Chapter 1c: Exploring Life
(section 1.5, first lab)

Experimentation

- Asking Good Questions
- Forming Hypotheses
- Testing Hypotheses

- As you can see by our continued emphasis of this third section on “testing hypotheses,” a great deal of a scientist’s time is spent hypothesis testing rather than hypothesis making
- This is why the caricature of scientists is a person in a lab coat working at a bench

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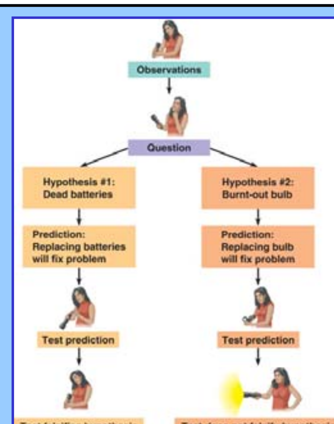
- As you can see by our continued emphasis of this third section on “testing hypotheses,” a great deal of a scientist’s time is spent hypothesis testing rather than hypothesis making.
- This is why the caricature of scientists is a person in a lab coat working at a bench.
- "The most creative aspect of science is designing a test of your hypothesis that will provide unambiguous evidence to falsify or support a particular explanation…
- …Scientists often design, critique, and modify a variety of experiments and other tests before they commit the time and resources to perform a single experiment."

Experimentation



Variables

- To understand how to successful perform experiments, one must understand how to successfully handle (and describe) variables
- A poorly run experiment often is as much a consequence of poor technique as a consequence of failure to properly identify and control variables
- We can divide variables into three general types:
  - Independent Variables
  - Dependent Variables
  - Controlled Variables
- Your goal will be to distinguish between and otherwise not confuse these three variable types

Dependent Variable

- Within an experiment, the *dependent variable* is that which is being measured, the variable that you hypothesize will change as a function of change in one or more independent variables
- Very often varying an independent variable will result in more than one measurable change in the system
- Consequently, it is routine for an experiment to have more than one possible *dependent variable*, though doing so will often increase the size and complexity of an experiment

Employing Dependent Variables

- If we were determining the effect of sunshine on plant growth, then some measure of plant growth would represent the *dependent variable*
- The hypothesis in this case may have been something to the effect of, "Exposure to sunshine causes plants to grow."
- Note that in order for this to be an effective experiment, the *dependent variable* must be measurable, the more precisely the better
- This need for measurability is what makes the "supernatural" off limits to science since the supernatural, by definition, is not a measurable quantity

Independent Variable

- The *independent variable* is that measure that is being purposefully varied in the course of an experiment to test whether such variation results in a change in the dependent variables
- For example, if we were determining the effect of sunshine on plant growth, then the degree to which we exposed a plant to sunshine would represent the *independent variable*
- The dependent variable need not vary with the *independent variable*
- Such a failure to vary with the *independent variable* would be termed a negative experimental result

Using Independent Variables

- An *independent variable* must vary over some range, preferably in a well-controlled or easily measured manner
- Note that it is possible to have more than one *independent variable* in an experiment, though using more than one *independent variable* typically increases the complexity of an experiment, or its size.
- On other hand, you may be able to save time by essentially doing more than one experiment simultaneously
- Having more than one *independent variable* typically is used as a productivity trick for those who are well practiced at a given protocol

Independent-Variable Limitations

- The requirement for control or measurement of *independent variables* limits the types of things that can be used as an *independent variable*
- The "supernatural" cannot serve as an *independent variable* by merely natural entities
- "Dinosaurs became extinct because a supernatural power was dissatisfied with their progress" therefore is not a very good hypothesis because, though we can measure the extinction of dinosaurs (the dependent variable), we cannot measure a supernatural power (the *independent variable*)
- We can also reject this hypothesis simply on the grounds that the term "progress" is too ambiguous:
- Just what is progress if the dinosaurs, in all of their glory, did not satisfactorily progress?

Level of Treatment

- *Treatment* refers to the independent variable
- Recall that the independent variable may be something that is applied, but also may be something that is inherent to the system and therefore only measured
- When considering *level of treatment*, be prepared for "treatments" that involve no treatment on the part of the experimenter
- That is, “treatment” is being used in a sense that differs from one’s usual understanding of “treatment.”
- Similarly, scientists use the word “theory” in a sense that is different from the lay understanding of the word.
- *Level of treatment* can be difficult to grasp due to the prejudices one brings to the table about the meaning of the term "treatment."
- *Level of treatment* is simply the degree or manner in which the independent variable is varied

Different Levels of Treatment

- Differences in drugs (or a drug versus a placebo).
- Differences in dates in the past (e.g., 50 million years ago versus 25 million years ago versus the present).
- Nothing at all (e.g., 50 minutes of doing nothing versus 90 minutes of doing nothing).
- Differences in what was sampled (e.g., an apple versus an orange).
- Different populations, etc.

Independent & Dependent Variables

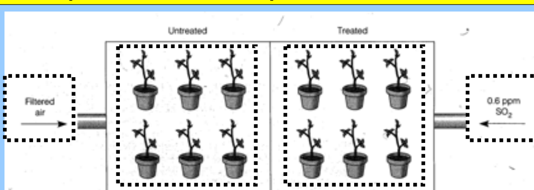


Figure 1.1.
Experimental design for soybean experiment. The experiment was repeated four times. Soybeans were fumigated for 4 hours.

What are the Levels of Treatment?
Dependent Variables include, e.g., # pods, # seeds/pod, pod weight

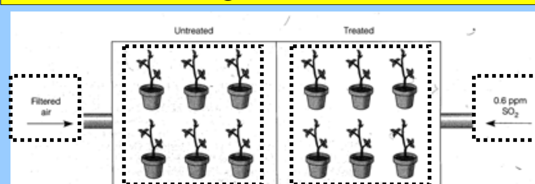
Controlled Variables

- The real world is a messy place
- Consequently, there typically exist many more variables within an experiment than just those we designate as the dependent variable and the independent variable(s)
- If we would like to see our experiment work (i.e., give us unambiguous results), we had better make sure that the only variables that actually vary are the independent variable(s)…
- …and should it vary with the independent variable(s), then the dependent variable as well
- These *other* variables that we attempt to keep from varying are called *controlled variables*

Controlling Controlled Variables

- Controlled variables are potential independent variables that, by design, are not varied in the course of an experiment
- Note that it is not always easy to determine and control all of the variables we would like to be our *controlled variables*
- One of the key difficulties (and challenges) of experimental science is designing and performing experiments in such a way that we keep our *controlled variables* actually under our control
- "The underlying assumption in experimental design is that the selected independent variable is the one affecting the dependent variable. This is only true if all other variables are controlled." your lab text

Accounting for all Variables



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Experimental design for soybean experiment. The experiment was repeated four times. Soybeans were fumigated for 4 hours.

What are the Controlled Variables?

The Protocol / Procedure

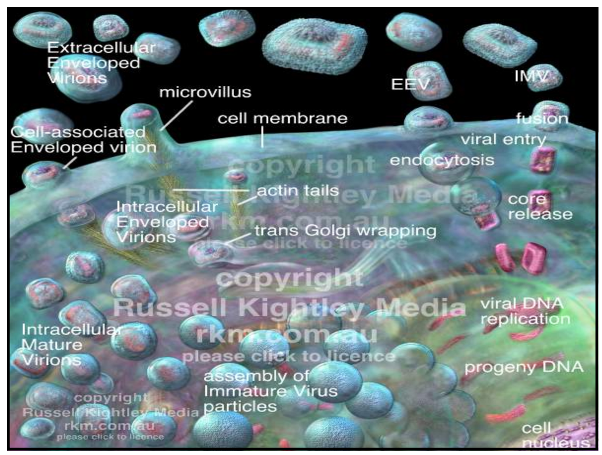
- "The **procedure** is the stepwise method, or sequence of steps to be performed for the experiment. It should be recorded in a laboratory notebook before initiating the experiment, and any exceptions or modifications should be noted during the experiment…
- The procedures may be designed from research published in scientific journals, collaborations with colleagues in the lab or other institutions, or by means of one's own novel and creative ideas. The process of outlining the procedure includes determining control treatment(s), levels of treatments, and numbers of replications." your lab text

How Much Detail is Enough?

- The *protocol* is the means by which an experiment is run
- The *protocol* must include all relevant details such that the original experimenter or others can repeat the experiment some time in the future
- In practice, however, often much is left unsaid in a given protocol with much knowledge on the part of the reader assumed
- Consequently, two individuals performing the "same" protocol do not always succeed in repeating each other's results
- In reality there is an art to including all "relevant" detail in a less than fully comprehensive protocol
- Minimally, one should write down what one did in a manner that will allow one to repeat the protocol some time in the future
- The more comprehensive the protocol, the less knowledge assumed of the reader

Replication

- "Scientific investigations are not valid if they base their conclusions on one experiment with one or two individuals. Generally, the same procedure will be repeated several times (**replication**) providing consistent results.
- Notice that scientists do not expect exactly the same results inasmuch as individuals and their responses will vary. Results from replicated experiments are usually averaged and may be further analyzed using statistical tests." your lab text



Replication Independence

- *Replications* can be performed that are more or less independent of one-another (in a statistical as well as actual sense of the term independent)
- Maximal independence is achieved by two different individuals performing two different experiments in two different laboratories, etc.
- Minimal independence is achieved, for example, by taking two measurements of the same variable from the same procedure during the same performance of that procedure (e.g., checking the temperature twice before writing it down)
- The latter guards against spurious mistakes while the former guards against more systemic errors
- Obviously, considerable independence between replications may be achieved without having your out-of-town buddies repeating your work for you

Control Treatment

- The *control treatment* can be as confusing as the concept of a level of treatment
- The *control treatment* represents what can be merely an arbitrarily chosen level of treatment (often zero if that is possible)
- This *control treatment* is used for comparison with other levels of treatment
- Sometimes *control treatments* are not used at all, though typically this is because two or more levels of treatment are being compared
- Including a"control treatment" *in these cases* would simply involve the semantics of calling one arbitrarily chosen level of treatment the *control treatment*

Control Treatment

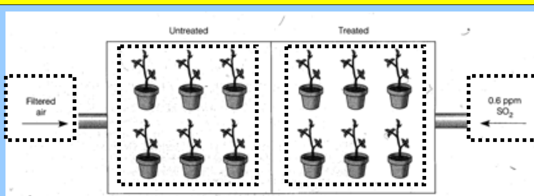
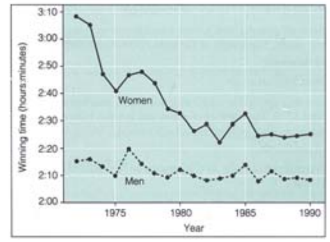


Figure 1.1.
Experimental design for soybean experiment. The experiment was repeated four times. Soybeans were fumigated for 4 hours.

Which is the Control Treatment?

Which is the Control Treatment?

Figure 1.5.
Winning times for the Boston Marathon for men and women from 1972 to 1990. (After Gould, 1996.)



Good question!

Controls

- In addition to the control treatment we can speak of controls
- One of the hardest things to do in designing experiments is including the proper controls
- Typically we distinguish controls into positive controls and negative controls
- Note that it is often quite possible to have more than one positive control and more than one negative control

Positive Control

- A *positive control* is a level of treatment that is expected to result in a change in the value of a dependent variable
- The purpose of the *positive control* is to serve as proof that the experiment can produce a positive result, i.e., a change in the value of a dependent variable
- If a *positive control* is not included in a protocol then *lack of change* in the value of a dependent variable may be due to:
  - A negative result
  - A protocol that is not capable of producing a positive result (a.k.a., systematic error)
  - Experimental error
- Experimental error could occur for individual measurements rather than for the entire experiment
- This is why one does more than one replication

Negative Control

- The *negative-control* level of treatment often corresponds with the control-treatment level of treatment
- The *negative control* is supposed to result in a lack of change in the dependent variable (or some baseline display value)
- The *negative control* also serves as proof that a given protocol is capable of giving baseline results
- We will know that a given treatment produces a change in the dependent variable only if we have something to compare the resulting value to

Negative & Positive Control

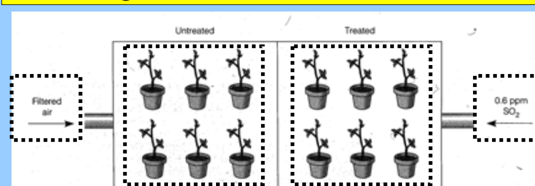


Figure 1.1.
Experimental design for soybean experiment. The experiment was repeated four times. Soybeans were fumigated for 4 hours.

Which is the Negative Control?

Which is the Positive Control?

Where is the Negative Control?

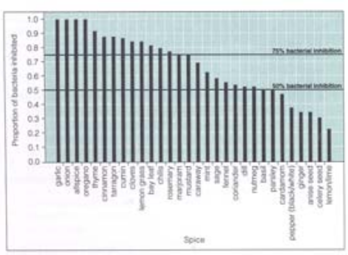


Figure 1.6. Bacterial inhibition by 30 spices tested in the laboratory. The number of bacterial species tested ranged from 4 to 31. (After Sherman and Billing, 1999.)

Prediction

- "The investigator never begins an experiment without prediction of its outcome. The **prediction** is always based on the particular experiment designed to test a specific hypothesis…
- Predictions are [formally] written in the form of if/then statements: 'If the hypothesis is true, then the results of the experiment will be…';...
- Making a prediction provides a critical analysis of the experimental design. If the predictions are not clear, the procedure can be modified before beginning the experiment...
- To evaluate the results of the experiment, the investigator always returns to the prediction. If the results match the prediction, then the hypothesis is supported. If the results do not match the prediction, then the hypothesis is falsified. Either way, the scientist has increased knowledge of the process being studied." your lab text

Negative Results

- Typically a *negative experimental result* is simply a result that is inconsistent with one's hypothesis and prediction
- Alternatively, a negative experimental result is no change in the dependent variable as a function of the independent variable
- Note that in the case of a hypothesis that predicts no change, a *negative experimental result* in the latter sense, could be a positive experimental result in the former sense
- Yes, this is confusing (but so is doing science)
- Note that an ambiguous result is *not* a negative result—instead it means that you don't know what the heck is going on
- Negative results, therefore, are preferable to ambiguous results—at least one can learn something from a negative result

Other Kinds of Results

- A *positive experimental result* is positive in terms of one's hypothesis and predictions and/or in terms of a change in the dependent variable as a function of the independent variable
- An *ambiguous experimental result* is one in which you cannot determine whether the experiment gave positive or negative experimental results
- (Time to go back and either repeat, or redesign and then repeat the experiment)
- A "Good" experimental result is a result that is *not ambiguous*
- Good experimental results are results that allow you to avoid wasting your time, even if they suggest that your cherished hypothesis is garbage

Exercise 1.3

Let's take some time
to read Exercise 1.3.

Who doesn't have a lab text yet?

Cardiovascular Fitness

- "Cardiovascular fitness can be determined by measuring a person's pulse rate and respiration rate before and after a given time of aerobic exercise…
- A person who is more fit may have a relatively slower pulse rate and a lower respiratory rate after exercise, and his or her pulse rate should return to normal more quickly than that of a person who is less fit…
- Your assignment is to investigate the effect of a well-defined, measurable, controllable independent variable on cardiovascular fitness." your lab text

Specific Questions

As a class, let's come up with questions concerning Cardiovascular Fitness

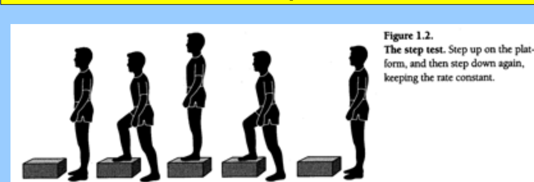
For example:
Is cardiovascular fitness greater in athletes than in nonathletes?

Associated Hypotheses

As a class, let's come up with hypotheses concerning Cardiovascular Fitness

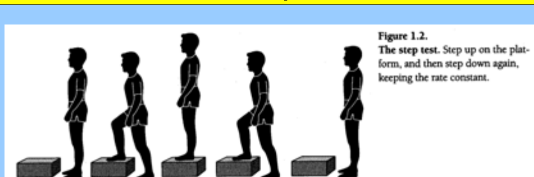
For example:
Athletic training improves cardiovascular fitness

The Step Test



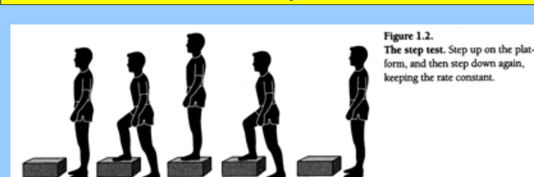
"The subject steps up and down on a low platform, approximately 8 in. from the ground, for 3 minutes at a rate of 30 steps per minute. (Using a metronome to count steps ensures that all subjects maintain a constant step rate.) The subject should step up and then step down again, keeping the rate constant."

The Step Test



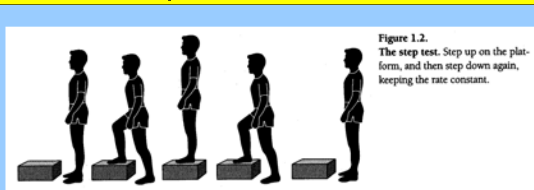
"The subject's pulse rate is measured before the test and immediately after the test. The subject should be sitting quietly when the pulse is counted. Use three fingers to find the pulse in the radial artery (the artery in the wrist, above the thumb). Count the number of beats per minute. (Count the beats for 30 seconds and multiply by 2.)"

The Step Test



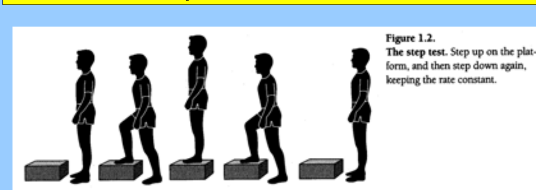
"Additionally, the pulse rate is measured at 1-minute intervals after the test until the pulse rate returns to normal (recovery time). Count the pulse for 30 seconds, rest 30 seconds, count 30 seconds, and rest 30 seconds. Repeat this procedure until the pulse returns to normal. Record the number of minutes to return to the normal pulse rate. (Do not record the pulse rate.)"

Dependent Variable



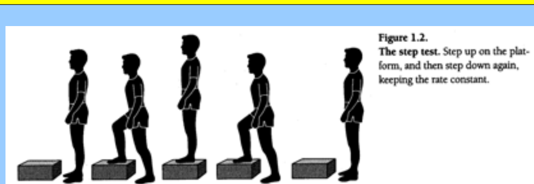
What is/are the Dependent Variable(s)?

Independent Variable



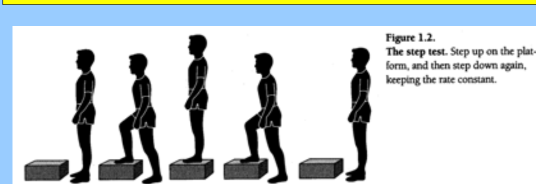
What is the Independent Variable?

Controlled Variables



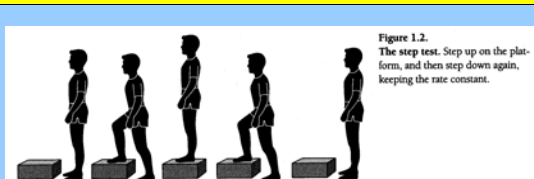
What are the Controlled Variables?

Controls



Are there any Controls?

Level of Treatment



What is/are the Level(s) of Treatment?

Prediction

For example:
If athletic training improves cardiovascular fitness, then the pulse rate of athletes will be lower after exercise and will return to normal more quickly than the pulse of nonathletes

Succeeding in Bio 113

1. Exam questions come from the notes that I hand out; *study* anything else (for the exam) and you are not being efficient
2. Reading is not necessarily the equivalent of studying.
3. Putting in more time is not necessarily as important as studying well, efficiently, or effectively (recall doing science with *rigor*)
4. You are not putting in a lot of time in until you are putting in excess of ~20 hours/week to studying biology/attending class and lab
5. If you don't learn/understand the material before going on to the next topic/material, when will you learn the old material?
6. Organizing the material is not equivalent to studying for the exams (though certainly it helps you prepare for studying)

Succeeding in Bio 113

7. If you want to do well, you must learn the majority of the material really, really well
8. Try triaging, i.e., concentrate on learning and memorizing that material that (a) you don't know/have memorized and (b) have some reasonable probability of learning
9. Part of studying for the exam should involved IDing that material to concentrate your studying on
10. If you can't at least make a reasonable attempt at knowing the material to the point where you can recite it from memory, then you are not doing an adequate job of studying for an exam
11. Don't put off learning the material until the night before the exam
12. Studying is not easy, no way, no how (and that's why you get summers off)

Succeeding in Bio 113 Lab

- Your lab grade in Bio 113 is based on two lab exams
- These exams are open lab text
- To do well you need to know your lab text
- Read assigned material, answer questions asked during reading, answer assigned questions, perform assigned tasks
- Your goal should be to conscientiously do the work, taking the time allotted during lab periods to get the work done
- By all means ask questions and do take notes.
- (Preferably take your notes in your lab text since that is what you will have with you during lab exams—not your lecture text, or other notes)
- Oh yes, and do bring in interesting CDs so that we can provide some sonic filler during labs that isn't based entirely on my own music collection

How Hard is Bio 113, Really?

- On a scale of 1 to 10, where intro Biology for non-majors is a 1 (I.e., Bio 101) and Organic Chemistry is a 10 (I.e., Chem 251), Bio 113 is something like a six or a seven
- In some ways Bio 113 is harder than Chem 121, in other was Bio 113 is easier than Chem 121
- Bio 113 is much, much harder than Chem 101 or Chem 102 (or Bio 101 or Bio 102)
- Bio 113 is harder even than Micro 509 (another course I teach)
- Bio 113 will bring most of you to your knees unless you keep on top of the material (and, in some cases, even if you do keep on top)
- If you can succeed at Bio 113, you will be well prepared to succeed at most higher-level biology courses

The End