Chemical Technology Program
General Education & Sciences Division
Cape Fear Community College
Wilmington Campus

C-TEACH ACADEMY
BRIDGING HIGH SCHOOL, COLLEGE, AND INDUSTRY

Lab Activity: Basics
USE OF MICROPIPETTES

Use of Micropipettes

Chemical Technology Program, Cape Fear Community College

This laboratory experiment is designed to introduce you to a micropipette. Micropipettes are commonly found in analytical laboratories to measure extremely small amounts of liquid. Typically, traditional volumetric pipettes are used to pipette 1.00 mL or more. Micropipettes should be used to deliver volumes smaller than 1.00 mL (1,000 uL). You will use micropipettes in many laboratory settings. While some of the procedures may seem trivial and the results obvious, this is a chance for you to develop good laboratory techniques.

Micropipette

Pipette Tips

Multichannel Micropipette

TERMINOLOGY:

Micropipette: *Devices commonly used to measure smaller volumes, in the 1 to 1000 uL (microliter; hence the name!) range. This is equivalent to 0.001 to 1.000 mL. Micropipettes may come in digital or automated forms. General Cost: $275.00 – $675.00 each.*

Fixed Micropipette: *A micropipette that is made to deliver a "fixed" amount of volume. You cannot change the amount of delivery. These micropipettes are normally cheaper for a laboratory to obtain.*

Variable Micropipette: *A micropipette that is made to deliver a range of volumes. You CAN change the amount of delivery. These micropipettes are more expensive but make more practical sense for a laboratory.*

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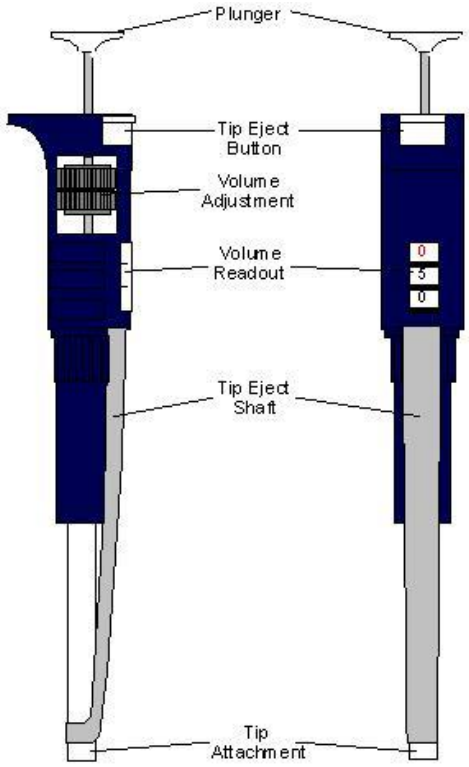
Multichannel Micropipette: *A micropipette that is used to simultaneously deliver the same volume to 8 wells/tubes at once. Typically used in biochemistry and biotechnology. General Cost: $650.00 to $950.00 each.*

Micropipette Tips: *All micropipettes require "tips" to be placed on the end. These tips hold the liquid you are measuring. The tips are made to be a "one-time" use (aka disposable).* ***There are different tips for different micropipettes; ensure that you are choosing the correct ones!***



NEVER OVERFILL THE PIPETTE!

PAY ATTENTION TO THE DELIVERY RANGE!

Improper use of micropipettes can result in liquids being accidentally drawn into the micropipette assembly. These liquids can destroy the gear mechanism that is responsible in delivering precise volumes. Additionally, these liquids can become a source of contamination for other samples. BE EXTREMELY CAREFUL WITH THE PLUNGER MECHANISM TO PREVENT THESE PROBLEMS FROM OCCURING.

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CLASS PROBLEM:
Suppose a technician is verifying the accuracy of a micropipette. The technician delivers 500 uLs of water, with a variable micropipette, a total of six times. He weighs each sample (density of water ~1.00 g per milliliter; therefore, the sample should weigh 500 micrograms or 0.500 g).

Weights:	0.491 g	0.501 g	0.483 g
	0.512 g	0.497 g	0.522 g

a. What is the average mass?
b. What is the standard deviation?
c. The micropipette has a "specification sheet" that states it should be delivering a volume of water within 2%. Does the analyst "pass" or "fail" on their delivery?
d. The precision (standard deviation) on the micropipette should be 0.010 or less. Does the analyst "pass" or "fail" on their delivery?

PART A - USE OF THE MICROPIPETTE

The micropipettes in the lab are probably the most precise, accurate and reliable pieces of equipment to deliver small quantities of volume. Although there are limits in the accuracy and precision of these micropipettes, most delivery errors are caused by incorrect handling of the sample or the micropipette. In this experiment you will practice using a micropipette and minimizing the errors associated with your technique.

GENERAL PROCEDURE FOR USING A MICROPIPETTE

1. Set the micropipette (if it is adjustable) to the desired volume. Be careful to NOT STRIP the gears.
2. Attach a disposable tip to the micropipette shaft. Press firmly.
3. While observing the tip and the sample, depress the plunger to the first stop, and place the tip in the liquid.
4. Allow the plunger to slowly return to its undepressed position as the sample is drawn into the tip. Wait 1 second to remove the micropipette from the liquid.
5. **NEVER ALLOW THE PLUNGER TO SNAP UP. THIS WILL CAUSE THE LIQUID TO ENTER THE GEAR MECHANISM AND RUIN THE PIPETTOR!**
6. Place the tip so that it touches the side of the container into which the sample will be expelled; depress the plunger to the SECOND stop.
7. Remove the micropipette, slowly, from the liquid while the plunger is still depressed.
8. Eject the tip using the third stop, tip ejector button, or other mechanism.

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In this procedure you will deliver various amounts of water into a container using a micropipette. The sample will be weighed. You will then report the weights to the laboratory instructor to ensure that you are properly delivering the correct amount of liquid. Unless instructed otherwise, you should handle the liquid sample with care! Measurements on the balance should be made to the nearest 0.1 mg (0.0001 g). **If your balance does not read to FOUR decimal places, please see a laboratory instructor.**

PROCEDURE: Using a Variable Micropipette to Deliver 1,000 uL (equivalent to 1.000 mL)

1. Place a weighing bottle on the balance and press TARE. The mass should read "0.0000 g".
2. Using the variable micropipette deliver 1,000 uL (equivalent to 1.000 mL) of the unknown liquid into the container. Record mass on your data sheet.
3. Repeat this process FOUR more times. You may simply "re-TARE" the container with the sample before the next addition. If you do this, you will not have to empty and dry out the container each time.
4. Make sure that you record your values in the data sheet.

PROCEDURE: Using a Variable Micropipette to Deliver 725 uL (equivalent to 0.725 mL)

5. Repeat the process above, using 725 uL instead of 1000 uL.

PROCEDURE: Using a Variable Micropipette to Deliver 515 uL (equivalent to 0.515 mL)

6. Repeat the process above, using 515 uL instead of 1000 uL.

PROCEDURE: Using a Variable Micropipette to Deliver 208 uL (equivalent to 0.208 mL)

7. Repeat the process above, using 208 uL instead of 1000 uL.

PROCEDURE: Using a Variable Micropipette to Deliver 100 uL (equivalent to 0.100 mL)

8. Repeat the process above, using 100 uL instead of 1000 uL.

PROCEDURE: Using a Variable Micropipette to Deliver 74 uL (equivalent to 0.074 mL)

9. Repeat the process above, using 74 uL instead of 1000 uL. A smaller micropipette may be needed in order to deliver this smaller volume requirement.

PROCEDURE: Using a Variable Micropipette to Deliver 26 uL (equivalent to 0.026 mL)

10. Repeat the process above, using 26 uL instead of 1000 uL. A smaller micropipette may be needed in order to deliver this smaller volume requirement.

PROCEDURE: Using a Variable Micropipette to Deliver 10 uL (equivalent to 0.010 mL)

11. Repeat the process above, using 10 uL instead of 1000 uL. A smaller micropipette may be needed in order to deliver this smaller volume requirement.

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ONCE YOU COMPLETE ALL THE VOLUME DELIVERIES ABOVE, PROVIDE YOUR DATA TO A LABORATORY INSTRUCTOR. IF YOUR DATA "PASSES" YOU HAVE COMPLETED THE EXERCISE. IF YOU DATA "FAILS", YOU WILL NEED TO RE-PIPETTE.

PROCEDURE: Test Your Skills!

1. Your instructor will have a 0.150 M solution of copper (II) sulfate pentahydrate [$CuSO_4 * 5 H_2O$] prepared (known as "stock"). Use the table below to make five solutions. The solutions should be prepared in small beakers or other suitable containers (such as a plastic conical tube). Use a micropipette for the preparation.
2. Once prepared, the five solutions will be analyzed on a UV-Vis instrument at 635 nm (instructor will aid in this process).
3. Record the absorbance (ABS) values from the instrument. Print off the data report and attached to your laboratory sheet.
4. Submit your data to the laboratory instructor to ensure that you are using the volumetric pipette correctly.

Test Tube	*1*	*2*	*3*	*4*	*5*
Volume of Stock (mL)	3.00	2.25	1.20	0.72	0.30
Volume of Water (mL)	0.00	0.75	1.80	2.28	2.70

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DATA SHEET

Part 1: Using a Variable Micropipette to Deliver 1,000 uL

Weight of 1st addition: ..... _____ g

Weight of 2nd addition: ..... _____ g

Weight of 3rd addition: ..... _____ g

Weight of 4th addition: ..... _____ g

Weight of 5th addition: ..... _____ g

Part 2: Using a Variable Micropipette to Deliver 725 uL

Weight of 1st addition: ..... _____ g

Weight of 2nd addition: ..... _____ g

Weight of 3rd addition: ..... _____ g

Weight of 4th addition: ..... _____ g

Weight of 5th addition: ..... _____ g

Part 3: Using a Variable Micropipette to Deliver 515 uL

Weight of 1st addition: ..... _____ g

Weight of 2nd addition: ..... _____ g

Weight of 3rd addition: ..... _____ g

Weight of 4th addition: ..... _____ g

Weight of 5th addition: ..... _____ g

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Part 4: Using a Variable Micropipette to Deliver 208 uL

Weight of 1st addition: ..... _____ g

Weight of 2nd addition: ..... _____ g

Weight of 3rd addition: ..... _____ g

Weight of 4th addition: ..... _____ g

Weight of 5th addition: ..... _____ g

Part 5: Using a Variable Micropipette to Deliver 100 uL

Weight of 1st addition: ..... _____ g

Weight of 2nd addition: ..... _____ g

Weight of 3rd addition: ..... _____ g

Weight of 4th addition: ..... _____ g

Weight of 5th addition: ..... _____ g

Part 6: Using a Variable Micropipette to Deliver 74 uL

Weight of 1st addition: ..... _____ g

Weight of 2nd addition: ..... _____ g

Weight of 3rd addition: ..... _____ g

Weight of 4th addition: ..... _____ g

Weight of 5th addition: ..... _____ g

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Part 7: Using a Variable Micropipette to Deliver 26 uL

Weight of 1st addition: ..... _____ g

Weight of 2nd addition: ..... _____ g

Weight of 3rd addition: ..... _____ g

Weight of 4th addition: ..... _____ g

Weight of 5th addition: ..... _____ g

Part 8: Using a Variable Micropipette to Deliver 10 uL

Weight of 1st addition: ..... _____ g

Weight of 2nd addition: ..... _____ g

Weight of 3rd addition: ..... _____ g

Weight of 4th addition: ..... _____ g

Weight of 5th addition: ..... _____ g

Part 9: Making Solutions Using a Micropipette: Print off UV Data Sheet and Generate Your Own Calibration Curve Using an Excel Spreadsheet (showing a Linear Regression Equation and Coefficient). Concentration should be plotted on the x axis. Absorbance values should be plotted on the y axis.

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