**Questions, Hypotheses, and Experimental Design**

Purpose

* Experiments are usually conducted with the intention of answering one or more **questions**; typically, these questions are “*does* a phenomenon occur?” or “*why* does a phenomenon occur?” For example: “does ambient temperature have an effect of the metabolic rate of organisms?”
* In order to design an experiment that will successfully test the question, scientists develop a **hypothesis** (a testable prediction). A hypothesis predicts the relationship between at least one *independent variable* and at least one *dependent variable*.
  + The **independent variable** refers to the variable that you think will *cause* an effect. It is the variable that you change. For example: if the hypothesis predicts that increasing temperature will result in an organism having heightened metabolism, then temperature is the independent variable.
  + The **dependent variable** refers to the variable that you think will *be affected* by the independent variable. It is the variable that you measure. In the above example, metabolic rate is the dependent variable.
* An experiment must be designed to address the hypothesis; in a **controlled experiment**, you manipulate the independent variable(s) and record data on the dependent variable(s), whereas in an **observational experiment**, you record data on all variables.

The Question

* The question is the beginning to any scientific study; it determines the type of hypothesis that you can come up with, which then determines the experimental design.
* Questions should always be informed by current knowledge in the topic you want to address. As such, you will need to review current scientific literature. Questions typically fall into four categories with respect to current knowledge:
  + You can address gaps in current knowledge: what have other studies not addressed, or have only addressed incompletely?
  + You can address novel components in a previous study: consider adjusting controlled variables and seeing if the same trends are observed.
  + You can address the effectiveness of previous experiments by questioning their methodologies. Use a different method to manipulate the independent variable or change how the dependent variable is measured.
  + You can address the effect of sample size: if a study found a trend with a small sample size and no statistical analysis, would that trend still exist with a larger sample size and stats?
* When coming up with a question, consider the life history of the organism(s) that you’re working with.
* Using the Discussion section of other studies is the easiest way to develop a question; often, scientists will include suggestions for future research directions in the Discussion.
* Not every study is a *completely* novel idea; in fact, virtually all studies are derived directly from information from previous works.

The Hypothesis

* The hypothesis is, in simple terms, a prediction about the question. It is sometimes phrased as an “if… then…” statement, but it doesn’t need to take this form.
* The hypothesis must *specifically* include both the independent and dependent variables.
  + If the question needs multiple independent variables to address, or requires many dependent variables, you may need multiple hypotheses. However, you should avoid overly complex experimental designs in this course, as you have limited time!
* The hypothesis must be *justified* with background information and sound rationale.
* Your hypothesis must be *testable*. If you are familiar with statistical analysis, this means that the hypothesis must be resolvable using a statistical test.

The Experiment

* The experiment is simply how you test your hypothesis and answer your question. The purpose is to determine the effect of the independent variable(s) on the dependent variable(s).
  + Because most experiments in this course are controlled experiments, you should consider how you will manipulate the independent variable(s). The methods should be feasible and relatively quick to perform.
  + You should consider how you’ll measure your dependent variables. In almost all experiments, you want to collect data on at least one **quantitative** dependent variable (meaning that the data is numeric).
* You also have the option of conducting a **methods study**, in which you address the possibility of faulty protocols or equipment.
  + *JIBI* articles often attribute abnormal results to faulty equipment, which means that you can test multiple different probes to determine if this claim is supported or not (in this case, the individual probes are the independent variable).
  + You also have the option of comparing different types of probes that can measure the same biological process: for example, both ethanol and CO2 probes can be used to estimate fermentation rates, so you could compare them.
  + Finally, you can design new protocols to test a biological process. Generally, you should justify doing this if the new protocol seems likely to be easier, quicker, cheaper, or more accurate than what is described in the lab manual.
* You should always try to have as large of a sample size as possible. Reviewers are instructed to deduct points if you have too small of a sample size. There are several ways to ensure you have a large sample size:
  + Make sure that you don’t have too many experimental groups. Each group should have a minimum of three samples, so if you have six experimental groups, you must have at least 18 trials.
  + Consider trial time. You want your trials to be long enough to collect meaningful data, but longer trials also mean fewer trials. In some investigations, the length of each trial is limited by animal care protocols.
  + Limit the number of independent variables you’re testing, usually to one. When testing multiple independent variables, you must avoid *confounding variables*, meaning you must test every single possible combination of treatments. If you have two independent variables, you must have a minimum of four experimental groups.