIONS:

¥ If you do not have 90 minutes for this lesson, you may want to complete it in cooperative groups. Have one group examine Suspect 1 and the evidence hair, one group examine Suspect 2 and the evidence hair, and so forth. Then have groups present their findings to the rest of the class.

¥ Ask students to research the differences between animal hair and human hair and the uses for hair analysis in forensics.

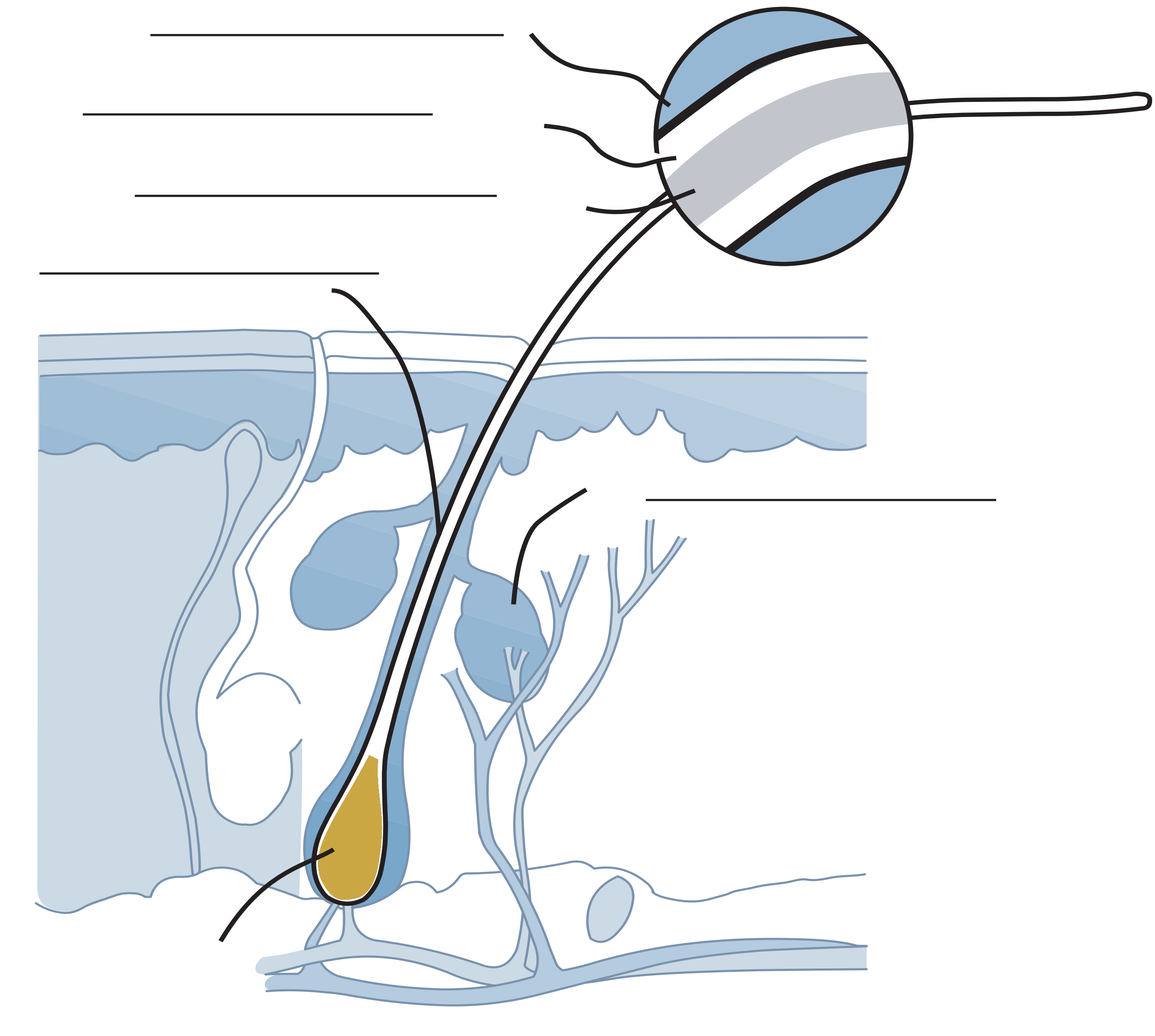
¥ Debate the pros and cons of admitting hair samples as evidence.

STUDENT HANDOUT

LESSON 3: HAIR IDENTIFICATION ACTIVITY SHEET

INTRODUCTION:

Hair is part of the integumentary system. It originates in the layer of skin called the dermis. The dermis contains numerous follicles from which hair grows. The hair follicle itself is nourished by blood vessels. Sebaceous glands are attached to the hair follicle to secrete oil for the hair and the skin. The unexposed part of the hair is called the root, while the part of the hair above the skin is the shaft.



UNIT 4

STUDENT HANDOUT

Hair Cuticles


Damaged hair cuticle


Healthy hair cuticle

The hair shaft is composed of three distinct areas. The outer layer of the hair shaft is the cuticle. The cuticle is composed of dead keratin-filled cells that form scales on the surface of the hair. The cuticle protects the hair from damage, but chemicals such as dyes and bleaches can damage the cuticle, therefore creating damaged scales. (See figures above.) The cortex of the hair shaft lies just inside the cuticle. Hair pigment (color) originates in the root and is scattered throughout the cortex. (Absence of pigment causes gray hair.) When hair is bleached, the natural pigment is removed from the cortex and the hair takes on a yellow color. The innermost area is called the medulla. The medulla looks like a tube running through the center of the hair. The size of the medulla is important in identifying the animal from which the hair originated.

Hair strands found at the scene of a crime can often provide clues for investigators. While hair analysis seldom provides definitive proof of a suspect s involvement, it can help to eliminate persons of interest and provide clues as to who might have been present at the scene of a crime. In our investigation, you will be making scale casts and wet mounts of hairs collected from the suspects and found at the crime scene. The scale casts will enable you to view the hair cuticles. You should be able to identify healthy hair and hair that may have been damaged by bleaching. The wet-mount slides will allow you to view the cortex of each hair to identify the color. In some cases, these slides will also allow you to view the medulla of the hair and identify any patterns visible in the medulla. It is important to understand that hair among one individual s can vary dramatically. Sometimes the medulla is visible and sometimes it is not. Hairs from the same individual can also differ in color. Keep this in mind as you complete the lab.

When forensic scientists collect hair or fibers from the scene of the crime, it is analyzed to determine its origin (animal, human, or carpet fiber) and then compared with the hair or fibers of the suspect(s).
Microscopic hair analysis is an important forensic tool, but it is a presumptive tool. Because the composition of many humans hair is similar, scientists can merely create possible connections between a suspect and a crime. Presently, scientists obtain more conclusive results by performing DNA tests on hair samples. You will combine microscopic hair analysis with other, more conclusive tests to determine the perpetrator(s) of this crime.

PROCEDURE:

(**Note:** *Be very careful not to mix up any of the hair samples provided in your envelopes. Be sure to label each slide.)*

Part I: Scale Cast Preparations

1. Using your permanent marker, label five slides 1, 2, 3, 4, and 5.
2. Pick up slide 1 and one coverslip. Place one drop of clear fingernail polish at one end of the slide.
3. Hold your coverslip at a 45-degree angle beside the drop of fingernail polish and allow the drop to run along the edge of the coverslip.
4. Now gently drag the coverslip across the slide so the polish covers the surface of the slide.
5. Quickly place three to four pieces of hair from Suspect 1 onto the polish so that the majority of the hair is lying on the slide (in the polish) and a small piece is hanging off the edge.
6. Place the slide someplace in an area where it will not be disturbed.
7. Now repeat steps 2—6 for the rest of the suspects hair samples. One partner can create slides 2 and 3 and the other partner can create 4 and 5.
8. Allow all slides to dry for 20 minutes while working on Part II.
9. After 20 minutes, use the forceps to pull the hair out of the polish (and off the slide) by quickly tugging it at the exposed ends.

Part II: Wet Mount Preparations

While you are waiting for your scale casts to dry, create a wet-mount slide for Suspects 1—5 using your second set of slides.

1. Using your permanent marker, label five slides 1, 2, 3, 4, and 5.
2. To create a wet-mount slide, simply place a few strands of hair on the slide and place one drop of water on the hair.
3. Hold your coverslip at the edge of the water drop and allow the water to run along the edge of the coverslip.
4. Gently lay the coverslip on top of the water, making sure no air bubbles form.
5. If you have an air bubble, gently tap it with your eraser until it exits the coverslip.
6. View your specimen first under low power, then medium, then high.
   ALL DRAWINGS SHOULD BE DONE WHILE USING THE HIGH-POWER OBJECTIVE.

Part III: Data Collection

Using the scale casts and wet-mount slides you have prepared, complete the data table below for the suspects hair samples. Make sure your sketches are done using high power! When you have completed your analysis of all suspect samples, your teacher will provide the evidence hair slides for you to examine.

STUDENT HANDOUT

HAIR ANALYSIS DATA TABLE:

	Sketch of Hair Shaft	Color of Hair	Drawing of Hair Scales	Is Hair Bleached or Natural?
Suspect 1				
Suspect 2				
Suspect 3				
Suspect 4				
Suspect 5				
Evidence				

ANALYSIS AND CONCLUSIONS:

1. *Describe the characteristics and the functions of the following structures:*

   a. hair cuticle:

   b. hair cortex:

   c. hair follicle:

2 *Which structures of the hair were closely analyzed in this lab?*

STUDENT HANDOUT

PROCEDURE:

3. *Why did we make casts of the hair?*

4. *Describe the scales of healthy hair versus chemically treated or damaged hair.*

5. *Based on your analysis, do any of the suspects hair types match the evidence hair? Does this provide any conclusive evidence with respect to the case?*

6. *Can you determine who committed the crime based solely on hair evidence? Why or why not?*

7. *What are some obstacles scientists might face when analyzing hair from the scene of a crime?*

8. *How is hair analysis useful in a criminal case?*

9. *For further exploration*
   Describe the differences between human hair found on the head and on other areas of the body.

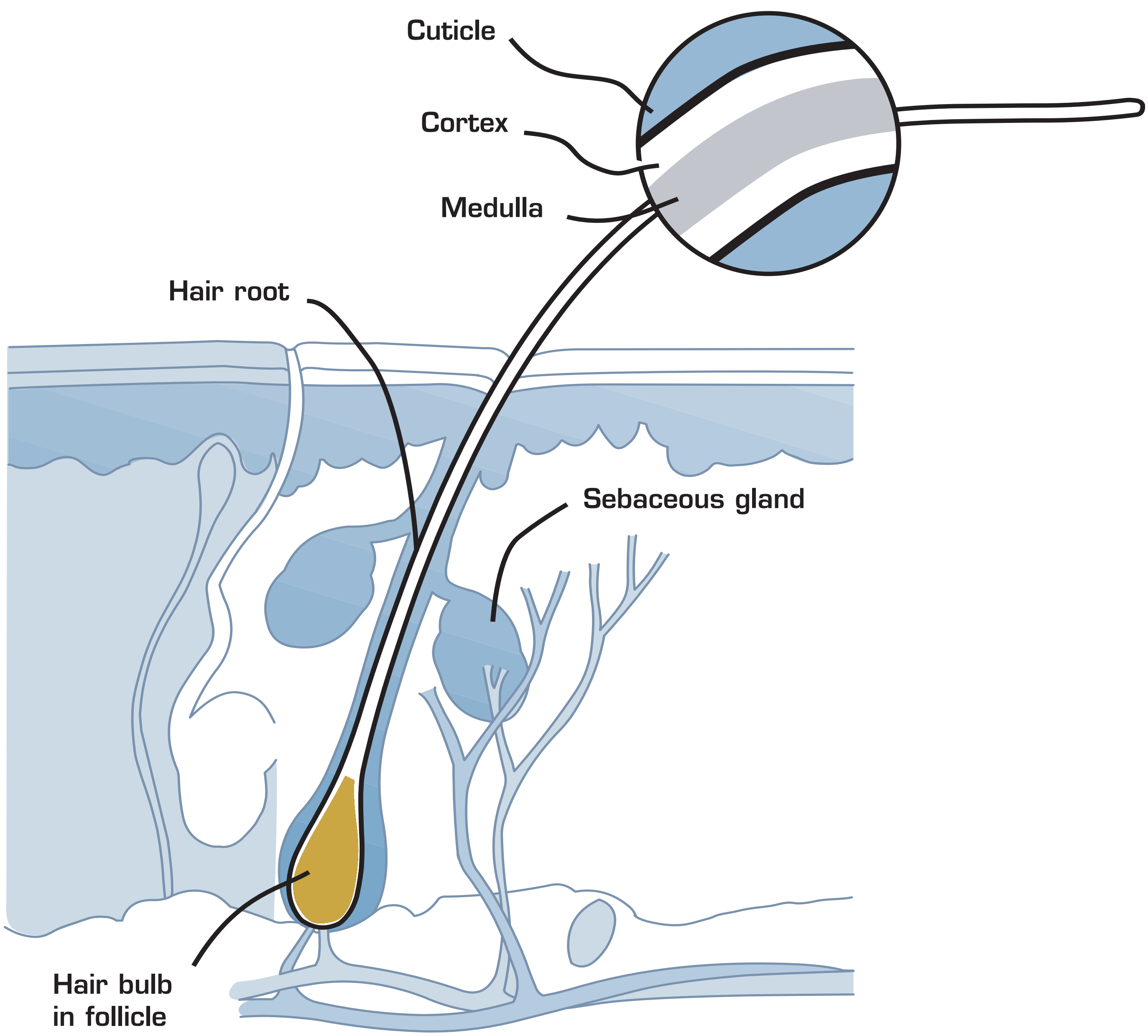
   Describe a criminal case in which hair was a pivotal piece of evidence in the trial of the criminal.

CONCLUSION:

Write two to three sentences summarizing the results of this activity as they relate to your investigation. You will need to refer to this information later in the investigation.

LESSON 3: HAIR IDENTIFICATION ACTIVITY SHEET

When completed, the hair identification diagram should appear as follows:



LESSON 4: BLOOD, CHROMOSOMES, AND DNA ANALYSIS

OBJECTIVE:

Students will learn how to use Punnett squares to determine the blood types of the suspects and will eliminate three suspects based on their blood type. Students will then construct karyotypes to determine chromosomal composition of the suspects and eliminate one more suspect from the field. Because the last two suspects will have the same karyotype, students will need to perform a DNA profile analysis. They will analyze the DNA profiles of the last two suspects to determine who cut the gas line in the cafeteria.

MATERIALS NEEDED:

Reproducibles

¥ *Blood Analysis Activity Sheet*
¥ *Part I: Blood Typing*
¥ *Part II: Karyotype Analysis*
Chromosome Spreads of the Two Suspects Blood and the Evidence Karyotype (optional)
¥ *Part III: DNA Profile Analysis*
¥ *Kastle-Meyer Color Test (Optional: see Appendix)*
¥ *Investigative Report*
¥ *Unit Epilogue*

Lab Supplies (For groups of 2)

¥ *Scissors*
¥ *Glue sticks*
¥ *Copy paper*

Note: *Any time you use chemicals in your classroom or lab, be sure to read and follow the safety instructions for use and disposal that are provided in the MSDS information that came with your lab materials.*

TIME REQUIRED:

¥ *Teacher Prep Time: 15 minutes*
¥ *Class Time: (longer if you choose to include the Kastle-Meyer test)*

LESSON DESCRIPTION:

In the previous lessons, students uncovered evidence that enabled them to bring the five suspects in for questioning. The hair analysis lesson provided information regarding the possible hair type of a person or person s at the scene of the crime, but it did not provide conclusive evidence as to who cut the gas line. In this lesson, students will use various methods of blood analysis to narrow down the suspects. If you wish to extend this lesson, students can perform the Kastle-Meyer Color Test before the blood analysis activities to determine whether the liquid on the gas line is blood. This optional lab is found at the end of the lesson. If you choose not to use the Kastle-Meyer test, then students will begin the lesson by analyzing the blood types of the parents of the five suspects to determine their sons possible blood types (using Punnett squares). Students will then compare those blood types to the blood type found at the crime scene. This will help determine which of the suspects could have cut the gas line and should be charged with the crime. This research will eliminate Suspects 1, 3, and 5. Next, students will cut out the chromosomes from the chromosome spreads for Suspects 2 and 4 and assemble them on blank paper to form a karyotype. They will then analyze the karyotypes and compare them to the karyotype of the crime scene blood. After discovering that Suspects 2 and 4 are both XYY males and that the blood at the scene was from an XYY male, students will then compare DNA profiles of the suspects and the evidence blood to determine that the person who cut the gas line is Suspect 4. They will then use the findings from each of their labs to write up an Investigative Report summarizing their conclusions.

BACKGROUND INFORMATION:

Before beginning this lesson, students should have a basic understanding of how to work Punnett squares for blood typing, how to analyze karyotypes for aneuploidies, and how to analyze and compare DNA profiles. If you are familiar with gel electrophoresis, describe this process to your students. If not, refer to the web sites listed below for in-depth explanations. You should also familiarize yourself with the Important Vocabulary Terms found at the beginning of the unit. It is helpful to clarify and reinforce these terms with your students, because they are used throughout the activity.

Before beginning this activity, it is important to adequately prepare for questions regarding blood typing, genetic diseases, and DNA profiling. To research additional information on this topic, refer to the following web sites:

¥ http://www.pbs.org/wgbh/nova/sheppard/analyze.html
¥ http://www.pbs.org/wgbh/nova/sheppard/courtroom.html
¥ http://science.howstuffworks.com/dna-evidence2.htm

There are many interactive DNA fingerprinting web sites available to students. You may want to browse through these or have your students visit them before this lesson.

LESSON STEPS:

Teacher Preparation

1. *Set out enough glue sticks, scissors, and copy paper for each person in the class.*

STUDENT HANDOUT

Lab Execution

1. If you choose to incorporate the Kastle-Meyer Color Test in this lesson, complete that activity first. (Activity procedures are included at the end of this lab.)
2. Distribute and review the Introduction and Part I of the Blood Analysis Activity Sheet. Allow students time in class to work the Punnett squares. Do not distribute Part II (the chromosome spreads for Suspects 2 and 4) until students complete the Punnett squares and determine that neither Suspect 1, 3, or 5 could have been the perpetrator. (If you give out the chromosome spreads at the beginning of the lesson, students will automatically know that Suspects 1, 3, and 5 have been eliminated.)
3. When students have finished the Punnett squares and have eliminated Suspects 1, 3, and 5, distribute the optional Part II: Karyotype Analysis. (Do NOT distribute the karyotype of the evidence blood until students have finished constructing the karyotypes for the suspects.) When students receive the evidence karyotype, they will determine that Suspects 2 and 4 are XYY males, as is the person whose blood was obtained from the crime scene.
4. Once students have completed the karyotype analysis and determined that either Suspect 2 or 4 could have cut the line, distribute Part III: DNA Profile Analysis. Instruct students not to discuss their findings with each other. (Note: This method of karyotype analysis is actually considered outdated in the world of forensics. However, it is still a quite common exercise in many high school biology labs, which is why it is included here. That said, the findings from these exercises are not required to solve the mystery, so if you would prefer to omit this step, you may do so.)
5. Once students have completed all parts of the lab, distribute the Investigative Report. Instruct students to compose their report based on results of all pieces of evidence they have examined. You can adjust the length and specificity of the report to your preference and students level and interest.
6. Once students have reported on their conclusions, provide the Epilogue to the mystery.

STUDENT HANDOUT

ACADEMIC EXTENSIONS/MODIFICATIONS:

- *If time permits, perform the Kastle-Meyer Color Test before the blood analysis lab to determine whether the liquid on the gas line is blood. This optional lab is found at the end of the lesson.*
- *If resources are available, you can order a simulated blood typing kit and substitute the Punnett square activity for actual blood typing. These kits include simulated or sterile blood samples and run between $25.00 and $30.00 for 50 students. Set up one type as the evidence blood and use the same type for Suspects 2 and 4.*
- *Have students create a pamphlet on the various types of genetic diseases that can be diagnosed by using karyotypes.*
- *Create an Internet lesson centered on DNA profiling. Use the web sites listed previously to help you locate these activities.*
- *Research the history of blood typing and transfusions. Identify the universal donors and recipients.*
- *To abridge this lesson, have students complete their Investigative Reports as homework.*
- *You may also choose to have students report their findings and present their conclusions to the officer who announced the crime.*

STUDENT HANDOUT

LESSON 4: BLOOD, CHROMOSOMES, AND DNA ANALYSIS

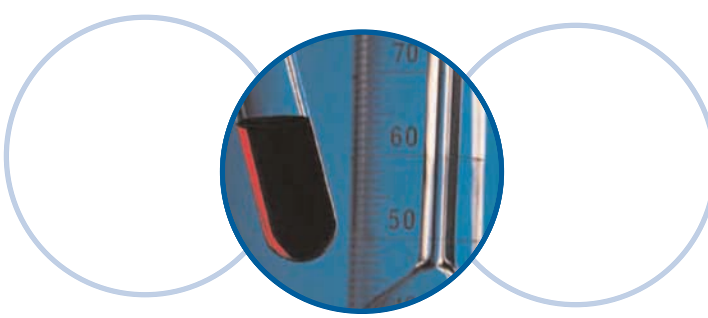
INTRODUCTION:

Blood analysis is a crucial part of modern forensic investigations. Since the discovery of the importance of DNA in the 1950s, investigators have relied more and more on DNA evidence. DNA can be isolated from hair roots, blood, and some bodily fluids. Once DNA is extracted, it is cut into smaller fragments using restriction enzymes. The same enzymes used to cut suspects DNA are used to cut evidence DNA. By cutting the suspect DNA and the evidence DNA at the same place, forensic investigators can compare DNA fragments. The resulting DNA fragments will either match or not. DNA fragments are analyzed by using gel electrophoresis and other methods to create a DNA profile. Except for identical twins, no two people have the same DNA. Although the DNA may be similar for a particular segment from two different persons, the more segments that are examined, the smaller the likelihood that there will be a match. Present DNA analysis uses enough fragments that the likelihood of finding a match between any two people is vanishingly small. Consequently, DNA evidence can be quite conclusive. This is not to say that errors in the analysis process cannot arise. Your teacher may provide web sites for you to further investigate the uses of DNA analysis in criminal investigations.

In this lesson, you will gather more conclusive information to help determine which of the five suspects cut the gas line in the cafeteria and should be charged with the crime. You will begin by determining the possible blood types of the five suspects using Punnett squares. You will be provided with the blood types of each suspect s parents. Cross the two parental blood types provided to determine the possible blood types of their children. (Because you have only the parents blood types, Punnett squares may provide 1, 2, or 3 possible blood types for each child.)

The 8 possible phenotypes for blood are A+, B+, AB+, O+, A-, B-, AB-, and O-. The + and — represents the presence or lack of the D antigen, which is one of the antigens in the Rh classification. If you are Rh positive, the D antigen is expressed. (This was named after a Rhesus monkey in which the marker was discovered.)

The alleles coding for types A (IA) and B (IB) are dominant with respect to the allele coding for type O(i), while the alleles coding for types A and B are codominant with one another. The Rh+ allele is dominant with respect to the Rh- allele. Each parent contributes one allele (gene) to each child; therefore, both parents determine the blood type of a child. Using this information, you will determine the possible blood types of the five suspects and compare them to the evidence blood type. Because many people share the same blood type, this test must be used only in conjunction with more conclusive tests.



STUDENT HANDOUT

PROCEDURE:

Use the following information to determine the blood types of the five suspects. If a parent is type A, remember that he or she can be IAi, or IAI. Also you must show all possible crosses. (More than one Punnett square has been provided for these situations.) Analysis of the crime scene blood has shown that the person who cut the gas line is Type O.

Suspect 1: Dad: AB Mom: B

Genotypes and Ratio:
Phenotypes and Ratio:

Genotypes and Ratio:
Phenotypes and Ratio:

Genotypes and Ratio:
Phenotypes and Ratio:

Genotypes and Ratio:
Phenotypes and Ratio:

*Possible Blood Types for Suspect 1*_______________

Suspect 2: Dad: B Mom: B

Genotypes and Ratio:
Phenotypes and Ratio:

Genotypes and Ratio:
Phenotypes and Ratio:

Genotypes and Ratio:
Phenotypes and Ratio:

Genotypes and Ratio:
Phenotypes and Ratio:

*Possible Blood Types for Suspect 2*_______________

STUDENT HANDOUT

Suspect 3: Dad: A Mom: AB

Genotypes and Ratio:
Phenotypes and Ratio:

Genotypes and Ratio:
Phenotypes and Ratio:

*Possible Blood Types for Suspect 3*________________

Suspect 4: Dad: O Mom: O

Genotypes and Ratio:
Phenotypes and Ratio:

*Possible Blood Types for Suspect 4*________________

Suspect 5: Dad: AB Mom: AB

Genotypes and Ratio:
Phenotypes and Ratio:

*Possible Blood Types for Suspect 5*________________

STUDENT HANDOUT

ANALYSIS AND CONCLUSIONS:

1. Can you eliminate any of the suspects based on their blood type? If so, who?

2. With this information, can you prove that one of these individuals cut the gas line? Why or why not?

3. On truTV, Mrs. Smith and Mrs. Jones were arguing about whether or not their babies were switched at birth. The following is known:

Baby Smith has type B blood.
Baby Jones has type O blood.
Mrs. Smith has type O blood and Mr. Smith has type B.
Mrs. Jones has type AB and Mr. Jones has type B.

Did a mixup occur?

STUDENT HANDOUT

PART II: KARYOTYPE ANALYSIS

Now that you have narrowed down the suspect field from five people to two, you will examine those two suspects more closely using a karyotype analysis. Karyotype analysis is often used to determine genetic diseases in utero before a baby is born. To create a karyotype, a picture is taken of a cell during metaphase while it is actively dividing. When a picture is taken of this phase, each of the 23 chromosome pairs can be identified. After the picture is taken, a scientist cuts out the chromosomes in the picture and arranges them to form a karyotype. Before computers, this work had to be done by hand. Fortunately, computers are now able to construct karyotypes directly from metaphase pictures.

In this activity, you will be constructing karyotypes of the two suspects chromosomes to determine any genetic abnormalities. Karyotypes normally identify males as XY for the 23rd pair of chromosomes and females as XX for the 23rd pair. But karyotypes can also reveal genetic diseases called aneuploidies, which involve the deletion (partial or full) or the addition of an X or Y chromosome. Some examples include Turner s female (only one X), Down syndrome (three instances of chromosome 21 instead of the normal pair), Super male (XYY), and Klinefelter s male (XXY).

Once you complete the karyotypes of the two suspects, you will then compare them to the karyotype of the blood found at the crime scene.

PROCEDURE:

(**Note:** *Each group member should do one karyotype.)*

1. *Ask your instructor for the chromosome spreads of the two possible perpetrators.*
2. *Cut out the chromosomes and paste them, in order, on a blank sheet of paper so that they form a karyotype.*
3. *Analyze the karyotype and identify any genetic disorders.*
4. *Now ask your instructor for the karyotype of the Evidence chromosomes. Compare the suspects karyotypes to the Evidence karyotype.*

ANALYSIS AND CONCLUSIONS:

1. Define autosomes and sex chromosomes.

2. What is an aneuploidy?

3. What causes aneuploidies?

4. Give an example of a sex chromosome aneuploidy and an autosomal aneuploidy.

5. Did either of the suspects have aneuploidies? If so, describe the aneuploidy and its symptoms.

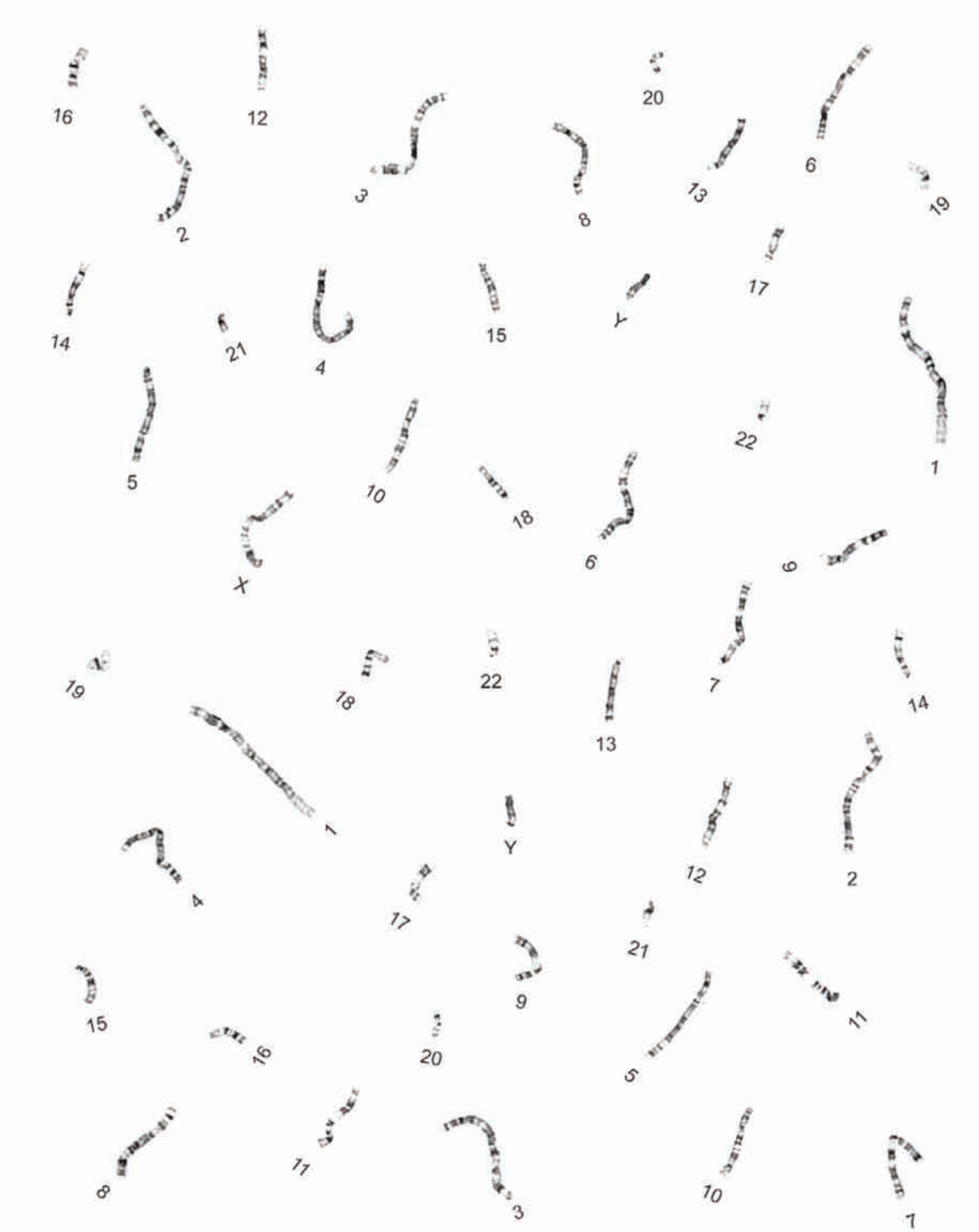
6. After comparing the suspects blood to the evidence blood, can you identify the perpetrator? Why or why not?

7. Name two diseases that cannot be diagnosed using a karyotype and explain why this is so.

STUDENT HANDOUT

CHROMOSOME SPREAD FOR SUSPECT 2:

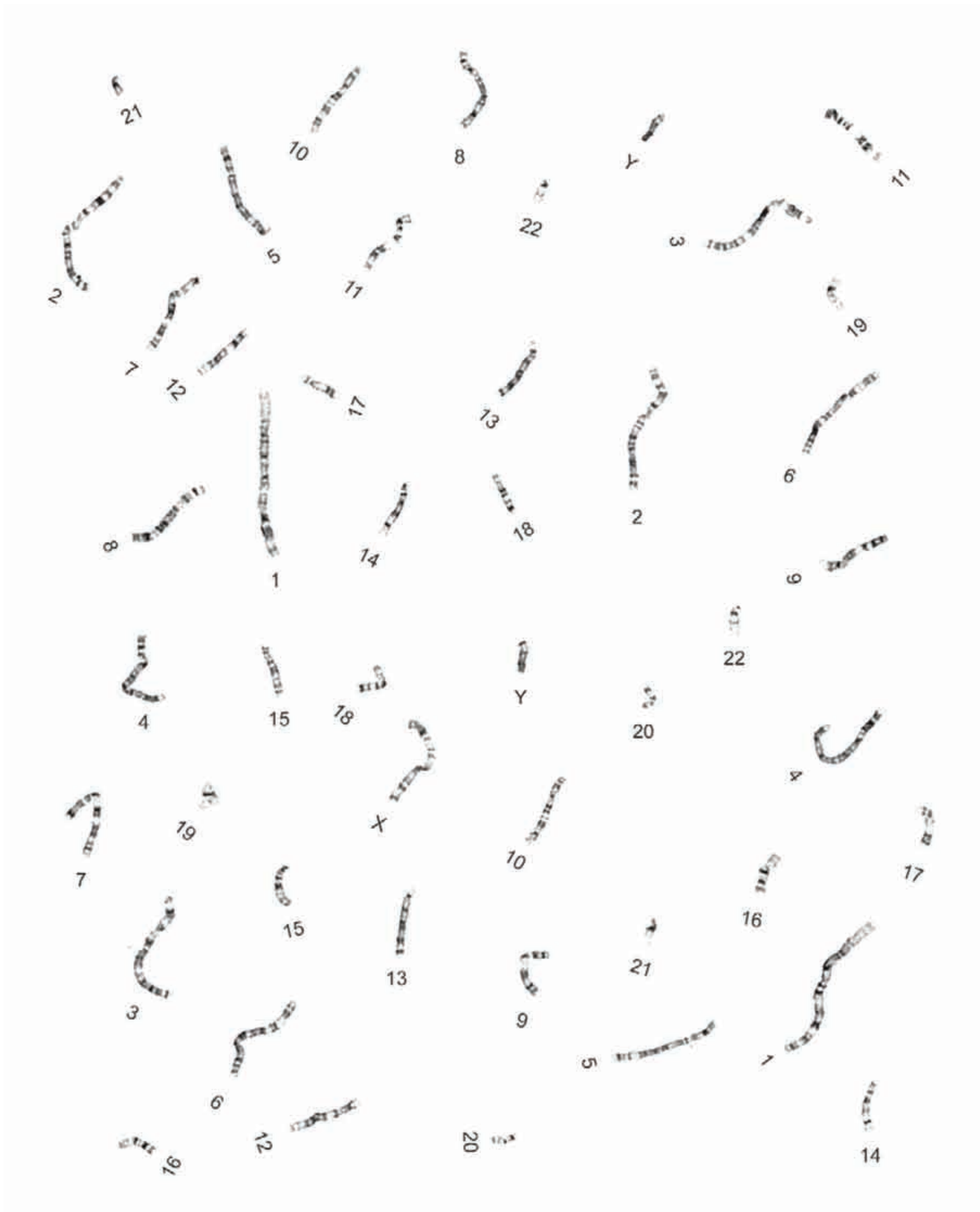
Cut out the individual chromosomes below and paste them, in order from left to right in a line, on a blank sheet of paper. Then analyze the karyotype and identify any genetic disorders indicated.



STUDENT HANDOUT

CHROMOSOME SPREAD FOR SUSPECT 4:

Cut out the individual chromosomes below and paste them, in order from left to right in a line, on a blank sheet of paper. Then analyze the karyotype and identify any genetic disorders indicated.



STUDENT HANDOUT

EVIDENCE BLOOD KARYOTYPE:

Permission granted for educational use only by Greenwood Genetic Center. ' 1995

PART III: DNA PROFILE ANALYSIS

The final phase of this lab includes a DNA profile analysis. Since a complete set of DNA is found in every nucleated cell of our bodies, we can extract DNA from any such cells we can find. Apart from tissue itself, these cells are present in hair roots, blood, and other bodily fluids. You will be analyzing profiles of DNA obtained from the two suspects and comparing them with the profile of the DNA extracted from blood found at the scene of the crime. This will be the final step in identifying the person who cut the gas line.

Recall that human DNA, spread over 23 pairs of chromosomes, has billions of base pairs of adenine bonded to thymine and of cytosine bonded to guanine. Trying to view all of these base pairs at one time is impossible. Fortunately, on each chromosome there are sequences of bases between genes that are not themselves genes. It is believed these sequences do not code for anything. (Remember, genes are sequences of bases that code for specific traits.) This non-coding DNA is sometimes called "junk DNA." Scientists have found the overall pattern of "junk DNA" to be unique to an individual, the only exception being for identical twins. Within these non-coding regions, there are repeating patterns of bases. For example, on a given chromosome the bases TAGA may be repeated 4 times in a row, producing a sequence of TAGATAGATAGATAGA. At the same site of the same chromosome in another individual, the bases TAGA may be repeated only 3 times in a row, producing a sequence of TAGATAGATAGA. All individuals have these repeating patterns, referred to as Variable Number of Tandem Repeats (VNTRs). Although the length of repeats at a VNTR locus may be exactly the same for a number of individuals, the likelihood that it will be exactly the same for any two individuals at 4 or 5 different VNTR loci is extremely small.

Using a method called PCR, or Polymerase Chain Reaction, millions of copies of a specific DNA fragment can be made from a single fragment, thus vastly increasing the quantity of DNA material available to be analyzed. This is especially helpful for forensic scientists, because the amount of DNA at a crime scene is usually very limited and indeed often too small for RFLP-based methods initially used in forensic DNA analysis. Fragments from several VNTR loci of a suspect s DNA are selected for amplification by PCR, and the same is done for the same VNTR loci of the evidence DNA. The resulting fragment lengths from the respective loci are then compared to determine, from DNA profiles, whether a "match" exists between the evidence DNA and that of any of the suspects.

DNA profile images are created using the following steps:

1. *DNA is collected at the crime scene from blood, semen, saliva, hair roots, or skin.*
2. *A specific VNTR locus on a specific chromosome is selected to analyze.*
3. *Enzymes are used to cut each end of the repeat fragment at a specific location. The section is then amplified using PCR, an overnight run generally being sufficient to produce a million copies of the selected fragment.*

STUDENT HANDOUT

4. *To get a useful profile for an individual, VNTR fragments from several different locations (loci) on the chromosomes are selected and analyzed. In this activity we are examining only 4. In the technique using shorter fragments, called Short Tandem Repeats, the standard number of loci examined is 13. VNTR fragments, being longer, display greater diversity from one person to the next, and so fewer loci are needed to obtain a profile for the individual that is highly unlikely to be repeated in another individual.*

5. *After the DNA fragments of interest have been amplified through PCR, the resulting material is then introduced at one end of a gel and covered with buffer solution, which causes the DNA fragments to take on an electric charge. An electric field is then applied to the gel in an electrophoresis chamber. The electric field causes the charged DNA fragments to move away from where the material was deposited, toward the opposite end of the chamber. The key to this technique is that the different lengths of fragments will move at different speeds; generally, the shorter ones moving faster. After a fixed time this results in a separation of the different-length fragments into a series of bands spread along the length of the gel. Various techniques exist for making the bands visible.*

6. *This procedure is done with both the evidence DNA and DNA from one or more known individuals, including but not limited to suspects. This results in several "ladders" of bands, one for each of the known DNA samples and one for the evidence DNA.*

In the image below, four different VNTR loci were chosen. Since each chromosome has two copies, two bands are created from each section for each sample (each individual). A VNTR-band ladder is presented for each of the two suspects DNA and for the evidence DNA, for the same four loci. A control ladder, with many bands, is also provided to offer a check and calibration for the analysis.

PROCEDURE:

Analyze the DNA profiles from the two suspects and from the evidence blood:

Fragment Size (Number of nitrogen bases in VNTR section)	**Control Ladder**	**Evidence DNA**	**Suspect #2**	**Suspect #4**
200	—	—		—
175	—		—	
150	—	—	—	—
125	—	—		—
100	—	—	—	—
75	—		—	
50	—	—		—
25	—	—	—	—

STUDENT HANDOUT

ANALYSIS:

1. Define VNTR.

2. Why is DNA an important tool in criminal investigations?

3. What is PCR and why is it an important procedure in criminal investigations?

4. After analyzing the DNA profiles of the suspects and the evidence, which individual can be eliminated as a suspect? Is this evidence conclusive in identifying the perpetrator? Why or why not?

CONCLUSION:

Write two to three sentences summarizing the results of this activity as they relate to your investigation. You will need to refer to this information as you complete the Investigative Report.

STUDENT HANDOUT

INVESTIGATIVE REPORT

To conclude your investigation, you need to summarize all of your results in an Investigative Report. A report such as this is intended to take all the evidence you have the results of the enzyme tests, hair analyses, and blood, chromosome, and DNA analyses to determine the identity of the guilty party. The Investigative Report is used by the district attorney s office during the prosecution of the criminal, so it needs to be as thorough as possible. You may also cite information provided by witnesses; however, your conclusions should rely strongly on the results of the scientific tests and physical evidence. Refer back to your notes in the Conclusion section of each unit activity for details.

Questions that should be answered by this report (in clear and concise sentences) include the following: What did the enzyme tests indicate about the contents of the liquid found on the cafeteria floor? What did the hair analysis show? Is this conclusive evidence? What unique abnormality did the karyotype analysis reveal? What does this mean with respect to the perpetrator? What evidence does the DNA profile analysis provide? Based on all the evidence at hand, which of the suspects cut the gas line in the cafeteria?

DATE: ______________________________

INVESTIGATOR NAME: ______________________________

LOG OF EVIDENCE RECEIVED: ______________________________

____

____

____

____

CONCLUSIONS: ______________________________

____

____

____

____

CONFIDENTIAL: This is an official report and is to be issued in connection with an official criminal investigation. Do not make public or reveal the contents hereof to any unauthorized person.The form of this report is found to be in compliance with NCGS 000001 and is hereby approved by the Attorney General.

STUDENT HANDOUT

EPILOGUE: UNIT CONCLUSION

David Host found himself back at the station. Like the last time he and the others were brought in, his mother was with him. David wondered why the others hadn t been called in this time. They all were there when it happened, he thought. It wasn t even his idea. He wondered now if that really mattered.

Detective Wulff looked down at David. "Philo Farnsworth was undoubtedly an interesting person," she began. "As far as I know, though, he was never a person of interest in a criminal investigation. In fact, David, you are no longer a person of interest yourself." David s heart raced. Maybe it wasn t as bad as he thought. The detective went on. "Nope, you re a full-fledged suspect, and I suspect that you ll be charged today."

David s mother cried out, asking David to tell the truth. David wanted to cry, too. "I don t know. I guess there s no point in denying it. I was part of the pranks and vandalism that occurred at the school. I broke some glass. I broke some trophies."

And you cut a gas line. You cut a gas line, David! Do you know how many lives you put in danger for that that prank you re responsible for?" said his mother.

David s first thought was that he hadn t really considered that before. His second thought was, how did they know he cut the line? And then, as if she were reading his mind, Detective Wulff said, "We found drops of your blood by the severed line."

David cast his eyes to the floor. "I was caught up in the moment, I guess. It s so rare that I m part of a group like that. It was great. The guys really liked me. We were having fun." And then it was as if David were getting caught up in the moment again just a different moment this time. He hit his fist on his knee. "We were taking out years of frustration on an educational system that claims to celebrate smarts but really doesn t. It celebrates celebrity. And celebrity and smarts don t dance together. They don t even speak the same language. Celebrity is sports. Celebrity is popularity. Celebrity is shallow."

And breaking trophies and cutting gas lines isn t?" Detective Wulff asked pointedly.

"Well, yes. Yes it is. But it did get us noticed."

"You re a bright kid," the detective said. She was not going to let him get away with his rant, even if parts of it were true. "That s why I can t imagine that this is the kind of notoriety you wanted, David. And maybe you re right that society doesn t give intelligence its due, but you ll have a hard time convincing me that your response to that was an intelligent one." David s expression told Detective Wulff that her "lecture" was not being lost on this particular Philophile.

STUDENT HANDOUT

His feeling idiotic wasn t enough for the detective, though. David needed to know that there were consequences to his actions and those of his fellow inductees. "Before this incident, I had never heard of Philo Farnsworth, David. Without question, it s true that he didn t get the recognition that he deserved. I can assure you, though, that you and your fellow vandals will get what you deserve. I don t know how Mr. Holden will punish you, but I can tell you that you could face criminal charges for vandalism, breaking and entering, and endangering the lives of students and school personnel by cutting that gas line. In addition to possible jail time, those offenses may carry many, MANY hours of community service to remind you just how unintelligent your choices were that weekend. The jury is still out on whether or not you ll get the celebrity status you desire."

Not long after word leaked out that the vandals had been caught thanks to the help of a student, more rumors began flying around Park Haven High. Spencer Rhodes had the feeling that some of those rumors involved him. He wasn t sure if the looks he seemed to be getting from other students were of admiration, disdain, or just his imagination. Spencer went back and forth between feeling like a big shot for having helped the detectives crack the case and feeling guilty over "ratting out" five guys he had once considered friends. In the final analysis, though, he knew that he had done the right thing by coming forward with the information. It s one thing to express yourself if you think that people are being treated unfairly, thought Spencer, but it s another thing if the form of expression you choose breaks the law and puts other people in danger. The Philophiles made a bad choice that put other people s lives at stake. Now they had to suffer the consequences of their actions. Besides, they all would have been caught eventually. It seemed to Spencer that a guilty person always says something, leaves something, or does something that indicates his or her guilt. In this particular case, Mr. Holden and Detective Wulff got lucky and had Spencer around to point them in the right direction.

When Spencer learned that all five guys had been expelled from school, he figured that it wouldn t go over well on their college applications. Maybe because their SAT scores were off the charts, schools might overlook their "youthful indiscretions." He doubted it, though, because colleges have plenty of students to choose from who have high test scores and no police record. Spencer was one of those students, and while the whole "ratting out" thing nagged at him a bit, he was comfortable about having come forward and pleased with his decision not to join the Philophiles. A wrong choice like that could have ruined his chances to get into MIT.

As he was mulling it all over, Jenny, a very smart and popular girl, walked over to Spencer. "Hey Spence," she said. "I heard you helped the detectives crack the case. Let s sit together at lunch today. I want to hear all about it."

LESSON 4: KASTLE-MEYER COLOR TEST

OBJECTIVE:

Students will be introduced to the story and will use the Kastle-Meyer Color Test to analyze what appears to be blood found at the crime scene.

MATERIALS NEEDED:

Reproducibles

- *Kastle-Meyer Color Test activity sheet (includes guidance for student observations and information about the reaction on which the test relies)*

Equipment and Chemicals

- *ethanol (95%) or distilled water*
- *Kastle-Meyer reagent**
  - *phenolphthalein***
  - *potassium hydroxide*
  - *absolute ethanol (100%)*
  - *zinc dust*
- *hydrogen peroxide (3%)*
- *sample of animal blood (can be obtained from a butcher) OR beef or calf liver (both contain high blood content), homogenized in a blender*
- *filter paper (cotton swabs may be used instead, if needed)*
- *latex/vinyl gloves*
- *chemical splash goggles*
- *droppers*
- *biohazard or other disposable bag*

** Premixed Kastle-Meyer reagent can be purchased from Antec, Inc.: 1-800-448-2954 or 1-502-636-5176*

*** Phenolphthalein can be purchased online from the following vendor, Sigma: http://www.sigmaaldrich.com or 1-800-325-3010; Product number P8903*

TIME REQUIRED:

- *Teacher Prep Time: 105 — 120 minutes*
- *Class Time: 1 class period (45 minutes)*

LESSON DESCRIPTION:

As part of the evidence analysis to determine whether the spot at the scene of the crime could be blood, students will first use a Kastle-Meyer Color Test to examine the residue for the presence of blood.

LESSON STEPS:

Lab Preparation

Kastle-Meyer (KM) presumptive test for blood

INGREDIENT	FINAL CONCENTRATION	AMOUNT
phenolphthalein	0.2%	2.0 g
potassium hydroxide	0.18 M	10.0 g
absolute ethanol (100%)	80%	800 ml
zinc dust	N/A	variable

Procedure

1. In an aluminum-foiled flask, dissolve the phenolphthalein in 200 ml deionized water.
2. Add potassium hydroxide. The phenolphthalein will dissolve.
3. Stir until clear (very light pink is OK).
4. Add the ethanol.
5. Add enough zinc dust to cover the bottom of the bottle.
6. Store between 2¡ and 8¡C in a dark or foiled bottle.

Prior to class, treat one set of filter paper samples with a 2:1 (water:blood) dilution. Label this set "Unknown from cafeteria." Prepare another set of paper samples with an undiluted blood sample and label it "Positive Control." Leave another set of paper samples untreated and label it "Negative Control." Each student should receive one sample from each set.

Lab Execution

1. Distribute the Kastle-Meyer Color Test reproducible. Familiarize students with the procedure and the purpose of testing a positive control (the sample that was known to contain blood) and a negative control (the sample that was known not to contain blood). Caution students about safety concerns associated with the lab. Specifically, they should wear clean gloves in order to avoid contaminating the wipes and chemical splash goggles to avoid eye irritation from hydrogen peroxide.

2. Using the procedures listed on the activity sheet, have your students perform the lab. Note that if they observe a pink color change prior to adding the hydrogen peroxide, other chemical oxidants may be present on the samples and are producing false positives.

3. Give students time to summarize their findings.

4. As a class, discuss the oxidation reaction that was observed.

5. Discuss with the class what the results suggest, namely that the presumed presence of blood in the cafeteria indicates that the person who cut the gas line could also have cut himself/herself in the process. Remind students, however, that the Kastle-Meyer test is a presumptive test. Reinforce the limitations of a presumptive test by having students define the term and indicate why the Kastle-Meyer test is presumptive.

KASTLE-MEYER COLOR TEST ACTIVITY

INTRODUCTION:

The Kastle-Meyer Color Test is commonly used to presumptively indicate whether blood is present on an object. It is fast and easy to use, making it an ideal test at a crime scene or for samples taken from a crime scene. The test relies upon the heme, the iron-containing portion of red blood cells, to catalyze the oxidation of phenolphthalein (also known as Kastle-Meyer reagent). Phenolphthalein is normally clear and colorless, but in the presence of blood and hydrogen peroxide it becomes pink. If results of a Kastle-Meyer test suggest the presence of blood, investigators then know that further analysis of the residue may provide additional useful information.

The disadvantage of the Kastle-Meyer test is that it is presumptive, not conclusive. If any chemical and plant oxidants from sources other than blood are present, the test can produce a false positive result. In this experiment, however, you will minimize false positive results by adding the hydrogen peroxide catalyst last. Other chemical oxidants that may be present will usually produce a pink color before the hydrogen peroxide catalyst is added.

The reaction that you will test for is described below:

Heme iron + phenolphthalein (clear) + hydrogen peroxide ⟶ oxidized phenolphthalein (pink) and water

(Heme is a catalyst and does not change. Hydrogen peroxide is reduced to water.)

SAFETY PRECAUTIONS:

Given the materials you will be working with for this test, safety goggles and latex gloves must be worn.

PROCEDURE:

1. *In order to determine that there are no other chemical oxidants present, first test the clean filter paper sample (Negative Control).*
2. *Apply one drop of ethanol or distilled water to the sample.*
3. *Apply one drop of Kastle-Meyer (KM) reagent to the same area on the sample and observe any color change. If there is a color change, record it in the chart provided. If not, record that no change occurred.*
4. *Apply one drop of 3% hydrogen peroxide to the sample area. Observe and record any color change. (Because there are no chemical oxidants present on the sample, you should observe and record no color changes.)*
5. *Repeat steps 2—4 on the Positive Control paper sample containing a known spot of blood. You should observe and record a pink color after adding the hydrogen peroxide.*
6. *Repeat steps 2—4 on the Unknown filter paper sample. The residue sample from the cafeteria has been wiped on it. Observe and record any color changes in the appropriate rows.*
7. *When finished, clean up your lab area and dispose of the bloody filter papers in the biohazard or other bag supplied by your teacher.*

OBSERVATIONS:

Enter the results of each test in the chart below. Make sure that you record precise descriptions of the color(s) that appear, and note when no color changes result.

	After Kastle-Meyer treatment	After hydrogen peroxide treatment
Negative control		
Positive control		
Unknown from cafeteria		

ANALYSIS:

Based on your recorded observations, answer the following questions:

1. *Did the wipes from the Unknown sample taken from the scene test presumptively positive for blood?*

2. *What was the purpose of performing the test on a known spot of blood and on clean filter paper?*

3. *Why is this test not conclusive with respect to the presence of blood?*

CONCLUSION:

Write 2-3 sentences summarizing the results of this test as they relate to your investigation. You will refer to this summary later in the investigation.