

Gel Loading Practice MiniLab Student's Guide

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Laboratory Safety

- 1. Exercise caution when working with reagents.
- 2. Never consume reagents or gels.
- 3. Gloves and eye protection should be used whenver needed as part of good laboratory practice.
- 4. Always wash hands thoroughly after handling biological material or reagents.



Objectives

To learn how to read and use a micropipette.

To learn how to load a gel.

To practice the aforementioned skills with good laboratory technique.

Background

The purpose of this laboratory is to introduce you to an important tool in biotechnology: the micropipette. This tool allows the researcher to transfer small and exact volumes of liquids when setting up experiments. For researchers working with DNA, they may need to analyze DNA samples by separating them with gel electrophoresis. Micropipettes are a crucial tool for them to transfer volumes of samples into a small cavity (called a well) in the gel.

Today, you will practice using a micropipette by loading colored samples into wells. Be careful! Gels are made of a fragile material called agarose and it is easy to pierce through the wells.



Plunger

Button

Part I: Using a Micropipette

Materials

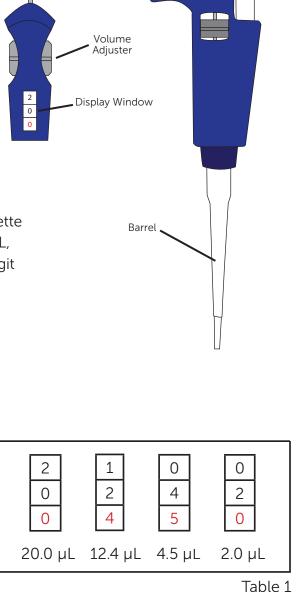
1 loading dye aliquot 1 micropipette (measures 2.0-20.0 μL) Pipette tips Practice sheet

Procedure

1. With the micropipette, review the parts of the tool. Locate the plunger button, tip ejector, volume adjuster, display window, and the barrel.

2. The display window shows the volume the micropipette will load or dispense. The top digit indicates 10's of μ L, the middle digit indicates 1's of μ L and the bottom digit indicates 1/10's of μ L.

By turning the plunger button, practice setting the micropipette to the four volumes listed in Table 1. (**Caution:** Never set a P-20 micropipette lower than 2.0 μ L or higher than 20.0 μ L. **It can seriously damage or destroy the tool.**)



Tip Ejector

minion ne.

Procedure (continued)

- 3. Load the pipette:
 - a. Set the micropipette to 10.0 µL.
 - b. Place a pipette tip on the micropipette by lowering the end of the barrel onto a tip and pushing firmly down. Do not touch the tip with your fingers to avoid contamination.
 - c. With your thumb, push the plunger button down to the first stop.
 - d. Insert the pipette tip vertically into the sample and **slowly** draw the plunger up.

Do not turn the tip up or lay the micropipette down.

- 4. Dispense the sample
 - a. Set the laminated practice sheet on a level surface.
 - b. Hold the loaded micropipette vertically over a spot on the practice sheet so that the end of the tip lightly touches the surface.
 - c. Slowly push the plunger button down to the first stop to dispense most of the sample. Continue pushing slowly to the second stop to dispense the rest of the sample.
 - d. While holding the plunger button down at the second stop, pull the tip up and out of the liquid. This prevents you from accidentally pulling liquid back into the tip.
 - e. Slowly release the plunger button.
 - f. Keep the tip on the barrel to practice more. When finished, eject the tip into the waste by pushing the ejector button.
- 5. Practice loading and dispensing samples with the volumes from Table 1.

Precision comes with practice! It is also important to practice dispensing samples without adding air bubbles.

Part II: Loading a Gel

Materials

1 loading dye aliquot 1 micropipette (measures 2.0-20.0 μL) Pipette tips Practice gel Water (enough to cover the gel)

Procedure

- 1. Take a practice gel. Gently peel and remove the seal.
- 2. Set the practice gel on a level surface, preferably a dark or a colored surface. Slowly pour water over the gel.

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3. With a micropipette, tips, and some samples, practice loading each well.

SYSTEMS

- a. Using good technique, load 10.0 μ L of sample into a pipette tip.
- b. Hold the pipette tip vertically over the gel.
- c. Steady your pipetting hand by placing your elbow on the table. For additional support, use your other hand.
- d. Gently insert the tip into the water and then slightly over a well opening. Do not touch the bottom or sides of the well to avoid breaking the gel.
- e. With good technique, dispense the sample. Because the sample is denser, it will easily sink down into the well. Avoid introducing air bubbles into the well to prevent any sample from being blown out.
- f. With the plunger still depressed, gently pull the pipette tip vertically up and out of the well. Do not touch the sides of the well to avoid breaking the gel.
- g. Practice loading samples across the gel. How many wells did you successfully load?







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