***Betta splendens***

**INTRODUCTION**

Ethology is the zoological study of animal behavior. Ethologists are generally interested in innate or instinctive responses ( genetically-programmed behaviors with little or no learned component). There are several advantages of instincts over learning. The animal does not require experience to perform the behavior; this is especially important if the initial response of the animal is vital to survival. Secondly, instincts do not require extensive neural coding for information storage and modification (and is therefore appropriate for animals with simple nervous systems). As an example, orb-weaving spiders build a perfect web on their first attempt (despite having no prior experience with webs for most species). Not to build a web, or to take the time learning to modify an imperfect web would result in starvation since spiders require webs to capture prey. A major cost of instincts over learning is their inflexibility in the face of environmental change. Innate responses control to one degree or another the feeding, sexual, and social behaviors of all animals (including man). For many animals these mechanisms drive them to choose a particular habitat or food. The study of communication is one of the more interesting aspects of animal behavior. Although much of our own language depends on learned responses, most organisms rely instead on genetically-coded rules of grammar and vocabulary. Three major forms of communication have been described among animals: chemical, visual, and mechanical (including touch and communication by sounds or vibrations). Of these three methods, transmitting messages by chemicals is probably the most primitive.

Wild *Betta splendens* are colorful and sexually dimorphic (i.e., males and females look different from one another), with males being more brightly colored than females. Because of their attractive colors and interesting behaviors, this species has been subject to many years of artificial selection (particularly the males) for mutant color morphs and very long fins and tails. Despite the domestic variety's showier appearance, its courtship and other intraspecific displays have remained relatively unchanged. Both wild and domestic *Betta splendens* will react strongly and aggressively to the appearance of another fish of the same species.

Several terms are commonly used in the ethological literature to describe the behavior patterns of animals. Fixed action patterns are genetically-determined sequences of movement. These stereotyped behaviors vary little from one individual to another and are often species-specific. Agonistic(from the Greek *agonistes*, meaning "champion") behavior in animals is defined as that exhibited during a contest, combat, escape, attack, or appeasement episode between two animals. The term is often used to describe Betta behaviors exhibited by male animals when they compete for mating opportunities with females. Releasers are found in the external environment and send out stimuli that release fixed action patterns. The courtship behavior displayed by honey bees vary little from one male to another and represents a fixed action pattern. The pheromone deposited by the female was the releaser for the males' courtship display. Releasers can be more complex than the simple presence or absence of a chemical as in the above example. In this exercise, we will explore the combination of complex visual stimuli that serve as releasers to trigger the fixed action pattern of aggressive (agonistic) display by male Siamese fighting fish (*Betta splendens*). The exchange of aggressive signals allows the fish to set up dominance-subordinate relationships. If the two combatants are not confined to a single bowl, the subordinate animal can flee and the conflict is resolved without the danger of bloodshed. As you shall see, visual communication is an effective way of showing varying levels of motivation and is an example of a graded response.

**MATERIALS NEEDED:**

* Male Siamese fighting fish (*Betta splendens*); one per group of two or three students.
* Wooden applicator sticks, transparent tape, brightly-colored construction paper, scissors, mirrors, and one- two liter flat-sided containers with water.



**Part I: Observation Procedure:**

1. Male fighting fish vary in their aggressive response and you will attain the best results will be from the more aggressive males. The sight of its own reflection in a mirror is enough to stimulate agonistic flaring in a sufficiently aggressive male (they do not recognize themselves, and think they see another male). To assess the relative aggressiveness of the animals at your station, and to observe the behaviors associated with agonistic display, slowly and carefully place a small mirror against the flat side of the bowl. Do not hold the mirror near the bowl for more than a minute or so. After a minute or so, you may reapply the mirror for an additional response. If your animal fails to display or the response is less intense than that of other fish, notify your laboratory instructor (you may need a more aggressive animal before continuing).

These are some of the components of agnostic displays to look for:(1) swimming, (2) swimming away, (3)pectoral fin movement, (4) raising of dorsal fin, (5) lowering of ventral fin, (6)expansion of tail, (7) increased coloring, (8)extension of gill covers, (9) facing the intruder

Record:

A.Which fins or other structures does the fish use for display?

B. What position does the fish assume with respect to its reflection (head-on? sidelong?)

C. What other movements or behaviors does the fish employ in his display?

D. Record any changes in the coloration of your fish (look for either "bleaching" or brightening in color).

E. Do the gill opercula open first; or are these structures one of the last to be displayed during an agonistic encounter?

**CAUTION:** Students with brightly-patterned clothing may excite some male fighting fish. Put on a lab coat or smock to hide these colors from the fish during the experiment. If the fish is over-stimulated, habituationto the stimulus will occur. This means that the animal has become accustomed to the stimulus, and will not respond as strongly (or respond at all) in subsequent trials. Do not position your fish where it can see neighboring animals.. Do not tap on the side of the fish bowl. Avoid abrupt movements when near the fish and speak quietly

**Part II: The Effect of Different Releasers on the Agonistic Display: Experimental Design.**

**DO NOT *EVER* PLACE TWO FISH IN THE SAME BOWL. SUCH IRRESPONSIBLE BEHAVIOR WILL RESULT IN YOUR BEING DISMISSED FROM THE LAB AND YOUR RECEIVING A ZERO FOR THIS LAB EXERCISE. FURTHER DISCIPLINARY ACTION IS ALSO POSSIBLE.**

Available at the front desk are materials such as paper cutout models ("puppets") of male and female Bettas of different colors, different sizes, and different positions. You can use these to pose questions about Betta agonistic behavior, devise null and alternative hypotheses, and then design and execute a rigorous experiment to test your ideas. Examine the puppets available, and use them to ask questions and formulate hypotheses.

Consider some of the following questions, and ask some of your own.

1. What releaser elicits the strongest response in your subject?

2. What fixed action pattern is used in response to particular releasers?

3. What is the possible evolutionary significance of the responses of the fish to various releasers?

Work in teams of four. Take the first 20 minutes of lab to consider *Betta splendens* natural history (feel free to do additional research before you come to lab!), and design an interesting, relevant experiment with the materials at hand. (If you require somethingelse, please ask. We might have it available.) Once your team has decided on

its course of action, complete the four outline items and draft a brief description of your experimental methods.

Observation:

Question:

Null Hypothesis:

Alternative Hypothesis:

Experimental Methods:

**General Instructions for a Successful Experiment**

1. Some individual fish are particularly sensitive or aggressive, and these may be stimulated even by the sight of brightly colored or patterned clothing. Avoid wearing such clothing to lab this week. Pale-colored clothing is the least likely to interfere with fish behavior.

2. Position your fish where it cannot see neighboring animals until you are ready to begin your experiment. Keep the visual barriers in place unless you are actually making observations of fish behavior.

3. Avoid abrupt movements when near the fish, and speak quietly. ***Do not tap on the side of the fish bowl.***

4. When recording the positions and movements of your Betta's display, note that the male will employ most of its fins, its gill opercula, and the associated branchiostegal membrane. A particularly energetic male may bend his body in tight angles. Note the orientation of the fish to its stimulus and record any changes in the coloration of your fish (for example, watch for color to fade or become brighter, or for color streaks to appear on various areas of the body.

5. Be sure to record the length of time of each behavior you are recording, as well as its subjective aspects (for example, you might rate the strength of the display ("-" for weak, "+" for medium, "++" for strong—or variations on that theme, perhaps using numbers to rank degree of energy shown by the fish.) Do not stimulate the fish for longer than one minute for each trial, as longer trials may result in habituation to the stimulus.

NR = No response

M = Mild response (not as strong as the response from the mirror)

S = Strong response (same response as mirror)

6. Wait least 5 minutes between trials, allowing the fish to calm down completely. Between trials, be sure to block your subject's view of other fish, and avoid fast movements or loud noises. (Yes, they can hear them!)

7. It may be helpful to record the sequence of movements the fish uses in a full display.

8. Replicate each trial at least 3 times. Note any differences between replicates. What might cause such differences as time goes on?

9. Paper fish models will also elicit a response from males, but because they are stationery, they will not elicit as strong a response. However, the subject will also not habituate as quickly to a static model as to a mirror (why might this be the case?)

10. When using a paper model, move it slowly up to the subject and then wave it slightly to attract the subject's attention. Try to use similar technique and movement in each trial, to avoid introducing human error into your experiment.

11. Obviously, male Bettas will respond strongly to the sight of another member of its own species, whether male or female. You may wish to measure the responses of your fish to same sex or different sex stimulus, and quantify the differences between the displays. The question, hypotheses and predictions are up to you!

12. Determining the relative aggressiveness of different animals can be done by

comparing displays recorded by different teams with different animals.

You may use the table below to record your data any way you wish, or make a different table of your own design. Be sure to note in the table legend (which you will provide) what you are measuring (nonparametric, such as "yes" or "no", or parametric, such as "seconds" or other discrete or continuous measurement) as well as their values.

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| --- | --- | --- | --- | --- | --- | --- | --- |
| Trial # | Dorsal  fin | Caudal  fin | Ventral  fin | Pelvic  fin | Branchiostegal  membrane | Orientation | Color change? |
|  |  |  |  |  |  |  |  |
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**The Lab Report**

Using the lab report format you have been given, write a lab report for the Betta Experiment.