

GETTING STARTED IN THE LAB

Introduction to Laboratory Equipment

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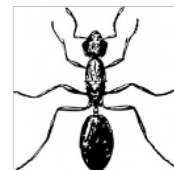
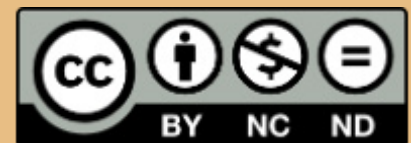


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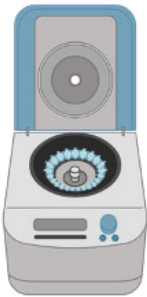
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Micropipettes are commonly used in laboratories to accurately measure and transfer small volumes of liquid.



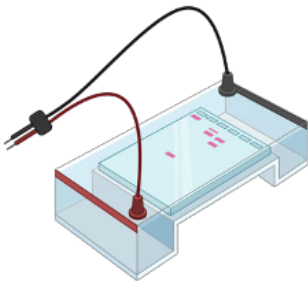
A **Vortexer** is an electric motor-powered device that makes quick, circular motions to thoroughly mix small vials or tubes of liquids.



A **Centrifuge** utilizes centrifugal force to spin vials or tubes at high speeds. It is often used to separate the contents of a solution based on density.



A **Thermal Cycler** heats tubes to specific temperatures in order to carry out PCR via a three-step process: denaturation, annealing, and extension.



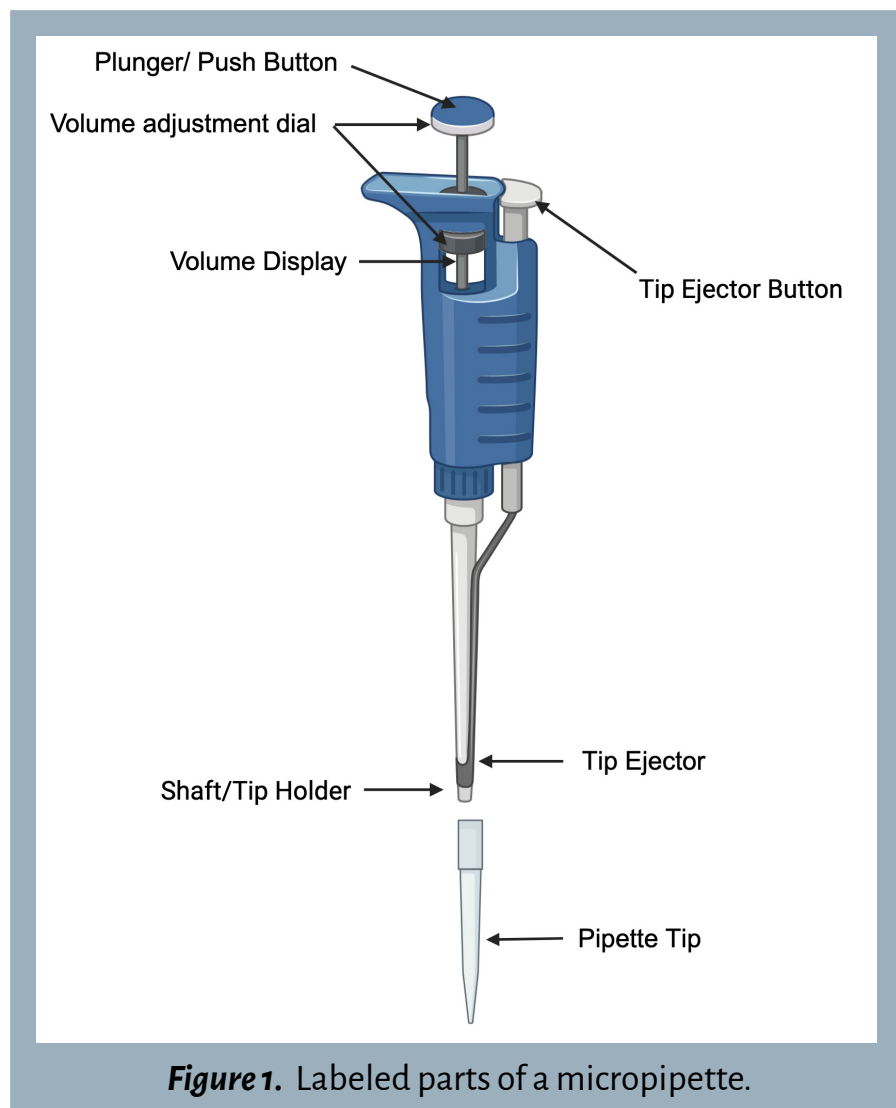
A **Gel Electrophoresis System** separates substances, such as DNA, based on the rate of movement while under the influence of an electric field.

Introduction

Pipettes are commonly used in laboratories to accurately measure and transfer a desired volume of liquid. The prefix micro- comes from the Greek word for “small” and is abbreviated as “ μ ”. A **micropipette** is different from a regular pipette in that it specifically measures and transfers small volumes of liquid from 0.1 μl up to 1,000 μl (or 1 ml). In this lab series, we will use the term “micropipette” interchangeably with “pipette” to refer to this device.

1,000 microliters (μl) = 1 milliliter (ml)

Before jumping into the lab exercises, familiarize yourself with the parts of a micropipette and review its proper use. **Figure 1** labels the parts of a common micropipette. Some pipettes may differ slightly from this illustration.



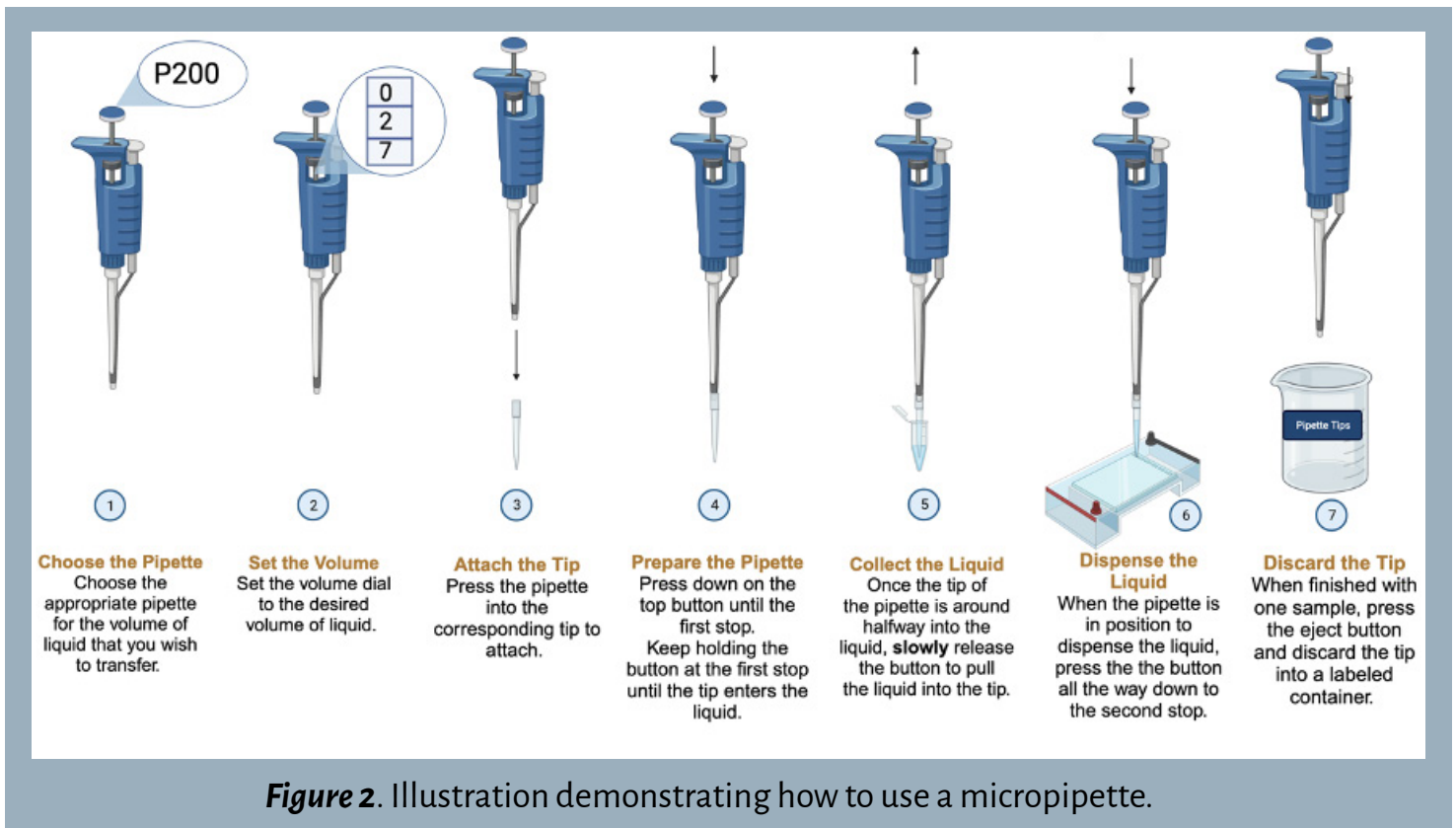
Getting Started

STEP 1:

Choose the pipette. After familiarizing yourself with the various parts of a micropipette, determine which pipette is best suited for your experimental needs. There are typically four options within a laboratory: P2, P20, P200, or P1000. Each number refers to the maximum volume of liquid - in microliters (μl) - that the pipette can measure. The volume range is printed on the side of the pipette and/or on the plunger button.

P2 = 0.1-2 μl
 P20 = 2-20 μl
 P200 = 20-200 μl
 P1000 = 100-1000 μl

The most accurate measurement is made using the smallest pipette within the recommended range. For example, the P20 pipet would be the best option to measure 13.5 μl of liquid because it is more than 2 μl but less than 20 μl .

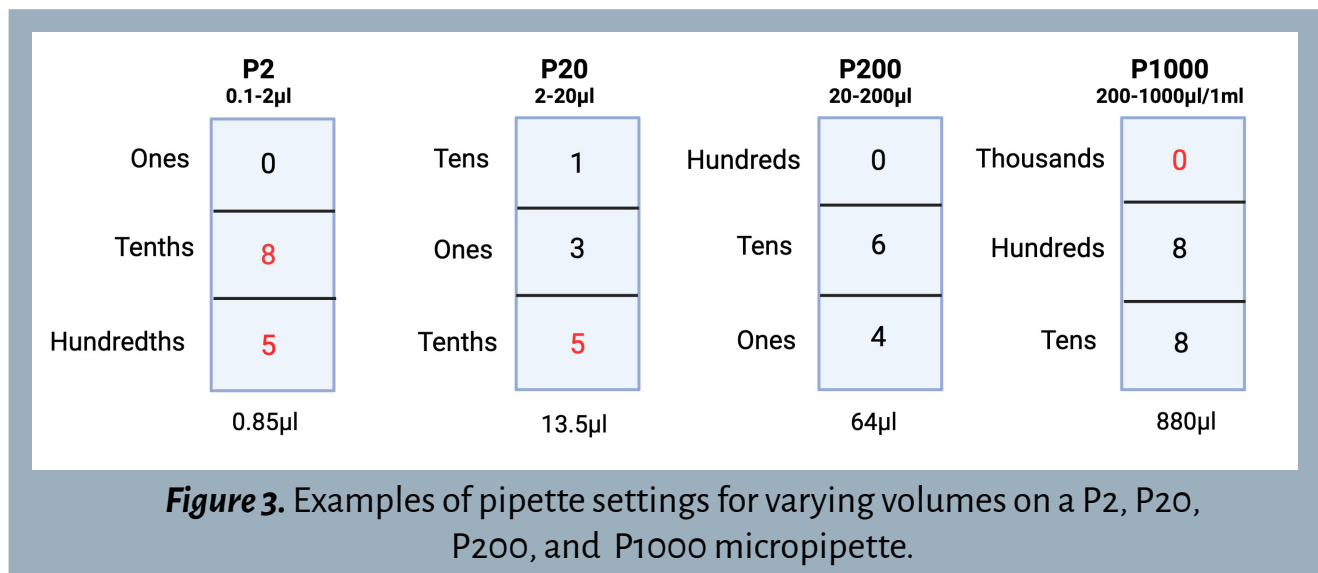


STEP 2:

Set the volume. It's important to understand how to set the volume dial correctly. Each pipette has a three-digit volume window to accommodate the largest indicated volume (see **Figure 3**).

- **P2:** The largest volume is 2 μl . The top digit represents the ones position, the middle digit represents tenths, and the bottom digit represents hundredths.
- **P20:** The largest volume is 20 μl . The top digit represents the tens position, the middle digit represents ones, and the bottom digit represents tenths.
- **P200:** The largest volume is 200 μl . The top digit represents the hundreds position, the middle digit represents tens, and the bottom digit represents ones.
- **P1000:** The largest volume is 1000 μl . The top digit represents the thousands position, the middle digit represents hundreds, and the bottom digit represents tens.

The number on the display is adjusted by turning the volume adjustment dial in either direction. Many pipettes have a lock located below the plunger button that must be switched to the unlock position while changing the number. Ensure that it is locked when pipetting.

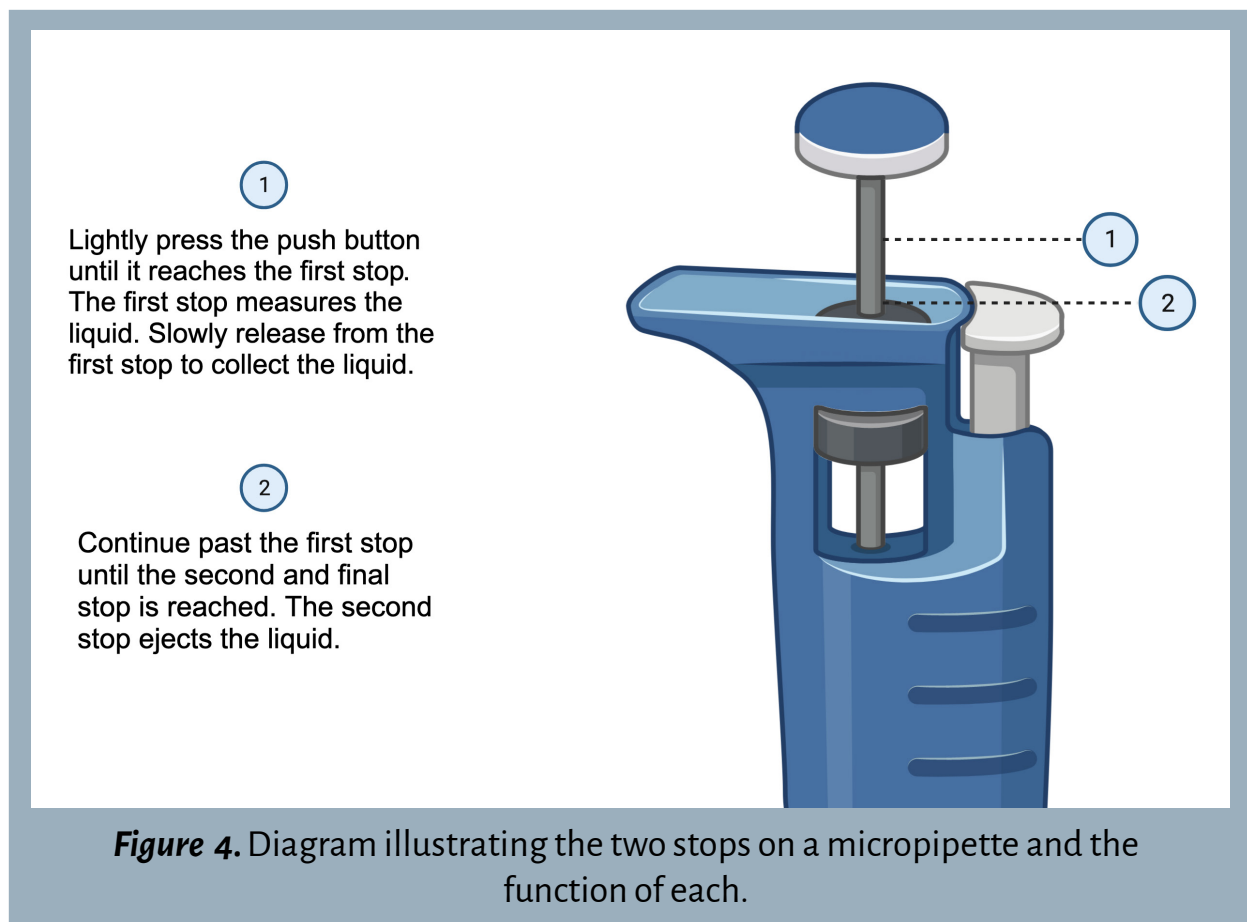


STEP 3:

Attach the tip. Each pipette requires a corresponding box of tips with the same volume range, as listed on the box. While most pipettes are compatible with universal tips, some brands require specialized tips. Once the correct tips have been selected, open the lid and press the end of the pipette into one of the tips. Press firmly enough to make a tight connection without breaking the plastic.

STEP 4:

Prepare the pipette. Press the plunger button down until there is resistance – this is the first stop out of two (**Figure 4**). Continue holding the button down at the first stop and insert the pipette into the liquid.



STEP 5:

Collect the liquid. Once the tip is about halfway into the liquid, slowly release the plunger button to pull the liquid into the pipette tip. Be sure that the tip is always submerged when pulling in the liquid. To avoid cross-contamination of samples, do not allow any other parts of the pipette to make contact with the liquid.

STEP 6:

Dispense the liquid. When ready to release the liquid, slowly press down the plunger button to the second and final stop (**Figure 4**).

STEP 7:

Discard the tip. After the liquid has been transferred, press the tip ejector button (see **Figure 1**) and dispose of the tip in a labeled waste cup. It's important to use a new tip for each sample to avoid cross-contamination. Later, the tips should be disposed of in a designated waste disposal area.

Helpful Tips

- Try to keep the lid to the box of tips closed when not in use. This will prevent contamination of the tips for future use.
- If in doubt that the correct volume was transferred, get a new tube, and repeat the steps. If this is not possible, make a note of the potential mistake in your lab notebook.
- It is normal to occasionally have an air bubble in the pipette tip. However, this may affect the accuracy of the measurement. If an air bubble is present inside the pipette tip, empty all liquid back into the container and try again.
- Pipetting too quickly can result in the liquid jumping into the top of the tip and may contaminate the micropipette shaft.

Practice Worksheet

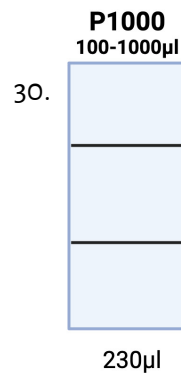
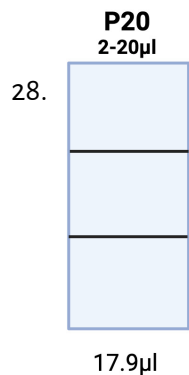
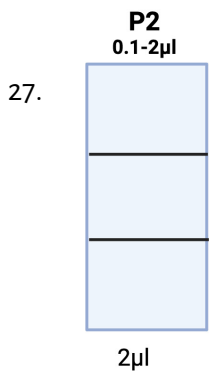
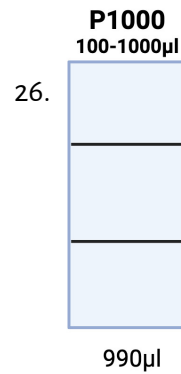
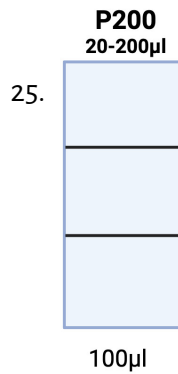
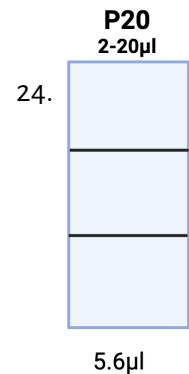
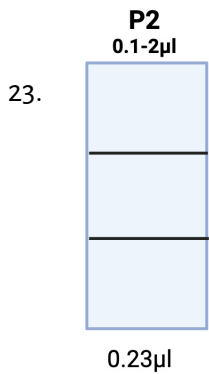
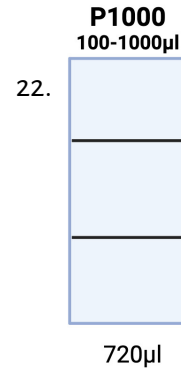
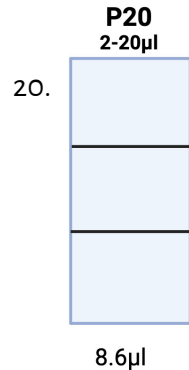
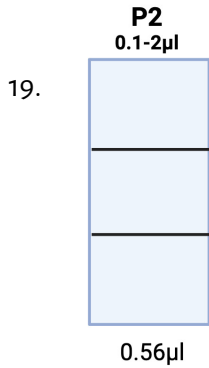
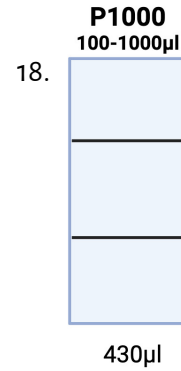
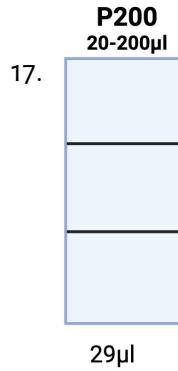
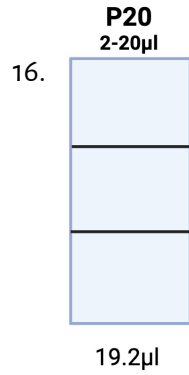
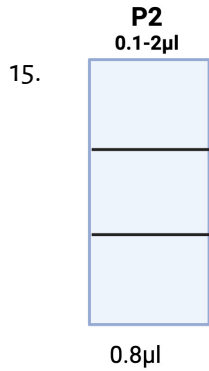
Choose the best pipette option to accurately transfer each volume of liquid.

- | | |
|---|---|
| <p>1. 835 μl</p> <p>a. P2</p> <p>b. P20</p> <p>c. P200</p> <p>d. P1000</p> | <p>4. 187 μl</p> <p>a. P2</p> <p>b. P20</p> <p>c. P200</p> <p>d. P1000</p> |
| <p>2. 35.5 μl</p> <p>a. P2</p> <p>b. P20</p> <p>c. P200</p> <p>d. P1000</p> | <p>5. 1000 μl</p> <p>a. P2</p> <p>b. P20</p> <p>c. P200</p> <p>d. P1000</p> |
| <p>3. 12 μl</p> <p>a. P2</p> <p>b. P20</p> <p>c. P200</p> <p>d. P1000</p> | <p>6. 2.45 μl</p> <p>a. P2</p> <p>b. P20</p> <p>c. P200</p> <p>d. P1000</p> |

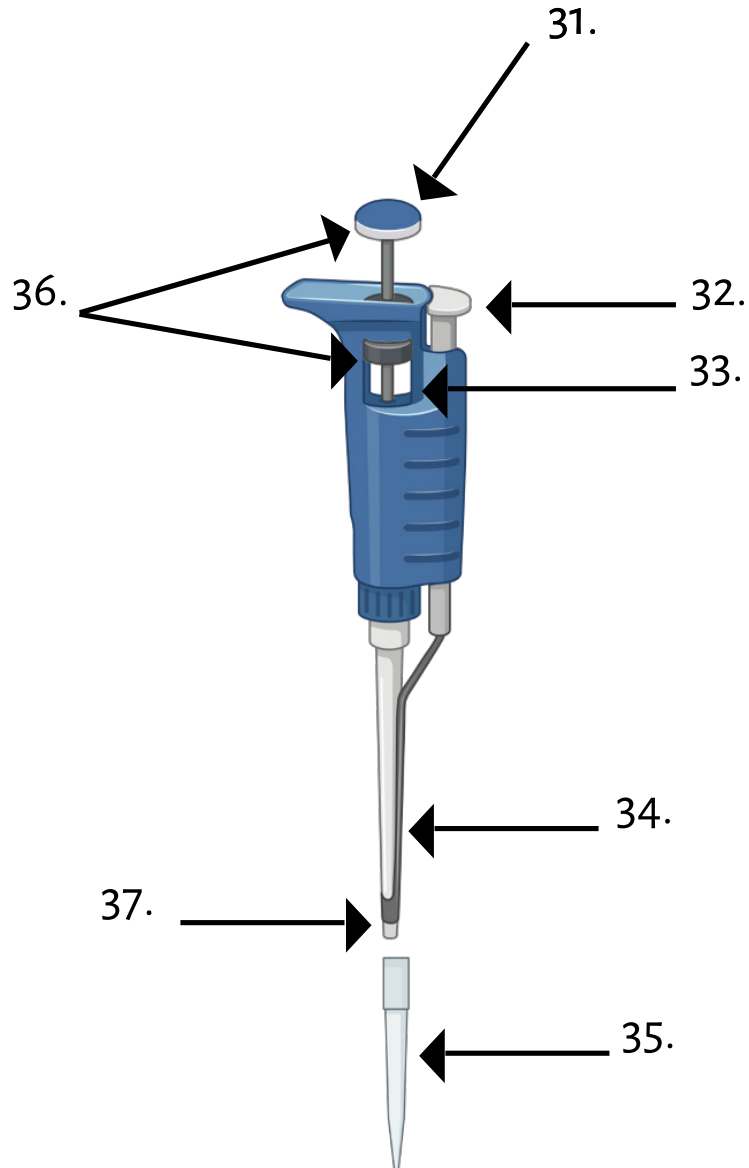
The rectangles below represent the volume dial on the indicated micropipette. Fill in the blanks with the volume that is displayed on each dial in microliters.

<p>7.</p> <p>P2 0.1-2μl</p> <table border="1" style="border-collapse: collapse; width: 60px; height: 100px; text-align: center;"> <tr><td>0</td></tr> <tr><td>6</td></tr> <tr><td>3</td></tr> </table> <p>_____ μl</p>	0	6	3	<p>8.</p> <p>P20 2-20μl</p> <table border="1" style="border-collapse: collapse; width: 60px; height: 100px; text-align: center;"> <tr><td>1</td></tr> <tr><td>8</td></tr> <tr><td>0</td></tr> </table> <p>_____ μl</p>	1	8	0	<p>9.</p> <p>P200 20-200μl</p> <table border="1" style="border-collapse: collapse; width: 60px; height: 100px; text-align: center;"> <tr><td>0</td></tr> <tr><td>2</td></tr> <tr><td>7</td></tr> </table> <p>_____ μl</p>	0	2	7	<p>10.</p> <p>P1000 100-1000μl</p> <table border="1" style="border-collapse: collapse; width: 60px; height: 100px; text-align: center;"> <tr><td>0</td></tr> <tr><td>4</td></tr> <tr><td>6</td></tr> </table> <p>_____ μl</p>	0	4	6
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3															
1															
8															
0															
0															
2															
7															
0															
4															
6															
<p>11.</p> <p>P2 0.1-2μl</p> <table border="1" style="border-collapse: collapse; width: 60px; height: 100px; text-align: center;"> <tr><td>1</td></tr> <tr><td>2</td></tr> <tr><td>5</td></tr> </table> <p>_____ μl</p>	1	2	5	<p>12.</p> <p>P20 2-20μl</p> <table border="1" style="border-collapse: collapse; width: 60px; height: 100px; text-align: center;"> <tr><td>0</td></tr> <tr><td>3</td></tr> <tr><td>9</td></tr> </table> <p>_____ μl</p>	0	3	9	<p>13.</p> <p>P200 20-200μl</p> <table border="1" style="border-collapse: collapse; width: 60px; height: 100px; text-align: center;"> <tr><td>1</td></tr> <tr><td>2</td></tr> <tr><td>4</td></tr> </table> <p>_____ μl</p>	1	2	4	<p>14.</p> <p>P1000 100-1000μl</p> <table border="1" style="border-collapse: collapse; width: 60px; height: 100px; text-align: center;"> <tr><td>1</td></tr> <tr><td>0</td></tr> <tr><td>0</td></tr> </table> <p>_____ μl</p>	1	0	0
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9															
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4															
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The rectangles below represent the volume dial on the indicated micropipette. Fill in the blanks on the volume dials with the corresponding volume of liquid in microliters.



Use the word bank to label the parts of the micropipette.



Word Bank

Plunger/Push Button
Volume Adjustment Dial
Volume Display
Tip Ejector Button

Tip Ejector
Shaft/Tip Holder
Pipette Tip

Lab Activity

Materials Needed

- 1.5 mL microcentrifuge tube
- Tube Rack
- Water
- Food coloring
- Parafilm or wax paper
- p2, p20, p200, and p1000 micropipettes
- Pippete tips
- Waste cup
- Ruler

Classroom Preparation:

- Review the Getting Started section on how to properly use a micropipette.
- Make a stock solution of water mixed with a few drops of food coloring. Aliquot at least 1 ml of the colored water into each microcentrifuge tube.
- Provide each lab station with at least one tube of colored water and a few squares of Parafilm, or wax paper.

Lab Activity:

Complete each step and record observations below. Change out tips – and possibly micropipettes – between each step.

STEP 1:

Place a piece of Parafilm, along with the paper overlay, wax side down on the bench. Use a blunt object, such as a coin or tip of a pen, to gently rub lines across the Parafilm until it is loosely attached to the bench. Remove the paper overlay. If using wax paper, cut out a flat piece of paper and use lab tape to secure to bench top.

STEP 2:

Transfer 2 ul of the liquid onto the Parafilm. Use a ruler to measure the size of the droplet and shade in the pipet tip to indicate relative volume.



Micropipette used: _____

Tip size used: _____

Diameter of the 2 ul drop: _____

STEP 3:

Transfer 20 ul of the liquid onto the Parafilm. Use a ruler to measure the size of the droplet and shade in the pipette tip to indicate relative volume.



Micropipette used: _____

Tip size used: _____

Diameter of the 20 ul drop: _____

STEP 4:

Transfer 200 ul of the liquid onto the Parafilm. Use a ruler to measure the size of the droplet and shade in the pipette tip to indicate relative volume.



Micropipette used: _____

Tip size used: _____

Diameter of the 200 ul drop: _____

STEP 5:

Transfer 500 ul of the liquid onto the Parafilm. Shade in the pipette tip to indicate relative volume.



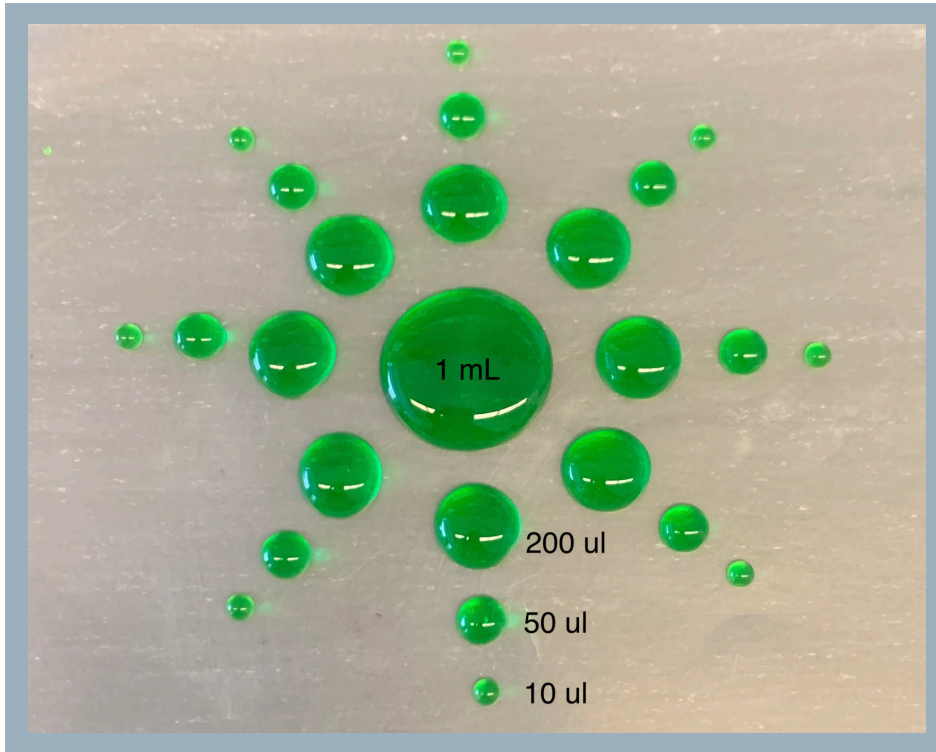
Micropipette used: _____

Tip size used: _____

Diameter of the 1000 ul drop: _____

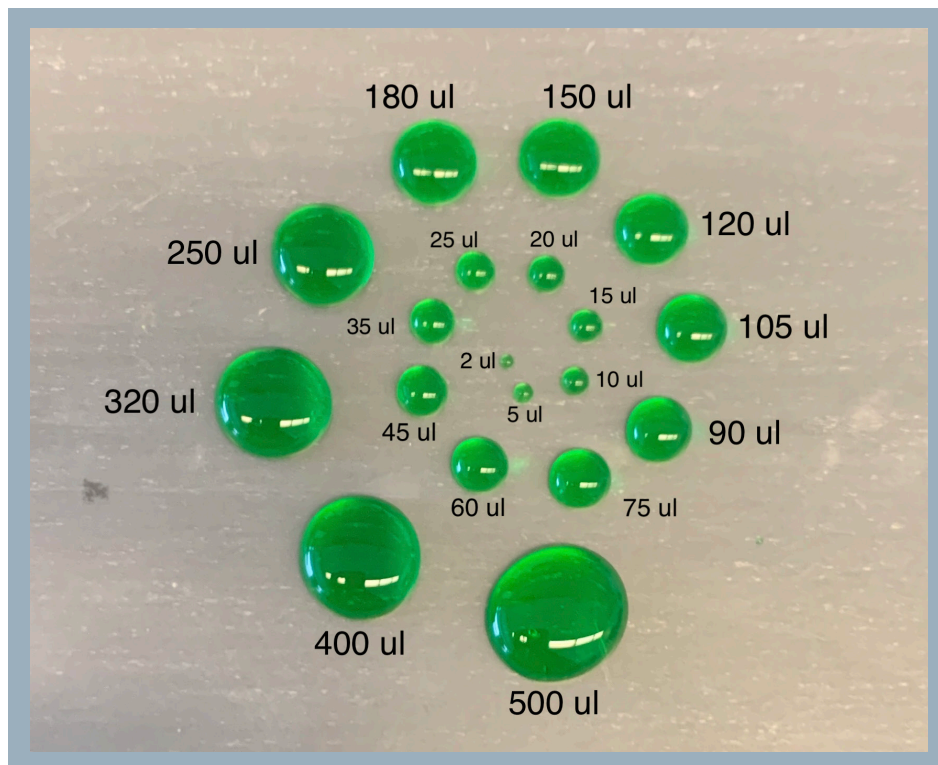
Bonus Activity: Pipette Art

Using the P2, P20, P200, and P1000 pipettes, recreate the designs below on a piece of Parafilm or wax paper using water dyed with food coloring.



We would love to see your artistic creations! Send images of pipette art to info@wolbachiproject.com to be featured on our social media accounts, website, and/or newsletter.

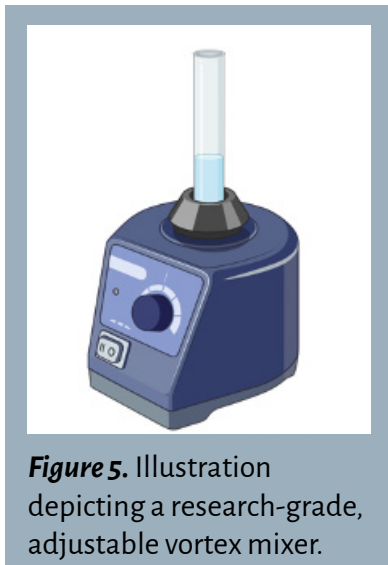
Would you like to create a new template for this activity? Indicate the droplet sizes and we will add your design(s) to this lab guide.



Introduction

Another tool commonly used in laboratories is a **vortexer**, or **vortex mixer**. This electric device makes quick, circular motions to thoroughly mix small vials or tubes of liquids. When used correctly, a vortex will be visible inside of the tube.

Vortex mixers are made in different shapes and sizes. An adjustable, research-grade vortexer is illustrated in **Figure 5**. Another common option is a mini vortex mixer, shown in **Figure 6**.



Mini vortex mixers are often touch-based and non-adjustable, operated by pressing the sample tube against the base. For larger vortex mixers, such as the Vortex Genie 2, there are usually two controls:

1. **On/off switch.** This often features “on” or “touch” options (see **Figure 7**). The “on” feature will automatically turn it on for continuous vortex, while the “touch” feature operates only when the tube or vial is pressed against the base.
2. **Variable speed control.** Some vortex mixers allow users to adjust the intensity. For the purposes of this lab series, set the dial set between 7 and 9 for high-speed mixing (see **Figure 8**).



Figure 7. Photo of the on/off/touch switch on a research-grade vortexer..

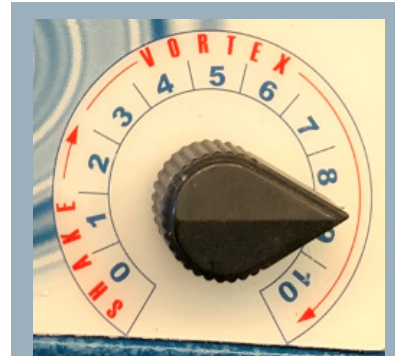


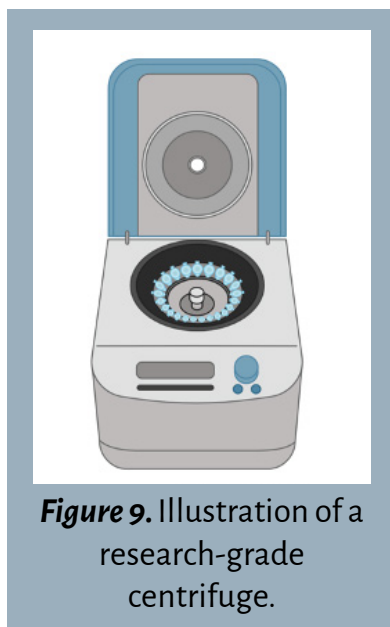
Figure 8. Photo of the adjustable dial on a research-grade vortexer.

Helpful Tips

- Completely seal each tube before using the vortex mixer to avoid spilling the sample and releasing aerosols into the environment.
- With a small number of tubes, it is acceptable to hold the tubes and press them against the base of the vortexer. A tube rack or adapter may be used to vortex multiple tubes at once.
- Make sure there is a visible vortex inside of the tube. If not, reposition the tube(s) on the platform to ensure efficient mixing.

Introduction

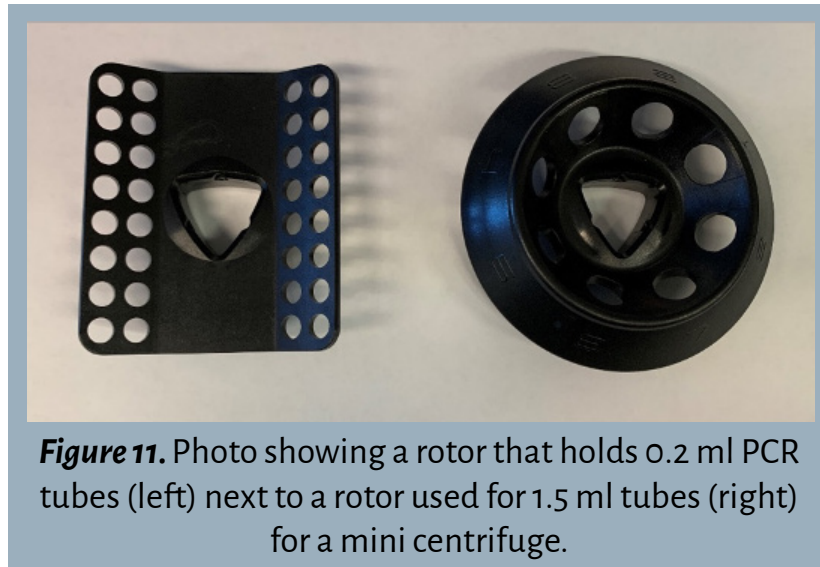
Centrifuges utilize **centrifugal force** to spin tubes very quickly. They are often used to “spin down” liquid so that it collects at the bottom of the tube, as opposed to sticking to the cap or the sides. Centrifugation also works to separate mixtures of liquids (or liquids and solids) of different densities, with the densest material settling at the bottom. For example, DNA extraction of an insect utilizes centrifugation to separate, or pellet, insect cellular debris at the bottom of the tube while DNA remains suspended in the liquid supernatant.



Similar to vortex mixers, centrifuges come in a variety of brands and sizes. Commonly, labs use adjustable centrifuges that hold large numbers of samples (**Figure 9**). These research-grade centrifuges are set to spin at a defined speed for a given amount of time. The speed, measured in either **Relative Centrifugal Force (RCF; $x g$)** or **Revolutions Per Minute (RPM)**, can be adjusted using a dial on the centrifuge. They often feature a lid covering the rotor, which must be removed before loading samples and screwed back on afterwards (particularly if spin columns are being used).

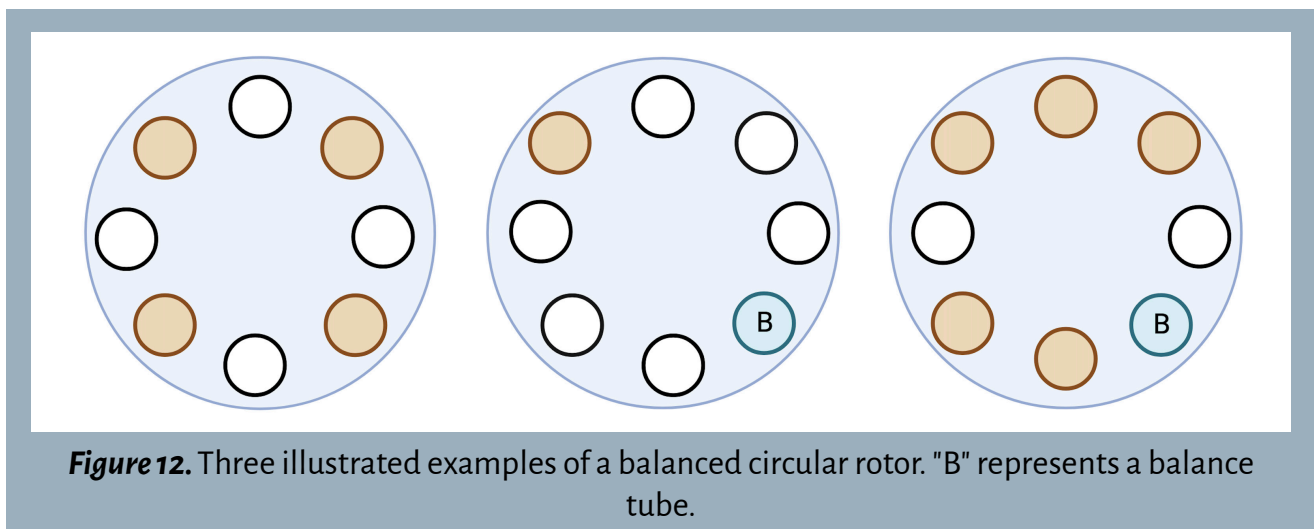
Alternatively, a mini centrifuge (**Figure 10**), holds a fewer number of tubes and allows for quick and convenient spins. Mini centrifuges often have a pre-determined speed that cannot be adjusted.

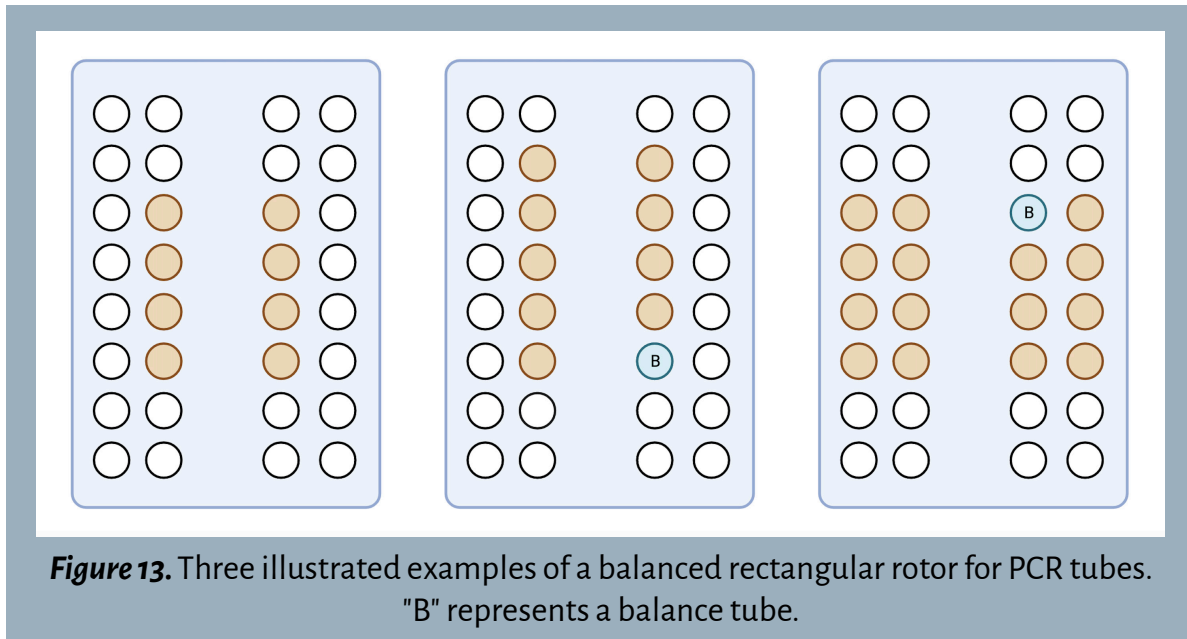
The **rotor** sits inside of the centrifuge and holds the tubes. Rotors are replaceable and come in different sizes to accommodate various sizes of sample tubes. For example, a different rotor is needed to spin 0.2 ml PCR tubes as opposed to 1.5 ml tubes.



The rotor must be balanced before turning on the centrifuge to ensure that it functions correctly. If the rotor is not balanced, the center of gravity will be off, causing a loud sound and potential damage to the equipment.

When loading tubes into the rotor, orient them in opposite positions. With an odd number of tubes, a **balance tube** (B) can be made using water filled to the same volume as the other tubes. See **Figure 12** and **Figure 13** for examples of balanced centrifuge rotors.





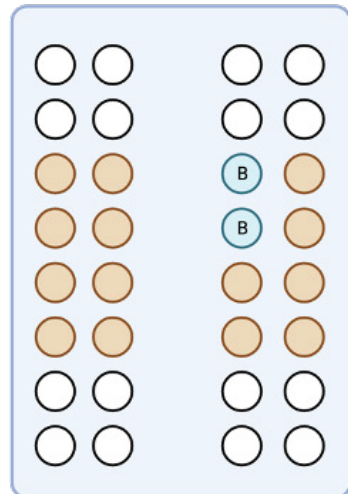
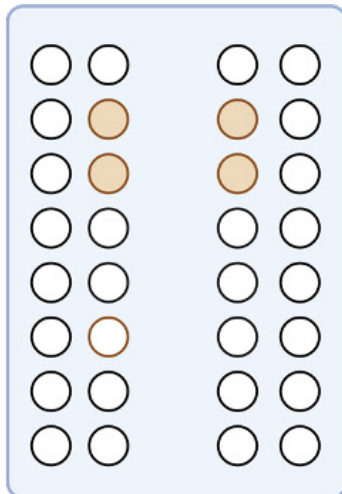
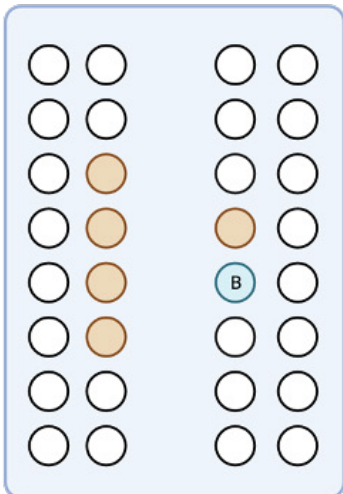
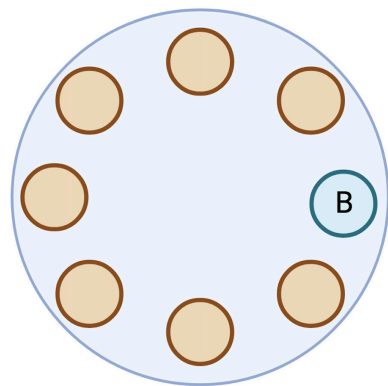
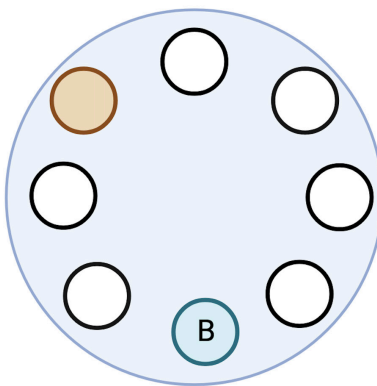
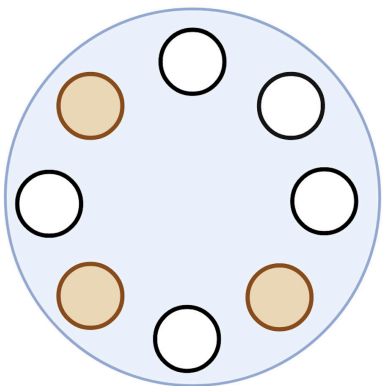
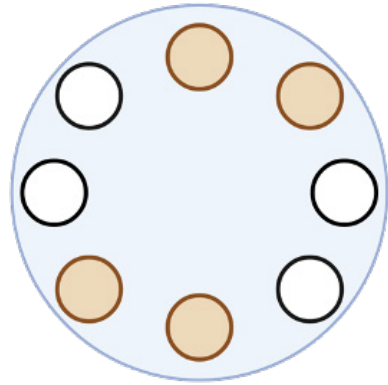
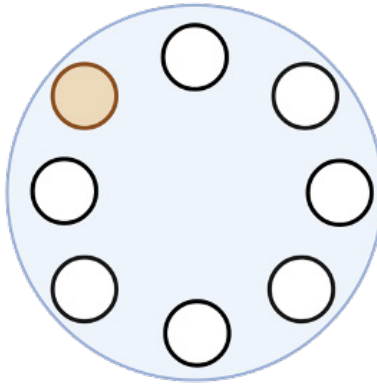
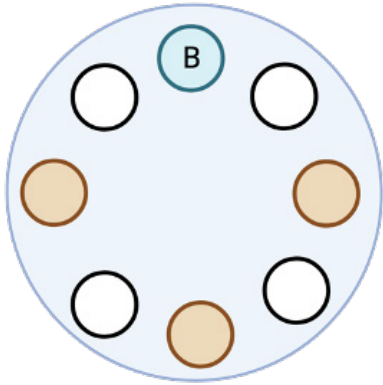
Helpful Tips

- Ensure that the lids are tightly sealed before centrifugation.
- There can be multiple ways to correctly balance a rotor.

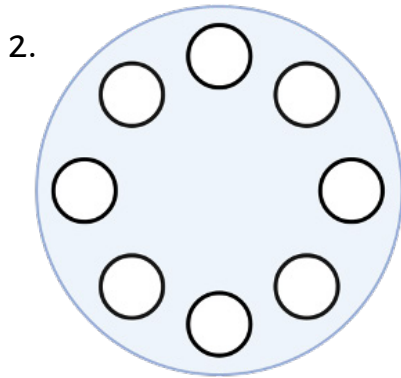
Practice Worksheet

Circle the rotors that are correctly balanced.

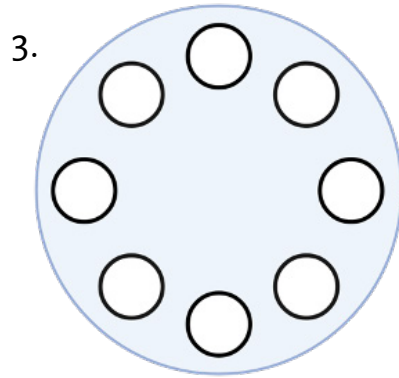
1.



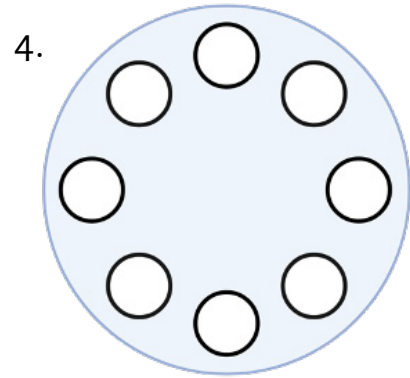
Fill in the circles on the rotors to represent how you would properly balance a centrifuge rotor. Write a “B” to represent balancer tubes. Keep in mind that there can be multiple ways to correctly balance a centrifuge.



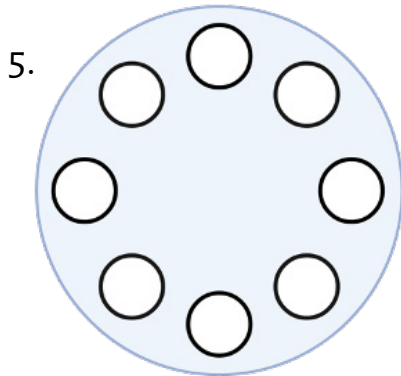
1 tube



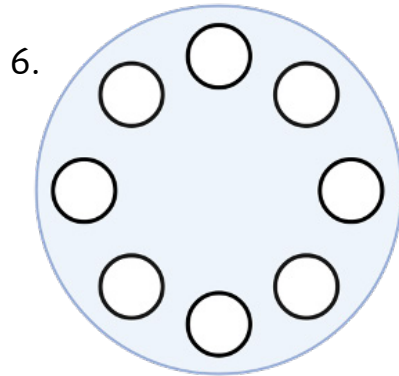
3 tubes



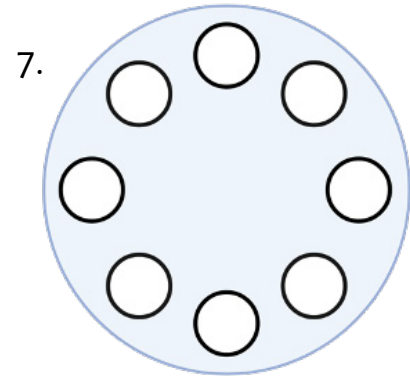
4 tubes



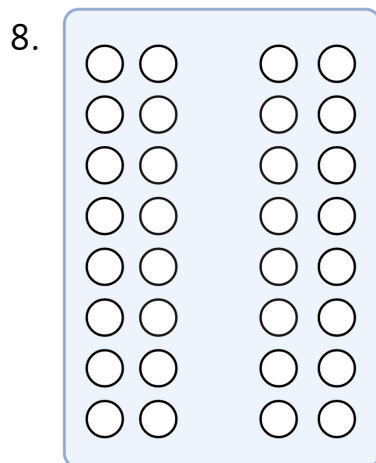
5 tubes



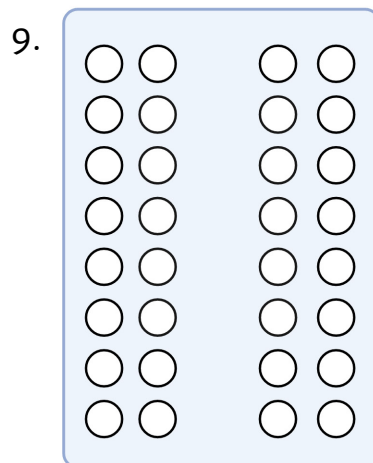
6 tubes



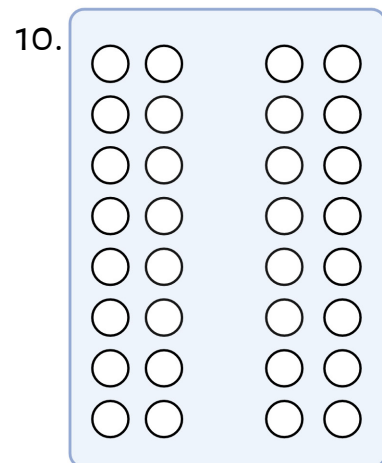
7 tubes



1 tube



9 tubes



16 tubes

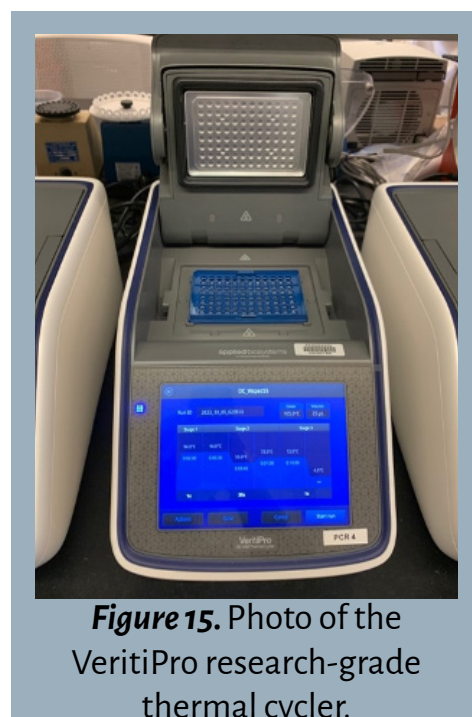
Introduction

A **Thermal Cycler**, otherwise known as a PCR machine, is most commonly used to amplify sections of DNA via polymerase chain reaction (PCR). To read more about PCR and its applications, see Lab 3 of the *Wolbachia* Project: <https://wolbachiaproject.org/pcr/>.

Thermal cyclers do exactly what their name implies; they fluctuate the temperature of samples over a given number of cycles. The temperature fluctuations allow for denaturation of the DNA, annealing of the PCR **primer**, and extension of the primer to the end of the amplicon. In general, PCR programs are based on the melting temperature (T_m) of the primers and length of DNA being amplified.

Thermal cyclers can also be used as **incubators**, as they can be programmed to hold samples at one temperature for a given amount of time. Often, the thermal cycler is programmed to hold DNA samples at 4C indefinitely (∞) after the run is complete to ensure preservation of the sample until it is removed from the machine. A thermal cycler can also be used for enzymatic assays when a sample must be incubated at precise temperatures.

The operation instructions for a thermal cycler vary depending on the model and size. Mini thermal cyclers, such as MiniOne (shown in **Figure 14**) and miniPCR, hold smaller numbers of tubes and are programmable with a mobile device. Larger thermal cyclers, such as the VeritiPro (shown in **Figure 15**), are programmed directly on the device, hold a greater number of tubes, and provide numerous customizable options.



Helpful Tips

- Thermal cyclers typically hold smaller tubes designed specifically for PCR. Be sure that your samples are in the correct tubes and lids are tightly sealed to prevent evaporation.
- Some thermal cyclers require adapters when using tubes rather than plates. Failing to use the adapters may result in melted tubes.
- Double check the program that you are running before starting the thermal cycler. If required, enter the correct sample volume (the total volume of liquid inside each tube).
- For PCR, make sure that your thermal cycler is set to hold samples at 4C after the program is complete.
- With some PCR machines, you can view the temperature fluctuate in real time as the cycle runs.

Introduction

Gel Electrophoresis is a technique that has been used for decades by scientists to separate, identify, and quantify DNA, RNA, or protein fragments. After DNA fragments have been amplified by PCR, for example, an agarose gel electrophoresis system is used to verify the presence and quantity of DNA in the sample.

The agarose gel electrophoresis system works by applying an electric current to an agarose gel with wells for each sample to be inserted. Because DNA is negatively charged, it will naturally move towards the positive end of the gel when the current is applied. The **agarose** gel, surrounded by a **buffer solution**, serves as a thick, porous material that slows the movement of DNA so that it can be observed.

Differently sized DNA fragments move at different speeds through a gel; smaller fragments move more quickly to the bottom of the gel while larger fragments move more slowly. This difference in movement speed allows fragments to be separated by size. Fragments of the same size will move at the same speed and clump together, forming a **band** in the gel. A **DNA ladder** containing DNA fragments of known sizes is loaded into one of the wells and serves as a visual reference to estimate the size of each band. The presence or absence of a band of a known size informs the researcher if the fragment that they are looking for is amplified. If no bands are present, either the specific DNA fragment is not present in the sample or something went wrong earlier in the experiment. For more explanation about this process, see the materials for Lab 4 of The *Wolbachia* Project: <https://wolbachiaproject.org/electrophoresis/>.

Like most lab equipment, agarose gel electrophoresis systems are available in different sizes and from different manufacturers. Systems vary in the number of samples that they can hold, the presence/absence of an integrated power supply, and the presence/absence of an integrated transilluminator. This guide will review the components of a standard agarose electrophoresis system as well as the MiniOne gel electrophoresis system.

Getting Started

Standard Agarose Gel Electrophoresis System

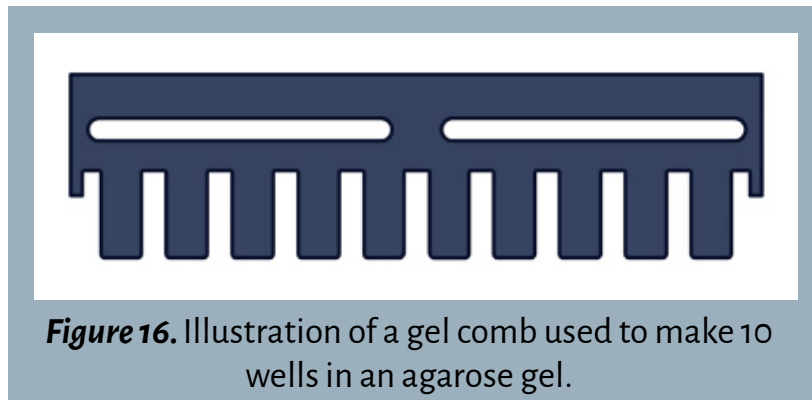
This system is most common in research laboratories and consists of a gel electrophoresis chamber, external power supply, gel casting tray, gel combs, and external imager.

STEP 1:

Prepare the agarose solution. Create an agarose solution by dissolving agarose powder in a buffer according to the instructions of the system. Allow the heated solution to sit for a few minutes before pouring.

STEP 2:

Prepare the casting tray. Ensure that all sides of the tray are sealed - this can be done by tightly wrapping lab tape around the two open sides, or by placing the tray inside a gel casting chamber, with the rubber pieces against both walls. Choose a gel comb based on the amount of samples being run, and insert the comb into the slots on the casting tray. Many trays allow for multiple gel combs, most commonly with one located towards the top of the gel and one towards the middle of the gel.



STEP 3:

Pour the gel. Pour the agarose gel solution into the casting tray and allow the gel to solidify for around 1 hour. Ensure that liquid is not leaking out of the tray.

STEP 4:

Assemble the electrophoresis chamber. After the gel is solidified, remove the comb(s), and place the tray with the gel into the **gel electrophoresis chamber**. The chamber, or box, is typically a plastic rectangular prism with a lid containing a **positive node (anode)** and **negative node (cathode)**. The nodes are connected to a power supply which sends an electric current through the gel and buffer. It's important that the positive end is oriented towards the bottom and the negative end is facing the top, so that the DNA moves in the correct direction through the gel.

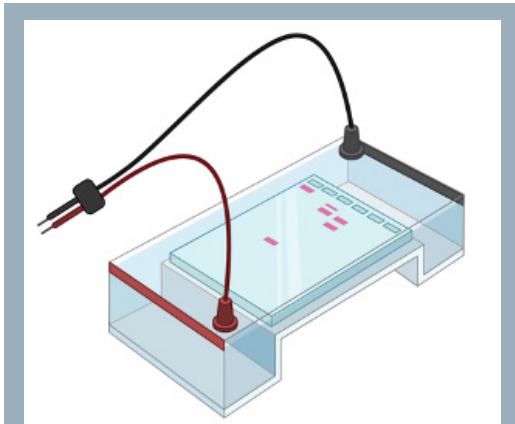


Figure 17. Illustration of a gel electrophoresis chamber with wires connected to the positive (red) and negative (black) nodes



Figure 18. Photo of a Fisher Scientific FB300 power supply with wires connected from the positive and negative nodes of a gel electrophoresis chamber.

STEP 5:

Add the running buffer. Before loading samples into the wells, add a buffer solution to the chamber up to the fill line to help conduct the current through the gel. Commonly used buffers are TAE buffer, TBE buffer, SB buffer, and LB buffer. The buffer added to the chamber should be the same as the one that was used to make the agarose solution in Step 1.

STEP 6:

Load the gel. Use a micropipette to add the DNA ladder and samples to the gel. Using a new pipette tip for each sample, carefully hover the tip directly above the well (inside of the buffer solution) and slowly eject the sample inside, being careful not to spill it outside of the well. To avoid introducing extra air, do not press the pipette plunger completely down to the second stop. Use caution to not puncture the gel with the pipette tip. It is vital that the tip is changed after loading each sample to avoid cross-contamination.

A **loading dye** is commonly added to the samples – either in the PCR master mix or directly to the PCR product - so that the samples are easy to see when loading the wells.

STEP 7:

Run the gel. After all of the samples are loaded, set the battery to run.

Power supplies are designed specifically for gel electrophoresis and can be set for a specific amount of time at the desired current. Keep in mind that the higher the current (or voltage), the quicker the sample will move through the gel. Voltage and run times are determined by the % agarose and type of buffer used to cast the gel. Higher percentage agarose gels feature smaller pores that allow for slower movement and enhanced discrimination between closely sized fragments relative to lower percentage agarose gels. TAE and TBE buffers require lower voltage which results in longer run times. SB and LB buffers, however, tolerate higher voltage and require shorter run times.

% AGAROSE

Lower % agarose

Larger pores, shorter run times

Higher % agarose

Smaller pores, longer run times

BUFFER

TAE and TBE buffers

Lower voltage, longer run times

SB and LB buffers

Higher voltage, shorter run times

STEP 8:

Stain the gel. After the gel has finished running, add a **DNA stain** in order to visualize bands. Commonly used DNA stains - ethidium bromide, GelRed, GelGreen, and SYBR Safe – vary in sensitivity, toxicity, and price. DNA stains can be added directly to the agarose gel as it is being prepared (pre-cast method) or the gel can be submerged in a stain solution following the gel electrophoresis run (post-stain method). Follow the instructions for the DNA stain available.

STEP 9:

Visualize the gel. Once staining is complete, the gel is ready to be photographed by an **imager** that exposes the dye to UV or blue light (depending on the type of DNA stain) to capture the DNA. The final image should show the DNA ladder alongside the loaded samples, with bands present if the desired DNA fragments were amplified during PCR (**Figure 19**).

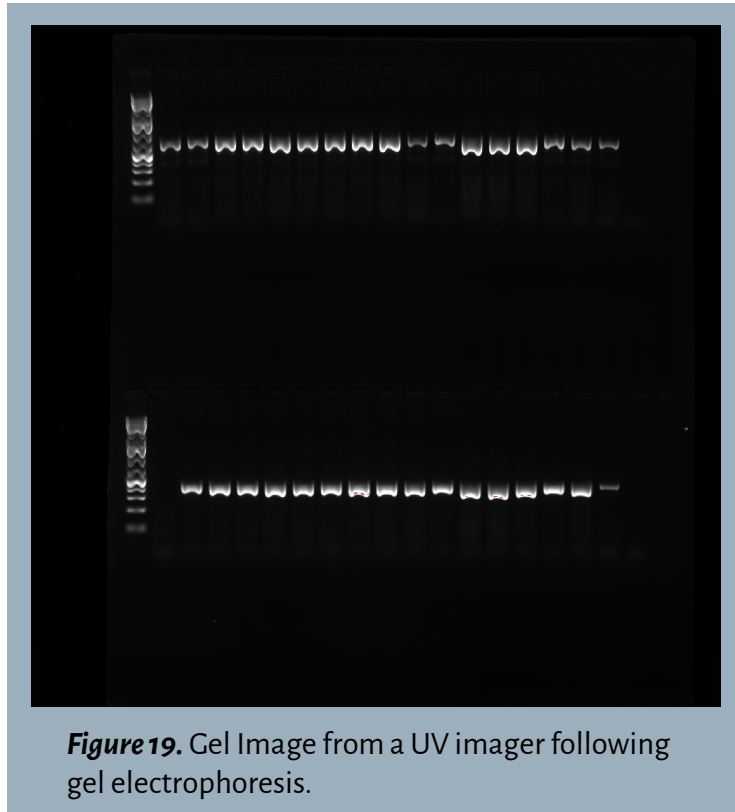


Figure 19. Gel Image from a UV imager following gel electrophoresis.

MiniOne Gel Electrophoresis System

The MiniOne Gel Electrophoresis System is an alternative device that is more affordable and easy to use for a small number of samples (up to nine at one time). Unlike the standard gel electrophoresis system, the MiniOne system incorporates the gel chamber, power supply, and imager into one portable device (see **Figure 20**). The system also comes with a casting tray and gel combs (Figure 21).

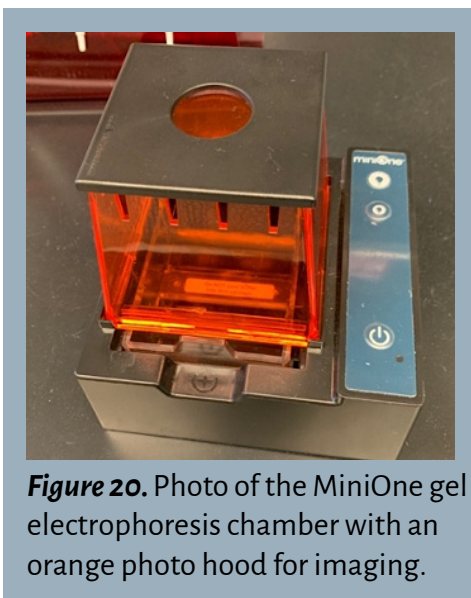


Figure 20. Photo of the MiniOne gel electrophoresis chamber with an orange photo hood for imaging.

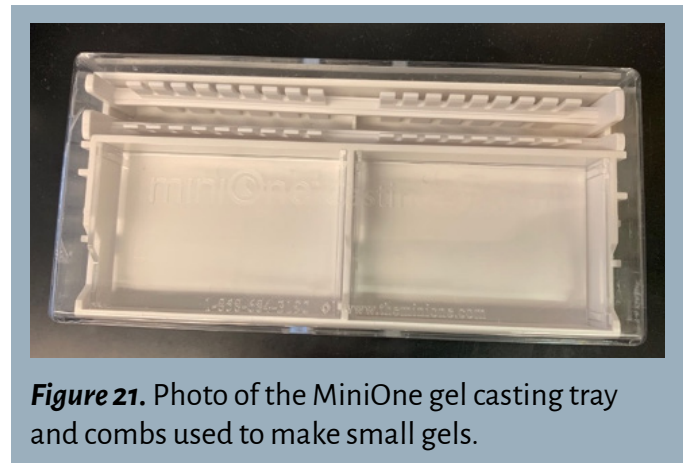


Figure 21. Photo of the MiniOne gel casting tray and combs used to make small gels.

STEP 1:

Prepare the agarose solution. Microwave the agarose gel solution according to the instructions on the GelCup. Allow the heated solution to sit for a few seconds before pouring.

STEP 2:

Prepare the casting tray. Insert the comb(s) into the designated slot on the mini casting tray.

STEP 3:

Pour the gel. Pour the agarose solution into the tray and allow the gel to solidify for around 10-20 minutes. Ensure that the gel has fully set, then carefully remove the comb(s) from the gel.

STEP 4:

Assemble the electrophoresis chamber. Remove the tray along with the gel and place it inside the gel electrophoresis chamber, ensuring the wells are oriented near the negative end.

STEP 5:

Add the running buffer. Locate the running buffer that comes with the GelCups and pour the diluted solution into one end of the chamber until it flows through the other side and up to the fill line.

STEP 6:

Load the gel. Load the DNA ladder and samples into the gel using a micropipette. This is done by hovering the tip of the pipette over the well (inside the buffer solution) and carefully ejecting the sample so that it collects inside the well, being careful not to spill it outside of the well. To avoid introducing extra air, do not press the pipette plunger completely down to the second stop. Use caution to not puncture the gel with the pipette tip. It is vital that the tip is changed after loading each sample to avoid cross-contamination.

This system uses the pre-cast method, where a DNA stain (Gel Green) is present in the agarose gel. Therefore, when the blue light is turned on, the user can see the DNA samples as they are loaded into the gel. It is best to use the dim blue light when loading samples (bottom lightbulb button) and save the bright blue light (top lightbulb button) for photographing the gel. This is because the DNA stain is photosensitive and will lose effectiveness after prolonged exposure to blue light.

STEP 7:

Run the gel. Once the samples are loaded, use the power button to activate the electric current. Periodically turn on the dim light throughout the run to check if any are bands present and if the DNA is moving through the gel.

STEP 8:

Visualize the gel. When enough time has passed, use a mobile device positioned on top of the orange photo hood to capture a photograph of the gel using the bright blue light.

Agarose: A polysaccharide purified from seaweed. When dry agarose is boiled in a buffer solution, it will harden into a flexible, gelatin-like slab as it cools.

Anode: A positively charged electrode.

Balance Tube: A tube that is filled with an equal volume of liquid for the sole purpose of balancing out the centrifuge when spinning an odd number of tubes.

Band: A clearly visible and defined mark on a gel, indicating DNA presence. The location of the band in relation to reference bands in the ladder indicates the size of the DNA product amplified in PCR.

Buffer Solution: A salt-containing solution used in gel electrophoresis to help conduct an electric current through gels.

Cathode: A negatively charged electrode.

Centrifugal Force: Inertial force experienced by objects moving in a circular motion.

Centrifuge: A piece of equipment that utilizes centrifugal force to separate fluids by density.

DNA Ladder: A mix of pre-cut DNA of defined sizes. Each ladder will have different “rungs” of pre-determined sizes; check the product information to find the sizes of DNA for each specific ladder.

DNA Stain: A dye that binds to the DNA, making it visible with UV or blue light.

Electrophoresis Chamber/Box: The box that contains the buffer solution, gel, and positive and negative nodes for gel electrophoresis.

Gel Electrophoresis: A method of separating substances based on the rate of movement while under the influence of an electric field. DNA is often separated and analyzed using agarose gel electrophoresis.

Gel Imager: A device that often uses UV light or blue light to illuminate stained DNA bands in a gel.

Incubator: A machine that holds samples at a constant temperature.

Loading Dye: Loading dye is added to aid in the loading of an agarose gel. It contains colored dye(s), often migrating with specific sizes of DNA, and glycerol.

Micropipette: Tool used to transfer small volumes of liquid (0.1-1000 μ l) from one place to another accurately and with minimized possibility for contamination.

Polymerase Chain Reaction (PCR): A common laboratory technique used to amplify DNA fragments. PCR consists of three steps: denaturation, annealing, and extension.

Primer: Approximately 18-30 nucleotides of DNA designed to bind and amplify a specific section of DNA. A PCR reaction contains both forward and reverse primers.

Relative Centrifugal Force (RCF; x g): Unit that describes the gravitational force, or g-force, that is exerted on each sample inside of a centrifuge. This measurement accounts for both the speed of rotation and the distance of the sample from the center of rotation.

Revolutions Per Minute (RPM): Unit that describes the speed at which the rotor revolves.

Rotor: The rotating unit of a centrifuge, the rotor houses the tubes to be centrifuged.

Thermal Cycler: The machine that heats tubes to specific temperatures to carry out PCR and incubations.

Vortexer/Vortex Mixer: Device that thoroughly mixes small vials or tubes of liquids by making quick, circular motions.

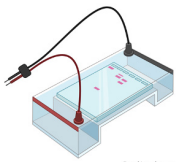
Practice Worksheet: Name that Device!

Micropipette
Thermal Cycler

Vortex Mixer
Gel Electrophoresis System

Centrifuge

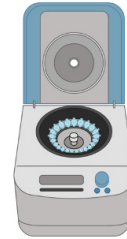
Use the Word Bank above to label each device.



1. _____



2. _____



3. _____



4. _____



5. _____

Below are excerpts from laboratory protocols. Indicate which device you would use to complete each activity.

6. _____

Transfer 23 μ l of PCR mix to each tube.

7. _____

Spin for 2 minutes at 16,000 x g.

8. _____

Incubate for 2 minutes at 35 °C followed by 5 minutes at 80 °C.

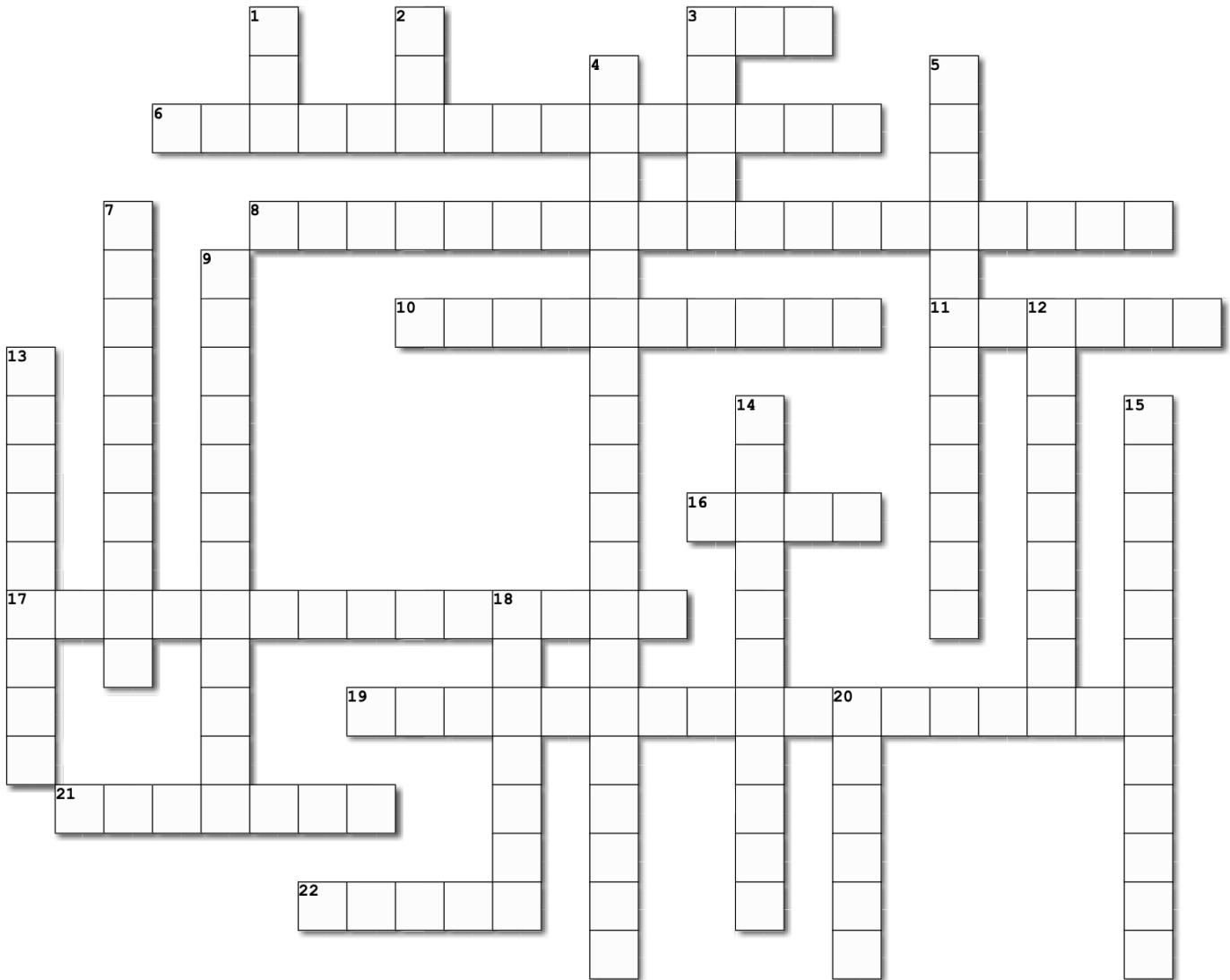
9. _____

Vortex each tube for 20 seconds.

10. _____

Run the agarose gel for 60 minutes at 70V.

Laboratory Equipment Crossword Puzzle



Created using the Crossword Maker on TheTeachersCorner.net

Use the definitions below to complete the Crossword Puzzle. Some answers contain two words. In this case, leave a blank space in the Crossword.

Across

3. Unit that describes the speed at which the rotor revolves.
6. A salt-containing solution used in gel electrophoresis to help conduct an electric current through gels.
8. A method of separating substances based on the rate of movement while under the influence of an electric field.
10. A piece of equipment that utilizes centrifugal force to separate fluids by density.
11. Approximately 18-30 nucleotides of DNA designed to bind and amplify a specific section of DNA.
16. A clearly visible and defined mark on a gel, indicating DNA presence.
17. The machine that heats tubes to specific temperatures to carry out PCR and incubations.
19. Internal force experienced by objects moving in a circular motion.
21. A polysaccharide purified from seaweed. Used to create the gel material used for gel electrophoresis.
22. A positively charged electrode.

Down

1. Unit that describes the gravitational force, or g-force, that is exerted on each sample inside of a centrifuge.
2. A common laboratory technique used to amplify DNA fragments. Consists of three steps: denaturation, annealing, and extension.
3. The rotating unit of a centrifuge. Houses the tubes to be centrifuged.
4. The box that contains the buffer solution, gel, and positive and negative nodes for gel electrophoresis.
5. Tool used to transfer small volumes of liquid (0.1-1000 ul) from one place to another accurately.
7. A device that often use UV light or blue light to illuminate stained DNA bands in a gel.
9. Device that thoroughly mixes vials or tubes of liquids by making quick, circular motions.
12. A machine that holds samples at a constant temperature.
13. A dye that binds to the DNA, making it visible with UV or blue light.
14. Added to aid in the loading of an agarose gel. Contains colored dye(s) and glycerol.
15. A tube that is filled with an equal volume of liquid to balance out the centrifuge.
18. A negatively charged electrode.
20. A mix of pre-cut DNA of defined sizes.

Laboratory Equipment Word Search

Z R P H D M O X H V W M T A E J Q S S A
 E D B O X R E D D A L A N D Z G E B E G
 E P U I N C U B A T O R F Z I J H D F G
 W V F I N U J M S M Z C W T L S O M E W
 W T F X G Y T U K E R O T O R H N L A B
 U C E C R O F L A G U F I R T N E C X T
 V O R T E X M I X E R Z Y A L L X O M T
 Z O S M A L L N K W W R C P E P O J D E
 E W O X Q R T H E R M A L C Y C L E R Z
 O G L T E E B Z E T I R T G C M L P E O
 T W U P E Q B M J V T R J V U O K V L M
 P E T F N H I U A L O E D N A B Z F L A
 D S I O I R C I T P U X P D P P S K O K
 Y O O E P R Z B H E L R I I I U G T B Y
 K R N K V D T O O J C N N E P C E X R N
 O A X U K M R N I J G N Z A N O D E E B
 Q G X A P E G J E D L H A T B E R M F D
 N A P R S W W Z Y C R S U L H L Z C G H
 I L E I P T L E L D N A S T A I N I I R
 D O S I B F G E L I M A G E R B H G U M

Agarose Band	RPM Vortex Mixer	Loading Dye Primer	Gel Imager Micropipette
Centrifugal Force	Anode	Rotor	RCF
DNA Stain	Buffer Solution	Balance Tube	Thermal Cycler
Incubator	Centrifuge	Cathode	
PCR	Gel Electrophoresis	DNA Ladder	