Lab Orientation

Principle or Rationale:

Working in a lab is a new experience for many Biology 210A students. All students need to learn about safety in the lab that includes familiarity with the safety rules and equipment, as well as how to behave to keep the lab environment safe for themselves and others.

Laboratory Safety Rules

Safety is the number one priority for work in any laboratory setting to ensure one’s safety and that of others. Below are some rules and procedures for you to adhere to throughout this course.

1. Learn the location of the following pieces of safety equipment and their proper use (note: eyewash does not have drain; for emergency use only):
   - Emergency Eyewash
   - Emergency Water Showers
   - Fire Extinguishers
   - Broom & Dust Pan.

2. Report any & all injuries immediately to the lab instructor.

3. REPORT ANY SPILLS TO YOUR INSTRUCTOR OR THE LAB TECH IMMEDIATELY.

4. Eye protection should be worn in the laboratory when working with potentially dangerous materials. Eye protection shall consist of goggles or glasses with side shields.

5. The laboratory is a place for work & study. Under NO circumstance should there be any horseplay or other hazardous activity in the lab. Failure to comply will result in dismissal from the laboratory.

6. NEVER put any substances or equipment from the laboratory into your mouth.

7. Always work under a fume hood when using substances, which have toxic or harmful fumes. Always use fume hoods when using any procedure, which might produce dense or hazardous fumes.

8. Always proceed in such a manner that laboratory reagents or substances do not contact your skin or clothing. When working with caustic or corrosive materials, lab clothing should be worn. In case of contact with the skin, immediately flush the area with water, using a faucet or the emergency showers. If something gets in the eye, use the emergency eyewash to flush the eye with large amounts of water.

9. Dispose of All organic or hazardous wastes in appropriately marked containers ONLY. If there is any question about where or how to dispose of waste materials, consult your instructor or the lab technician.
10. DO NOT EAT OR DRINK IN THE LAB. SMOKING OR CONSUMPTION OF ANY FOOD OR BEVERAGE IN THE LABORATORY IS PROHIBITED AT ALL TIMES (Actually smoking is not wise any time). Place all food and drink containers beneath your lab table.

11. Wear your SHOES AT ALL TIMES IN THE LAB. No “open-toed” shoes will be allowed.

12. Report any breakage of glassware to your instructor or the lab tech immediately. If the glassware did not contain or contact chemical substances, dispose of it in the appropriately marked container.

13. Dispose of all blood products in the appropriately marked container ONLY. Blood is a bio-hazardous substance. Carefully follow guidelines given by your instructor. When in doubt, please ask your instructor or the lab tech.

14. Wipe down your lab table tops with a mild disinfectant before AND after each lab.

15. Keep all personal items off the lab table during lab. Store your items in the cabinet or under the lab table.

16. In case of a fire alarm, go outside immediately and wait for further instructions.

I, __________________________ (write your name here), have read the rules and safety procedures of this laboratory. I understand them and I will abide by all.
Scientific Method

Learning Objectives
- To distinguish between reductionism and systems biology as two strategies for the study of biology
- To compare and contrast discovery and hypothesis-driven scientific investigations
- To understand the power and limitations of the scientific method and to identify the features of scientific questions it can address.
- To learn the steps of hypothesis-driven investigations: observations, question, hypothesis, experimentation, data gathering and presentation, result analysis and conclusions.
- To define and identify the difference between experimental variables: independent variable, dependent variable, controlled, and uncontrolled variables.
- To apply the scientific method to the study of cardiovascular fitness and to report in writing on the study and its findings as required in a scientific report.

Principle or Rationale:

There are two complementary strategies for the study of biology: Reductionism and Systems Biology. Following a reductionist strategy scientists reduce or break apart the complex biological system into its components for study. For example, in the context of the topic of this course (Cellular and Molecular Biology), to understand the cell one can break it into its sub-cellular compartments and study isolated individual genes, single proteins, or the chemistry and functional biochemistry of one sub-cellular organelle at a time. On the other hand, since life emerges from complex systems with inter-dependent components one can focus on the study of the emergent whole. For example, by genetically or environmentally manipulating the cell one can study the impact on the whole and can understand the interrelations and roles of different cellular components.

Irrespective of the strategy, it is important to always remember that scientific knowledge results from a collection of answers to questions obtained in a specific way, that of the scientific method. In our human quest to understand ourselves and the world around us, we seek answers to many worthy questions, some of which do not lend themselves to investigation using the scientific method and as such are outside the realm of science. For example, despite the eternal human strive to answer two fundamental questions of “who” created the universe and “why”, biological scientist focus on the mechanism of life ranging from its chemical and molecular bases to the global study of the Earth.

Biologists learn about the world around them by gathering information and seeking explanation using the scientific method. Biological scientists conduct two types of scientific investigation or inquiry: discovery science that describes nature and hypothesis-driven science that seeks to provide explanation for biological systems. To arrive at conclusions scientists conducting both types of scientific investigations use logic and reasoning.

**Discovery Science**

**Hypothesis-driven Science**

**Discovery** science involves recording of observations that are either qualitative or quantitative (require measurement) in nature about a novel aspects of Biology that was previously unknown. Examples of discovery science include: observation of chimpanzee behavior in the wild, early studies describing of the cell structure, building databases of DNA sequences, and description of a previously unknown organism.
**Hypothesis-driven** science seeks natural causes and explanations for existing observations. It is conducted following the defined steps of scientific inquiry:

1. Asking a defined **question** based on observation or researching relevant literature
2. Proposing a possible answer or explanation (**hypothesis**)
3. Formulating a **prediction** addressing one experimental variable
4. Designing a **controlled experiment** to test the selected variable
5. Conducting the **experiment** and collecting the **results** (data)
6. **Analyzing** the results
7. Drawing a logical **conclusion** as to whether the results support the prediction and hypothesis

Examples of hypothesis-driven science include: investigation of whether the level of acidity (pH) affects the action of digestive enzymes or whether there is a relationship between heart disease and gender

The following is a comparison between discovery science and hypothesis-driven Science

<table>
<thead>
<tr>
<th>Discovery</th>
<th>Hypothesis-driven</th>
</tr>
</thead>
<tbody>
<tr>
<td>Conducted by few pioneer scientists in subjects about which there is no pre-existing knowledge</td>
<td>Conducted by most scientists who seek explanations</td>
</tr>
<tr>
<td><strong>Steps:</strong></td>
<td><strong>Steps:</strong></td>
</tr>
<tr>
<td>1. Observing a novel phenomenon</td>
<td>1. <strong>Encountering</strong> a specific problem</td>
</tr>
<tr>
<td>2. Recording of data (qualitative or quantitative)</td>
<td>2. Formulating a <strong>question</strong> based on pre-existing knowledge (Scientific Literature)</td>
</tr>
<tr>
<td>3. Analysis of data</td>
<td>3. <strong>Proposing</strong> a hypothesis (possible explanation)</td>
</tr>
<tr>
<td>4. Using inductive reasoning to arrive to a logical conclusion</td>
<td>4. Making a <strong>prediction</strong> statement</td>
</tr>
<tr>
<td>5. Testing hypothesis and prediction by setting-up a controlled experiment manipulating one variable only*</td>
<td>5. Testing hypothesis and prediction by setting-up a controlled experiment manipulating one variable only*</td>
</tr>
<tr>
<td>6. Collecting data</td>
<td>6. Collecting data</td>
</tr>
<tr>
<td>7. Analysis of data</td>
<td>7. Analysis of data</td>
</tr>
<tr>
<td>8. Concluding whether the results do, or do not, <strong>support prediction and hypothesis</strong></td>
<td>8. Concluding whether the results do, or do not, <strong>support prediction and hypothesis</strong></td>
</tr>
<tr>
<td>Using deductive reasoning to arrive to a logical conclusion</td>
<td>Using deductive reasoning to arrive to a logical conclusion</td>
</tr>
</tbody>
</table>

(*) It is important that controlled experiments are not the only means of testing hypothesis. Other means include additional observations, experimentation, and synthesis of information found in and supported by data from other sources.

Scientific knowledge is self-correcting and modification of ideas is subject to technological innovation that often leads to novel experimental tools that can provide for better understanding of biology.

**Considerations for Application of the Hypothesis-Driven Scientific investigation**

In the laboratory, we will focus on applying the scientific method to hypothesis-driven scientific investigations that starts with a **new defined and testable question** about a topic for which a body of previous knowledge, acquired by many others, is available. The elements of the phenomenon under study must be **controllable** and **measurable** within the experimental set-up. One possible explanation or answer to the question, known as a **hypothesis**, is then proposed, which is neither right nor wrong, as all proposed answers are valid. To support or falsify a particular explanation or hypothesis an **experimental set-up** is thoughtfully considered and designed that would lead to a **predicted test result**. Prediction statements are always written using the following format:

**If** the hypothesis is true (**restatement of the hypothesis**), **then** the results of the experiment will show (**introduction of the experiment and the anticipated result**)

A controlled **experimental set-up** is designed after consideration of all experimental **variables** and **level of treatment** that can impact the test measurement, with appropriate positive and negative **controls**, as warranted.

There are four separate variables for consideration:

1. The **independent variable** is the element of the biological phenomenon that is manipulated or
measured against by the scientist to obtain a test measurement.

2. The **dependent variable** is the test measurement made for every change in the independent variable.

3. The **controlled variables** are elements of the biological phenomenon different from the independent variable that can impact the measurement of the dependent variable and are included or excluded from both the test and control of the experimental set-up.

4. The **uncontrolled variables** are elements of the biological phenomenon different from the independent and controlled variables that can impact the measurement and cannot be included or excluded from both the test and control of the experimental set-up.

In the design of any experiment, the procedure and measurement are repeated multiple times the results are averaged before conclusions are made. No meaningful conclusions can be made using results that cannot be reproduced by the original investigator and others who may repeat the experiment under the same experimental set-up.

**Lab Procedure**

I. **Application of the Scientific Method to the Study of Cardiovascular Fitness**

Your entire class will be divided in teams to conduct a single scientific investigation relating to cardiovascular fitness using a hypothesis-driven study that is built on previous knowledge of the topic.

**Introduction - Background and known scientific facts about cardiovascular fitness**

Cardiovascular fitness refers to the ability of the heart to use its muscle tissue efficiently without excessive strain under conditions of higher oxygen demand. Under resting conditions with no exercise, each person has a resting heart beating rhythm known the resting heart rate (RHR) that is defined as the number of beats per minute (BPM). Physical exercise creates a higher demand for oxygen and as a result the heart beats faster to accommodate the demand. The level of cardiovascular fitness level can be assessed based on two measures: 1) increase in HR after exercise and 2) how quickly the heart recovers and goes back to RHT after exercise. The heart rate of individuals with high cardiovascular fitness shows a lower increase in BPM and a faster return to normal heart rate after exercise than others with lower cardiovascular fitness.

The **step test** is a simple exercise that is commonly used to measure cardiovascular fitness. It starts with determining the resting heart rate of the subject(s) undergoing the test. In this test, individuals step on a low platform above the ground at a constant rate of 30 steps per minutes, for 3 minutes.

**Formulating a question for investigation**

1. Discuss with your team members factors that may impact cardiovascular fitness such as cigarette smoking etc…. Enter in the provided space two factors that you would like to investigate

2. Your instructor will work with the whole class to determine which factor will be investigated. Follow your instructor’s direction.

Write down the question that the class will address

**Proposing a hypothesis**

Write down the simple answer to the question.

The simple answer to the question above is **your hypothesis**.

**Formulating your own prediction**
Based on your hypothesis and what you know about cardiovascular fitness and the step test from the introduction above, fill in the blanks of the following prediction statement:

If _______________ have a higher cardiovascular fitness than _______________, then they will show _______________ and _______________ after _______________.

**Experimental set-up to test the hypothesis and prediction**

1. The first step of the staircases outside the lab side of the building will be used as the low platform.
2. Each team will have two subjects that will perform one step test to collect data regarding increase in heart rate and return time to resting heart rate. Enter the original data of both measures in table 1, below.
3. You will be performing one experimental procedure to measure the two variables.
4. Measure the RHR of both subjects while sitting quietly by counting the pulse rate for 30 second then multiply by 2. Enter the results in Table 1. You can use either the artery in the wrist above the thumb or the artery in the neck (Follow your instructor’s directions)

5. Proceed with the step test using the following procedure and timing
6. Record, in Table 1, the heart rate of your team's subjects exercise a 0 minutes after 3 minutes.
7. Continue measuring HR in subjects until both regain their resting HR. Record the number of minutes in Table 1. (You don’t need to enter the actual heart rate in the table).

**Table 1. Group Step Test results**

<table>
<thead>
<tr>
<th></th>
<th>Pulse Rate (Beats per Minute)</th>
<th>Recovery time to resting HR (in minutes)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Resting- Before step test</td>
<td>After step test</td>
</tr>
<tr>
<td>Subject 1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Subject 2</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

8. Enter your team’s data in Tables 2 and 3, calculate the % increase in HR after step test, and fill-in the appropriate cells.
9. Explain why calculation of % increase in heart rate is necessary.

Table 2. Class results of increase in HR after Step Test for individuals designated as subjects 1.

<table>
<thead>
<tr>
<th>Subject</th>
<th>Pulse Rate (Beats per Minute)</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Resting HR- Before step test</td>
<td>HR After step test</td>
<td>Change in heart rate (HR after test- resting HR)</td>
<td>% increase in HR (Change in HR/resting HR x 100)</td>
</tr>
<tr>
<td>Group 1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 3</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 4</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 6</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 3. Class results of increase in HR after Step Test for individuals designated as subjects 2.

<table>
<thead>
<tr>
<th>Subject</th>
<th>Pulse Rate (Beats per Minute)</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Resting HR- Before step test</td>
<td>HR After step test</td>
<td>Change in heart rate (HR after test- resting HR)</td>
<td>% increase in HR (Change in HR/resting HR x 100)</td>
</tr>
<tr>
<td>Group 1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 3</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 4</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Group 6</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

10. Enter your team’s data of HR recovery time in Table 4.

Table 4. Class results of HR recovery time after Step Test for individuals designated as subjects 1
and 2.

<table>
<thead>
<tr>
<th>Subject 1</th>
<th>Recovery time to resting HR (in minutes)</th>
<th>Subject 2</th>
<th>Recovery time to resting HR (in minutes)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1</td>
<td>Group 1</td>
<td>Group 1</td>
<td></td>
</tr>
<tr>
<td>Group 2</td>
<td>Group 2</td>
<td>Group 2</td>
<td></td>
</tr>
<tr>
<td>Group 3</td>
<td>Group 3</td>
<td>Group 3</td>
<td></td>
</tr>
<tr>
<td>Group 4</td>
<td>Group 4</td>
<td>Group 4</td>
<td></td>
</tr>
<tr>
<td>Group 5</td>
<td>Group 5</td>
<td>Group 5</td>
<td></td>
</tr>
<tr>
<td>Group 6</td>
<td>Group 6</td>
<td>Group 6</td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td>Average</td>
<td>Average</td>
<td></td>
</tr>
</tbody>
</table>

**Conclusion statement**

________________________________________________________________________
________________________________________________________________________
________________________________________________________________________

14
The Scientific Method Report

What was the question of the lab exercise? ____________________________________________

Hypothesis: (General Statement)
________________________________________________________________________________

Prediction:
________________________________________________________________________________

Experimental Design:
While conducting your investigation of cardiovascular fitness, what were the two Dependent Variables?
________________________________________________________________________________

List:
- The Independent Variable(s) of your study? __________________________________________
- Two controlled variables (list 2): _________________________________________________
- Two uncontrolled variables (list 2): _______________________________________________
- Level of treatment:
  Activity ___________________________ Duration _____________________________

Number of replicates:
a. # of subjects/team __________
b. # of times the experiment was conducted by the whole class ________________

Were there control groups in this experiment? Why or why not _________________________

Present your data as a figure in graph form (Choose a graph format that best describes your conclusion).
Attach 2 graphs:
Where did you plot the dependent variable(s) on the graph?
Where did you plot the independent variables on the graph?

Discussion & Conclusion: (Your graph should illustrate your conclusion).
Start by presenting the rationale behind your study. Restate your hypothesis and prediction. Summarize your results. State whether the results you obtained did or did not support your hypothesis? End with a concluding single sentence that is the answer to your original question. (You can use a separate sheet to continue this section)
We investigated ...........