



Micropipetting and Density Experiment

MEASURING ACCURATELY WITH MICROPIPETTORS

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Introduction

In this activity, you will learn to use micropipettes to accurately measure small volumes of food dye. You will then compare your expected measurements to observed measurements using a digital balance. Pay attention to technique and cleanliness to ensure your data is as accurate as possible!

Materials

- P-200 μL micropipette per participant
- 5 micropipette tips per participant
- Food Dye
- Water (for rinsing, does not need to be DI)
- Paper towels
- Small Plastic Cups
- Digital Balance (accurate to 0.001 g)
- Calculator

Part 1: Micropipette Use

Micropipetting: Step-by-Step

1. **Set the Volume:** Adjust the dial on the micropipette to the desired volume. *Ensure the volume is within the range of the pipette!*
2. **Attach a Tip:** Firmly press the micropipette onto a new pipette tip.
3. **First Stop:** Press the plunger down to the *first stop*. This is the amount of air that will displace your desired volume.
4. **Aspirate:** Immerse the tip into the food dye. Slowly release the plunger to draw the liquid into the tip.
5. **Dispense:** Touch the tip to the side of a clean plastic cup. Press the plunger to the *second stop* to expel all the liquid.

6. **Eject the Tip:** Press the tip ejector button to discard the used tip into an appropriate waste container. **Do not reuse tips!**

Part 2: Calculating Expected Mass

The density of food dye is typically 1.025 g/mL. Recall that density = mass/volume. Calculate the expected mass (in grams) for each of the following volumes. Show your work. Remember that 1 μL = 0.001 mL.

- 40 μL = _____ g
- 70 μL = _____ g
- 100 μL = _____ g
- 150 μL = _____ g

Part 3: Measuring Observed Mass

1. Zero the Balance: Place a clean, dry plastic cup on the digital balance. Wait for the reading to stabilize. Press the "tare" or "zero" button to zero the balance.
2. Pipette Food Dye: Using the micropipette, carefully dispense the specified volume of food dye into the tared plastic cup.
3. Record the Mass: Record the mass displayed on the balance in the "Observed Mass" column of the data table.
4. Repeat: Use a *new* or rinsed/dried cup for each measurement to avoid cross-contamination. Repeat steps 1-3 for each volume (40 μL , 70 μL , 100 μL , and 150 μL).

Data Table

Volume (μL)	Expected Mass (g)	Observed Mass (g)
40		
70		
100		
150		

Analysis

1. Calculate the Percent Error: For each volume, calculate the percent error using the following formula:
 - Percent Error = [(Observed Mass - Expected Mass) / Expected Mass] * 100
2. Discuss Sources of Error: What factors might have contributed to any differences between the expected and observed masses?

Extension Activity (AP Biology)

Chi-Square Analysis of Pipetting Accuracy

1. **Null Hypothesis:** State a null hypothesis about your pipetting accuracy. For example: "*There is no significant difference between the expected and observed mass of food dye dispensed by the micropipette.*"
2. **Chi-Square Calculation:**
 - Combine your data with at least two other lab groups to increase your sample size and statistical power. Sum the *Expected* mass values in one column, and sum the *Observed* mass values in another column.
 - Calculate the Chi-Square (χ^2) statistic using the following formula:
3. **Degrees of Freedom:** Determine the degrees of freedom (df).
4. **Critical Value:** Using the Chi-Square Distribution Table (below), find the critical value for your chosen significance level ($\alpha = 0.05$) and degrees of freedom.
5. **Conclusion:**
 - If your calculated χ^2 value is greater than the critical value, reject the null hypothesis. There is a statistically significant difference between the expected and observed masses.
 - If your calculated χ^2 value is less than the critical value, fail to reject the null hypothesis. There is no statistically significant difference between the expected and observed masses.
6. **Alternative Hypotheses (if null hypothesis is rejected):** If you reject the null hypothesis, develop one or two alternative hypotheses to explain the discrepancies observed in your data. What could be contributing to the significant difference?

$$\chi^2 = \sum \frac{(O-E)^2}{E}$$

Chi-Square Table

p value	Degrees of Freedom							
	1	2	3	4	5	6	7	8
0.05	3.84	5.99	7.82	9.49	11.07	12.59	14.07	15.51
0.01	6.64	9.21	11.34	13.28	15.09	16.81	18.48	20.09

Next Steps

1. Clean up your workstation by discarding used pipette tips and used cups.
2. Wash and return any glassware to their appropriate place.
3. Analyze the data you collected and discuss the possible sources of error in a group discussion.