Title: BEHAVIOR OF THE SIAMESE FIGHTING FISH  
(Betta splendens)

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Appropriate Level: General Biology, Regents Biology and Advanced Biology

Abstract: This laboratory exercise is designed to give high school students experience in using the scientific method. Students design and conduct their own research studies on the behavior of Siamese fighting fish (Betta splendens) that are kept in classroom aquaria. Included in the laboratory materials is a "Study Proposal Form" that students use to structure their research designs.

Time Requirement: for completion of exercise: four to six 45-minute lab periods  
to prepare lab equipment: 2 to 3 hours
Additional Teacher Information

Materials:

Per Team

- 2 gal. aquarium (67-4332 or 67-4334 Carolina Biological) (any small transparent container with vertical sides will do)
- 1 male Siamese fighting fish (purchase a variety of colors)
- fish food (live or frozen brine shrimp are best, some fish will eat dry fish food)

Class Equipment (all of these are optional and may be supplemented by what is available)

- extra males and a few females to be introduced into experimental animals aquaria (use plastic bags to hold & protect introduced animals)
- plastic hand mirrors with colored backs
- colored paper (construction and transparent)
- plastic poker chips
- fish nets
- timers
- plastic pipettes
- strips of cardboard for isolating fish
- colored lights
- clear plastic bags or plastic dividers
- thermometers
- large pails for aging water
- scissors

Introduction

1. Objectives:

In this laboratory exercise, students will design and conduct their own research studies on the behavior of the Siamese fighting fish (*Betta splendens*). Through this exercise, students will gain a greater understanding of, and appreciation for, the scientific method. Students will undertake all of the following: hypothesis derivation, research design, data collection, and scientific writing. At the same time, students will learn something about animal behavior and develop their ability to think creatively, an often overlooked skill in the teaching of science.

From the students, this exercise demands creativity, active involvement, and independent work, rather than direction-following and passive learning. Consequently, students will find that their task
is demanding and, compared to most of their other schoolwork, ambiguous. The teacher's role is therefore, equally demanding, for he or she must actively guide each student's work on an individual basis, without being overly directive.

2. Information students must have prior to beginning lab:
   - Familiarity with the steps of the scientific method.
   - Basic introduction to the field of animal behavior.
   - Some prior experience collecting data.

Tips for Laboratory Set-Up:

1. Set up the tanks about 10 days prior to the beginning of the lab. This allows for the water to age before the fish are introduced and for the fish to acclimate to their surroundings before the experiments begin.

2. Introduce fish slowly to the tanks by placing the bag with the newly purchased fish in the aquarium for a few minutes before releasing them.

3. NOTE: The male fish must be kept physically separate from one another, and the aquaria should be arranged so that no male fish can see another one when in its own tank. Boxes or pieces of cardboard work well in keeping the tanks isolated.

4. Siamese fighting fish are surprisingly hardy. Therefore, unless the school temperature falls dramatically at night, the fish will be able to tolerate the normal temperature swings of the aquarium without a heater. However, please note that the transfer of fish from tank to tank is stressful for the fish. Therefore, encourage students to perform their experiments with a minimum of fish transfers.

5. Keep the set-ups simple to ease preparation and simplify student experiments.

6. Preparation of the tanks will take approximately 1 to 2 hours.

7. After the experiments are completed, the teacher may want to introduce a female Siamese fighting fish to one or more of the males for observation of breeding behavior. Place the male and the female in one of the small tanks. The female may be removed after she lays her eggs (400-500). The male is responsible for watching the brood and should be removed from the tank 2-3 days after the eggs are laid. The tiny fry should be fed dry powdered food.
8. Giving students some role in the maintenance and feeding of the fish is easy and can help teach responsibility, as well as help ensure that students treat the fish with respect and care during their experiments.

9. So that each fish can be individually identified, be sure to label each fish's home tank and advise students to keep careful track of the fish they are using in their experiment.

**Instructional Flow Chart**

Below is an outline of a suggested overall strategy for teaching this lab. Part III of the Instructor's Materials contains a more detailed discussion. This is just a suggested format, the teacher may adapt it as desired, including shortening it substantially if necessary.

**BEFORE LAB**

Step 1: Distribute copies of the lab to students including 2 copies of the Study Proposal Form. Have students read the Introduction and answer the prelab questions prior to coming to the first lab session.

**CLASS 1**

Step 2: Collect or discus the prelab questions (5 min.)

Step 3: Have students read Section #1a and #1b of the Procedure portion of their lab materials. Introduce the fish and have students make preliminary observations (5 min.)

Step 4: Have students read Section #1b of the Procedure section. Demonstrate the aggressive behavior of Siamese fighting fish while students record observations. (10 min.)

Step 5: Make materials available and give students time to experiment informally with the fish. (5 min.)

Step 6: Briefly describe brainstorming. Have students brainstorm individually and then collectively on the problem asked in Section #1d. (10 min.)

Step 7: Have the students read the Introduction to the Study Proposal Form section in the Procedure portion of their labs. (Can be assigned as homework.)

**CLASS 2**

Step 8: As a class, use the Study Proposal Form to write a sample study proposal. Or, alternately, present an already-completed study proposal as a guideline for student work. (20 min.)
Step 9: With teacher guidance, students prepare study proposals in groups of 2 or 3. (30 min., or this could be done as homework)

BEFORE NEXT CLASS

Step 10: Teacher review of study proposals.

Step 11: Study proposals amended as necessary. (Variable time)

CLASS 3 AND/OR 4 (depending on time)

Step 12: Data collection by students, with teacher guidance. (45 min.)

IN CLASS OR HOMEWORK

Step 13: Data analysis and lab report writing. Teacher should clearly explain, either verbally or in writing, the format that student should use in their reports. (45 min. or completed as homework)

**Detailed Instructions for the Flow Chart**

Step 2: Answers to the prelab questions:

1. A variety of answers are possible. Any answer that makes sense should be accepted. Example: "Aggressiveness may give a male fish access to a greater amount of food and to more females than would submissiveness."

2. This question is intended to give students an opportunity to begin thinking creatively. The teacher may want to reinforce this process by calling on students to share one of their ideas with the class. Be particularly careful to be positive and supportive of the student's ideas. Some of their suggestions will be quite unusual or impractical. Shape these ideas into more useable ones in order to show that even "crazy" thoughts can be starting points for further constructive thinking.

3. This statement anthropomorphizes the fish. "Jumping for joy" refers to an inner experience that we cannot with confidence apply to the minds of fish.

Though not a necessary part of this laboratory, teachers may want to use this opportunity to introduce students to one or more of the following terms: territoriality, ethology, instinct, courtship, dominance hierarchy, aggressive behavior, submissive behavior.
Step 3: The upward-pointed mouth helps the fish feed at the surface of the murky waters of its native habitat. It also allows the male to gulp air for building a bubble nest.

When not exposed to other male fighting fish, male Siamese fighting fish tend to be somewhat placid, slow-moving fish.

Check student descriptions of fish behavior to be sure that they are precise, complete, and not unduly anthropomorphic.

Step 4: To demonstrate aggressive behavior, the teacher can either use a mirror, or place two fish, one either side, of a transparent divider. When two fighting fish are placed on opposite sides of the transparent divider in the small tank, they will usually begin to respond to one another with aggressive displays within about a minute. If they fail to respond, nudge one closer to the other until the fish notice one another's presence. When exposed to his own mirror image (the mirror can be placed inside or outside the tank), a male fighting fish will usually respond even more rapidly than he does to another fish.

If two fighting fish are placed in a small tank without the divider present, the experimenter must be ready to remove one of the fish quickly in case of an attack. An attack can be sudden and can result in loss of fins. DO NOT allow students to do this without close supervision.

Step 5: Give students time to experiment independently with the fish and some of the available equipment. The knowledge gained during this free inquiry can form the basis for the formal research conducted later by the students.

Step 6: Brainstorming is a method of creative group problem-solving. Group members take turns suggesting ideas. The "wilder" the idea, the better! As ideas are generated, criticism is not allowed. Group members are encouraged to use other people's ideas to develop their own (a process called piggybacking). After a large number of ideas are produced, the group can then identify the best ones and then test their usefulness.

Brainstorming is an enjoyable teaching technique that can be used to develop creativity. If time permits, students can be asked to brainstorm on the possible uses for a variety of objects, such as a fermentation tube, a mousetrap, a necktie, a paper clip, or just about any household or laboratory object.

At this point in the lab, you may want to use brainstorming in the following fashion:

1. Have students read Section #1d.
2. Tell them to think creatively about the problem. Emphasize that wild ideas may be good ideas. Take a couple of minutes to allow them to think about the question and to write down a few ideas.

3. Call on students to suggest ideas. Emphasize that criticism is not allowed. If a student has trouble thinking of ideas, remind them of previous suggestions and suggest that they should "piggyback" on one of them. Reward unusual suggestions, especially those that break out of habitual ways of thinking.

Step 7: If students have not recently discussed the scientific method, this section may be particularly difficult and abstract. After students have read it, find out which parts are particularly difficult and address the difficulties. For General Biology students, this reading will be nearly meaningless without a concrete example for reference.

Step 8: The purpose of this step is to provide students with a concrete example to use as a guideline for writing their own study proposals. There are two ways that the instructor can provide this example:

1. The teacher can give students an already-written sample proposal. Such a sample proposal can be found on the next page. Note that the example is better designed and more organized than expected for a typical student proposal.

2. The teacher can work with the class to design a sample proposal together during class time. This second option is more challenging for both the teacher and the students than is the first option, but gives students more experience in proposal writing and is, therefore, more instructive.
STUDY PROPOSAL FORM -- SAMPLE

Group Members: John Doe, Richard Roe

Date: Nov. 22, 1992

Title: The Effect of Light Intensity on Aggressive Behavior in Male Siamese Fighting Fish

I. Research Question: What effect does light intensity have on aggressive behavior in male Siamese fighting fish?

II. Hypothesis: As light intensity increases, the response time by male Siamese fighting fish to threatening stimuli will decrease.

III. Test of Hypothesis:

   • We will define specific criteria to determine whether a fish acted aggressively or submissively.

   • We will place a mirror in one of the small tanks.

   • In part I of the experiment, we will place each of the 6 male fish, at a designated spot in the tank with the mirror. Each time, we will use a stopwatch to determine how long it takes the fish to respond aggressively or submissively toward its reflection. If the fish does not respond within 3 minutes, we will remove that fish and write "no response" on our data table. In part I, the experiments will be conducted at normal light intensity of the classroom.

   • In part II of the experiment, we will conduct the same trials as in part I, except this time they will be conducted in the storage room in the back of the science lab with the shades drawn and the lights out.

   • For each trial, we will keep all of the environmental variables the same except for light intensity. Temperature, amount of time spent by the fish out of the water, position of the scientists around the tank, and placement of the fish in the tank will all be held constant.
IV. Data Tables: Time Taken to Respond Aggressively (seconds)

<table>
<thead>
<tr>
<th>Fish</th>
<th>Full Light</th>
<th>Reduced Light</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
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<tr>
<td>2</td>
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<td>4</td>
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<td>5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

V. Proposed Plan to Analyze Data: We will calculate the average amount of time required in full light and in reduced light. If the time required in normal, full light is at least 10% greater than the time required in reduced light, then the hypothesis will be accepted. Otherwise, the hypothesis will be rejected.
Steps 9 and 10: Students will need a great deal of help in the writing of their research proposals. Be encouraging and supportive. Be sure that all group members participate and that criticism amongst lab partners is not excessive.

Groups will vary widely in the time taken to complete their proposals. As the quicker groups finish their work, the teacher can begin reviewing their proposals. Expect the proposals to be confusing and incomplete, particularly the first drafts. It is best, therefore, to review the proposals with the group members present so that you can understand what it is they are trying to do and so that they can fully understand your critiques.

When reviewing student Study Proposal Forms, keep the following notes in mind:

1. Students have trouble writing hypotheses. Their hypotheses will tend to be either too narrow or too broad.

2. Check to be sure that the research designs the students propose really test their hypotheses.

3. The "Test of Hypothesis" section will tend to be very disorganized. A moderate amount of disorder is to be expected and should be tolerated on such a challenging task.

4. Check to be sure that the sample size is clearly specified and is reasonable given your time constraints.

5. Unless the teacher intends to introduce some simple statistical tests, informal common-sense methods of data analysis should be used. In the example proposal, the "10% or more" figure in the Data Analysis section is such a common-sense method. Let students know that scientists have developed sophisticated statistical tests for evaluating their data.

6. On the Study Proposal Form, students should sketch the data tables into which they propose to record their data. After their proposal is reviewed and accepted, students should then draw new, larger data tables into which their data will actually be recorded.

Step 12: Active guidance by the teacher is essential during data collection. Carelessness will be common and a number of problems will arise. Don't hesitate to help students amend their proposals.
Step 13: The write-up is an important but difficult part of this exercise. General Biology students will have particular trouble with this step. A simplified format may be necessary for them. Requiring the submission of a rough draft may be helpful.

The format of the research report should be determined by the teacher and clearly presented to the students. One possible format is the following:

Abstract
Introduction (includes the research question)
Hypothesis
Procedure
Data (a well-presented data table)
Results (includes the data analysis)
Discussion (an analysis of any problems, possible errors, and proposed further study)
BEHAVIOR OF THE SIAMESE FIGHTING FISH

Objectives

Upon completion of this exercise, you should better understand . . .

• the use of the scientific method
• how to make careful observations
• how to record accurate experimental results
• how to use your data to support your conclusion
• the aggressive behavior of fish

Background Information

The Siamese fighting fish (*Betta splendens*) is a large, attractive, and hardy tropical fish that lives up to its name as a "fighting" fish. In its native Thailand, this species has been used in fish fights for centuries and has even been selectively bred for aggressiveness. Only the colorful males display this aggressive, agonistic behavior. The male fish makes a bubble-nest of mucus-coated, floating bubbles to which he attempts to attract the drab female to lay her eggs. When the territory of his nest is threatened by another male, the male Siamese fighting fish is particularly likely to show aggressive behavior. The aggressive display is characterized by the following behaviors (see diagram below to help identify anatomical structures):
Aggressive Behavior

- Rapidly fluttering pectoral fins
- Flared gills and gill covers
- Brightened colors
- Lunging toward the threatening stimulus and attempted biting
- Raised and spread fins which increase the apparent size of the fish
- Positioning of the fish's body perpendicular to the threatening stimulus (this behavior is called lateral display)

In response to an aggressive display, some fish will show submissive behavior.

Submissive Behavior

- Folding of the fins
- Drooping of the body
• Darkening of the colors

• Movement away from the threatening stimulus

The field of animal behavior attempts to understand such phenomena as aggressiveness in the Siamese fighting fish. In this laboratory, you will be the animal behaviorist and the Siamese fighting fish will be your subject of study. With your teacher's guidance, you will find a research problem, develop an hypothesis, design an experiment that tests your hypothesis, observe and collect data and, finally, write a research report on your activities. You will be using the scientific method to gain knowledge about the Siamese fighting fish.

One final word of caution. There is a tendency for newcomers in animal behavior to give to the animals they are studying human traits the animals do not have. The error is called "anthropomorphizing" and may lead to incorrect observations and conclusions. For example, it is wrong to attribute such emotions as "joy" or "friendship" to fish, for we cannot directly observe such emotions. Instead, the behaviorist should attempt to describe observed behaviors with simpler, more objective language that doesn't anthropomorphize the subjects of study.
Procedure:

Introduction to the Study Proposal Form

On the last page of this lab guide is a copy of the Study Proposal Form. Your team will use this form to write a proposal for your research. In this section of the laboratory instructions you will find information that will help you use the Study Proposal Form.

Project Title: This should be clear and concise, but not so short that you exclude an important part of the study. You may want to write this last!

Research Question: This is the problem that you want to investigate. It should be of interest to you and one you are able to test.

Hypothesis: This is an educated guess that answers the research question. The hypothesis is tested by the research that you conduct. An hypothesis should be based on your first observations of the fish and whatever knowledge you have about fish behavior from your experience or reading. It should not be so broad that your research cannot fully test it, nor should it be so narrow that it does not answer the research problem.

Test of Hypothesis: In this section, you present the details of the experiment that you will be conducting. Note the following:

a. In a properly designed scientific experiment, the procedure should be carefully planned ahead. Therefore, plan all the parts of your study as precisely and as thoroughly as possible. Think ahead about what it is you want to do and include these details in your proposal.

b. In experimental science, all variables, except for the one you are studying, should be controlled. In other words, all environmental conditions should be identical every time you conduct your experiment except for the one variable that you consciously change. Include in your proposal how you will control these other variables.

c. Keep the experiment as simple as possible. Test just one variable. Be specific about what the variable is. Can you measure or count something to make your observations more objective?

d. Include in your design the number of times your experiment will be repeated with one fish and the number of fish to be tested, and, if appropriate, the length of time each test
will last. In general, the greater the sample size the more confidence you can have in your results.

e. A diagram of your experimental set-up may be helpful.

f. If you need additional materials to conduct your study, ask your teacher if he or she can provide them for you.

Data Table: The recording of data in a clear, organized, and complete format is essential. Therefore, you should plan what data you will be collecting and have a table prepared ahead in which you can record the data as it is collected.

Data Analysis: Once you have collected data, you must then use it to come to a conclusion about your hypothesis. If the data support the hypothesis, then the hypothesis is accepted. If the data don’t support the hypothesis, then you must reject the hypothesis. In this section of the Study Proposal Form, you must explain how you plan to use your data to decide whether to accept or reject the hypothesis. Possible methods of data analysis include using the data to create a graph or the calculation and comparison of the average values for the different experimental conditions that you studied.

Writing the Research Proposal:

Now that you’re familiar with the Study Proposal Form, you can use it to create your own research design. As you work with your group, keep the following in mind:

a. A good proposal requires imagination and insight. NOTE: When thinking creatively, there is no such thing as a bad idea. All ideas have potential. Therefore, work together in a helpful way. Criticism and rudeness only stunt creativity.

b. Begin by brainstorming about possible research problems. "Brainstorming" means to work as a team to come up with a variety of ideas. After brainstorming, narrow your ideas down to the one you wish to pursue.

c. As you work out the details of your proposal, the need for changes will become evident. Therefore, do not be surprised if more than one draft of the proposal is necessary. If you need special materials, see if your teacher can provide them.

Your teacher will review your Study Proposal Form and return it to you with suggested changes.
Data Collection

Once your teacher has approved your proposal in its final form, collect the data, recording the results in your data table. Unforeseen difficulties may arise. If necessary, ask your teacher how to adjust your plan to cope with these problems.

Data Analysis

Analyze the data as planned. Conclude by rejecting or accepting your hypothesis.

Write-Up

The presentation of experimental results in a scientific research paper is an essential part of the scientific method. No scientist works in a vacuum. Each builds on the work of other scientist. Your research is no different. It is of little use if not presented formally. Your teacher will give you a format for your report.
BEHAVIOR OF THE SIAMESE FIGHTING FISH

Prelab Questions:

1. What function(s) do you think aggressive behavior in male Siamese fighting fish might serve? In other words, how can aggressiveness help a male survive and reproduce?

2. Suggest two research questions you may want to study concerning the aggressiveness of Siamese fighting fish. Use your imagination!

3. What is wrong with writing in a scientific report, "The male fish jumped with joy when the female laid her eggs in his bubble-nest"?
BEHAVIOR OF THE SIAMESE FIGHTING FISH

Procedure

Observation of the Fish

1a. With your teacher's guidance, observe your male Siamese fighting fish. Be sure you can identify the fins indicated in the diagram on page 1. Look for a collection of bubbles floating at the surface of the tank. This is the bubble-nest that the male has created in which a female would lay her eggs. Note that the fish's mouth is directed upward.

What do you think is the purpose of the upward-pointed mouth?

b. Briefly describe, in the space below, the behavior of the Siamese fighting fish when alone in the tank (position of fins, gill covers, color of fins & gill covers, is he swimming around or staying in one location, where is he staying, how often does he gulp air, what is the water temperature, does he have a nest):
c. When everyone is ready, your teacher will demonstrate aggressive behavior in the male Betta. Your teacher will carefully lower the mirror into the aquarium and leave it in place for 10 minutes. If there is a nest, the mirror will be lowered close to the nest and turned so male Betta can see himself.

Briefly describe, in the space below, what occurred when the male Betta became aware of the mirror. Include a description of the aggressive and submissive behaviors that you saw. Be sure you can identify both aggressive and submissive behaviors!

d. Animal behaviorists are often interested in determining the environmental variables (stimuli) that trigger certain behaviors. Use your imagination to think of three environmental stimuli that may either increase or decrease the frequency of aggressive behavior in male Siamese fighting fish. You might give some thought to what senses, i.e. vision, smell, hearing, feeling water movements, a fish might use to receive stimuli. (Note: there are no wrong answers here, so be creative. Anything outside the fish's body that can affect its behavior can be considered an environmental stimulus.)
BIOLOGY

NAME:__________________________________

EXERCISE # _____ DATE:_______________

BEHAVIOR OF THE SIAMESE FIGHTING FISH

STUDY PROPOSAL FORM

Team Members:__________________________________________

Title:__________________________________________________________________

I. Research Question:

II. Hypothesis:

III. Test of Hypothesis:
IV. Data Tables:

V. Proposed Plan to Analyze Data: