Build a Gel Electrophoresis Apparatus

Lab III-6

EQUIPMENT AND MATERIALS

You'll need the following items to complete this lab session. (The standard kit for this book, available from *www.thehomescientist.com*, includes the items listed in the first group.)

MATERIALS FROM KIT

- Leads, alligator clip (2)
- Ruler

MATERIALS YOU PROVIDE

- Aluminum foil
- Batteries, 9V transistor (5, 7, or 9)
- Gel casting comb materials (see text)
- Marking pen (Sharpie or similar)
- Plastic containers (see text)
- Scissors
- Tape (electrical or masking)

BACKGROUND

There's a time-honored custom in science: we scientists build our own apparatus. Not always, of course, nor even most of the time. Nowadays, anyway. Sometimes it's cheaper, easier, and faster just to buy what we need. But when we need something to complete an experiment and that something isn't available or we don't have the budget to buy it, we make do. If that involves designing and building a piece of equipment, so be it. For the following lab session, we needed a gel electrophoresis apparatus and power supply, and we didn't feel like spending $300 or $400 to buy one. So we designed and built our own, at a total cost of about $10.

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Like paper chromatography, gel electrophoresis is used to separate mixtures of chemical compounds. (In biology, gel electrophoresis is often used to separate proteins or DNA fragments.) The similarities do not end there. In paper chromatography, the fixed phase (matrix) is a strip of paper; in gel electrophoresis, the matrix is a bed or column of gel. In both cases, the matrices provide resistance to the movement of molecules. In paper chromatography, capillary action (wicking) provides the force required to move the mobile phase (solvent) and analyte(s) through the matrix; in gel electrophoresis, the force is supplied by an external electrical current, with positively charged molecules attracted to the negative electrode and vice versa.

Although gel electrophoresis is conceptually simple, commercial gel electrophoresis setups sell for $300 and up, and the gels, buffers, and other supplies they require are not cheap. If you can afford a commercial apparatus and supplies, there's no question you'll find it easier and faster to run gels. However, most homeschoolers and hobbyists can't justify spending several hundred dollars on a gel electrophoresis apparatus and supplies. (Check with your homeschool group or co-op, which may have such an apparatus available to borrow.) Fortunately, with a few minutes' work, you can assemble your own apparatus and supplies for only a few dollars, using items from the kit and some common household items. Here's what you'll need:

Shallow plastic container

You'll need a shallow, easily cut plastic container to serve as the gel casting bed. We used the base of a one-quart Gladware container, trimmed to about 1 cm tall. You can also use a soap dish or similar container. If you want to cast multiple gels in one pass, make several of these gel bed containers.

Deeper container

You'll need another plastic container that's larger and deeper than the gel bed. We used a sandwich container, but you can substitute any suitable container. This container needs to be at least as long and wide as the gel bed container—although, ideally, not all that much longer and wider—and at least a centimeter or so taller.

Tape

We'll cut the ends of the gel bed container off, but we need to be able to seal them temporarily back in place while casting gels. We used plastic electrical tape, but you can substitute ordinary masking tape. Once the gel solidifies, you simply peel off the tape to expose both ends of the gel. Tape may also be useful in making the comb that's used to produce wells in the gel.

Comb

No, not the kind you use on your hair. A gel electrophoresis comb is a comb-like assembly with teeth that are placed in the gel casting container to produce small wells in the gel as it solidifies. You can buy a commercial comb for an outrageous price, or you can improvise your own with common household materials.

We've made combs from (a) cardboard and aluminum foil or disposable coffee stirrers, (b) wooden paint stirrers and nails or disposable chopsticks, and (c) Lego blocks. The only important things are that the prongs be relatively smooth (so as not to tear up the solidified gel when they're removed), roughly 3 mm to 4 mm in diameter (or the equivalent area if the teeth are square or rectangular), long enough to reach the bottom of the gel casting container (or nearly so), and spaced at intervals of roughly 1 cm across the width of the gel casting container. Use your imagination and materials at hand.

Aluminum foil

The apparatus requires an electrode at each end of the larger container. We'll use two pieces of aluminum foil to make these electrodes. You may also use the aluminum foil to make the teeth in the comb used to produce wells in the gel.

9V transistor batteries

The apparatus requires a DC power supply, which can be anything from 30 or 40 VDC to 150 VDC or more. The lower the voltage, or the larger the gels, the longer they take to run.

Five 9V transistor batteries connected in series provide 45 VDC, which is enough to run small gels in a reasonable time. Using seven 9V batteries for 63 VDC or nine 9V batteries for 81 VDC lets you run gels faster, or larger gels in the same amount of time.

Why the odd numbers? Because we'll connect the batteries in series by snapping the positive connector on one battery to the negative connector on the next battery, as shown in Figure III-6-1.

For reasonably fast run times, use at least 3V per centimeter of gel length. For example, if your gel bed is 15 cm long, your power supply should be at least 45 VDC. (With these parameters, running a gel may require two or three hours or more, depending on the type and concentration of the gel.)

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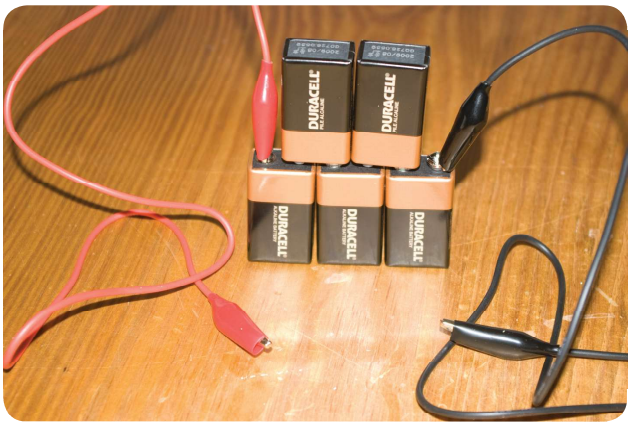
WARNING

Be careful with 9V batteries connected in series. Depending on your skin resistance and other factors, even 45 VDC may produce a painful and potentially dangerous electrical shock. **Never** touch the power leads of the battery assembly or touch the liquid in the gel electrophoresis apparatus while power is connected to it.

Never close the circuit (that is, once they are connected to the battery stack, never allow the free ends of the positive (red) and negative (black) leads to contact each other. That short circuit can rapidly weld the connection and cause the batteries to overheat and explode. (It's safe to connect the positive terminal of one battery to the negative terminal of the next because the circuit has not been closed unless you allow the final open positive and negative terminals to contact each other, such as by allowing the free end of the connected red lead to contact the free end of the connected black lead.)

If you run many gels, the cost of 9V batteries starts to add up fast, so it's cheaper in the long run to buy a DC power supply. For smaller gels, any DC power supply that provides fixed-, multiple-, or variable-output DC voltages in the range of 50 to 70 VDC or higher at a few hundred mA will suffice. We've seen suitable single-voltage units on eBay for $30 to $90, depending on voltage.

Figure III-6-1: *A 9V transistor battery stack*



PROCEDURE III-6-1: MAKING THE GEL CASTING CONTAINER AND COMB

1. To begin, place the marking pen flat on a table and measure the distance between the table surface and the point of the pen. If it is less than 1 cm, place magazines or other spacers between the table surface and the pen until the tip of the pen is about 1 cm above the table surface.

2. Holding the pen firmly in place, place the smaller plastic container flat of the table, and bring its surface into contact with the tip of the pen. Move the plastic container to draw a line around its exterior that is 1 cm above its bottom, as shown in Figure III-6-2.

Figure III-6-2: *Marking the gel casting tray for depth*

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3. Use scissors to trim the excess from the plastic container, converting it to a shallow plastic tray.

4. Use the scissors to remove most of both ends of the tray, as shown in Figure III-6-3. Retain these cut ends for future use.

Figure III-6-3: *Removing the ends from the gel casting tray*

5. Hold the ends of the tray in position, and tape them securely back in place, as shown in Figure III-6-4. These taped ends will hold the liquid gel in the tray until it's solidified. Before running the gel, you'll remove the ends to allow current to flow through the gel.

Figure III-6-4: *Taping the ends of the gel casting tray into place*

Place the gel tray aside for now. If you want to cast multiple gels in one pass, make several of these trays before continuing. The next step is to make the comb. Here's one way to do it (but feel free to experiment with other methods...).

6. Cut a strip of stiff cardboard about 3 cm wide and 2 cm longer than the width of your gel tray. Draw a centered line the length of the strip. Place tick marks on that line, beginning 1.5 cm from each end of the strip and then evenly spaced at about 1 to 1.5 cm intervals across the width of the gel bed container.

7. Twist pieces of aluminum foil to form stiff cylinders about 2.5 cm long by 3 mm in diameter. (The number you need varies by the width of your gel tray; make one of these cylinders for each of the tick marks across the tray width.) Try to make the surface of the teeth as smooth as possible.

8. Make a 90° bend in each cylinder 1 cm from one end, forming an "L" shape, with one leg 1 cm long and the other 1.5 cm long.

9. Place each cylinder with the bend on a tick mark, and press the longer leg flat against the cardboard. Securely tape the longer leg of each cylinder to the cardboard, with the 1 cm long leg projecting vertically from the surface of the cardboard.

Calculate the volume of the tray by measuring and multiplying its three dimensions. For example, if your tray is 7 cm wide by 10 cm long by 1 cm deep, its total volume is (7 * 10 * 1) = 70 cm^3, which is the same as 70 mL. When you cast gels, you'll fill the tray to about 0.6 cm (6 mm) to 0.8 cm deep, so each gel tray requires something between 42 mL (0.6 * 70 mL) and 56 mL of gel.

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PROCEDURE III-6-2: ASSEMBLE THE APPARATUS

1. Fold aluminum foil over each end of the larger container and connect one end of an alligator clip lead to each of the foil electrodes. (Don't connect the other ends of the alligator clip leads to the battery stack until you're actually ready to run a gel.)

2. Place the gel bed container inside the larger container, centered within the larger container. Verify that there is at least a few millimeters of clearance between each end of the gel bed container and the corresponding electrode on the larger container. Also verify that all sides of the larger container are a centimeter or so higher than the sides of the gel bed container.

At this point, you're ready to cast and run gels, which you'll do in the next lab. Incidentally, believe it or not, the apparatus you've just built is fully capable of running gels just as good as those produced by expensive professional apparatus. It's clumsier to use, certainly, but if you use the same agarose and stains that are used on professional apparatus, the resulting gels will be indistinguishable.

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