



Gel Loading Practice Kit

Cat# M3002

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Laboratory Safety

1. Exercise caution when working with reagents.
2. Never consume reagents or gels.
3. Always wash hands thoroughly after handling reagents.

Practice Kit Components

Supplies Included:

Practice Gels	20
Practice Transparency Sheets	20
Loading Dye	20 mL
1.5 mL Microcentrifuge Tubes	4 tubes x 6 colors (24 total)
Micropipette Tips	60 tips

Additional Materials Required

Micropipettes*	1 per group
*Not included with this kit	

Teacher's Guide

Experiment Objective

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.

Before the Lab

Prepare Loading Dye Aliquots

The kit provides 20 mL of loading dye at a working concentration (1X). With the provided microcentrifuge tubes, aliquot 500 μ L for each group.

Other Materials (Each group will need)

Micropipette (measures 2.0-20.0 μ L)

1 loading dye aliquot

1 practice sheet

1 practicing gel

Pipette tips

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, display window, and the barrel.

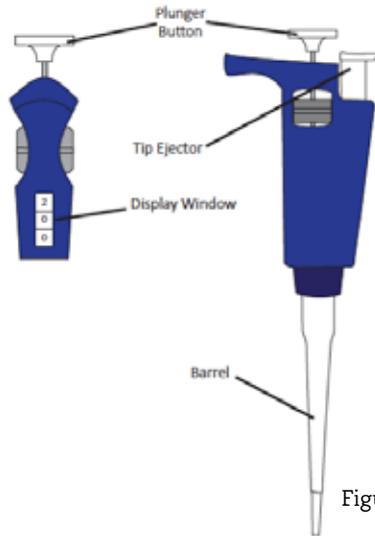


Figure 1

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

By twisting the plunger button, practice setting the micropipette to the four volumes listed in Figure 2. (**Never** set a P-20 micropipette lower than 2.0 μL or higher than 20.0 μL . It can seriously damage or destroy the tool.)

2	1	0	0
0	2	4	2
0	4	5	0
20.0 μL	12.4 μL	4.5 μL	2.0 μL

Figure 2

Procedure (continued)**3. Load the pipette:**

- a. Set the micropipette to 10.0 μL .
- b. Place a pipette tip on the pipette 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 pipette down.

Note: If the plunger is released too fast, the sample can shoot up into the pipette past the tip and contaminate the equipment. If the tip is not securely on the barrel, laying the pipette sideways or turning it upside down can also move the sample into the pipette.

4. Dispense the sample:

- a. Set the laminated practice sheet on a level surface.
- b. Hold the loaded pipette 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.

Note: Always dispense slowly to avoid introducing air bubbles into your sample. Practice this good technique especially for loading gels.

5. Practice loading and dispensing samples with the volumes from Figure 2. 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 lightly 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.

Note: Adding water serves several purposes. Water simulates the running buffer used in electrophoresis. It fills wells so that dispensed samples will easily fall to the bottom of wells (Loading dry wells is hard because samples may be drawn up well walls by capillary effect). Water also simulates the visibility of gels in the buffer of an electrophoresis running tank. Dark or colored surfaces help students to see the wells for loading because the gel is made of an opaque, fragile material.

3. With a pipette, tips, and some samples, practice loading each well.
 - a. Using good technique, load 10.0 μL 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.

Note: Samples that are blown out may float into a neighboring well during an electrophoresis run and affect results. Less sample remaining in the original well may also result in dimmer bands.

- 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?

Note: If a well is pierced on the bottom from students inserting the tip too far in, samples may be lost out of the well. If a well is nicked on the side walls, samples may not settle evenly and during an electrophoresis run, sample bands will not look sharp and clean. If a well wall is broken, samples can also leak into a neighboring well and affect results.

Student Worksheet

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Part I: Using a Micropipette

Materials

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Procedure

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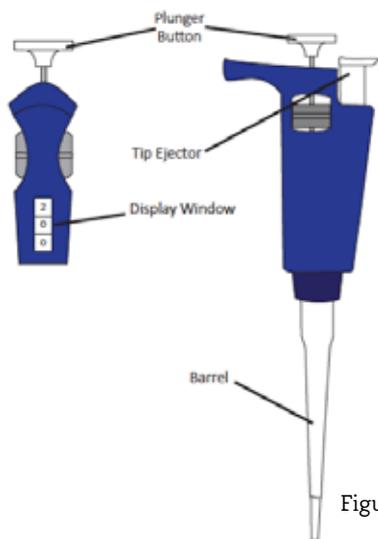


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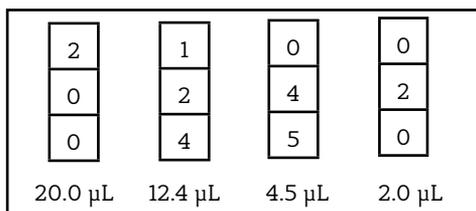


Figure 2

Procedure (continued)

3. Load the pipette:
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 - 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 Figure 2. Precision comes with practice! It is also important to practice dispensing samples without adding air bubbles.

Part II: Loading a Gel

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 - 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?

Additional MiniOne Products

MiniOne Equipment

Catalog #	Description	List Price
M1000	MiniOne Electrophoresis Unit	\$379.00
M1010	MiniOne Electrophoresis Classroom Package of 10	\$3380.00
M2031	MiniOne Microcentrifuge	\$350.00

MiniLabs

Catalog #	Description	Shelf Life	List Price
M3001	Electrophoresis 101 - The "fun"-damentals of electrophoresis (10 groups)	6 months	\$39.00
M3002	Pipette Practicing Kit - Master the skills to load a gel (20 groups)	6 months	\$49.00
M3003	PTC Genetics - Mendelian inheritance and taste blindness (10 groups)	6 months	\$69.00
M3004	DNA Fingerprinting - Help a whale calf find her father (10 groups)	6 months	\$69.00
M3005	CSI Forensics - Solve the crime using DNA & other evidence (10 groups)	6 months	\$69.00
M3006	Forensic Science - Foodborne Outbreak Investigation AP Bio (10 groups)	6 months	\$120.00

Consumables

Catalog #	Description	Shelf Life	List Price
M3101 TBE	Tris-Borate EDTA (TBE) Concentrate, 500 mL	12 months	\$18.00
M3102 TBE	1% GreenGel-in-a-Cup with 100 mL TBE Concentrate, 10 cups	6 months	\$21.00
M3103 TBE	2% GreenGel-in-a-Cup with 100 mL TBE Concentrate, 10 cups	6 months	\$21.00
M3104	MiniOne DNA Marker, 500 µL/ 1 vial, 100, 300, 500, 1000 and 2000 bp	12 months	\$35.00
M3105	Electrophoresis Grade Agarose, 5 g	24 months	\$15.00
M3106	Electrophoresis Grade Agarose, 25 g	24 months	\$45.00
M3108	0.6 mL Microcentrifuge Tubes, Assorted Colors, 200/pk	N/A	\$10.00
M3110	1.7 mL Microcentrifuge Tubes, Assorted Colors, 200/pk	N/A	\$10.00
M3111	1 - 200 µL Universal Fit Micropipette Tips, 250/pk	N/A	\$9.50
M3112	1 - 10 µL Micropipette Tips, 250/pk	N/A	\$9.50
M3113	GelGreen DNA Stain, 10,000X stock, 50 µL	12 months	\$20.00
M3114	GelGreen DNA Stain, 10,000X stock, 500 µL	12 months	\$100.00
M3115	5X Sample Loading Dye w/ Orange G and Xylene Cyanol, 10 mL	12 months	\$10.00
M3116	1 Kb DNA Ladder, 1000 µL	12 months	\$65.00
M3117	100 bp Ladder, 1000 µL	12 months	\$65.00

Pipettes

Catalog #	Description	U.S. List Price	10+ Units
M2008	Variable Volume, 2-20 µL	\$89.00	\$79.00
M2010	Variable Volume, 20-200 µL	\$89.00	\$79.00
M2011	Variable Volume, 100-1000 µL	\$89.00	\$79.00
M2012	Variable Volume, 1-10 µL	\$89.00	\$79.00

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