

## The effect of variable environments on aggressive behavior in *Betta splendens*

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### ABSTRACT

Sixteen adult *Betta splendens* were tested for aggressive behaviors based on environmental changes. Two groups of eight fish were randomly assigned. The two groups both experienced both treatments of control and variability. The variable group experienced structural changes during the treatment to produce variability. They were then tested through an ethogram after each treatment to measure aggressive characteristics. The procedure used was focused on the already natural aggression *Betta splendens* have. Placing a mirror in the tank for a duration of fifteen minutes with observation triggers this aggressive behavior. An ethogram was made to make observational characteristics measurable and quantitative during this mirror observational period. The action of changing the structural environment within each tank comparatively to the same fish being in a controlled structural environment did not change aggressive behavior.

Keywords: *Behavioral Science, Betta splendens, Environmental Science, Ethogram, Siamese Fighting Fish*

### INTRODUCTION

In our ever-changing world today, the environment is no exception. The deforestation and destruction of many rural areas have consequently led to the destruction of living organisms' ecosystems. Another major factor is crop rotation performed by many farmers in which the land is used for more than one type of crop yearly. This leads to the organisms living in these areas to go through extreme change in their environment. These environmental disturbances range from machinery destroying or the change directly to new implication of environmental factors.

How constantly fluctuating environments affect organisms and whether this has an effect on their survival and conservation is an important aspect to research. One response to variable environments could be aggression.

The Siamese fighting fish, *Betta splendens*, is currently studied by the scientific community for its aggressive behavior. They are native to Thailand, Cambodia, Malaysia, and Myanmar (Srikrishnan 2017). Within those geographic areas, their natural habitat is in large, heavily vegetated marshes, rice paddies and slow-moving streams. These fish can be territorial and are prone to high levels of aggression towards each other because of this (Meliska 2004). In contrast, females are not as aggressive as males, due to the competition and natural selection in the wild. *Betta splendens* are listed as vulnerable by the IUCN. The Siamese fighting fish is an ideal subject to study aggression in animals, and the variables contributing to that behavior.

The experimental study of *Betta splendens* is used very often due to how easy it is to identify signals of aggression along consistent protocols used to invoke these displays are practiced. Opercular flaring will be measured according to the number of repetitions,

along with the strikes accosted with the flaring (Meliska 2004). Orientation is also measured to identify how "curious" or "timid" their behavior traits are. These behavioral attributes of the Siamese fighting fish conclude them to be ideal models to study the factors affecting aggression.

Some things we know as a scientific community are; *Betta* fish can efficiently control the population of bacteria (Lima et al 2010). Exposed to mirror image for a long duration, they become accustomed and show low signs of aggression (Baenninger 2014). Initially, *Betta* fish will show high levels of aggression toward mirror image even when dosed with Fluoxetine (Bogdan 2010). An audience of other *Betta splendens* will create experimental issues when studying behavior, this is due to their emitter-reciever dyadic interactions and networking with one another (Doutrelant 2001). These receivers' interactions or simply the visibility between fish will therefor result in altercation of results. Also, Carotenoid Pigments are responsible for the color of a *Betta* fish (Clotfelter 2007). They show examples and non-verbal communication of sexual attractiveness between two fish. Lastly, fish utilize learning and memory to orientate within their natural environments (Way 2015).

My particular research will focus on the effects an environment can have on *Betta* levels of aggression. More specifically, it will focus on the effects of variable structural landscapes. These fish and many other organisms in our vast world face new structural challenges such as trash, or pollution and some even face the opposite or removal of structures due to deforestation, or domestication. This experiment will result in data to show the true impact many organisms are facing and how society/variations are changing

their behaviors.

## MATERIALS AND METHODS

### Fish Care and Maintenance

Sixteen *Betta splendens* were purchased for this study from Petco in Wichita, KS. Each fish was set up in their own individual fish tank that was blocked off from each other, due to prone aggression towards each other (Doutrelant 2001). Sixteen new and clean tanks were prepared with water and fish pH water conditioner. They were left to sit for five days to complete conditioning and adjust completely room temperature. Next, within each tank, three vegetation mimicking structures were placed in the corners, along with an air stone connecting to the main filtering air pump. The fish were then each added to their respectively labeled tanks and once again allowed to fully settle in with an acclimation period of three days.

### Experimental Setup

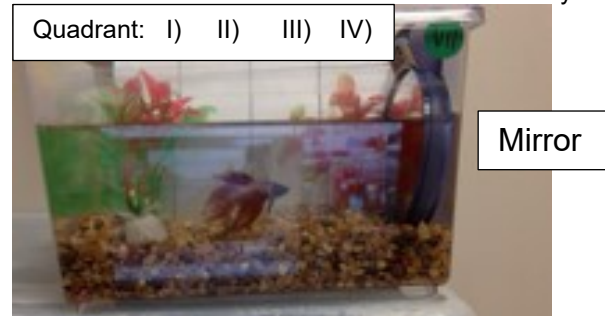
Each of the 16 fish experienced both experimental treatments in this repeated measure design. The two experimental treatments were: 1) Control treatments, which will not undergo any structural changes within their tanks. This group will remain constant and only be slightly moved during feeding, and 2) Variable treatment, which will undergo a structural change every day of the treatment period. The structural change was made by replacing current and artificial vegetation with a new colored and new shaped vegetation block. This replacing will occur on one of the three already in place artificial vegetation blocks at random.

After the acclimation period, half of the sample population will be identified as the "Control-1" group, and half will be identified as the "Variable-1" group. We will need a separate living area for each fish in each of their designated (completely random) groups. Each tank will have three structures of vegetation in three corners. The "Variable" group will go through structural change in their environment, specifically the replacement of a new structure for an old one every single day. The control group will live in a stationary, non-changing, environment for the entire experiment. From prior knowledge, we know they will lose aggression towards a mirror-ethogram study if done for a long duration of time. So, we will conduct this experiment for only two and a half weeks. \*Not including set-up or take-down time.

Following the initial trials, each fish was given three days before the next treatment began. Each treatment lasted seven days, and were performed equivalently.

Prior to experimental observation, I set up a camera to record the trial and then placed the mirror in the tank (set-up shown in Figure 1). All observations and actions of the fish, including number of opercular flares, time spent flaring, time oriented to mirror, color

change, time spent in each quadrant of tank, and if immediate cover was taken was recorded and analyzed.



**Figure 1.** Observational tank setup and quadrants

ed in the ethogram structure for duration of the 15-minute trial. After this, the fish will return to its original location in the laboratory and relabeled for the vice versa scenario in the experiment. i.e. Control to Variable or Variable to Control. Another acclimation period is then allowed for three days for the experiment to settle.

A paired t-test was used to compare the results found. The comparison between one fish in both treatments will conclude in direct relation of structural change and aggression. The t-test comparison was used to compare the number of opercular flares, time spent oriented to mirror, and time spent in each quadrant of tank.

## RESULTS

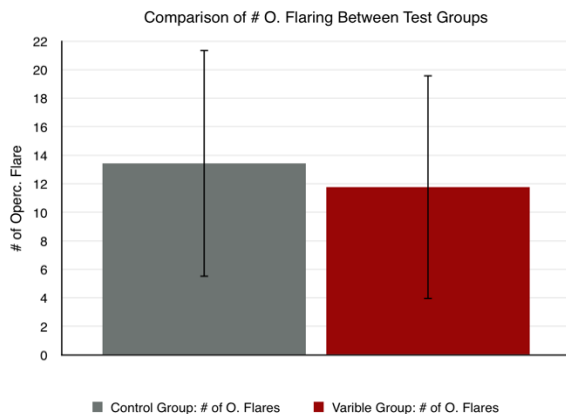
The number of opercular flares did not differ between control groups and variable groups (Figure 2;  $t=0.557$ ,  $df=15$ ,  $p=0.586$ ). The next specified and measured characteristic of these fish was the time spent oriented to the mirror. The time spent oriented to the mirror did not differ across the environmental treatments (Figure 3;  $t=0.629$ ,  $df=15$ ,  $p=0.539$ ). The time spent in each and all quadrants did not differ across the environmental treatments: Quadrant I (Figure 4;  $t=0.069$ ,  $df=15$ ,  $p=0.964$ ). Quadrant II (Figure 4;  $t=0.954$ ,  $df=15$ ,  $p=0.355$ ). Quadrant III (Figure 4;  $t=0.008$ ,  $df=15$ ,  $p=0.994$ ). Quadrant IV (Figure 4;  $t=0.834$ ,  $df=15$ ,  $p=0.417$ ). The respective comparisons and results are also shown on the following page.

## DISCUSSION

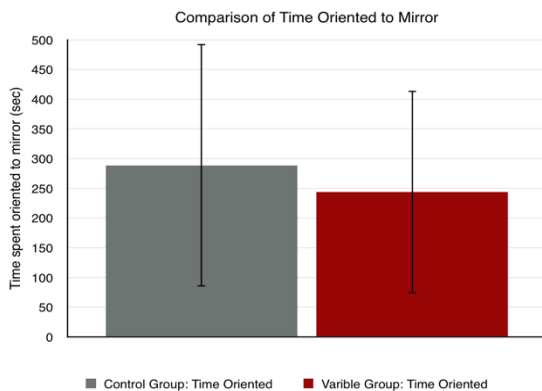
Based on the results, it can be concluded that aggression to be not affected by the structural change of their environment for this specific study.

Due to the lack of an effect on aggression in this study, it is suggested to redo this experimental procedure with accommodating changes. The fish used in this experiment were derived from local stores and pet carriers. This was due to the availability but could have in fact pre-neutralized these fish to

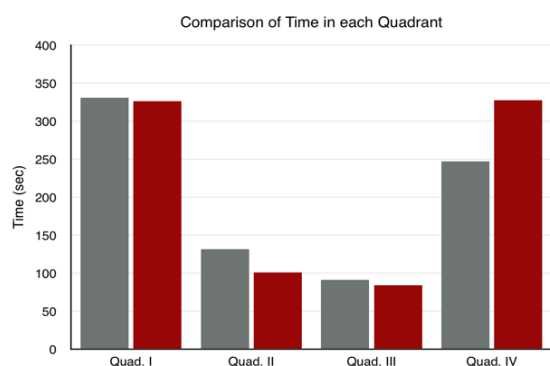
changes in their environment. From previous research, we know the possibility of these fish already



**Figure 2.** Number of opercular flares between two testing groups, control and variable. ( $t=0.557$ ,  $df=15$ ,  $p=0.586$ )



**Figure 3.** Comparative results of time spent oriented to mirror between two testing groups, control and variable. ( $t=0.629$ ,  $df=15$ ,  $p=0.539$ )



**Figure 4.** Time spent in each quadrant during trial, comparatively between two testing groups, control and variable. Quadrant I ( $t=0.069$ ,  $df=15$ ,  $p=0.964$ ). Quadrant II ( $t=0.954$ ,  $df=15$ ,  $p=0.355$ ). Quadrant III ( $t=0.008$ ,  $df=15$ ,  $p=0.994$ ). Quadrant IV ( $t=-0.834$ ,

$df=15$ ,  $p=0.417$ )

being exposed to brain stimuli that corrupted natural aggressive judgments (Bogdan 2010). Another uncertain aspect of this research was the visibility between fish at all time. There were small gaps of time during experiment where these fish could have made visible contact, and this could have skewed the results.

Another possible source of explanation based on our results is called acclimation (Kozłowski 2002). This is the process of an organism becoming accustomed to a new climate or condition. This is an extremely important explanation of our result. *Betta splendens* need the ability to adapt or acclimate to a new environment to prolong organism life. Without the ability to adapt these fish would not be efficient or effective in the wild.

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