AMGEN Biotech Experience

Scientific Discovery for the Classroom

Student Guide















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ABOUT THE AMGEN BIOTECH EXPERIENCE

Genetic engineering is a branch of biotechnology that uses special procedures and techniques to change an organism's DNA. This ability has had a huge impact on the field of medicine, as genetically modified bacteria can make human insulin (the hormone responsible for regulating glucose levels in the blood) and other life-saving products. It's rare for high school students to have the chance to learn about and actually practice the procedures and techniques that are the foundation of the biotechnology industry—but in this program, you will have just that opportunity. As you work in the laboratory and carry out the very experiments that led to breakthroughs in biotechnology, you will gain hands-on experience with producing genetically modified bacteria.

The procedures in this program were developed through a series of discoveries that led to important breakthroughs in biotechnology. Some of the pioneering scientists who made these discoveries received Nobel Prizes in Physiology or Medicine in 1978 and Chemistry in 1980 and 1993. (The Nobel Prize is the highest distinction awarded to scientists in these fields from around the world.) The work that you are about to do is based on this Nobel Prize-winning science—science that is significant and will continue to play an important role in the development of biotechnology and medicine. You will follow in the footsteps of the many scientists who have pushed and continue to push the boundaries of biotechnology. There are many advances still to be made—and students who decide to continue studying this field may contribute to those advances.

In science, the ability to keep track of what you are doing and communicate about your work is extremely important. To demonstrate that you performed an experiment, either so that it can be duplicated and verified by others or if you want to apply for a patent—you need to have a very accurate record of what you've done. As you carry out this program, carefully record your notes, ideas, observations, results, and answers to questions in a science notebook, in pen. (For scientific purposes, it is important to keep a record—even of your mistakes.) If possible, use a separate bound composition notebook and organize the labs with a table of contents at the front. Since you will use a pen to write with, you'll need to cross out any mistakes you make—and it is good practice to simply "X" out the section you want to change (so that it can still be read) and to note

why you've done so. Following these best practices will make this program even better preparation for you!

The Amgen Biotech Experience (formerly Amgen-Bruce Wallace Biotechnology Lab Program) had humble beginnings 30 years ago with visionary scientists and teachers who shared passion and energy for imparting their knowledge with students. Bruce Wallace, one of Amgen's first staff members, wanted all students to experience the joy of discovery and the excitement of having science at their fingertips. A desire for more robust science education at schools near Amgen's global headquarters led to involving area high school teachers and, later, a college professor, in developing curriculum and educator training in biotechnology. The program grew through word of mouth and teacher interest, and expanded over time to other states and countries.

Visit the ABE website at www.amgenbiotechexperience.com.



PROGRAM INTRODUCTION AMGEN BIOTECH EXPERIENCE



WHAT IS BIOTECHNOLOGY?

At its simplest, biotechnology is the use of biological systems to create products. The use of yeast to make bread is one of the earliest examples of humans using a biological process (fermentation by yeast) to create a desired product (food).

It was not until the 1970s that the science of biotechnology really took off when scientists made two key discoveries about bacteria. The first discovery was that bacteria contain tiny circles of *DNA* (deoxyribonucleic acid, a double-stranded biomolecule that encodes genetic information), called *plasmids* within them. The second was that bacteria also contain *proteins* (large biomolecules that carry out essential functions in cells) called *restriction enzymes* that can cut DNA at very specific places.

The findings made by basic research often lead to fundamental understandings about the nature of life. In some instances, these findings can also lead to new tools and technologies that can improve life. With the discovery of plasmids and restriction enzymes, for example, a whole new era of biotechnology using recombinant DNA technology was launched. *Recombinant DNA* refers to DNA that contains sequences or genes from two or more sources—sometimes even from two different species! By harnessing natural biological processes, scientists can generate products that can contribute to human society in ways never before imagined.

Modern biotechnology is now used to develop hundreds of products and technologies—to create fuels to power the world, to develop better systems for the production of food, and to improve human health.

STUDYING HUMAN BIOLOGY TO TREAT DISEASES

Biopharmaceutical (biopharma) researchers study human biology to better understand how to develop solutions to improve the lives of people who suffer from serious diseases. To do so, these researchers study a disease closely,

exploring its mechanisms and the changes it causes to the human body. Based on this research, scientists can develop biopharmaceutical therapies that take advantage of biological systems to treat or cure these diseases.

The biopharma industry ushered in a new wave of protein-based medicines that are made through the marriage of science and the molecular machinery of *cells* (the basic units of any living organism that carry on the biochemical processes of life). The earliest biotech drugs were genetically engineered versions of human proteins—large molecules far too intricate to assemble through chemical processes but which could be made by harnessing cells with strategically-engineered DNA. Today, protein engineers can reconfigure nature's building blocks to design innovative structures that fight disease in a more sophisticated manner.

What is the relationship between DNA and proteins? Both are *biomolecules*, large molecules made by living cells. When scientists investigated *traits* (genetically determined characteristics) in organisms, they found that proteins were responsible for traits and that DNA was responsible for creating proteins. For example, consider a plant that has the trait of red flowers. The flowers' red pigment is produced by the action of an *enzyme* (a protein that increases the rate of a chemical reaction). The DNA in that plant contains instructions for making proteins, including that enzyme. The part of a DNA molecule that has the instructions for making a particular protein is called a *gene*.

THE FUTURE OF BIOPHARMA

With our advanced understanding of the human genome and the wealth of human genome data available today, biopharma researchers are finding new ways to identify the genetic basis of diseases and individual responses to treatments so that they can target therapies to specific people. Doctors can identify patients for whom certain medicines are ineffective because of their genetic profile, and instead choose options that will work better for that individual. The examination of the human genome and its variations allows researchers to better understand the disease-related genetic differences of diverse populations of people and then use that understanding to develop better medicines.

Biopharma researchers are also working on developing new mechanisms for treating disease. New "targeted" cancer drugs, for example, hold tremendous promise within the biotech industry. Chemotherapy drugs—the traditional treatment for cancer—target and destroy rapidly dividing cells. Unfortunately, these drugs often cause significant "collateral damage" because they are unable to differentiate between cancerous rapidly-dividing cells and normal rapidly-dividing cells. Chemotherapy can destroy healthy blood cells, hair follicles, and the cells lining the stomach and digestive tract, causing patients experience

debilitating side effects from these medications. Researchers are working hard to create drugs that will effectively eliminate cancerous cells but spare healthy tissues. Doctors are especially optimistic about the future of several recently developed immunotherapy drugs, which allow a patient's own immune system to fight their cancer. One such drug is a type of synthetic antibody, which is attracted only to proteins located on tumor cells. Once attached to a tumor cell, these antibodies release several proteins that both induce programmed cell death (apoptosis) and cause the cell to burst. The ability to selectively eliminate cancerous cells without damaging healthy cells would be an enormous step forward in treating cancer.

The field of *genetic engineering* (the process of altering the genetic material of cells or organisms to enable them to make new substances or perform new functions) that began in the 1970s has revolutionized medicine. With each passing year, the pace of discovery quickens and our understanding of the role of genetics in human health grows. Technology that allows us to quickly and efficiently edit DNA is being applied to the development of 42 new pharmaceuticals and is even being explored as a way to replace defective genes in human somatic cells—for example, to replace a defective gene that causes cystic fibrosis with a functional gene. Another recent advance allows researchers to reprogram adult cells into embryonic stem cells and then induce those cells to become any type of cell. These cells can be used to make model organs on which drugs can be tested outside of the human body. These technologies, and others that haven't yet been envisioned, are changing the future of medicine and providing dramatic improvement in human health and disease treatment.

DID YOU KNOW?

The DNA Code

DNA information is encoded by the arrangement of *nucleotides*, small molecules that join together to form the DNA molecule. A DNA molecule has millions of nucleotides. There are four different kinds of nucleotides, and they are arranged in a specific *sequence* (order). A specific sequence of nucleotides in the DNA (i.e., a gene) is a code for how to make a specific protein. Think of a sequence of nucleotides as similar to a sequence of written musical notes—the code for how to play music. Just as different sequences of notes encode different songs, different sequences of nucleotides encode different proteins.



THE TOOLS AND TECHNIQUES OF BIOTECHNOLOGY

For the next few days, you will explore the science of biotechnology and the tools used by scientists to create products. Your first task is to try out two of the tools used in biotechnology, the micropipette and gel electrophoresis.

When carrying out any scientific experimentation, you will find that accuracy and precision are important as is ensuring that you follow procedures carefully. Throughout your experience with ABE, your goal should be to learn about how and why the tools and techniques you're learning are used.

USING THIS STUDENT GUIDE

Icons are used throughout the Student Guide to draw attention to various aspects of the curriculum. The following is a list of those icons and their meanings.

lcon	Meaning
?	DID YOU KNOW? : Background information about concepts covered in the chapter.
	STOP AND THINK: Questions about the lab protocols.
	CONSIDER: Questions about important biological concepts.
	SAFETY: Reminders of key lab safety techniques.
	LAB TECHNIQUE: Useful lab techniques to improve efficiency and results.

PROGRAM INTRODUCTION GLOSSARY

Biomolecule: A molecule produced by living cells. Examples include proteins, carbohydrates, lipids, and nucleic acids.

Cells: The basic units of any living organism that carry on the biochemical processes of life.

DNA (deoxyribonucleic acid): A double-stranded biomolecule that encodes genetic information.

Enzyme: A protein that increases the rate of a chemical reaction.

Gene: The part of a DNA molecule that contains the instructions for making a particular protein.

Genetic engineering: A branch of biotechnology that uses specific procedures and techniques to change an organism's DNA.

Nucleotides: Small molecules that join together to form the DNA molecule.

Plasmid: A circular molecule of DNA.

Protein: A large biomolecule. Proteins carry out essential functions in cells, from forming cellular structures to enabling chemical reactions to take place.

Recombinant DNA: DNA that contains sequences or genes from two or more sources.

Restriction enzyme: A protein that can cut DNA at a specific sequence.

Sequence: A set of related events, movements, or items (such as nucleotides) that follow each other in a particular order.

Trait: A genetically determined characteristic. DNA codes for proteins, which determine traits.



CHAPTER 1 SOME TOOLS OF THE TRADE

INTRODUCTION

The year 1978 marked a major breakthrough in medicine. For the first time ever, scientists were able to engineer bacteria capable of producing human proteins. They achieved this by strategically inserting small pieces of human DNA into bacterial cells. This new technology, termed genetic engineering, can be used to make proteins that treat the symptoms of certain *genetic diseases* (those caused by a change in DNA, often inherited from parents). Genetic engineering, also called genetic modification, is the direct manipulation of an organism's genes using biotechnology.

To carry out genetic engineering, you need good laboratory skills. In this chapter, you'll focus on gaining practice in the use of *micropipettes* (instruments used to transfer small volumes of liquid) and *gel electrophoresis* (a technique for separating and identifying biomolecules)—two critical skills for biotechnology. You will complete two labs, using instruments and supplies that are identical to the ones used in biotechnology research laboratories. These labs are the first step in building the skills you'll need to be successful in biotechnology.

CHAPTER 1 GOALS

By the end of this chapter, you will be able to do the following:

- Correctly use micropipettes and the technique of gel electrophoresis
- Explain the importance of micropipettes and gel electrophoresis in genetic engineering
- Describe how gel electrophoresis separates DNA
- Explain how genetic engineering can be used to treat some genetic diseases

WHAT DO YOU ALREADY KNOW?

Discuss the following questions with your partner, and write your ideas in your notebook. Be prepared to discuss your responses with the class. Don't worry if you don't know all the answers. Discussing these questions will help you think about what you already know about biotechnology.

- 1. What tools and techniques of biotechnology have you used before? What did you use them for?
- 2. Why is precision important when you are carrying out biotechnology procedures?

LABORATORY 1.1: HOW TO USE A MICROPIPETTE

The purpose of this laboratory is to introduce you to an important tool used in genetic engineering: the micropipette, shown in **Figure 1.1**. A micropipette is used to transfer very small and exact volumes of liquids in either milliliters (mL, thousandths of a liter) or microliters (μ L, millionths of a liter), which are the measurements of volume most often used in genetic engineering. This laboratory will give you the chance to learn how to use the micropipette and to see the relative size of different amounts of solution measured by this very precise tool and how precise the amounts that you can measure with it are.

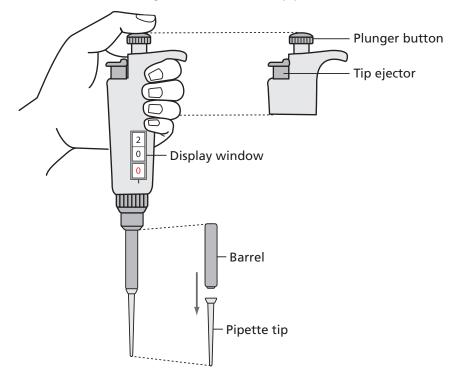


Figure 1.1: A P-20 micropipette

BEFORE THE LAB

Respond to the following with your group and be prepared to share your responses with the class.

- 1. Why do you think it is necessary to use very small and exact volumes of material in biotechnology?
- 2. Read through the *Methods* section on pages 17 through 19 and briefly outline the steps, using words and a flowchart.

MATERIALS

Reagents

• A plastic microfuge tube rack with a microfuge tube of red dye solution.

Equipment and Supplies

- P-20 micropipette (measures 2.0–20.0 μL)
- Tip box of disposable pipette tips
- Laminated micropipette practice sheet
- Waste container for used tips and microfuge tubes (will be shared among groups)

SAFETY:

- All appropriate safety precautions and attire required for a science laboratory should be used, including safety goggles. Please refer to your teacher's instructions.
- Wash your hands well with soap after completing the lab.

METHODS

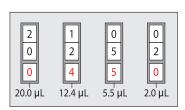
- 1. Check your rack to make sure that you have the reagent listed in *Materials*.
- 2. Review the parts of the micropipette shown in (see Figure 1.1 on page 16).
- 3. Find the display window on the handle of the micropipette.
- 4. Different micropipettors are adjusted in different ways. Many have a wheel that adjusts the volume. Others are adjusted by turning the plunger. In most cases, you turn right to increase the volume and left to decrease it.
- 5. **Figure 1.2** shows four micropipette volumes. Practice setting the micropipette to these volumes.

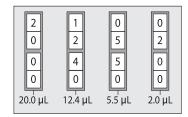
LAB TECHNIQUE: Never set the P-20 micropipette lower than 2.0 μ L or higher than 20.0 μ L or you could damage the equipment.





Figure 1.2: Four volumes shown on two different P-20 micropipettors





The display window of a micropipette shows how much fluid it will load and dispense. Four examples of displays and the corresponding amounts are shown.

- Review the laminated micropipette practice sheet. Each group member will
 pipette five drops of different volumes onto the sheet. Pipetting consists of
 two parts: loading the liquid into the micropipette, and dispensing the liquid
 from the micropipette.
- 7. Load the micropipette with 20.0 μ L of red dye (RD) by doing the following:
 - a. Set the micropipette to 20.0 μL.
 - b. Open the tip box. Lower the micropipette onto a tip and press down firmly (do not touch the tip with your fingers). Close the box when done.
 - c. Bring the micropipette and the RD tube to eye level.
 - d. Use your thumb to press the plunger to the first stop position, which is your first point of resistance.

LAB TECHNIQUE: When loading the micropipette, only press the plunger to the first stop or you will draw too much solution into the pipette tip.

e. Put your pipette tip into the RD and slowly release the plunger to draw up the solution.

LAB TECHNIQUE: Do not lay down a micropipette with fluid in the tip or hold it with the tip pointed upward. If the disposable tip is not firmly seated onto the barrel, fluid could leak back into the pipette.

- 8. Dispense RD onto the laminated sheet by doing the following:
 - a. Place the pipette tip over the 20.0 μL circle.
 - b. Use your thumb to press the plunger to the first stop position and then press down to the second stop.

LAB TECHNIQUE: When dispensing liquid from the micropipette, press the plunger to the first stop to dispense most of the liquid and then press the plunger to the second stop in order to dispense the last little bit.

c. With the plunger still depressed, pull the pipette away from the paper—this prevents you from accidentally pulling the liquid back into the tip.









- 9. Without setting down the micropipette, twist the plunger button to set it to 15.0 μ L and repeat steps 7b–8c, dispensing over the 15.0 μ L circle.
- 10. Without setting down the micropipette, set it to 10.0 μ L and repeat steps 7b–8c, dispensing it over the 10.0 μ L circle when dispensing the liquid.
- 11. Without setting down the micropipette, set it to 5.0 μ L and repeat steps 7b–8c, dispensing it over the 5.0 μ L circle.
- 12. Without setting down the micropipette, set it to 2.0 μ L and repeat steps 7b–8c, dispensing it over the 2.0 μ L circle.
- 13. Use the tip ejector to place your pipette tip into the waste container.

STOP AND THINK:

- When loading or dispensing a solution, why is it important to actually see the solution enter or leave the pipette tip?
- You were instructed to avoid contact with the pipette tips—for example, you were asked to put the pipette tip on without using your hands, to avoid setting down the micropipette, to use the ejector button to remove the tip, and to keep the tip box closed. If you were working with plasmids and bacterial cells, why would these precautions be important?
- 14. Using the micropipette practice sheet, each person in your group should have a chance to load and dispense the five drops of different volumes, with each person using a new pipette tip.
- 15. When everyone in your group has had a chance to dispense RD onto the micropipette practice sheet, draw the approximate sizes of each drop in your notebook (or take a photograph and tape it into your notebook) and label them with the amounts.



LABORATORY 1.2: GEL ELECTROPHORESIS

The purpose of this laboratory is to give you experience with gel electrophoresis, which is used to separate and identify a mixture of biomolecules including DNA; the size of the components of each mixture can then be identified by their location in the gel. Biomolecules are too small to see, and estimating their exact size is very difficult. Gel electrophoresis allows scientists to easily visualize information and compare various biomolecules. Gel electrophoresis works based on the fact that many biomolecules have a negative charge, which means that they will move in response to an electric charge. The biomolecules move through a gel, and the distance they travel varies primarily according to their size, although molecular shape and degree of charge also influence their movement.

The electrophoresis setup consists of a box containing an agarose gel and two electrodes that create an electric field across the gel when the box is attached to a power supply. The negative electrode is black, and the positive electrode is red. *Wells* are depressions in the agarose that can hold small volumes of samples. The molecules in a sample will travel through the gel and be sorted based on their properties of charge and size. Samples of negatively-charged biomolecules are pipetted into wells near the negative (black) electrode. The samples move through the gel toward the positive (red) electrode, as shown in **Figure 1.3**, because opposite charges attract and like charges repel.

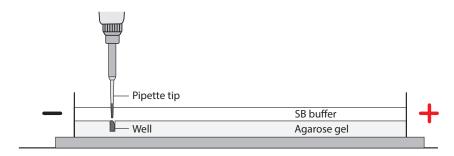


Figure 1.3: The gel electrophoresis unit

The gel that the biomolecules move through is composed of *agarose*, a polysaccharide (complex sugar) found in seaweed. Its structure is a porous matrix (like a sponge) with lots of holes through which the solution and biomolecules flow. **See Figure 1.4**.

Porous gel

Porous gel

Porous gel

Figure 1.4: How biomolecules, including DNA, move through the agarose gel matrix in gel electrophoresis

BEFORE THE LAB

Respond to the following with your group, and be prepared to share your responses with the class.

- 1. In what circumstances might it be important to use gel electrophoresis to separate and identify plasmids and short linear pieces of DNA?
- 2. Read through the *Methods* section on pages 22 through 25 and briefly outline the steps for Part A and for Part B, using words and a flowchart.

MATERIALS

Reagents

- A plastic microfuge tube rack with the following:
 - Microfuge tube of red dye solution
 - Microfuge tube of dye solution 1 (S1)
 - Microfuge tube of dye solution 2 (S2)
 - Microfuge tube of dye solution 3 (S3)
- 50-mL flask containing 1x sodium borate buffer (1x SB) (shared with another group)

Equipment and Supplies

- P-20 micropipette (measures 2.0–20.0 μL)
- Tip box of disposable pipette tips
- 2 pipetting practice plates loaded with 0.8% agarose gel



- Electrophoresis box loaded with 0.8% agarose gel (will be shared among groups)
- Microcentrifuge (will be shared among all groups)
- Waste container for used tips and microfuge tubes (will be shared among groups)

SAFETY:

- All appropriate safety precautions and attire required for a science laboratory should be used, including safety goggles. Please refer to your teacher's instructions.
- Wash your hands well with soap after completing the lab.

METHODS

PART A: PIPETTING INTO WELLS

You will practice pipetting RD into preformed wells in an agarose gel.

- 1. Check your rack to make sure that you have the red dye (RD) tube.
- 2. Fill the two pipetting practice plates with 1x SB buffer to a level that just covers the entire surface of the gel. If you see any "dimples" over the wells, add more *buffer* (a solution that can maintain a nearly constant pH. In gel electrophoresis, it prevents the gel's pH from changing due to the electrical current).
- 3. Set the P-20 micropipette to 10.0 µL and put on a pipette tip.
- 4. Draw up 10.0 μL of RD in to the pipette.

LAB TECHNIQUE: Do not lay down a micropipette with fluid in the tip or hold it with the tip pointed upward.

- 5. Dispense RD into a well in one of the practice plates by doing the following:
 - a. Place your elbow on the table to steady your pipette hand. If needed,
 also use your other hand to support your

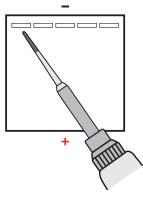
pipette hand.

b. Lower the pipette tip until it is under the buffer but just above the well.

LAB TECHNIQUE: Be careful not to place your pipette tip into the well or you might puncture the gel, which will make the well unusable.

c. Gently press the plunger to slowly dispense the sample. To avoid getting air into the buffer, do not go past the first stop. The sample will sink into the well.







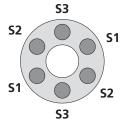
- Repeat steps 4 and 5 until all the practice plate wells have been filled.
 Everyone in your group should get an opportunity to practice pipetting into the wells.
- 7. Eject the pipette tip.

PART B: SEPARATING DYES WITH GEL ELECTROPHORESIS

Now you will use gel electrophoresis to separate different dyes. First you will add dyes into wells in the gel electrophoresis unit. You will then turn the unit on in order to move the negatively charged dyes through the gel. (You will share the electrophoresis boxes with one other group; your teacher will tell you which wells your group should use.)

- 1. Check your rack to make sure that you have the three dye solutions (\$1, \$2, and \$3).
- 2. Review **Figure 1.4** on page 21. Check to make sure that the wells in the gel are located near the negative (black) electrode.
- 3. Fill the box with 1x SB buffer to a level that just covers the entire surface of the gel. If you see any "dimples" over the wells, add more buffer.
- 4. Centrifuge the S1, S2, and S3 tubes.

LAB TECHNIQUE: Distribute the tubes evenly in the microcentrifuge so that their weight is balanced.



- 5. Make a drawing in your notebook that shows the location of the wells in the electrophoresis box. Record which solution you will place in each well.
- 6. Set the P-20 micropipette to 10.0 μ L and put on a pipette tip.
- 7. Draw up $10.0 \mu L$ of S1 into the pipette.
- 8. Dispense the S1 into the well you've designated for that solution by doing the following:
 - a. Place your elbow on the table to steady your pipette hand. If needed, also use your other hand to support your pipette hand.
 - b. Lower the pipette tip until it is under the buffer but just above the well.

LAB TECHNIQUE: Do not puncture the gel with the pipette tip or it will become unusable; the sample will sink into the hole below the gel instead of moving through the gel.





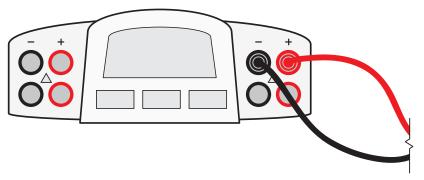


c. Gently press the plunger to slowly dispense the sample. To avoid getting air into the buffer, do not go past the first stop. The sample will sink into the well.

LAB TECHNIQUE:

- While the plunger is still depressed, pull the tip out of the buffer so that you don't draw up the solution or buffer.
- Use a fresh pipette tip for each sample.
- 9. Using a new pipette tip with each solution, repeat steps 7 and 8 for S2 and S3.
- 10. When all the samples have been loaded, close the cover tightly over the electrophoresis box. (Carefully close the cover so that samples don't spill.)
- 11. Connect the electrical leads to the power supply. Connect both leads to the same channel, with cathode (–) to cathode (black to black) and anode (+) to anode (red to red). See Figure 1.5

Figure 1.5: Leads from electrophoresis box connected to correct channel in power supply



- 12. Turn on the power supply and set the voltage to 130–135 V. (You will see bubbles form in the buffer at the red [+] end of the electrophoresis unit.)
- 13. After two or three minutes, check to see if the dyes are moving toward the positive (red) electrode. You should begin to see the purple dye (bromophenol blue) beginning to separate from the blue dye (xylene cyanole).



STOP AND THINK:

- Study your gel electrophoresis results. Which solution sample contained a single dye: S1, S2, or S3? How do you know?
- What electrical charge do the dyes have? Explain your reasoning.
- The dyes that you are separating are orange G (yellow), bromophenol blue (purple), and xylene cyanole (blue). If the molecular shape and electric charge of all three dyes are similar, what is the order of the dyes from heaviest to lightest molecules, based on your initial results? Why do you think this is the correct order?

- 14. In approximately 10 minutes, or when you can distinguish all three dyes, turn off the power switch and unplug the electrodes from the power supply. Do this by grasping the electrode at the plastic plug, NOT the cord.
- 15. Carefully remove the cover from the gel box, and observe the dyes in the gel.
- 16. In your notebook, draw the relative location of the bands and their colors in each of the lanes containing your samples.
- 17. Leave the gels in the gel box.

CHAPTER 1 QUESTIONS

- 1. Why would it be beneficial to use a micropipette to measure reagents in biotechnology rather than another measuring instrument?
- 2. What do the results of gel electrophoresis tell you about genetic material?



DID YOU KNOW?

Gel Electrophoresis in DNA Fingerprinting

DNA fingerprinting uses gel electrophoresis to distinguish between samples of genetic material. In DNA fingerprinting, human DNA molecules are treated with enzymes that chop them at certain characteristic points, thereby reducing the DNA to a collection of smaller and more manageable pieces. The DNA fragments are loaded into a gel and placed in an electrical field, which sorts the DNA fragments into various bands. These bands can be colored with a radioactive dye to make them visible to imaging techniques. Methods of DNA identification have been applied to many branches of science and technology, including medicine (prenatal tests, genetic screening), conservation biology (guiding captive breeding programs for endangered species), and forensic science. In the latter discipline, analysis of the pattern of DNA fragments that results from the action of restriction enzymes enables us to discriminate between suspects accused of a crime or between potential fathers in a paternity suit.

CHAPTER 1 GLOSSARY

Agarose: A polymer made up of sugar molecules that is used as the matrix in gel electrophoresis procedures.

Buffer: A solution that can maintain a nearly constant pH. In gel electrophoresis, it prevents the gel's pH from changing due to the electrical current.

Gel electrophoresis: The movement of charged molecules toward an electrode of the opposite charge; used to separate biomolecules. When used to separate DNA fragments, electrophoresis will separate the fragments by size, with smaller fragments moving faster than larger fragments.

Genetic disease: Those diseases caused by a change in DNA. Genetic diseases are often inherited from parents.

Micropipette: A laboratory instrument used to measure, dispense, and transfer very small amounts of liquid.

Well: A depression in the agarose that can hold small volumes of samples.