ESA21
Environmental Science Activities for the 21st Century

Experimental Analysis

Instructors: If your institution does not have the Fish Farm computer simulation, contact the project directors for information on obtaining it free of charge. The ESA21 project team is indebted to Dr. Bob Kosinski for making this resource available.

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Testing Factors that Affect Growth in Fish

Central to good science are accurate observations, testable hypotheses, well-designed experiments or other tests, and reasonable data analyses. The purpose of Laboratory 1 is to introduce you to the basics of designing and analyzing experiments. The following two laboratory exercises will provide you with further steps in organizing and analyzing data. Many interesting experiments are impossible to do in a normal undergraduate science laboratory setting. For this reason, your introduction to designing an experiment that has relevance in "the real world" involves a computer simulation. The program involves a growing industry, that of farming fish.

What is your favorite kind of fish to eat? Catfish? Salmon? Perhaps a nice, sleek trout? Chances are pretty good that the fish you ate last, unless you or a friend caught it, was the product of a fish farm.

One of the most promising technologies for providing the growing human population with high quality protein foods is aquaculture, sometimes called "fish farming". Aquaculture is the managed production of plants and animals that live in bodies of water, such as lakes and ponds. Salmon, trout, shrimp, clams, oysters, and seaweed are all common products. Many of the fish served in restaurants or sold in grocery stores were grown in such "fish farms". Although it has been practiced since ancient times, aquaculture has become particularly significant in the last forty years as the rapid increase in the world population has expanded the need for inexpensive, high-yield foods. Over the last ten years, world aquaculture production has expanded nearly 11% per year and it is now one of the fastest growing segments in agriculture[1]. This rapid increase is due, in part, to an expansion in the number of aquaculture businesses, but it is also aided by a growing understanding of the optimum conditions needed for producing more and larger fish.

(1) http://www.aps.uoguelph.ca/~aquacentre/faq/overview.htm

Experimenting with Fish Growth
How might changing conditions affect the yield of a fish farming operation? There are at least three factors that can lead to production differences: (a) a change in the number of fish produced, (b) a change in the number of fish that die before harvesting, and (c) a change in the weight of each fish. The first two factors affect population size and the last affects the size of individual fish. Environmental conditions could influence any or all of these.

In this lab, we will test whether different conditions affect the weight of fish through the use of a computer simulation. In the process, we will also discuss the basics of designing an experiment. We will begin by discussing, running, and analyzing one experiment. You will then design, run, and analyze a second experiment.

Hypothesis
Experiments are designed to test particular hypotheses. A hypothesis is a reasonable explanation for an observation, an "educated guess". To be considered a scientific hypothesis, it must be testable; that is, there must exist the possibility of disproving it. Many hypotheses may be proposed to explain a particular phenomenon, just as we gave three possible explanations for how different conditions change yield of fish in aquaculture. One way to state the explanation, or hypothesis, we have chosen to test is:

Hypothesis: Environmental conditions affect fish growth.

Predictions
To test a hypothesis, predictions are made and experiments are performed. A prediction is a statement of what will happen under certain circumstances if the hypothesis is true. It is a necessary consequence of the hypothesis. For example, in testing this hypothesis, we would predict that if we make some change in the environment then there would be a change in the growth of the fish.

There are many environmental factors we could test – temperature, amount of food, living space available per fish, acidity of the water, and so forth. If we changed them all at the same time, we would not know which one of them caused any observed changes. Hence, we will test only one factor at a time. One of the environmental conditions that is easy to change is temperature, so in our first experiment, that will be the environmental factor we manipulate.

How will we measure fish growth? Growth could be measured by looking at changes in one of several different parameters such as length, width, weight, or surface area of the fish. The particular program we are using begins with fingerlings (young fish) that are all about the same weight and reports on the average weight per fish after six weeks of growth. This means that weight would be a good variable for us to measure.

Given this information, it should be easy to see that, if the hypothesis is correct, then we should be able to see differences in the average final weight of fish that are kept at different temperatures.

Prediction 1: Fish grown in different temperatures will have different weights.

Procedure

Fish Farm is a simulation of fish growth in a variety of conditions. One possibility is to "grow" fish for fifty-four days in tanks equipped to control a variety of environmental factors. For the test of the first prediction, tanks will be held at different temperatures during that time.

Variables

There are three types of variables to consider in conducting a scientific experiment. The independent variable is the factor that is intentionally made different at the beginning of the experiment, and at least two different cases (temperatures) of that factor must be examined. More cases will usually support your conclusion better. In the first prediction, the independent variable is temperature, so we will "grow" the fish in several different temperatures. How will we decide which ones? Most living things exist between 0^{o}C and 40^{0}C (note that scientists use the metric scale for measuring dimensions and the Celsius scale for measuring temperature.) With this in mind, we can limit the temperatures used to that range. If we test 5^{o} intervals, that will give us eight different cases – enough to see any basic patterns of change emerge.

A second type of variable is the dependent variable. This is the variable that is measured to see if changing the independent variable had an effect. It is the kind of data that is being collected. In the first prediction, the dependent variable is the average weight of the individual fish at the end of the experiment. Since the fish all began at about the same weight, growth can be compared by looking at the final weight.

The third type of variable is a controlled variable, and any given experiment will have several. Controlled variables are the factors that might affect the dependent variable and that therefore must be consistent for all cases of the independent variable tested. In testing prediction 1, for example, all of the tanks used are identical, the amount of oxygen and food provided is the same, the young fish are all of the same species and age, and the lighting conditions are identical. Anything that might affect growth in the fish, other than the temperature, must be the same for all temperatures tested.

Experiment 1: General Comments

A computer simulation will be used to test each of the predictions. The particular program you will use has been programmed with data from several fictitious species, although these species all resemble actual types of fish. The program was written to test fish in two different situations -- ponds and tanks. Because the conditions can be more accurately controlled in tanks, you will simulate experiments performed in tanks. Each run simulates a trial that runs for six weeks, from March 21 to April 30. The species studied usually mature in October. Young fish are used because they grow more rapidly than more mature fish, and anything affecting growth rates will most likely have the greatest affect on young fish. All environmental variables are controlled except temperature, oxygen levels, the protein content of food, and the amount of food provided. You can set each of these for each experiment. The program was written primarily to measure the profitability of each set of conditions, and the focus of the data given, therefore, is cost per gram of fish produced. However, the data in which you are interested are the average weights of the fish produced.

All units in the program are in the metric system. The size of the fish, for example, is measured in grams. A gram is a unit of mass, and, to give you an estimate of its size, the average soup can holds about 430 grams of veggies and broth.

Oxygen content, in this program, is given in milligrams per liter (ml/l). There are a thousand milligrams in a gram, so a milligram is a very small quantity. The liter is a measure of volume approximately equivalent to a quart.

Temperatures are given in degrees Celsius. Room temperature is usually about 25^{o}C, while the freezing point of water is 0^{o}C and the boiling point is 100^{o}C.

To test the first prediction, you will run several different tanks, each at a different temperature. You will keep all other environmental conditions the same. Your data will be recorded on a data table (Table 1-1 in the Data section of this lab).

Accessing Fish Farm:
Fish Farm is not available online, and so will be made available on a local computer network. Check with your instructor for directions on accessing the application.

Running Fish Farm:	
	Once you are on the first screen:
(1.)	Press the space bar to continue.
(2.)	Do not change the speed. Go to the next screen.
(3.)	Press the letter of your assigned fish species. Use species C if one was not assigned.
(4.)	Read about your fish -- it may give clues to the best growing conditions. Go to the next screen.
(5.)	Skip this screen.
(6.)	Press "1" for tank experiments.
(7.)	Press "A" to choose the temperature.
(8.)	Press "5" to set the temperature and press "enter".
(9.)	Press "B" and set the oxygen content at 10 mg/l. Press "enter".
(10.)	Press "C" and set the protein feed at 50%. This means that half of the fish food will be protein. Press "enter".
(11.)	Choose constant rate.
(12.)	Set the feeding rate at 10%. This means that the fish will receive 10% of their body weight in food daily.
(13.)	Press "esc" to run this test.
(14.)	Wait for the data screen and record the average weight of the fish at 5^{o}C, Trial 1, on Table 1-1. (If all of the fish die, record a zero.) Press the space bar.
(15.)	Press either A or D -- the graphs produced by this program records only the cost per gram of fish produced and are not useful to us.
(16.)	Press the space bar to go to the next screen.
(17.)	Press "1" for a new experiment.
(18.)	Press "1" to choose a tank experiment. Notice that the conditions set are the same as those for the test you just ran. Do not change the conditions.
(19.)	Press "esc" to run the same test again. Did the answer change? It may, because the program adjusts for the possibility of slightly different genetic stock or uncontrollable conditions for each run. It is a very realistic program in this respect. Record your data on Table 1-1 for 5^{o}C, Trial 2.
(20.)	Repeat steps 15 - 19 for a third run, recording the results on Table 1-1 in the column for Trial 3.
	Continue in this fashion, changing the temperature every three runs and using the temperatures listed on Table 1-1.

Experiment 1: Data Tables
Data tables are a good way to record and organize data. In this experiment, you will record two kinds of data. The first is raw data, the data you are taking directly from your experiment. Often, this is sufficient, but there are times when data must be further processed to be useful. In this case, random errors may result in different masses for each trial at a given temperature. Finding the mean (average) of these data will minimize this random error. For example, in Table 1-1 add the three values for the three trials done at 5^oC, divide by 3, and record the answer in the last column. This new value is more accurate and is the one you will use in discussing and graphing the results.

Data tables are set up after the experiment has been designed and before the data are taken. Each should have a title that includes both the independent and the dependent variable and that reflects the experiment. Columns and rows should also have appropriate labels. A data tables for the first prediction is supplied for you to use.

Experiment 1: Graphs
Although a table is very useful for organizing data, graphs can often help to clarify the relationships between the independent and the dependent variables by depicting them visually. When graphing the results of an experiment, the dependent variable is represented on the vertical or Y-axis and the independent variable on the horizontal or X-axis. The title of a graph is similar to that of a table and should include both the independent and dependent variables. Both axes should be labeled. Be sure to include the units of measurement. In this case, the titles and labels for the first prediction and test are supplied for you, but you must complete these items for later graphs. You should decide on the scale for each of the three graphs. Remember to spread the scale for each axis over the space allotted rather than using only the bottom half or the left half of the graph.

Data Analysis
Once you have completed an experiment, you can determine whether or not the data support or do not support the hypothesis. In this case, for example, if the final weights of the fish varied significantly at different temperatures, the hypothesis has been supported. If the fish weighed about the same in every case, then the hypothesis has not been supported.

Once you have completed taking the data for an experiment, you must decide if it supports the hypothesis. To report this to someone else, your analysis must include a bit more than just your conclusion – it should also include your reasoning. For this reason, a good analysis: (1) begins with a statement of the hypothesis, (2) indicates the kind of data that would support the hypothesis (the prediction), (3) cites the relevant data that were collected (gives examples that illustrate the trend you see), and (4) draws a conclusion as to whether or not the hypothesis was supported. Remember, a hypothesis is only a possible explanation that is being tested and the purpose of this argument is to convince the reader that the evidence either does or does not support the hypothesis.

Evaluating the Design of an Experiment
Analyzing the data from an experiment is only one important aspect in evaluating what it says about the hypothesis. It is also important that the experiment be a good one. If an experiment is flawed, the data cannot be completely trusted and may therefore lead the investigator to an incorrect conclusion.

While doing a complete analysis of the experimental design may take an expert, there are four common criteria we can apply:
(1.) Is there only one independent variable and why do you think it is an appropriate one for testing the hypothesis?
(2.) Is the dependent variable accurately measured and why do you think it is an appropriate one for testing the hypothesis?
(3.) Are all of the other potentially important variables controlled (maintained the same in all cases of the independent variable)?
(4.) Is there enough data?

Notice that none of these criteria judge the experiment on the basis of whether or not it supported the hypothesis. An experiment is a way to ask if the hypothesis is correct. The validity of the experiment is independent of the conclusion. Let's look again at each of these criteria, see what they mean, and use them to examine the first experiment from this lab. Then you will be asked to evaluate the experiment you designed. In answering these questions, it will be helpful to remember the hypothesis and the first prediction.

Hypothesis: Environmental conditions affect fish growth

Prediction 1: Fish grown in different temperatures will have different weights.

(1.) Is there only one independent variable and why do you think it is an appropriate one for testing the hypothesis? In answering this question you should identify the independent variable and explain why you think manipulating it is a case of changing environmental conditions. In the first experiment, temperature is the independent variable and it is the only variable we purposely changed. We chose temperature because it is clearly an environmental condition.

(2.) Is the dependent variable accurately measured and why do you think it is an appropriate one for testing the hypothesis? In answering this question, you should identify the dependent variable, indicate why you think it was or was not accurately measured, and indicate why you think measuring it is a way to measure the dependent variable of the hypothesis. In the first experiment, the dependent variable was weight of the fish. This is appropriate because it is a commonly accepted way of measuring growth. We cannot comment on accuracy because the computer program does not give us any information on this.

(3.) Are all of the other potentially important variables controlled (maintained the same in all cases of the independent variable)? In answering this question, indicate what variables were controlled and how. In testing the first prediction, you know that oxygen levels, feeding levels, and protein content of the food were controlled by providing the same levels to all tanks. You also know that growing room and effects of other living organisms in the environment were controlled by keeping all fish in the same size tanks with the same initial number of fish.

(4.) Were enough data collected and why do you think this? There must always be enough data collected to reflect the actual situation. Questions to ask yourself include whether there are enough replicates of each test as well as whether or not enough cases of the independent variable were tested. In Prediction 1, for example, are three replicates sufficient for each temperature tested? Probably not if this were a study for the fish industry, but in this case we can accept it because the three replicates, in each case, came up with very similar final weights. Another question to ask is whether or not enough different temperatures tested and was a wide enough range used. Since most organisms live within a range of 0^{o}C and 40^{o}C, that range should be sufficient. Ten equally spaced temperatures were tested and provided a clear trend in the data.

Assumptions of an Experiment

Another factor that affects the conclusions we draw from an experiment are whether or not the assumptions are reasonable. Assumptions are factors that are thought to be true for the investigation but that have not been verified or controlled. They may include information that is commonly accepted and seems to need no further confirmation, such as that young fish will grow in appropriate conditions. Sometimes assumptions are variables that are thought to be held constant but that are not checked, such as the salt content of the water. They may be factors that are beyond the control of the investigator because of technical or time considerations. For example, if we are doing this experiment in order to find the set of conditions that will provide the largest mature fish, we are assuming that the trends observed over six weeks would be the same as those observed if we ran the test for eight months. Incorrect assumptions invalidate the investigation. All scientific tests are based on some assumptions that must be identified and considered in the analysis.

Deciding if the assumptions are reasonable is called justifying the assumptions. To justify an assumption involves indicating (a) why it is important to the experiment and (b) why it probably is or is not true.

In the first experiment, the following are among the assumptions and justifications:
(1.) *The responses of the fish species examined are typical and representative of all fish.* The hypothesis is a generalization meant to apply to most species of fish. If the species examined is not typical, the results will be true only for a few select species rather than for most species. The assumption is reasonable to accept, however, because the species used are fictitious ones based on a variety of typical commercially-raised fish.
(2.) *The set of environmental conditions that are optimum for growth do not change as the fish mature .* The fish used are young, which is usually a time of rapid growth. If the conditions that are optimum for fish growth change as a fish matures, the trends detected are valid only for young fish and are not predictive of growth that occurs later. Experience with other species of organisms indicates that, in the majority of species, the effects of a particular condition on growth do not change greatly until the organism has reached maturity. It is therefore reasonable to accept this assumption.
(3.) *Fish confined to a tank demonstrate the same growth patterns as those raised in a stock pond.* If this assumption is false, any conclusions we reach cannot be applied beyond the conditions of the experiment. The primary factor here is the size of the container (tank or pond) in which the fish are raised. This factor generally affects fish only when the fish are large with respect to the size of the container. The fish used in this investigation are small enough that any effects of container size should be negligible.

At this point, refer to the lab printout sheet and complete the section for Experiment 1.

Experiment 2: Procedures

A hypothesis that has been tested only once usually remains a hypothesis until it has been tested many times. If the tests provide convincing evidence that the hypothesis is correct, than it is considered to be true until (and if) convincing contradictory evidence emerges. If the tests provide convincing evidence that the hypothesis is not correct, it is either dropped from consideration or changed and retested. In other words, a reasonable hypothesis is usually tested many times.

For the second part of this laboratory, you are being asked to use the Fish Farm program to choose and test a second prediction from the original hypothesis. Recall, the hypothesis is:

Hypothesis: Environmental conditions affect fish growth.

There are three other factors you can choose to test using this program – oxygen concentration, the amount of food, and the proportion of the food that is protein. Pick one of these, make a prediction about it, and design an experiment. Using the Fish Farm program, run your experiment. Remember that you will have to use tank experiments and set each of the elements in the program (temperature, etc) to the desired level.

Refer to the lab printout sheet and complete the section for Experiment 2.

ESA 21: Environmental Science Activities

Activity Sheet
Experimental Analysis

Name:

Instructor:

Experiment 1:

Table 1-1: The Effects of Temperature on the Average Weight of Fish

Temperature	Average Weight of Fish after Six Weeks			
	Trial 1	Trial 2	Trial 3	Mean (Average)
5^o C				
10^o C				
15^o C				
20^o C				
25^o C				
30^o C				
35^o C				
40^o C				

Answer these questions for the first experiment examining the relationship between the temperature of the water and the final weight of the fish.

1. What is the independent variable for this experiment?

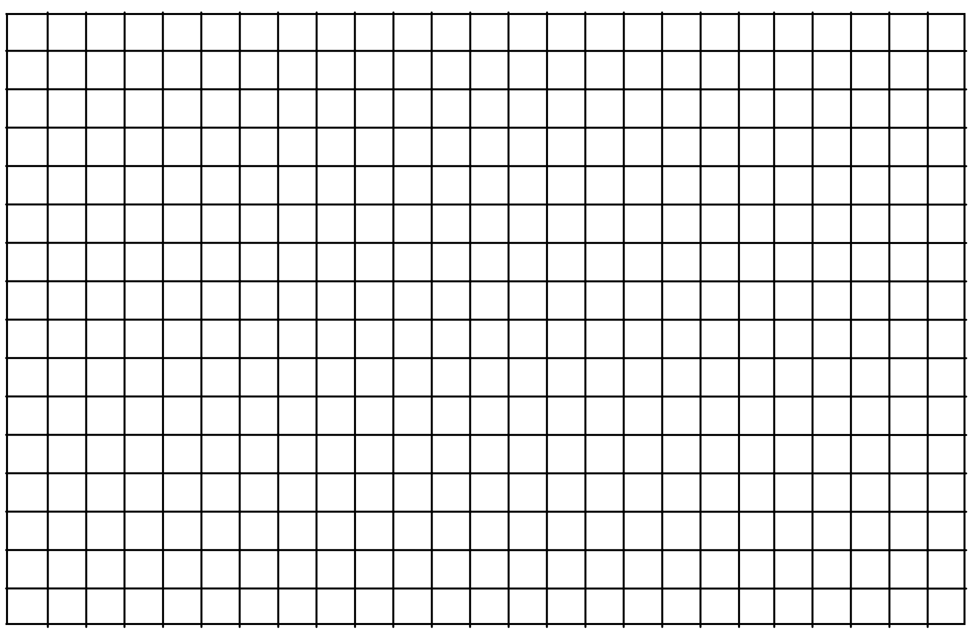
2. What is the dependent variable for this experiment?

3. List at least four controlled variables for this experiment

4. Using the data you collected for your experiment, complete the graph below for this experiment.

Graph 1-1 The Effects of Temperature on Average Weight of Fish

Average
Weight
of Fish
(grams)



T
emperature in ^{o}C

Experiment 2:

In the second part of the laboratory, you designed your own test of the hypothesis: **Environmental conditions affect fish growth.** All of the questions on this sheet refer to the experiment you designed.

5. What was your prediction?

6. What is the independent variable for your experiment?

7. What is the dependent variable for your experiment?

8. List at least four controlled variables for your experiment

9. Using the data you collected for your experiment, complete the table and graph on the other side of this paper, then write an analysis of your data here.

Table 1-2

TITLE: ____

Independent Variable is: ___________	Average Weight of Fish after Six Weeks			
	Trial 1	Trial 2	Trial 3	Mean (Average)

Graph 1-2

Title: ____

Y-Axis
Label:

______

____

Label x-axis: ____