NAME: _____ DATE: _____

PARTNER: _____

INTRODUCTION TO THE BIOLOGY LABORATORY
Measurements, Microscopes and Dichotomous Keys

This exercise is intended to provide the student with a review of some basic methodologies and equipment that will be used in the laboratory over the course of the semester. The activities will include a review of measurement methods and factors of conversion within the metric system. The data collected will then be used to generate tables and graphs to properly present the information that was collected. The student will also become familiarized with the proper use and care of the microscope.

I. MEASUREMENTS & STATISTICS

In the first part of this activity, students will determine their height, compile a table of the combined class data and determine the averages and standard deviations for the male and female students in the class.

A. Data Collection - Height Measurement

This activity will provide an opportunity to make some basic measurements and conversions using the metric system. As we will be measuring height (distance), some relevant terms and conversion factors that we will use during this course include the following:

Measurement	Units per Meter	Other Comparisons
micrometer (μm)	1,000,000.000	(1 millionth of a meter; 1/1000 of a millimeter)
millimeter (mm)	1,000.000	(1/1000 of a meter; 1000 micrometers; 1/10 of a centimeter)
centimeter (cm)	100.000	(1/100 of a meter; 10 millimeters;)
meter (m)	1.000	(1000 millimeters; 100 centimeters; 1/1000 of a kilometer)
kilometer (km)	0.001	(1000 meters)

1. With your partner, determine your height to the nearest 0.5 cm with the meter sticks positioned along the walls of the room.
   a. What is your height in centimeters (cm)? _____
   b. What is your height in meters (m)? _____
2. Record your height (in meters) in the appropriate column (male / female) in the table at the front of the classroom.
3. Record the total class data in the space below.

Female Student Heights (m)	Male Student Heights (m)

B. Data Analysis – Mean & Standard Deviation

In this part of the activity, you will use simple statistics to analyze the class data.

1. Determine the mean (average, $\bar{x}$) for each of the data sets (males vs. females). This is done by taking the sum (Σ) of all of the values (adding them up) and then dividing by the number of individual data points (sample size, n).
   a. What is the average height (in meters) of the female portion of the class? (Use two numerals after the decimal point.) _____
   b. What is the average height (in meters) of the male portion of the class? (Use two numerals after the decimal point.) _____
2. Determine the standard deviation for each of the data sets. This provides a measure of the amount of variation in each of the data sets. The formula is as follows:

$$s = \sqrt{\frac{\sum (x - \bar{x})^2}{n - 1}}$$

s = standard deviation; n = sample size; x = individual sample; $\bar{x}$ = mean

It may be easier to simply use a spreadsheet with a statistical package (such as Microsoft Excel) to perform the calculations. See a tutor or instructor for assistance.

   a. What is the standard deviation of the female heights? _____
   b. What is the standard deviation of the male heights? _____
   c. What can you conclude regarding heights of men and women in your laboratory section? Use your data to support your conclusion. _____

_____

_____

_____

_____

_____

_____

C. Results Presentation - Tables & Figures

In this part of the activity, you will generate data tables and graphs in order to present the data summary (not individual data points) in proper scientific format. Use the handout on graph and table format as a guide. These should be constructed on a computer using a program such as Microsoft Excel. You will likely need to see a tutor or instructor for assistance. Print your table and figure and staple to your packet.

1. Construct a single table showing the mean and standard deviation (SD) values for both the male and female class members. Sample sizes should be noted in footnote.
2. Construct a column graph comparing the mean height of the male and female class members. Include SD error bars on figure and note sample sizes and SDs in the legend.

II. MICROSCOPE CARE AND USE

Throughout the course of this lab, we will use the process of bright field microscopy to observe our specimens. This term refers to the way in which light passes through the specimen relative to the surrounding field. Since the specimen is more dense than its surroundings, it absorbs and reflects some of the light so that it produces an image that appears darker than the surrounding field. The microscopes we will use are also considered binocular compound microscopes since they have two eyepieces and two sets of lenses (oculars and objectives).

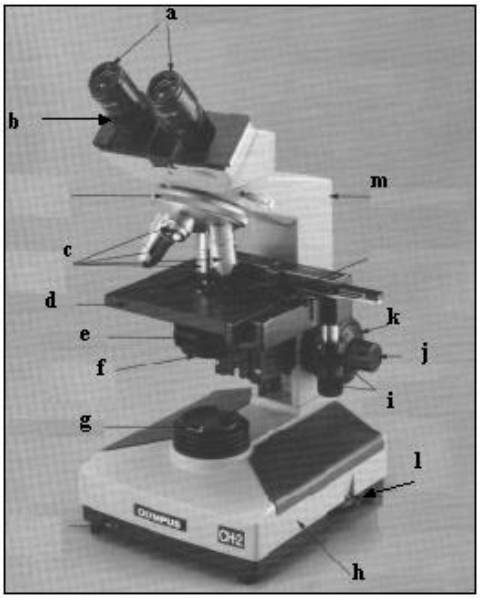
A. General Use and Care

- When carrying, always use both hands and support the microscope by the base and arm.
- Always begin at a low power (4x or 10x) and use the coarse focus adjustment to bring the image into focus. Because these are parfocal microscopes, when moving to higher powers only the fine focus adjustment should be needed.
- The condenser should be positioned at its maximum position (nearly even with stage).
- Adjust the iris diaphragm down to improve contrast.
- If needed, use the cleaning kits supplied (detergent & lens paper) to clean the microscope lenses. To avoid scratching the lenses, only lens paper should be used when cleaning.
- When using oil immersion for the first time on a slide, proceed (focusing at each step) from 10x, 40x, apply oil, 100x. After oil has been applied, be careful not to rotate the 40x objective back through the immersion oil.
- When finished, be sure the stage is centered, the 100x lens has been cleaned of oil and the 4x objective is in the working position. Also, if the head was moved, be sure it is rotated so that the eyepieces extend over the stage.

B. Parts of the Microscope

Use the diagram to locate the following parts of your microscope. Check parts off the list as you locate them.

1. ___ ocular lenses (a)
2. ___ objective lenses (c)
3. ___ diopter adjustment ring (b)
4. ___ iris diaphragm (e)
5. ___ stage (d)
6. ___ condenser (f)
7. ___ light source (g)
8. ___ base (h)
9. ___ light regulator (l)
10. ___ mechanical stage adjustment knobs (i)
11. ___ fine focus adjustment (j)
12. ___ coarse focus adjustment (k)
13. ___ arm (m)



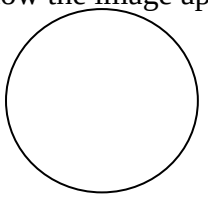
C. Magnification

Magnification refers to the degree to which an object is visually enlarged or enhanced. It is determined by multiplying the power indicated on the ocular lens by the power indicated on the objective lens in use (i.e., magnification = ocular x objective).

1. What is the power of the ocular lenses? _____ x
2. What are the powers of the objective lenses? _____ x, _____ x, _____ x, _____ x
3. What would the <u>total</u> magnification be if you were looking at an object using the 40x objective lens? _____

D. Focusing

1. Adjust the eyepieces so that they are even with the width of your eyes. If this is set correctly, the field of vision should appear as a single circle.
2. Adjust the objectives so that the 4x lens is in the viewing position.
3. Adjust the focus of the ocular lenses for each eye by taking the following steps:
   a. Place a slide of the letter "e" on the stage so that <u>the letter (on the glass portion) is oriented as it would be written</u>. Use the mechanical stage adjustment knobs to position the slide so that the "e" to be viewed is centered in the stage opening.
   b. Place an index card over the <u>right</u> eyepiece and adjust the focus so that the image is in perfect focus for your left eye.
   c. Now cover the <u>left</u> eyepiece with the index card. <u>DO NOT</u> move the focus adjustments! Instead, turn the focus ring (diopter adjustment ring) on the right eyepiece until the image is in clear focus for your right eye.
   d. Look through the microscope with both eyes. You should see a clear image.
4. In the space provided, <u>draw</u> how the image appears under the microscope.



Describe the orientation of this image: _____
_____.

This is called a virtual image. Explain what that means: _____
_____.

Note: this is the same way the lenses in our eyes focus the light reflected off of objects we see.

5. Now slide the 10x objective into viewing position. Because these microscopes are parfocal, the image should be nearly in focus. If not, use <u>only</u> the <u>fine</u> focus adjustment to focus the image. Now try 40x. <u>DO NOT</u> use the 100x objective. This requires a special immersion oil for proper viewing.
6. Obtain one of the other specimen slides on display. Focus on the object using the 40x objective (be sure to start with the 4x or 10x and then move up). Signal your instructor for confirmation that you can locate and focus an object.

specimen _____ , instructor initial _____

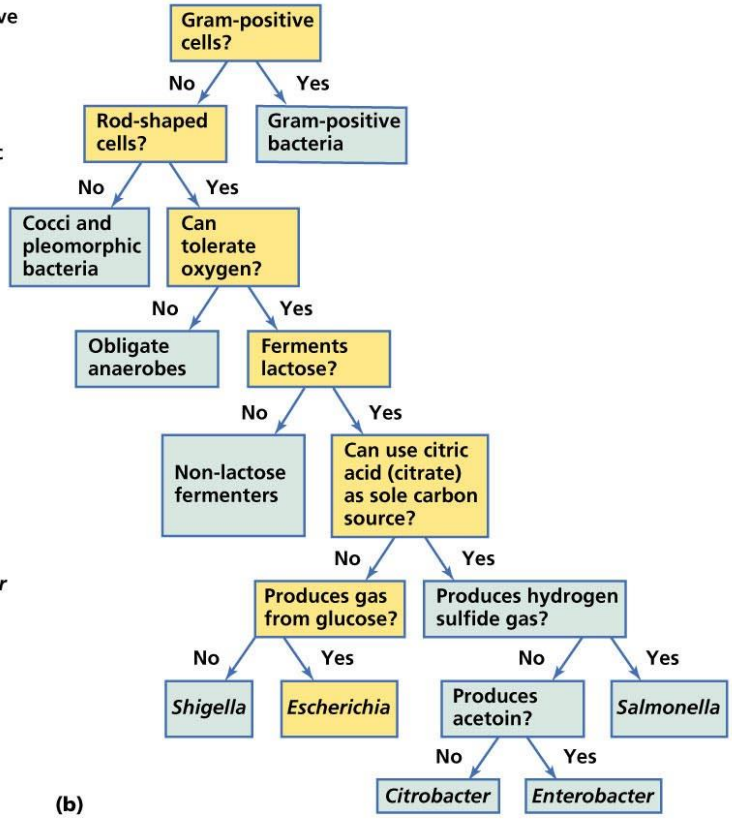
III. **DICHOTOMOUS KEYS**

One of the primary activities that biologists engage in is to classify organisms by species. This includes both newly discovered species and well-described species that are encountered in field work. A dichotomous key is a table or diagram of a series of paired choices. Working through the series of choices leads to identification of a previously-described, but unknown to the researcher, organism. An example of table form, followed by an example of graphical form, is given. You will then develop a dichotomous key to classify a collection of organisms found on the back bench of the lab. Note that each pairing should be based on a visible characteristic – this is an important difference between a dichotomous key and a cladogram. Also, avoid using identification type pairings – identifications should come only once you have narrowed the identification down to one type of organism

A. Examples for identification of bacteria

1a. Gram-positive cells	Gram-positive bacteria
1b. Gram-negative cells	2
2a. Rod-shaped cells	3
2b. Non-rod-shaped cells	Cocci and pleomorphic bacteria
3a. Can tolerate oxygen	4
3b. Cannot tolerate oxygen	Obligate anaerobes
4a. Ferments lactose	5
4b. Cannot ferment lactose	Non-lactose fermenters
5a. Can use citric acid as a sole carbon source	6
5b. Cannot use citric acid alone	8
6a. Produces hydrogen sulfide gas	*Salmonella*
6b. Does not produce hydrogen sulfide gas	7
7a. Produces acetoin	*Enterobacter*
7b. Does not produce acetoin	*Citrobacter*
8a. Produces gas from glucose	*Escherichia*
8b. Does not produce gas from glucose	*Shigella*

(a)



(b)

B. Develop your own dichotomous key

First, make a list of all the types of organisms found that require classification. If you do not know what something is, consult your instructor, or search online to figure it out. Use species names (or group names if the species cannot be determined). Record your list here:

_____

_____

_____

_____

Next, select one characteristic you can use to divide the whole group of organisms into two sub-groups of approximately equal numbers. What characteristic did you use? _____

_____

Record this in the table below, using it to develop your key. Your key should direct readers to go to either segment 2 or 3, depending on the presence or absence of the characteristic you chose.

Select one subgroup, and in segment 2, select another characteristic you can use to create further subgroups. Record as before. The other subgroup should be placed in segment 3 and subdivided similarly. If there are more than one organism in the group, subdivide further, and given instructions to the reader regarding which segment to go to. When you arrive at a point where only one organism is in a subgroup, you have identified that organism. Instead of further instructions, simply write the name of the organism in that blank. You may not need all the blanks, depending on the way you organize your information. If you need additional blanks, add them. When you are finished, check your key against your initial organism list to ensure you have identified all organisms thoroughly. To check your work, ask a classmate to go through your dichotomous key to try to identify all the organisms.

Organism Dichotomous Key

1. A. _____......_____
   B. _____......_____

2. A. _____......_____
   B. _____......_____

3. A. _____......_____
   B. _____......_____

4. A. _____......_____
   B. _____......_____

5. A. _____......_____
   B. _____......_____

6. A. _____......_____
   B. _____......_____

7. A. _____......_____
   B. _____......_____

8. A. _____......_____
   B. _____......_____

9. A. _____......_____
   B. _____......_____

10. A. _____.....
    B. _____.....

11. A. _____.....
    B. _____.....

12. A. _____.....
    B. _____.....

13. A. _____.....
    B. _____.....

14. A. _____.....
    B. _____.....

15. A. _____.....
    B. _____.....

16. A. _____.....
    B. _____.....

Extra credit (1 point): Construct a diagram of your dichotomous key in the space below