FISHER SCIENCE EDUCATION

HEADLINE DISCOVERIES

DNA: Expanding forensic science

It's the spellbinder of primetime television, appearing in shows from CSI to The New Detectives. It's genetic science applied to criminal investigation. Sleuthing around cells. Lt. Columbo, meet the double helix—forensic DNA testing.

For forensic scientists, a key characteristic of DNA is that its configuration is the same in all cells of an individual. While all humans' DNA bears marked similarities, the exact order of the base pairs makes each person (or animal) unique. Scientists use analytical methods to analyze these patterns. The analysis doesn't render an individual "fingerprint" per se, but tests can determine conclusively whether two DNA samples are from the same person, related people, or non-related people.

Clues at the crime scene

At one time, it would have been inconceivable that a tiny speck of blood or hair could mean the difference between a suspect's conviction or release. But, thanks to current DNA identification procedures, drawn from classic life science techniques, those specks are playing a crucial role in many trials. Increasingly, too, DNA analysis is being used to vindicate wrongly convicted people.

More than 60 prisoners in the United States have been exonerated as a result of post-conviction DNA testing, a fact that may turn one's notion of the legal system topsy-turvy. Now, all 50 states admit DNA evidence to some extent, and 34 states have post-incarceration DNA evidence laws.

Changing the face of forensics

Forensic use of DNA analysis in criminal cases began in 1986. Since then, DNA identification, popularly known as "DNA fingerprinting," has evolved rapidly into the foremost forensic technique for identifying perpetrators, eliminating suspects, and clearing people who were wrongly convicted.

Advances in DNA testing have further revolutionized the forensic analysis of blood, hair, saliva, fingernail scrapings and other bodily tissues and fluids left at a crime scene. Now widely found in life science labs, the Polymerase Chain Reaction (PCR†) method, an enzymatic process, is used to copy a section of DNA over and over, increasing the total amount of DNA available to be analyzed. This molecular copying technique makes it possible to generate reliable data from extremely small amounts of DNA found at the crime scene. Whereas previous methods required sample sizes as large as a dime-sized drop of blood, PCR testing of nuclear DNA can be done on samples as small as a visible dot of blood or single strand of hair, even if the sample is old or degraded. PCR testing of mitochondrial DNA (mtDNA), can be performed even on samples containing extremely old or highly degraded DNA, such as hair shafts and dried bones or teeth.

Match game

Scientists interpret DNA test results as an inclusion, exclusion, or inconclusive. An inclusion means that the results obtained from the suspect or other individual are consistent with, or show the same pattern as, the DNA results obtained from a crime-scene sample. In other words, the test yields a "match," which shows that the individual may be a source of the DNA found in the crime-scene sample. How likely that match is can be expressed using statistics generated from various population groups. If a match is contested, then more in-depth testing can be performed with remaining evidence and/or DNA.

An exclusion means that the DNA fingerprint pattern result yields a "nonmatch"; thus, the individual is eliminated as a source of the DNA found in the crime-scene sample. A DNA test is inconclusive when partial or no results indicate that there is too limited an amount of suitable human DNA, or no DNA samples for comparison to crime-scene samples.

Balancing science and justice

With rapidly evolving DNA technology, the role of forensic DNA analysis has profound implications. Advances in DNA testing have revealed imperfections in the criminal justice system, shaking the very foundation of our legal system.

But performance of DNA analysis, like any other laboratory activity, has an error rate. Occurrences of false positives (false matches) due lab error have highlighted the need for evidence control, validation, calibration, proficiency testing and an overarching program for reviewing and auditing laboratories—in short, the need for national laboratory standards. These have been developed and are required of labs to receive federal funding or participate in the FBI CODIS (COmbined DNA Index System) program, which allows law enforcement agencies to search for matches from pertinent databases.

Elementary, my dear Watson?

What would a Sherlock Holmes or Jane Marple have said about forensic DNA? That's one mystery that will remain unsolved, but genetic science will continue to unlock crime scene secrets for decades to come—helping to keep the scales of justice in balance.

MORE!➤

†The Polymerase Chain Reaction (PCR) process is covered by U.S. Patents 4,683,202; 4,683,195; and 4,965,188 or their foreign counterparts owned by Roche Molecular Systems, Inc. and F. Hoffmann-La Roche Ltd.

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In this activity, students will extract DNA from onion cells and examine some of its physical characteristics.

A CLOSER LOOK AT DNA

No other substance is as important as DNA—deoxyribonucleic acid. It contains the set of chemical instructions that controls all of a cell's activities. The process of DNA extraction is an essential first step in the analysis and manipulation of DNA. Scientists extract DNA to detect gene defects, produce DNA fingerprints and create recombinant organisms. This activity allows students to extract DNA and observe its physical characteristics.

PROCEDURE:

STEP 1: Breaking down the cell wall using a detergent/salt solution

Prepare the cell lysing solution by adding 10mL of light-colored liquid detergent or shampoo and 1.5g table salt to 100mL bottled water. Slowly stir the mixture (to prevent foaming) until the salt is completely dissolved.

Remove the skin and first few layers of a large onion. Coarsely chop and place the onion pieces in a 125mL beaker. Add enough of the detergent/salt solution to cover the onion pieces.

The detergent causes the cell membranes to lyse, releasing DNA into the solution. The salt contained in the solution pulls the phosphate ends of the DNA molecule closer together, making it easier to remove (precipitate) DNA out of solution.

STEP 2: Heating the extracted DNA solution to degrade DNase enzymes

Place the beaker in a water bath containing an inch of warm water.

Carefully heat water to 55–60˚C for about 10 minutes. Heating degrades the DNase enzymes that break the DNA strands into small fragments and make it difficult to spool.

While the mixture is heating, press the chopped onion against the side of the beaker with a Popsicle™ stick.

Cool the onion extract in an ice bath for 5 minutes while continuing to press the chopped onion against the side of the beaker. Rapid cooling will slow the breakdown of DNA.

STEP 3: Separating the onion extract

Pour the onion/alcohol mixture through a strainer lined with a #6 coffee filter or four layers of cheesecloth and capture the liquid portion (extract) of the mixture in a 50mL beaker. Keep this solution refrigerated or on ice.

STEP 4: Extracting DNA

Bend the end of a 10-inch piece of thin wire upward into the shape of a fishhook. This will serve as a DNA spooler.

Measure 5mL of onion extract into a test tube.

Holding the test tube at a 45° angle, use an eyedropper to slowly add drops of ice-cold isopropyl alcohol down one side of the test tube as shown in Figure 1.

The alcohol will form a distinct layer on top of the onion extract solution. Avoid shaking or mixing the layers. Let stand undisturbed for 2–3 minutes.

The alcohol will cause the DNA to come out (precipitate) of solution. Look carefully and you will see DNA floating in the alcohol layer.

Lower the wire DNA spooler into the test tube until the hooked end is at the boundary between the onion extract solution and the alcohol layer. Gently swirl the spooler to wind the DNA around it—similar to winding spaghetti around a fork.

Lift the DNA spooler out of the test tube and carefully observe the extracted DNA.

EXTENSION

All long linear chain molecules, like DNA, have the same physical characteristic of visco-elasticity—a resistance to flow and the ability to resume their original size after being stretched. To demonstrate this, carefully place the extracted DNA on the lip of the test tube and tug on one end of the mass. When the DNA is stretched, it forms thin, hair-like threads. If it is pulled too far, it snaps back.

Test other obviously viscous materials such as syrup, molasses, or white glue. Are any of these substances long, linear chain molecules?

SAFETY:

- **Safety goggles (HS47617W, student sizes available)**
- **Polyethylene gloves (HS47330)**
- **Wear goggles and gloves when handling ethyl alcohol**
- **95% ethyl alcohol is a flammable liquid—keep away from excessive heat and open flames.**

MATERIALS:

- Light-colored detergent or shampoo, Dawn™ or Suave™
- Water, bottled
- Table salt
- Onion, large
- Knife
- Beaker, 50mL (HS63251)
- Beaker, 250mL (HS63254)
- Water bath setup (HS52601)
- Beaker tongs (HS50275)
- Thermometer (HS63341)
- Balance (HS40075)
- Ice bucket (HS66126) with ice
- Strainer
- Popsicle stick
- Coffee filter (#6) or cheesecloth (HS47252)
- Test tube (HS63285)
- Graduated cylinder, 10mL (HS63305)
- Eyedropper (HS18750)
- Denatured ethyl alcohol (95%) (HS73979), 5mL stored on ice in a test tube
- Wire, thin (10-inches) (HS49083)

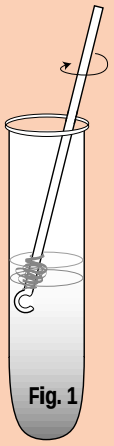
Fig. 1

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