EXERCISE 1 Name ____________________________

How do scientists collect and analyze data?

Objectives

After completing this exercise, you should be able to:

- identify the names and functions of the laboratory items on display in the lab
- explain what a random variable is, and distinguish between discrete and continuous random variables
- use SI units when making measurements in lab
- convert between different SI units
- use the correct number of significant digits when reporting a measurement
- identify the number of significant digits in a reported measurement
- choose an appropriate measuring device when measuring distance, weight, or volume; and use it correctly
- use the correct number of significant digits when reporting computed values
- display data in a well-organized and clearly labeled data table
- use a calculator to calculate the mean, percent error of the mean, and standard deviation of a list of measurements; and explain what these statistics tell us
- use the appropriate statistics to compare the precision and accuracy of different measuring devices
- explain what the Student's t-test is used for, and how to interpret the results
- use a spreadsheet program such as Excel to calculate the mean, percent error of the mean, and standard deviation of list of measurements
- use a spreadsheet program such as Excel to carry out the Student's t-test

Prelab

Before you come to lab, read this entire exercise. You must also answer all questions and complete all assignments on the first 14 pages of this exercise. Your instructor will give you directions on when and where to turn in your work.

Scientific investigation is based on **observations**. Observations refer to any information we collect about the physical universe using our senses (sight, sound, smell, taste, and touch.) The factors or conditions being observed are called **random variables** (or experimental variables) because our observations of them can **vary** from one trial to the next. A random variable can be either **discrete** or **continuous**. For example:

1. A coin is tossed ten times. The random variable X is defined as the number of tails that occur. Because X can only take certain values (0, 1, 2, 3, 4, ….10) it is a **discrete random variable**.

2. The length of a sheet of paper is measured. The random variable Y is defined as its length in centimeters. Because Y can take any positive real value, it is a **continuous random variable**.

Your Turn

Give an example of a discrete random variable: ____

____

Give an example of a continuous random variable: ______________________________________

____

A major advance in science occurred when scientists realized the advantage of describing observations of random variables **quantitatively** (using specific **numbers**) rather than **qualitatively** (descriptions without the use of specific numbers). For one thing, quantitative observations tend to be more objective than qualitative observations. In addition, quantitative observations can be analyzed using various mathematical and statistical techniques that can give us important information about the random variables we are studying, and the exact relationships between those variables.

Your Turn

Give a quantitative description of your weight: ____

____

Give a qualitative description of your weight: ____

____

Is weight a continuous random variable or a discrete random variable? ____________________________

Measuring is simply a way of quantifying our observations of a random variable (i.e. assigning a specific numerical value to each observation) through the use of a **measuring device**. Usually, when we make measurements, each observation is described in terms of BOTH a **number** AND a **unit of measurement**. The information that is recorded to describe our observations (either quantitatively or qualitatively) is referred to as **data**. This lab focuses on a number of important principles and techniques involved in measuring random variables and in analyzing quantitative data.

A. Measuring Random Variables

When measuring a random variable, what units of measurement should I use?

Measurements of a random variable almost always consists of BOTH a number AND a unit. Failure to use appropriate units along with the numerical value makes your measurement incorrect. Scientists use the International System of Units (SI units). SI is the modern version of the "metric system". This system is convenient for several reasons. For one, it is the standard used around the world, which makes it easy for scientists to compare their results with each other. For another, because SI units are related to each other by units of 10, it is easy to convert from one unit to another.

Below is a review of several important SI units and their prefixes.

International System of Units

Random Variable	***Unit***	**A*bbreviation***
Distance	**meter**	**m**
Weight	**gram**	**g**
Volume	**liter**	**L**
Time	**second**	**s**
Amount of a substance	**mole**	**mol**

SI prefixes for units of measurement

Prefix	***abbreviation***	***relationship to basic SI unit***	***means multiply by: (to convert to basic SI unit)***
Micro-	**μ**	**one-millionth**	**10^{-6} or 0.000001**
Milli-	**m**	**one-thousandth**	**10^{-3} or 0.001**
Centi-	**c**	**one-hundredth**	**10^{-2} or 0.01**
Deci-	**d**	**one-tenth**	**10^{-1} or 0.1**
Deca-	**da**	**ten times**	**10**
Kilo-	**k**	**one thousand times**	**1,000**
Mega-	**M**	**one million times**	**1,000,000**

What are the SI units for measuring distance?

The basic unit of distance is the meter (m). You should be able to rapidly convert measurements between meters, centimeters, and millimeters.

One meter (m)	=	100 centimeters (cm)	=	1,000 millimeters (mm)
One centimeter (cm)	=	10 millimeters (mm)	=	0.01 meter (m)

To convert meters to centimeters, multiply by 100. To convert centimeters to meters, divide by 100:

$$3 \text{ m} \times 100 \text{ cm/m} = 300 \text{ cm}$$

and

$$\frac{650 \text{ cm}}{100 \text{ cm/m}} = 6.50 \text{ m}$$

To convert centimeters to millimeters, multiply by 10. To convert millimeters to centimeters, divide by 10:

$$12 \text{ cm} \times 10 \text{ mm/cm} = 120 \text{ mm}$$

and

$$\frac{170 \text{ mm}}{10 \text{ mm/cm}} = 17 \text{ cm}$$

(Notice how the decimal point is moved to the **right** when converting from larger to smaller units, and to the **left** when converting from smaller to larger units.)

Your Turn

Express the following measurements in meters:

340 cm _______________ 23 cm ________________ 4,250 mm ____________

Express the following measurements in centimeters:

8.54 m _______________ 0.0006 m _____________ 390 mm _______________

Express the following measurements in millimeters:

0.0007 m _____________ 0.24 m _______________ 0.62 cm _______________

What are the SI units for measuring weight?

The basic unit of weight is the gram (g). You should be able to rapidly convert measurements between grams, milligrams, and micrograms.

One gram (g)	=	1,000 milligrams (mg)	=	1,000,000 micrograms (µg)
One milligram (mg)	=	1,000 micrograms (µg)	=	0.001 gram (g)

To convert grams to milligrams, multiply by 1,000. To convert milligrams to grams, divide by 1,000:

$$3\ \text{g} \times 1{,}000\ \text{mg/g} = 3{,}000\ \text{mg}$$

and

$$\frac{650\ \text{mg}}{1{,}000\ \text{mg/g}} = 0.650\ \text{g}$$

To convert milligrams to micrograms, multiply by 1,000. To convert micrograms to milligrams, divide by 1,000:

$$12\ \text{mg} \times 1{,}000\ \mu\text{g/mg} = 12{,}000\ \mu\text{g}$$

and

$$\frac{170\ \mu\text{g}}{1{,}000\ \mu\text{g/mg}} = 0.170\ \text{mg}$$

Your Turn

Express the following measurements in grams:

340 mg ______________ 23 mg ________________ 4,250 µg _____________

Express the following measurements in milligrams:

8.54 g ________________ 0.0006 g ______________ 390 µg ________________

Express the following measurements in micrograms:

0.0007 g ______________ 0.24 g ________________ 0.62 mg _______________

What are the SI units for measuring volume?

The basic unit of volume is the liter (L). You should be able to rapidly convert measurements between liters, milliliters, and microliters.

To convert liters to milliliters, multiply by 1,000. To convert milliliters to liters, divide by 1,000:

$$2\ \text{L} \times 1{,}000\ \text{mL/L} = 2{,}000\ \text{mL}$$

and

$$\frac{300\ \text{ml}}{1{,}000\ \text{mL/L}} = 0.300\ \text{L}$$

To convert milliliters to microliters, multiply by 1,000. To convert microliters to milliliters, divide by 1,000:

$$0.600\ \text{mL} \times 1{,}000\ \mu\text{L/mL} = 600\ \mu\text{L}$$

and

$$\frac{1{,}500\ \mu\text{L}}{1{,}000\ \mu\text{L/mL}} = 1.5\ \text{mL}$$

Your Turn

Express the following measurements in liters:

250 μL ________________	1,500 μL_______________	37 mL _______________

Express each of the following measurements in milliliters:

2.56 L ________________	0.0008 L ______________	1,037 μL ______________

Express each of the following measurements in microliters:

0.347 mL _____________	0.00004 L _____________	0.062 L _______________

When reporting a measurement, how many significant digits should I use?

When making measurements, you must report your results using the correct number of **significant digits** (also called significant figures.) The significance of a digit has to do with whether or not it represents a true measurement. Any digit that can be accurately measured or reasonably estimated with the particular measuring device you are using is considered significant.

It is easy to determine the number of significant digits when using a digital measuring device; simply assume that all digits shown on the display are significant. For example, if you are weighing an object on an electronic balance and the display shows 20.00 g, record all of the digits and assume that the balance can accurately estimate the weight to the nearest one-hundredth of a gram. In this case there are 4 significant digits

When using an analog measuring device, the decision is a little more complicated. In this case, the first **estimated** digit should be the last significant digit reported in the measured value. For example if you are measuring temperature with a thermometer where the smallest calibration marks are 5 degrees apart, then the first **estimated** digit would be the nearest whole degree and that would be the last significant digit reported in your value. Such a thermometer could be used to report a temperature of 37 ^{o}C but not 37.0 ^{o}C or 37.00 ^{o}C. On the other hand, if the smallest calibration marks on the thermometer are one degree apart, then the first **estimated** digit would be tenths of a degree and that should be the last significant digit reported in your value. Such a thermometer could be used to report a temperature of 37.0 ^{o}C but not 37 ^{o}C or 37.00 ^{o}C.

How do I determine the number of significant digits in a reported measurement?

To determine the number of significant digits in a reported measurement, we need to look at two cases:

A. Numbers with Indicated Decimals

1. All non-zero digits (1-9) are counted as significant.

2. Only zeros that have non-zero digits somewhere to the LEFT of them are considered significant – all other zeros are place holders

For example, in the value **0.0012010 g**, only the last 2 zeros have non-zero digits to their left and are counted as significant. The first 3 zeros are merely place holders and are not significant digits. Therefore, the total number of significant digits in this value is 5.

B. Numbers without Indicated Decimals

1. All non-zero digits (1-9) are counted as significant.

2. Zeros that have non-zero digits somewhere to the RIGHT of them are considered significant, but there is no way of knowing whether any other zeros are significant

For example, in the value **100,500 g**, the first 2 zeros are significant because there is a non-zero digit to their right. However, we really don't know if the 3^{rd} or 4^{th} zeros are significant. This ambiguity can be avoided by writing the number with a decimal or by using scientific notation. When using scientific notation, all digits before the multiplication sign are considered significant. For example, to write **100,500 g** using scientific notation:

If the 3^{rd} and 4^{th} zeros are not significant, you could write 100.5×10^3 g (4 significant digits)

If the 3^{rd} zero is significant but not the fourth, you could write 100.50×10^3 g (5 significant digits)

If both the 3^{rd} and 4^{th} zeros are significant, you could write 100.500×10^3 g (6 significant digits)

Your Turn

1. You are measuring the migration distance of protein bands in a gel using a ruler where the smallest calibration marks are 1 mm apart. Which of the following measurements have the correct number of significant digits for these measurements (circle ALL correct answers):

   23 mm 23.0 mm 2.3 cm 2.333 cm 2.34 cm 23.64 mm 0.0231 m

2. How many significant digits are there in each of the following measured values?

23.0167 g __________	0.0034 g _______________	45.0021 g ______________
104.50 g ___________	0.0010 g _______________	3.00×10^4 g ___________

When measuring a random variable, how do I know which measuring device to use?

1. Distance

In lab, you will generally use a 15 cm ruler to measure distance. The numbers on the ruler represent centimeters, and the smallest calibration marks represent millimeters

2. Weight

Weight will be measured using **top-loading electronic balances**. These balances have a digital read-out to display the weight of the object, and measure to 2-place accuracy, meaning to the nearest 0.01 g (much less than the weight of a drop of water). Never place the substance being weighed directly on the pan; always use a weigh boat or weigh paper. The tare button will reset the zero point after placing anything on the pan. In order to get accurate measurements, the balances MUST be calibrated each time they are turned on. Electronic balances are expensive instruments that are easily damaged, so treat them with care. They should be kept dry and should not be jarred. Good measurements on an electronic balance are hindered by vibrations, drafts and air currents, and deviations in temperature. Balances should be placed on a level and stable surface, and samples to be measured should be the same temperature as the balance. Examine one of the top-loading balances set out in lab, then carefully read the instruction sheet so you are familiar with the different controls on the balance and the correct way to use them.

3. Volume

You will often be required to measure the volume of liquids in this course. The choice of a suitable device for measuring volume will depend on both the amount of liquid you want to measure, and the level of accuracy needed. In general, you should use the smallest measuring device available that is large enough to hold the volume of liquid you wish to measure. Using a pipet or graduated cylinder over an over again (because it is too small) compounds errors in the measurement each time it is used. Conversely, using a pipet or graduated cylinder that is too big also reduces accuracy. **Accuracy will be greatest when a measuring device is used to measure volumes over 50% of its capacity.**

When you measure water and aqueous solutions in glass pipettes or graduated cylinders, the top surface of the liquid will rise slightly towards the edges, forming a curved surface called the **meniscus.** You should always line up the **lowest** point of the meniscus with the appropriate calibration mark on the measuring device. Make sure that your eye is level with the meniscus, as well, so that the bottom of the meniscus lines up properly to the calibration mark.

a. **Beakers and Flasks –** Beakers and flasks are designed primarily to hold or store liquids, not to measure them. Although these containers sometimes have calibration marks on them, these marks are not as accurate as calibration marks on graduated cylinders or pipettes. Calibration marks on beakers and flasks are generally only “friendly suggestions” of the true volumes, and should not be used to estimate volumes unless a very low level of accuracy is needed.

b. **Graduated Cylinders –** Graduated cylinders come in a variety of sizes, usually ranging from 10 mL up to about 2 L. Remember, accuracy of a device for measuring volume will be greatest when it is used to measure volumes over 50% of its capacity. Accordingly, graduated cylinders are available to appropriately measure volumes between 5 mL and 2 L.

c. **Pipettes –** Pipettes commonly come in sizes of 1 mL, 2 mL, 5 mL, and 10 mL. Therefore, they are suitable for measuring volumes of as little as 0.5 mL and as much as 10 mL.

d. **Micropipettors (or automatic pipettors) –** Micropipettors are generally used to measure volumes of 1 mL or less. Because they measure such small volumes, they are calibrated in microliters (μL) rather than milliliters.

How do I choose the correct micropipettor to use?

When using micropipettors, it is essential to choose the correct size for the job. Three sizes are available:

- pipettors that measure volumes between 2 μL and 20 μL
- pipettors that measure volumes between 20 μL and 200 μL
- pipettors that measure volumes between 100 μL and 1,000 μL

⇒ **IMPORTANT: Never attempt to use a micropipettor to measure a volume beyond the range it was designed to measure.**

Your Turn

Write 2 – 20 μL, 20 – 200 μL, or 100 – 1,000 μL to indicate which micropipettor should be used for each of the following measurements:

50 μL ______________________ 300 μL ______________________

0.015 mL ______________________ 0.6 mL ______________________

1.0 mL ______________________ 0.05 mL ______________________

Test Your Knowledge:

Assume that you have the following measuring devices available:

50 mL beaker	5 mL pipette
50 mL graduated cylinder	100 – 1,000 μL automatic pipettor
25 mL graduated cylinder	20 – 200 μL automatic pipettor
10 mL pipette	2 – 20 μL automatic pipettor

Fill in the name of the device that will most accurately measure each of the following volumes:

0.25 mL ______________________________ 37 mL ______________________________

2.5 mL ______________________________ 0.08 mL ______________________________

21 mL ______________________________ 8.5 mL ______________________________

0.007 mL ______________________________ 0.016 mL ______________________________

0.8 mL ______________________________ 0.04 mL ______________________________

B. Analyzing Quantitative Data

As mentioned previously, one of the major advantages of describing observations **quantitatively** rather than qualitatively is that quantitative observations can be analyzed using mathematical and statistical techniques. Such analysis can give us valuable information about the random variables we are studying, and the exact relationships between those variables. In this part of the Prelab, we will focus on a number of important principles and techniques involved in analyzing quantitative data.

How do I organize data in a table?

Often, one of the first steps in analyzing data is to organize it in a table. A table is an orderly presentation of data aligned in columns and rows. In the table below the data are organized in 2 columns and 6 rows. Tables often help us see relationships between the variables being studied. The table below, for example, clearly shows that as time increases, culture density also increases:

Table 1. Change in cell density of *E. coli* when cultured at 37^{o}C in nutrient broth

Time (minutes)	culture density (cells/mL)
0	100
20	198
40	422
60	796
80	1605
100	3212

⇒ **IMPORTANT: All tables MUST have a descriptive title that explains the data in the table, and each column MUST have a descriptive header complete with the units of measurement.**

Your turn

Suppose you are studying the relationship between surface area and volume, and have done the following calculations: A cube of 1 cm x 1 cm x 1 cm has a volume of 1 cm^3 and a surface area of 6 cm^2 . The ration of surface area to volume is 6.00. A cube of 2 cm x 2 cm x 2 cm has a volume of 8 cm^3 and a surface area of 24 cm^2 . The ratio of surface area to volume is 3.00. A cube of 3 cm x 3 cm x 3 cm has a volume of 27 cm^3 and a surface area of 54 cm^2. The ratio of surface area to volume is 2.00. A cube a 4 cm x 4 cm x 4 cm has a volume of 64 cm^3 and a surface area of 96 cm^2. The ratio of surface area to volume is 1.50. A cube of 5 cm x 5 cm x 5 cm has a volume of 125 cm^3 and a surface area of 150 cm^2. The ratio of surface area to volume is 1.20. A cube of 6 cm x 6 cm x 6 cm has a volume of 216 cm^3 and a surface area of 216 cm^2. The ratio of surface area to volume is 1.00. A cube of 7 cm x 7 cm x 7 cm has a volume of 343 cm^3 and a surface area of 294 cm^2. The ratio of surface area to volume is 0.86. A cube of 8 cm x 8 cm x 8 cm has a volume of 512 cm^3 and a surface area of 384 cm^2. The ratio of surface area to volume is 0.75.

Make a table with four columns and nine rows. The first row is for the column headings that describe the type of information found in each column. Your column headings will be "Dimensions of cube (cm)", "Surface Area (cm^2)", "Volume (cm^3)", and "Surface to Volume Ratio". Fill in your table with the calculations in the previous paragraph. **Make sure your table has a title that makes it clear what kind of information is displayed in the table.**

Which is the easier to study and understand: the paragraph or the table?

____

When reporting a computed value, how many significant digits should I use?

Calculations performed on measurements often produce results with more digits than the original measurements. Suppose, for example, that you use a calculator to determine the average of 3.0, 3.2, 3.3, 3.2, and 3.1 mm, and the result displayed on the calculator is 3.13333 mm. This number implies that the average is accurate to within a hundred thousandth of a millimeter! Unfortunately, your data does not become more accurate simply because you carried out a series of calculations. Therefore, computed values should not have more significant digits than the original data. In this example, the average should be rounded off to 3.1 mm.

1. Rule for reporting the correct number of significant digits when you multiply or divide

When you multiply or divide, your answer should have the same number of significant digits as the number with the fewest. For example, if you want to calculate the volume of a cube by multiplying the length of its sides, it might look like this:

(3.4 mm) x (56.8 mm) x (2.435 mm) = 470.2472 mm^3 (on your calculator)

However, the answer you report should have the same number of significant digits as the measurement with the fewest (3.4 mm), so you should only report 2 significant figures: 4.7×10^2 mm^3.

2. Rule for reporting the correct number of significant digits when you add or subtract

The rules change when you are adding or subtracting numbers. In this case, you look at the number of **decimal places** to determine the accuracy of the measurement. The final answer can have no more digits to the right of the decimal than the number involved in the operation with the fewest digits to the right of its decimal. For example, if you are adding volumes to get a total, it might look like this:

(2.4 mL) + (351 mL) + (0.543 mL) = 353.943 mL (on your calculator)

If you look at the decimal places, however, you will notice that the 351 mL measurement has no digits to the right of the decimal. Therefore, you should report the total volume as 354 mL.

Your Turn

a. Calculate the answer to the following problem and express your answer with the correct units and the correct number of significant digits.

$$\frac{(34.0\ g) \times (0.003g)}{(1.90g) \times (20.0g)} = \_\_\_\_\_\_\_\_\_\_$$

b. Calculate the answer to the following problem and express your answer with the correct units and the correct number of significant digits.

12.098g - 11.6g = __________

How do I use a hand-held calculator to determine the mean, percent error of the mean, and standard deviation of a random variable?

Uncertainty, or experimental error, is always involved when making a measurement. If two people independently took several measurements of a physical quantity such as the weight of an object, it is unlikely that both would come up with exactly the same results every time. Perhaps the instrument used to make the measurements is out of calibration or is influenced by variations in temperature and line voltage. Or perhaps the instrument can't discriminate well between two very similar values. Or maybe personal error, carelessness, or bias is involved. For reasons like these, measurements are never perfect; they are only approximations of some true value that is being measured.

The **accuracy** of a measurement refers to how close the measured values come to the true value. In other words, it is a measure of the correctness of the result. The **precision** of a measurement refers to how variable the measured values are when the same quantity is measured several times. A very precise measurement would be one that did not vary much over several trials, although it may or may not be accurate. In this section we will examine three statistics that scientists commonly use to evaluate the accuracy and precision of their measurements.

Mean

The **mean** is a statistic that is used to express the "average" or "center" of a set of numbers. When the same quantity is measured several times, the mean of the replicated measurements is a more precise measure of the quantity than any of the individual measurements.

You should have a scientific calculator that automatically calculates the mean for a list of numbers. Use the instructions that came with your calculator to learn how to calculate the mean. If you no longer have the instructions, you can use the instructions in Appendix B of this Manual, provided you are using the TI-36X. If you are using a different calculator, it is fairly easy to find instructions for most calculators on the Internet (check the manufacturer's Web site or do a Google search). If you are still having problems, get help from one of the math tutors located in the Tutoring Labs at any ACC campus.

Percent Error of the Mean – used to express accuracy

Accuracy refers to how close a series of measurements are to the true or target value. It is often expressed in terms of the **percent error of the mean**. In general, the closer the measurements are to the true or target value, the lower the percent error of the mean, and the better the accuracy of the measurements. The percent error of the mean is calculated using the following formula:

$$\text{Percent error of mean} = \frac{|\text{calculated mean} - \text{true or target value}|}{\text{true or target value}} \times 100$$

In the formula above, the notation $|$**number**$|$ means "the absolute value of the number." The absolute value refers to the numerical value of a number without regard to sign. Thus, 7 is the absolute value of both +7 and -7.

For example, suppose you want to know if you have a realistic sense of how long one minute is. You check your sense of time by saying "start" and "stop" while a lab partner measures the actual time that passes with a stop watch. Here are your results: 45 sec, 52 sec, 50 sec, 47 sec, and 53 sec. The mean number of seconds that you thought was one minute is 49 sec **(note that the calculated mean cannot have more decimal places than the original measurements.)** The target value is 60 sec. Using the formula for calculating percent error of the mean:

$$\frac{|49 - 60|\ \text{sec}}{60\ \text{sec}} \times 100 = 18\%$$

So, in this case your sense of the length of one minute is 18% inaccurate.

Standard Deviation – used to express precision

Precision refers to how close replicate measurements are to each other. It is often expressed in terms of the **standard deviation**. In general, the less variation there is among the values, the lower the standard deviation, and the better the precision of the measurements.

Use the instructions that came with your calculator to learn how to calculate standard deviation. If you no longer have the instructions, you can use the instructions in Appendix B of this Manual, provided you are using the TI-36X. If you are using a different calculator, you can find instructions for most calculators on the Internet (check the manufacturer's Web site or do a Google search). If you are still having problems, get help from one of the math tutors located in the Tutoring Labs at any ACC campus.

How can I determine how accurately a given volume of water has been measured?

Pure water has a density of 1.00 g/mL at room temperature. Therefore, we can easily convert a given volume of water into its corresponding weight and vice versa. For example, 5 mL of deionized water weighs 5g, and 5g of water has a volume of 5 mL. Because weight can be easily measured to the nearest 0.01g on the electronic balances in lab, the weight of a given volume of water can be used to check how accurately a the volume was measured to the nearest 0.01 mL.

Your turn

NOTE: Make sure you use the correct units and the correct number of significant digits in all of your answers below.

A student uses a 100 mL beaker to measure 80.0 mL of deionized water (dH_2O). She then weighs the water with an electronic balance. Following this procedure 6 times, she obtains the following results: 82.89 g, 80.23 g, 81.27 g, 81.92 g, 82.03 g, and 80.17 g. What is the mean **volume** of water delivered by the 100 mL beaker?

____

During the second part of her experiment, the student repeats her measurements using a 100 mL graduated cylinder instead of a beaker. This time she obtains the following results: 80.79 g, 79.33 g, 80.25 g, 79.82 g, 80.13 g, and 80.19 g. What is the mean **volume** of water delivered by the 100 mL graduated cylinder?

____

What is the percent error of the mean for the measurements made using the beaker? _______________

What is the percent error of the mean for the measurements made using the graduated cylinder? _________

What is the standard deviation ($\sigma_{x\ n-1}$) for the measurements made using the beaker? ___________

What is the standard deviation ($\sigma_{x\ n-1}$) for the measurements made using the graduated cylinder? ________

Which measuring device seems to be more accurate? (Explain your answer.) ____________________

____

Which measuring device seems to be more precise? (Explain your answer.) _____________________

____

What is the Student's t-test?

In the previous section, when comparing measurements made using a beaker and graduated cylinder, we asked, "Which measuring device **seems** to be more accurate?" Why did we use the weasel-word "**seems**"? If one measuring device has a smaller percent error of the mean than another, can't we say it **is** more accurate? Unfortunately, the answer is **no**. To help understand why, imagine that the same student repeated her experiment, only this time instead of using a beaker for the first set of measurements and a graduated cylinder for the second set, she used a single graduated cylinder for both sets of measurements. In this case, would you expect to find exactly the same percent error of the mean for both sets of measurements? Although this is possible, it is extremely unlikely. In fact, if the experiment were repeated many times, chances are that sometimes the first set of measurements would have the lower percent error of the mean and sometimes the second set would. In each experiment, the two sets of measurements, taken with the same graduated cylinder, probably would have different percent errors of the mean because of **chance**, not because the graduated cylinder is actually becoming more or less accurate from one trial to the next.

As another example, let's say you toss a coin 100 times and get 59 "heads". Next, you toss the same coin 100 more times and during the second trial you get only 49 "heads". Why did you get a different result the second time? One possibility is that the difference was simply caused by chance.

So, let's consider the experiment from the previous section where we measured 80 mL of dH_2O using a beaker and a graduated cylinder. If the mean volume measured by the beaker were 82.83 mL and the mean volume measured by the graduated cylinder were 80.13 mL, how would we know if the graduated cylinder is closer to the target value because it is more accurate, or if the difference between the two means was simply caused by chance? **Unfortunately, there is no way to be sure!** No matter how large the difference between the 2 means is, there is no way we can rule out the possibility that it was caused by chance alone. However, what we can say is that the larger the difference between the means, the less likely it was caused by chance alone. In fact, scientists can use statistical tests to determine the **probability** that we would get a difference as large or larger than the one we observed by chance alone. This probability is called the **p value**.

The **Student's t-test** is a statistical test that can be used to test the null hypothesis that the difference between 2 means was caused by chance alone. By convention, if the p value is greater than 0.05 we conclude that the difference between the 2 means is NOT significant (i.e. there is a relatively high probability that it was caused by chance alone.) On the other hand, if the p value is less than 0.01 we conclude that the difference between the 2 mean is highly significant (i.e. there is a very low probability that it was caused by chance alone.) A p value between 0.01 and 0.05 is considered a borderline region, the difference is considered significant but not highly significant. In this case we would probably want to collect more data before we make a conclusion.

IMPORTANT

In science, when we **accept** a hypothesis, this does **NOT** mean we have decided that the hypothesis is correct or that it is probably correct. It simply means that we do not have convincing evidence to show that the hypothesis is wrong. (Just like in our legal system where we assume someone is innocent until proven guilty, in science we **accept** our hypothesis until we have convincing evidence to show it is false.)

For example, let's say we want to test the null hypothesis that when measuring 190 μL of water, there is no difference in the accuracy of the 20 – 200 μL automatic pipettor and the 100 – 1000 μL automatic pipettor. We measure out 190 μL of water 8 times with each instrument and weigh each sample. The mean weight of the water delivered by the 20 – 200 μL automatic pipettor is 0.19 g and the mean weight of water delivered by the 20 – 200 μL automatic pipettor is 0.20 g. The Student's t-test gives a p value of 0.32. Because $p > 0.05$ we conclude that the difference in means is not significant. Therefore, we **accept** our hypothesis that the 2 pipettors are equally accurate. However, **this does not mean that we have decided that the 2 pipettors are equally accurate**; it simply means that our experiment did not provide sufficient evidence to conclude that they aren't equally accurate. Of course, it is quite possible that if we repeated our experiment using more trials or using a more accurate scale (one that could read to the nearest 0.001 g or the nearest 0.0001 g) we would find sufficient evidence to show that one pipettor is more accurate than the other.

How do I use a spreadsheet program such as Excel *to organize data in a table?*

Rather than draw tables by hand and calculate statistics with a hand-held calculator, it is usually easier and faster to organize and analyze your data with a spreadsheet program, such as Microsoft's *Excel*. Like all Microsoft programs, *Excel* has an extensive help feature that is useful for people who are somewhat familiar with the program. However, if you have never used *Excel* before, it is probably more helpful to consult one of the numerous *Excel* tutorials that are available on the Internet. To locate these tutorials, do a Google search for **excel tutorial.** You can add additional terms to your search if you wish to find tutorials on specific features of *Excel*.

To help you learn the features of *Excel* required for this class, three *Excel* quizzes have been posted at the following address:

http://www.austincc.edu/biology/labmanuals/manualsindex.html

Introduction to Excel 1 describes the basic features of *Excel* and explains how to enter data into a table (or spreadsheet) made up of horizontal rows and vertical columns. **Introduction to Excel 1** also explains how to enter formulas into your spreadsheet. You can find another description of how to enter data into an Excel spreadsheet in Appendix B, at the end of this manual.

Your turn

As part of this Prelab, you must complete **Introduction to Excel 1**. To complete this quiz, go to the Web address listed above, click on **Introduction to Excel 1**, and follow the directions.

To complete the quiz you will need a computer that has a copy of *Excel* and an Internet connection. If you do not have a computer with *Excel* and an Internet connection, go to one of the ACC computer labs, or check with your instructor. After you have completed the quiz, save your spreadsheet and email a copy to your instructor. Check with your instructor about the deadline for submitting your quiz.

How do I use a spreadsheet program such as Excel to:

a. determine the mean, percent error of the mean, and standard deviation of a random variable?
b. carry out the Student's t-test?

Introduction to Excel 2 explains how to use *Excel* to calculate statistics such as mean, percent error of the mean, and standard deviation. It also describes the Student's t-test and explains how to use *Excel* to carry out the Student's t-test. You can find another description of how to calculate mean and standard deviation with *Excel* in Appendix B at the end of this manual.

Your turn

As part of this Prelab, you must complete **Introduction to Excel 2**. To complete the quiz, go to the Web address listed above, click on **Introduction to Excel 2**, and follow the directions. After you have completed the quiz, save your spreadsheet and email a copy to your instructor. Check with your instructor about the deadline for submitting your quiz.

Lab Procedures

I. Familiarize yourself with the equipment on display in the lab

Several items that you will be using during the semester are on display in the lab:

Pasteur pipet and bulb	test tube	beaker
blow-out pipet	Erlenmeyer flask	Spec 20 cuvette
delivery pipet	top loading balance	disposable cuvette
transfer pipet	centrifuge tube	Petri dish
pipet filler	Eppendorf tube	squeeze bottle liquid dispenser
micropipettor	Eppendorf tube holder	Parafilm
graduated cylinder	magnetic stir bar	weigh boats

- ♦ Examine each item and make sure you are able to recognize and name it.
- ♦ Make a simple sketch of any unfamiliar items in your laboratory notebook for study purposes.

II. Compare the accuracy and precision of volumetric measuring devices

In this part of the lab, you will use a 100 mL beaker, a 100 mL graduated cylinder, a 10 mL graduated cylinder, a 5 mL pipet, and two different micropipettors (20-200 μL and 100-1000 μL) to measure specified volumes of deionized water. After measuring each volume, you will weigh the water and record the results. Later, you will carry out some statistical tests to compare the accuracy and precision of your measurements.

⇒ **IMPORTANT: Before you begin, copy the following table into your lab notebook so that you can use it to record your results. Data collected during lab should always be recorded directly into your lab notebook, NOT into your lab manual or on loose sheets of paper. Recording data into your lab manual or on loose sheets of paper and then copying it into your notebook increases the chance that the data will be lost or that errors will be made during the transfer.**

Weight of Water Measured by Various Volumetric Measuring Devices

Trial	100 mL beaker (g)	100 mL graduated cylinder (g)	10 mL graduated cylinder (g)	5 mL pipet (g)	20–200 μL pipettor (g)	100-1000 μL pipettor (g)
1	_______	_______	_______	_______	_______	_______
2	_______	_______	_______	_______	_______	_______
3	_______	_______	_______	_______	_______	_______
4	_______	_______	_______	_______	_______	_______
5	_______	_______	_______	_______	_______	_______
6	_______	_______	_______	_______	_______	_______
7	_______	_______	_______	_______	_______	_______
8	_______	_______	_______	_______	_______	_______

⇒ **IMPORTANT: Before you make any measurements, make sure you calibrate your electronic balance. In addition, as you carry out your experiments, make sure every member of your group has a chance to practice using each measuring device.**

A. How accurately and precisely do beakers measure volume?

1. Measure 60 mL of dH_2O using a 100 mL beaker. When making your measurement, be sure the lowest point of the meniscus just touches the 60 mL calibration mark on the beaker. You can use a Pasteur pipette with a small bulb to add or remove water, adjusting the volume as close to 60 mL as possible.

2. Place a large (at least 10 cm x 10 cm) plastic weigh boat on your electronic balance and tare the balance to zero. Carefully pour all of the dH_2O from the beaker into the weigh boat, and record the weight, to the nearest 0.01 g, in your data table. Finally, discard the dH_2O into the waste receptacle provided, place the weigh boat back on the balance, and re-tare the balance.

3. Repeat this procedure seven more times using the same beaker, for a total of eight measurements.

B. How accurately and precisely do graduated cylinders measure volume?

1. Measure out 60 mL of dH_2O using a 100 mL graduated cylinder. When making your measurement, make sure the lowest point of the meniscus just touches the 60 mL calibration mark on the graduated cylinder. You can use a Pasteur pipette with a small bulb to add or remove water, adjusting the volume as close to 60 mL as possible.

2. Determine the weight of the dH_2O measured with the 100 mL graduated cylinder. Follow the same procedure you used to weigh the dH_2O from the beaker. Record the weight, to the nearest 0.01 g, in your data table. Finally, discard the dH_2O into the waste receptacle provided, place the weigh boat back on the balance, and re-tare the balance.

3. Using the same graduated cylinder, measure and record the weights of seven more 60 mL volumes of dH_2O, for a total of eight measurements.

4. Now, repeat the procedure eight more times using a 10 mL graduated cylinder to measure 3 mL volumes of dH_2O. Record the results in your data table.

C. How accurately and precisely do pipets measure volume?

1. Pipet fillers are designed to draw liquid into a pipet and accurately deliver specific volumes. Before you make any measurements, learn how to use the pipet filler by carefully reading the instruction sheet provided. Ask your instructor for help if you do not understand the instructions.

2. Insert the top (blunt) end of a 5 mL blow-out pipet into the pipet filler. The pipette should be attached firmly, but do not push it in too far or it will be difficult to remove when you are done.

3. Immerse the tip of the pipet into a container of dH_2O and practice using the pipette filler to slowly fill and empty the pipet. If you are having difficulty, ask your instructor for help.

   ⇒ **IMPORTANT: Never allow liquid to be drawn into a pipet filler since that could damage the filler and certainly would contaminate the liquid being delivered!**

4. Use the pipet filler to slowly fill the pipet with slightly more than 3 mL of dH_2O. (Note that pipets are usually calibrated in reverse, so that the zero mark is near the top and the calibration marks increase towards the tip. In this case, the calibration marks show how much liquid has been delivered, assuming the pipet was originally filled to the zero line, not the amount the pipet contains. Therefore, if a 5 mL pipet is filled slightly above the 2 mL line, it will contain slightly more than 3 mL.)

5. Raise the tip of the pipet out of the water and use the pipet filler to slowly release dH_2O back into the container until the bottom of the meniscus exactly reaches the 2 mL line on the pipet (i.e. the pipet contains exactly 3 mL of dH_2O). Touch the tip of the pipet to the inside of the container to remove any water on the outside of the tip. You will next transfer this measured volume to a weigh boat. Be sure the pipet remains vertical and no water escapes during this transfer.

6. Place a weigh boat on an electronic balance, tare the balance to zero, and release all 3 mL of dH_2O from the pipet into the weigh boat. **[IMPORTANT: After the water drains out of the pipet, follow the instructions on the instruction sheet to expel the small amount of water remaining in the tip. If necessary, ask your instructor for help.]** After all water has been expelled from the pipet, touch the tip of the pipet to the weigh boat to remove any water suspended from the tip. Record the weight in your data chart and then re-tare the balance.

7. Using the same pipet, measure and record the weights of seven more 3 mL volumes of dH_2O, for a total of eight measurements.

D. How accurately and precisely do micropipettors measure volume?

Three different sized micropipettors are available in lab. The button color and the numbers written on the button indicate their measuring range. Examine each micropipettor, and then fill in the table below:

Button Color	Measuring Range of Micropipettor
____________________	____________________
____________________	____________________
____________________	____________________

When making measurements, you must use the correct size micropipettor. In this exercise, you will be measuring 0.17 mL of dH_2O. Which size micropipettor should you use to measure this volume?

____________________ or ____________________

The following descriptions are for Pipetman micropipettors. Other brands may work differently. Ask your instructor if you are not clear on how to use your micropipettor. To adjust a micropipettor to the desired volume, turn the wheel in the handle until the desired volume, in microliters, is displayed. Although most micropipettors have a 3-digit display, these digits represent different values in different sized micropipettors. For example, if the digits 093 appear in the micropipettor display, this represents different volumes:

2 – 20 µL micropipettor		20 – 200 µL micropipettor		100 – 1,000 µL micropipettor	
display	value in µL	display	value in µL	display	value in µL
0	tens	0	hundreds	0	thousands
9	ones	9	tens	9	hundreds
3	tenths	3	ones	3	tens

If the three different sized micropipettors were adjusted as shown in the table above,

What volume will the 2 – 20 µL micropipettor deliver? ______________

What volume will the 20 – 200 µL micropipettor deliver? ______________

What volume will the 100 – 1,000 µL micropipettor deliver? ______________

⇒ **NOTE: Have your instructor check your answers before you continue with this exercise!**

To help you remember how to read the displays, the 2 – 20 μL micropipettor has a red line between the second and third digits. This line represents a decimal point in the microliters displayed. For example, "200" means 20.0 μL, and this is the largest number that should be displayed because it is the maximum this micropipettor can measure. On the other hand, the 100 - 1000 μL micropipettor has a red line between the first and second digits. This line represents a comma in the microliters displayed. For example, "100" means 1,000 μL, and this is the largest number that should be displayed because it is the maximum this micropipettor can measure.

⇒ **IMPORTANT: Never try to set an automatic micropipettor beyond the range it was designed to measure. If the dial resists turning, STOP TURNING IT! At best, you will get an inaccurate measurement, and you may permanently damage the micropipettor!**

To measure 0.17 mL (170 μL), you can use either the 20 – 200 μL micropipettor or the 100 – 1,000 μL micropipettor. Let's try both sizes and then compare how accurately and precisely they measure 170 μL of dH_2O. We'll begin by making measurements with the 20 – 200 μL micropipettor.

1. Adjust the 20 - 200 μL micropipettor to deliver 170 μL of dH_2O. Ask your instructor to check your work.

2. Next, attach an appropriate plastic disposable tip. These tips are color coded: yellow or white tips are used on micropipettors with a yellow button and blue tips are designed for micropipettors with a blue button. Make sure the tip is attached snugly so it is air-tight.

   ⇒ **IMPORTANT: Always use the micropipettors with a disposable tip attached, and never get any liquid into the micropipettor itself. This would cause permanent damage to the micropipettor!**

3. To get a feel for how a micropipettor handles, grasp the handle of the micropipettor with your thumb resting on the top button. Now slowly push the button down until it meets resistance. This is referred to as the "first stop". If you push down harder past the first stop, the button will move down a little further, until it reaches the "second stop". Now, SLOWLY release the button until it returns to its original position. Practice pressing down on the button and releasing it until you can easily feel the first and second stops.

4. The correct procedure for filling and emptying the micropipettor tips is as follows:

   a. First, push the button down to the FIRST STOP ONLY.
   b. Next, immerse the end of the tip of the micropipettor into the liquid you wish to measure and SLOWLY release the button, drawing the liquid up into the plastic tip.
   c. Lift the tip just above the level of the liquid and gently touch the side of the container with the end of the tip to remove any hanging droplets from the tip.
   d. Insert the tip into the container that will receive the liquid and SLOWLY press the button ALL THE WAY DOWN TO THE SECOND STOP to expel all the liquid from the tip.
   e. Gently touch the side of the container with the end of the tip to remove any hanging droplets, and remove the micropipettor tip before slowly releasing the button again. You have just transferred the correct amount of liquid from the first container to the second.

   ⇒ **IMPORTANT: The button should be pushed down to the FIRST STOP when the tip is being filled, while it should be pushed all the way to the SECOND STOP when the tip is being emptied. This will expel any liquid that sticks to the tip through capillary action. If you use the second stop to fill the tip, your volume will be inaccurately measured!**

   ⇒ **IMPORTANT: Keep your micropipettor upright at all times so that none of the solution in the disposable tip slides down into the micropipettor---this could permanently damage this expensive instrument!**

5. Place a small plastic weighing boat on an electronic balance and tare the balance to zero.

6. Transfer the 170 μL of dH_2O to the weigh boat from your 20 – 200 μL micropipettor. Record the weight in the appropriate column of your table, and re-tare the balance to prepare it for the next measurement.

7. Measure, weigh, and record the weights for seven more 170 μL volumes of dH_2O in the same way, for a total of eight measurements.

8. Now set the 100 – 1000 μL micropipettor to measure 170 μL of dH_2O. The process for measuring and transferring liquid with this micropipettor is the same, except for the color of disposable tips. Make eight measurements with this micropipettor and record their weights in your table.

E. What is the best way to measure 2.7 mL of dH_2O?

You are working in a lab where you need to put 2.7 mL of dH_2O into a test tube. One member of your group thinks it would be best to use a 5 mL pipet to measure this volume. Another member of your group thinks it would be best to use the 100 – 1000 μL micropipettor three times, measuring out 0.9 mL each time. You think it doesn't make any difference. Design and carry out an experiment to see who is correct. Make sure you repeat each method for measuring 2.7 mL at least eight times.

F. How does incorrect use of the micropipettors affect their accuracy and precision?

According to the instructions for using the micropipettors, it is very important to press the button down to the FIRST stop when getting ready to fill the tip, and all the way down to the SECOND stop when emptying the tip. (Note: detailed instructions for using the micropipettors are on the previous page.)

A lab tech working at a medical lab, who did not pay close attention during her cellular and molecular biology class in college, has been using the micropipettors incorrectly. Her procedure is to press the button all the way down to the second stop both when getting ready to fill the tip and when emptying the tip. Design and carry out an experiment to determine how her mistake affects the accuracy and precision of her measurements. Make sure you repeat each method for measuring (correct and incorrect) at least eight times.

Clean up

Wash all regular glassware:

1. Pour contents into an approved container (Ask your instructor if you are unsure of how to discard a particular chemical.)
2. Rinse with tap water.
3. Scrub with soapy water and a test tube brush.
4. Rinse 3 times with tap water.
5. Rinse 3 times with distilled water.
6. Store inverted over absorbent towels until dry.

All disposable glassware goes into the special glass disposal receptacle.

All instruments should be turned off and unplugged.

Wipe off your workspace with a damp paper towel.

Make sure everything that you have used is clean, put away, or discarded. Leave your work area in the same order that you found it in.

Ask your instructor to check your work area before you leave.

Postlab

1. Calculate the mean, the percent error of mean, and the standard deviation for the volumes of water delivered when using the 100 mL beaker, the 100 mL graduated cylinder, the 10 mL graduated cylinder, the 5 mL pipette, and the automatic pipettors. Display your calculated values in a clearly labeled table. Give the table an appropriate title, and be sure to use the correct number of significant digits and appropriate units when reporting your results.

2.
   a) When measuring 60 mL of dH_2O, which measuring device seems to be more accurate, the 100 mL beaker or the 100 mL graduated cylinder? Be sure to **explain** your answer.

   b) When measuring 60 mL of dH_2O, which measuring device seems to be more precise, the 100 mL beaker or the 100 mL graduated cylinder? Be sure to **explain** your answer.

   c) Use the Student's t-test to test the null hypothesis that there is no difference in the accuracy of the 100 mL beaker and the 100 mL graduated cylinder when measuring 60 mL of dH_2O. Be sure to show your p value and explain your conclusions. **Remember, if your data support your hypothesis you should NOT conclude that your hypothesis is correct.**

3.
   a) When measuring 3 mL of dH_2O, which measuring device seems to be more accurate, the 10 mL graduated cylinder of the 5 mL pipet? Be sure to **explain** your answer.

   b) When measuring 3 mL of dH_2O, which measuring device seems to be more precise, the 10 mL graduated cylinder or the 5 mL pipet? Be sure to **explain** your answer.

   c) Use the Student's t-test to test the null hypothesis that there is no difference in the accuracy of the 10 mL graduated cylinder and the 5 mL pipet when measuring 3 mL of dH_2O. Be sure to show your p value and explain your conclusions. **Remember, if your data support your hypothesis you should NOT conclude that your hypothesis is correct.**

4.
   a) When measuring 0.17 mL of dH_2O, which measuring device seems to be more accurate, the 20 – 200 μL micropipettor or the 100 – 1,000 μL micropipettor? Be sure to **explain** your answer.

   b) When measuring 0.17 mL of dH_2O, which measuring device seems to be more precise, the 20 – 200 μL micropipettor or the 100 – 1,000 μL micropipettor? Be sure to **explain** your answer.

   c) Use the Student's t-test to test the null hypothesis that there is no difference in the accuracy of the 20 – 200 μL micropipettor and the 100 – 1,000 μL micropipettor when measuring 0.17 mL of dH_2O. Be sure to show your p value and explain your conclusions. **Remember, if your data support your hypothesis you should NOT conclude that your hypothesis is correct.**

5. Describe the experiment you carried out to answer the question in part E of the lab procedures. Display your results in a clearly labeled table. Explain your conclusions and support them by referencing your data and any statistical tests you performed. **Remember, if your data support your hypothesis you should NOT conclude that your hypothesis is correct.**

6. Describe the experiment you carried out to answer the question in part F of the lab procedures. Display your results in a clearly labeled table. Explain your conclusions and support them by referencing your data and any statistical tests you performed. **Remember, if your data support your hypothesis you should NOT conclude that your hypothesis is correct.**

7. Show the 3 digits that would appear in the display of each micropipettor when it is set to measure its minimum and maximum volumes. (Note: you need to draw 2 diagrams for each micropipettor, one showing the display when it is set to measure the minimum volume and another showing the display when it is set to measure the maximum volume.)