AP BIOLOGY NAME_____
BIOCHEMISTRY
ACTIVITY #8 DATE_____HOUR_____

# DESIGNING A CONTROLLED EXPERIMENT

## STEP 1: DEFINING THE PROBLEM

Every scientific investigation begins with the question that the scientist wants to answer. The questions addressed by scientific inquiry are based on observations or on information gained through previous research, or on a combination of both. Just because a question can be answered doesn't mean that it can be answered *scientifically*.

1. Discuss the following questions with your lab group and decide which of them you think can be answered by scientific inquiry.

   - What is the cause of AIDS?
   - Are serial killers evil by nature?
   - Why is the grass green?
   - What is the best recipe for chocolate chip cookies?
   - When will the Big Earthquake hit San Francisco?
   - How can the maximum yield be obtained from a peanut field?
   - Does watching television cause children to have shorter attention spans?

2. How did you decide what questions can be answered scientifically?

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## STEP 2: IDENTIFYING THE DEPENDENT VARIABLE(S)

The **dependent variable** is what the investigator measures (or counts or records). It is what the investigator thinks will vary during the experiment. For example, he may want to study peanut growth. One possible dependent variable is the height of the peanut plants.

3. Name some other aspects of peanut growth that can be measured.

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All of these aspects of peanut growth can be measured and can be used as dependent variables in an experiment. There are different dependent variables possible in an experiment. The investigator can choose the one he thinks is most important, or he can choose to measure more than one dependent variable.

## STEP 3: IDENTIFYING THE INDEPENDENT VARIABLE

The **independent variable** is what the investigator deliberately varies during the experiment. It is chosen because the investigator thinks it will affect the dependent variable.

4. Name some factors that might affect the number of peanuts produced by peanut plants.

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In many cases, the investigator does not manipulate the independent variable directly. He collects data and uses the data to evaluate the hypothesis, rather than doing a direct experiment. For example, the hypothesis that more crimes are committed during a full moon can be tested scientifically. The number of crimes committed is the dependent variable and can be measured from police reports. The phase of the moon is the independent variable. The investigator cannot deliberately change the phase of the moon, but can collect data during any phase he chooses.

Although many hypotheses about biological phenomena cannot be tested by direct manipulation of the independent variable, they can be evaluated scientifically by collecting data that could prove the hypothesis false. This is an important method in the study of evolution, where the investigator is attempting to test hypotheses about events of the past.

The investigator can measure as many dependent variables as he thinks are important indicators of peanut growth. By contrast he must choose only one independent variable to investigate in an experiment. For example, if the scientist wants to investigate the effect that the amount of fertilizer has on peanut growth, he will use different amounts of fertilizer on different plants; the independent variable is amount of fertilizer.

5. Why is the scientist limited to one independent variable per experiment?

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Time is frequently used as the independent variable. The investigator hypothesizes that the dependent variable will change over the course of time. For example, he may want to study the diversity of soil bacteria found during different months of the year. However, the units of time used may be anywhere from seconds to years, depending upon the system being studied.

6. Identify the dependent and independent variables in the following examples.

   a. Height of bean plants is recorded daily for 2 weeks.

      Dependent variable:_____

      Independent variable:_____

   b. Guinea pigs are kept at different temperatures for 6 weeks. Percent weight gain is recorded.

      Dependent variable:_____

      Independent variable:_____

   c. The diversity of algal species is calculated for a coastal area before and after an oil spill.

      Dependent variable:_____

      Independent variable:_____

   d. Light absorption by a pigment is measured for red, blue, green, and yellow light.

      Dependent variable:_____

      Independent variable:_____

   e. Batches of seeds are soaked in salt solutions of different concentrations, and germination is counted for each batch.

      Dependent variable:_____

      Independent variable:_____

   f. An investigator hypothesizes that the adult weight of a dog is higher when it has fewer littermates.

      Dependent variable:_____

      Independent variable:_____

## STEP 4: IDENTIFYING THE STANDARDIZED VARIABLES

A third type of variable is the **standardized variable**. Standardized variables are factors that are kept equal in all treatments, so that any changes in the dependent variable can be attributed to the changes the investigator made in the independent variable.

Since the investigator's purpose is to study the effect of one particular independent variable, he must try to eliminate the possibility that other variables are influencing the outcome. This is accomplished by keeping the other variables at constant levels, in other words, by *standardizing* these variables. For example, if the scientist has chosen the amount of fertilizer as the independent variable, he wants to be sure that there are no differences in the type of fertilizer used. He would use the same formulation and same brand of fertilizer throughout the experiment.

7. What other variables would have to be standardized in this experiment?

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## STEP 5: WRITING THE HYPOTHESIS

A scientific question is usually phrased more formally as a **hypothesis**, which is simply a statement of the scientist's educated guess at the answer to the question. A hypothesis is usable only if the question can be answered "no". If it can be answered "no", then the hypothesis can be proven false. The nature of science is such that we can prove a hypothesis false by presenting evidence from an investigation that does not support the hypothesis. But we cannot prove a hypothesis true. We can only support the hypothesis with evidence from *this particular investigation*.

Scientific knowledge is thus an accumulation of evidence in support of hypotheses: it is not to be regarded as absolute truth. Hypotheses are accepted only on a trial basis. Future investigations may be able to prove any hypothesis false. Current scientific studies you read about in the newspaper (for example, investigations of the effects of caffeine) are sometimes quite preliminary and therefore tentative in nature. Often, studies are published whose results contradict each other. However, this does not mean that scientific knowledge is flimsy and unreliable. Much of the information in your textbook, for example, is based upon many experiments carried out by numerous scientists over a period of time.

The scientific method, then applies only to hypotheses that can be proven false through experimentation. (There are other types of scientific investigation, such as observation and comparison that do not involve hypothesis testing.) It is essential to understand this in order to understand what is and is not possible to learn through science. Consider, for example, this hypothesis: More people behave immorally when there is a full moon than at any other time of the month. The

phase of the moon is certainly a well-defined and measurable factor, but morality is not scientifically measurable. Thus there is no experiment that can be performed to test the hypothesis.

8. Propose a testable hypothesis for human behavior during a full moon.

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9. Which of the following would be useful as scientific hypotheses? Give the reason for your decisions.

a. Plants absorb water through their leaves as well as through their roots.

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b. Mice require calcium for developing strong bones.

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c. Dogs are happy when you feed them steak.

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d. An active volcano can be prevented from erupting by throwing a virgin into it during each full moon.

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e. The higher the intelligence of an animal, the more easily it can be domesticated.

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f. HIV (human immunodeficiency virus) can be transmitted by cat fleas.

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The investigator devises an experiment or collects data that could prove the hypothesis false. He should also think through the possible outcomes of the experiment (whether the hypothesis is supported or proven false) and make predictions about the effect of the independent variable on the dependent variable in each situation. For example, a scientist has made the following hypothesis: Increasing the amount of fertilizer applied will increase the number of peanuts produced. He has designed an experiment in which different amounts of fertilizer are added to plots of land and the number of peanuts yielded per plot is measured. The predictions should state specifically how the dependent variable will change in relation to the independent variable and must be stated as an If … Then statement. The general format for an If … Then statement is "if the independent variable is changed in this way, then the dependent variable will change this way." For example, if the amount of fertilizer applied to a field is doubled, then the number of peanuts produced will double. Or, if the temperature of the reactants in a chemical reaction increases, then the rate of the reaction will increase.

10. Write a hypothesis for each of the following:

a. Guinea pigs are kept at different temperatures for 6 weeks. Percent weight gain is recorded.

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b. Batches of seeds are soaked in salt solutions of different concentrations and the number of seeds that germinate is counted for each batch.

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## STEP 6: SETTING THE LEVELS OF TREATMENT

Once the investigator has decided what the independent variable for an experiment should be, he must also determine how to change or vary the independent variable. The values set for the independent variable are called the **levels of treatment**. For example, an experiment measuring the effect of fertilizer on peanut yield has five treatments. In each treatment, peanuts are grown on a 100-$m^2$ plot of ground, and a different amount of fertilizer is applied to each plot. The levels of treatment in this experiment are set as 200 g, 400 g, 600 g, 800 g, and 1000 g fertilizer/100 $m^2$.

The investigator's judgment in setting levels of treatment is usually based on prior knowledge of the system. For example, if the purpose of the experiment is to investigate the effect of temperature on weight gain in guinea pigs, the scientist should have enough knowledge of guinea pigs to use appropriate temperatures. Subjecting the animals to extremely high or low temperatures can kill them and no useful data would be obtained. Likewise, the scientist attempting to determine how much fertilizer to apply to peanut fields needs to know something about the amounts typically used so he could vary the treatments around those levels.

## STEP 7: IDENTIFYING THE CONTROL TREATMENT

It is also necessary to include **control treatments** in an experiment. A control treatment is a treatment in which the independent variable is either eliminated or is set at a standard value. The results of the control treatment are compared to the results of the experimental treatments. In the fertilizer example, the investigator must be sure that the peanuts don't grow just as well with no fertilizer at all. The control would be a treatment in which no fertilizer is applied. An experiment on the effect of temperature on guinea pigs, however, cannot have a "no temperature" treatment. Instead, the scientist will use a standard temperature as the control and will compare weight gain at other temperatures to weight gain at the standard temperature.

11. Tell what an appropriate control treatment would be for each of the following examples.

    a. An investigator studies the amount of alcohol produced by yeast when it is incubated with different types of sugar.

       Control Treatment:_____

    b. The effect of light intensity on photosynthesis is measured by collecting oxygen produced by a plant.

       Control Treatment:_____

c. The effect of NutraSweet sweetener on tumor development in laboratory rats is investigated.

Control Treatment:_____

d. Subjects are given squares of paper that have been soaked in a bitter-tasting chemical. The investigator records whether each person can taste the chemical.

Control Treatment:_____

e. A solution is made up to simulate stomach acid at pH 2. Maalox antacid is added to the solution in small amounts, and the pH is measured after each addition.

Control Treatment:_____

## STEP 8: DETERMINING REPLICATION

Another essential aspect of experimental design is **replication**. Replicating the experiment means that the scientist repeats the experiment numerous times using exactly the same conditions to see if the results are consistent. Being able to replicate a result increases our confidence in it. However, we shouldn't expect to get exactly the same answer each time, because a certain amount of variation is normal in biological systems. Replicating the experiment lets us see how much variation there is and obtain an average result from different trials.

A concept related to replication is **sample size**. It is risky to draw conclusions based upon too few samples. For instance, suppose a scientist is testing the effects of fertilizer on peanut production. He plants four peanut plants and applies a different amount of fertilizer to each plant. Two of the plants die.

12. Can he conclude that the amounts of fertilizer used on those plants were lethal? What other factors might have affected the results?

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## STEP 9: WRITING THE METHOD

After formulating a hypothesis and selecting the independent and dependent variables, the investigator must find a method to measure the dependent variable; otherwise, there is no experiment. Methods are learned by reading articles published by other scientists and by talking to other scientists who are knowledgeable in the field. For example, a scientist who is testing the effect of fertilizer on peanuts would read about peanut growth and various factors that affect it. He would learn the accepted methods for evaluating peanut yield. He would

also read about different types of fertilizers and their composition, their uses on different soil types, and methods of application. The scientist might also get in touch with other scientists who study peanuts and fertilizers and learn about their work. Scientists often do this by attending conferences where other scientists present results of investigations they have completed.

In this course, methods are described in the labs.

**QUESTIONS**

13. A group of students hypothesizes that the amount of alcohol produced in fermentation depends on the amount of glucose supplied to the yeast. They want to use 5%, 10%, 15%, 20%, 25%, and 30% glucose solutions.

    a. What is the independent variable?

    _____

    b. What is the dependent variable?

    _____

    c. What control treatment should be used?

    _____

    d. What variables should be standardized?

    _____

    _____

14. Having learned the optimum sugar concentration, the students next decide to investigate whether different strains of yeast ferment glucose to produce different amounts of alcohol. Briefly explain how this experiment would be set up.

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15. A group of students wants to study the effect of temperature on bacterial growth. To get bacteria, they leave Petri dishes of nutrient agar open on a shelf. They then put the dishes in different places: an incubator (37°C), a refrigerator (10°C), and a freezer (0°C). Bacterial growth is measured by estimating the percentage of each dish covered by bacteria at the end of a 3-day growth period.

    a. What is the independent variable?

    _____

    b. What is the dependent variable?

    _____

    c. What variables should be standardized?

    _____

    _____

    _____

16. A team of scientists is testing a new drug, XYZ, on AIDS patients. They expect patients to develop fewer AIDS-related illnesses when given the drug, but they don't expect XYZ to cure AIDS.

    a. What hypothesis are the scientists testing?

    _____

    b. What is the independent variable?

    _____

    c. What is the dependent variable?

    _____

    d. What control treatment would be used?

    _____

    e. What variables should the researchers standardize?

    _____

    _____

17. A group of students decides to investigate the loss of chlorophyll in autumn leaves. They collect green leaves and leaves that have turned color from sugar maple, sweet gum, beech, and aspen trees. Each leaf is subjected to an analysis to determine how much chlorophyll is present.

    a. What is a reasonable hypothesis for these students?

    _____

    _____

    b. What is the independent variable?

    _____

    c. What is the dependent variable?

    _____

    d. What would you advise the students about replication for this experiment?

    _____

    _____

18. A scientist wants to study mating behavior in crickets. She hypothesizes that males that win the most male-vs.-male contests mate with the most females. She observes the crickets to obtain data. For each male, she counts the number of male-male fights he wins and the number of females he mates with.

    a. What is the independent variable?

    _____

    b. What is the dependent variable?

    _____

    c. What constitutes replication in this experiment?

    _____

19. Design a controlled experiment to determine the effect of exercise on heart rate.

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| **PROBLEM** (in the form of a question) | |
| **Dependent variable** | |
| **Independent variable** | |
| **Standardized variables** (list at least 4 factors that must be kept equal in all treatments) | |
| **HYPOTHESIS** | |
| **Levels of treatment** (values set for the independent variable) | |
| **Control treatment** (independent variable either eliminated or set to standard value) | |
| **REPLICATION** | |

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|---|---|
| **METHOD** (what steps will you follow) | |
| **Expected results** | |
| **Explanation** of expected results | |