UC Berkeley
Student Research Papers, Fall 2011

Title
BEHAVIORAL SYNDROMES IN JUVENILE DASCYLLUS TRIMACULATUS (THREE-SPOT DASCYLLUS) IN MO'OREA, FRENCH POLYNESIA: INDIVIDUAL VARIATION IN SHYNESS/BOLDNESS

Permalink
https://escholarship.org/uc/item/3k0228kq

Author
Parkhouse, Tess

Publication Date
2011-12-12

Undergraduate

eScholarship.org Powered by the California Digital Library
University of California

BEHAVIORAL SYNDROMES IN JUVENILE *DASCYLLUS TRIMACULATUS* (THREE-SPOT *DASCYLLUS*) IN MO'OREA, FRENCH POLYNESIA: INDIVIDUAL VARIATION IN SHYNESS/ BOLDNESS

TESS A. PARKHOUSE

Integrative Biology, University of California, Berkeley, California 94720 USA

Abstract. Behavioral syndromes can provide an evolutionary advantage because they allow a population to quickly adapt to dynamic selective pressures. This study used field manipulations to determine whether a behavioral syndrome is present in juvenile *Dascyllus trimaculatus.* Within- and between- individual consistency was found in individuals' responses to a predator model, a non-predator model, and a novel object. These tests were completed in the wild in Mo'orea, French Polynesia and filmed for further analysis. Behavioral syndromes are shaped by a wide array of factors, so studies in this topic contribute both to a better understanding of a species' behavior, as well as the selective pressures it faces.

Key words: animal behavior; Dascyllus trimaculatus; *behavioral syndromes; individual variations; bold/shy continuum; Mo'orea, French Polynesia*

INTRODUCTION

Behavioral responses differ between individuals in a species or population, which can provide crucial understanding about the selective pressures that shape behavior (Dzieweczynski *et al.* 2011). Individuals in a given population must trade off potential risks (e.g., exploring a novel object, defending territory from a threat) for possible gains in resources (Scharnweber *et al.* 2011). Research in various animal taxa, including mammals, birds, reptiles, amphibians, fish, arachnids, crustaceans, insects, and cephalopods, has shown that there are consistent differences in behavioral patterns between individuals (Smith 2008). Dynamic selective pressures maintain individual variation, allowing species or populations to better adapt to their environment and reduce the risk of extinction (Smith 2008). Therefore, the study of individual variations in behavior, called *behavioral syndromes*, is a key aspect of both behavioral and conservation biology.

Previous studies have shown that individuals may vary in the level of risk they will take along a bold/shy continuum (Scharnweber *et al.* 2011). This trait, like many others, is represented as a continuum with two extremes represented as tendencies. Boldness represents the way an individual reacts to a risky situation. In this trait, "bold" and "shy" are two extremes rather than different traits (Réale *et al.* 2007). These variations in boldness may be a result of a wide array of abiotic or biotic factors, including limited habitable space, increased predator density, amount of food available, etc. When there are correlations between an individual's behavior across multiple contexts or situations, a behavioral syndrome is thought to exist (Sih *et al.* 2004b). The presence of a behavioral syndrome is significant because it indicates limited plasticity that carries over to multiple contexts, and shows the significance of behavioral variation for the survival of a population (Sih *et al.* 2004a).

There are two aspects of a behavioral syndrome: within-individual consistency and between-individual consistency. Within-individual consistency refers to the tendency for any given individual to exhibit consistent behavior across observations, while between-individual consistency refers to consistent differences among individuals in behavior expressed statistically as behavioral correlations (Sih *et al.* 2004b). In this study, both aspects of a behavioral syndrome will be examined in juvenile *Dascyllus trimaculatus*.

Juvenile *Dascyllus trimaculatus*, commonly known as three-spot *Dascyllus*, are readily found around the anemone *Heteractis magnifica* in Mo'orea, French Polynesia. The three-spot *Dascyllus*, like many reef fish, have a planktonic larval stage (Bernardi *et al.* 2001), after which they settle on anemones and mature into adults within a year (Holbrook and Schmitt 2005). Upon reaching maturity, they leave the anemone and settle into reef crevices (Bernardi *et al.* 2001). The juvenile three-spot *Dascyllus* is an ideal study organism when examining behavioral syndromes because of its use of anemones for safety. They cluster near the base of the anemone when a perceived threat is near, and venture outward when the threat no longer exists (Personal Observation). This system allows bold behavior to be defined as distance and time spent away from anemone. There have been studies on the bold/shy continuum in other fishes in the past, however, many of them have been completed in laboratory contexts rather than natural environments (Scharnweber *et al.* 2011, Dzieweczynski *et al.* 2011). This natural measurement allows for observations to be made through contextual manipulations in the field. In order to fully understand the role of the environment in shaping individual variations in behavior it is critical to carry out experiments under natural conditions (Brown *et al.* 2005).

This study examined the bold/shy continuum in *Dascyllus trimaculatus* to determine if a behavioral syndrome is present in this species. Based on previous observation and similar studies done for other species, it was hypothesized that a behavioral syndrome would in fact be present. Group size and individual size were also briefly examined. It was predicted that larger group size would increase the average level of boldness, but that the size of an individual would have no effect.

METHODS

Study site

The fieldwork for this study was Cook's Bay, directly in front of the Richard B. Gump Research Station in Mo'orea, French Polynesia (17°29'25.69"S, 149°49'33.12"W). This location was chosen because of the high numbers of three-spot *Dascyllus* fish there, which ensured a large sample size. In Cook's Bay, there is a fringing reef that quickly drops off into deeper water. Most of the anemones that host the three-spot *Dascyllus* are located near or on the edge of the coral fringing reef. Snorkel was used to conduct the research, though some free diving was necessary for the experimental set up. The average depth of the anemones that the field experiments were conducted at was 2.1 m.

Experimental setup

In this experiment, video was used to document the behavior of the three-spot *Dascyllus* while a series of tests were performed in the field. The video camera was placed on a tripod and weighed down with dive weights to the depth of the anemone. It was placed about 75 cm away from and directly in front of the anemone's oral surface, or as close to this set up as possible depending on the bottom conditions. Once the video started, there was a 5 minute acclimation period before beginning each test to minimize any effect from the presence camera or researcher on behavior. Three tests were completed to get a full picture of the individual's behavior across multiple contexts, with an hour break between each test. The tests included the introduction of a known predator model, a novel object, a known non-predator model. The tests were done in a different order for each of the three replicates to avoid bias.

The models were made out of thin plastic material which was then painted with a gloss enamel. The predator was a *Myripristis amaena* (brick soldierfish), which are known to feed on juvenile three-spot *Dascyllus* (Holbrook and Schmitt 2002). The non-predator was a *Chaetodon lineolatus* (lined butterflyfish), which feeds on coral polyps, benthic invertebrates, and algae (Randall 2005). The novel object was a blue square that was similar in size to the fish models (12 cm). The fish models were attached to a thin wooden pole and placed in the water at the height of the anemone, and the novel object was hung with fishing line at the height of the anemone. Different methods were used with the fish models and novel object due to limited supplies, however, a thin wooden pole without any models on it was also tested in the field to ensure it did not create a confounding variable. Each object was placed 20 centimeters away from the anemone and held in place for ten minutes. Each test was completed three times. Once the first set of tests were done, the following tests were each completed two days after the test before until all three replicates were complete.

Video analysis

Once the tests were complete, video analysis was used to follow each individual, see if its boldness level remained consistent between each test, and determine where in the bold-shy continuum the individual should be placed. Several methods were used to quantify boldness. The fish were observed several times before the testing began and their behaviors were defined and transcribed into an ethogram (Table 1). Once this was complete, certain behaviors were determined to be more or less bold based on the amount of risk it entailed. All the behaviors in the ethogram were a normal part of the fishes' movements except for the hiding behavior, which only happened when a perceived threat occurred. Based on this information, less time spent in hiding was defined as more bold for this study.

Table 1: Ethogram of relevant behaviors

Behavior	Definition
Hiding	Taking cover behind an anemone or in rock crevices.
Chasing	Quickly swimming at a fish.
Darting	Rushing forward or to either side.
Exploring/ venturing	Swimming further than 10 cm away from the anemone.
Hovering	Remaining stationary in the water column.
Retreating	Maintaining body position while using pelvic fins to move backward.
Positioning	Facing intruder while remaining relatively stationary in the water.
Approaching	Moving toward another fish or object.

Several factors were documented during the process of video analysis. Emergence time after the models were presented, as well as reemergence time once the models were removed were noted for each test. The percentage of time that each fish spent out of hiding was also calculated for each test. The distance that each fish traveled away from the anemone was quantified using four categories: hiding— the individual remained out of sight for over ninety percent of the test, close— the individual remained immediately adjacent to the anemone, medium— the individual did not go further than half the video screen distance from the anemone, and far— the individual went to the edge of the screen or beyond. Once these data were collected for each individual, the information was used to determine if there is variation between individuals.

Statistical analysis

Once the boldness level was numerically quantified, it was possible to statistically analyze the data that had been obtained. JMP version 7 statistical package was used for all statistical calculations. First, a MANOVA analysis was run to determine whether or not there was variation between each individual. That determined whether the three-spot *Dascyllus* met the between -individual consistency requirement of a behavioral syndrome. Next, between individual consistency was tested by quantifying behavioral repeatability (Lessells & Boag, 1987), and Spearman rank correlations were calculated between each context. Repeatability values ranged from 0 to 1 with higher values indicating that more variation was explained by between- than within individual differences (Dzieweczynski *et al.* 2011). Lastly, the effect of group size was analyzed by testing for correlations between the number of fish in each group and the average level of boldness.

RESULTS

Overall means

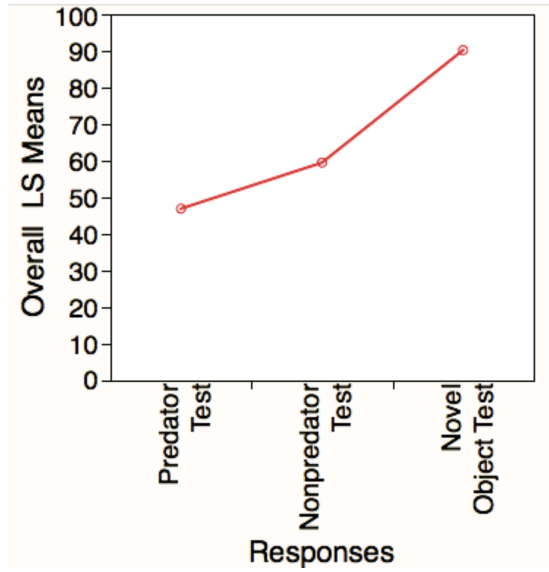

Fig. 1. Mean least squares of percentage of time spent out of hiding in each context by all 14 *Dascyllus trimaculatus* individuals in Mo'orea, French Polynesia.

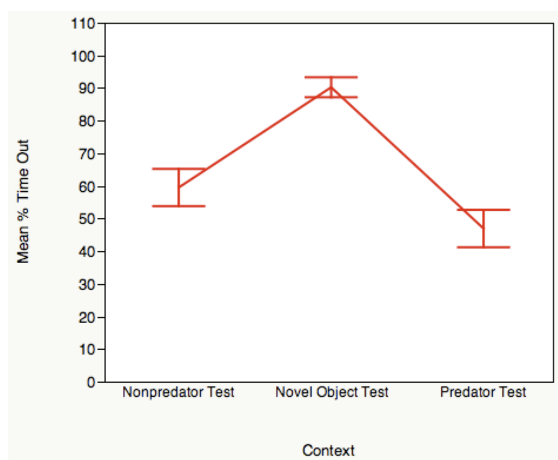

Fig. 2. Mean percentage of time spent out of hiding in each context by all 14 individuals in Mo'orea, French Polynesia. Each error bar is constructed using one standard error from the mean.

On average, the three-spot *Dascyllus* spent more time in hiding when the predator model was present than when the non-predator and novel object were present (Fig. 1). When the predator model was present, the fish spent an average of 46.92% of the time in hiding. The novel object had the smallest effect on behavior; during this test all individuals remained out of hiding for the majority— an average of 90.22%— of the testing period.

Individual variations

There are distinct, consistent differences in response between individuals across contexts. Some of the fish tested spent a higher percentage of time out of hiding in each context than others (Fig. 3). For example, individual 8 spent 94.2% of the time during the predator test, 89.2% of the time during the non-predator test, and 98.9% of the time during the novel object test out of hiding, making it the boldest individual. In contrast, individual 13 spent 0.35% of the time during the predator test, 4.5% of the time during the non-predator test, and 45.2% of the time during the novel object test out of hiding, making it the shyest individual across contexts.

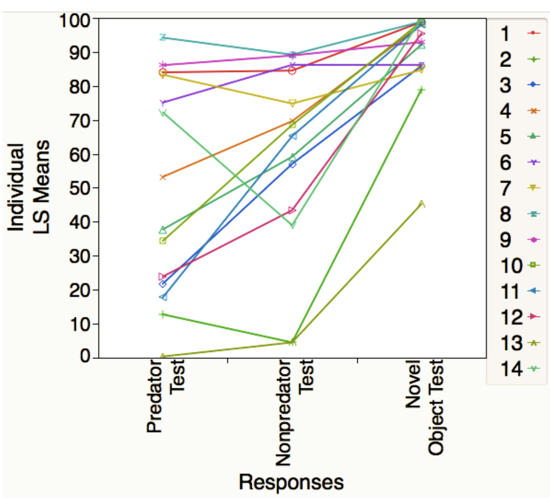

Fig. 3. Mean least squares of percentage of time spent out of hiding in each context by 14 *Dascyllus trimaculatus* individuals in Mo'orea, French Polynesia. Percentage of time varies between individuals.

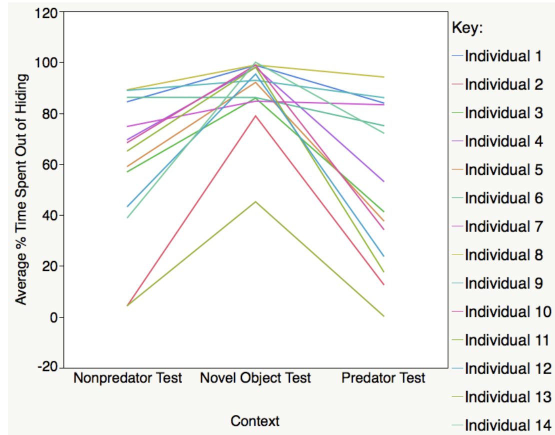

Fig. 4. Average percentage of time spent out of hiding in each context by 14 *Dascyllus trimaculatus* individuals in Mo'orea, French Polynesia.

MANOVA analysis was used to determine that there are differences between individuals (F=3.958, NumDF=13, p=0.0003*). There are also differences in response between each context (F=5.322, NumDF=2, p<0.0001*). Individual behavior remained consistent across the three contexts (Wilks' Lambda=0.182, NumDF=26, p=0.0426*), but the response of some individuals may have varied more in one context than another.

Repeatability and behavioral consistency

Repeatability was examined to assess consistency within individuals. Behavior in the predator test was found to be highly repeatable (r=0.906) and slightly repeatable in the non-predator test (r=0.219). However, in the novel object test behavior was not found to be repeatable (r=0.096).

Behavioral consistency across contexts was reported using Spearman rank correlations. Behavior was consistent between all three tests. The predator and non-predator tests were the most highly correlated (Spearman ϱ=0.6019, p=0.0002*) followed by the novel object and non-predator tests (Spearman ϱ=0.4832, p=0.0044*) However, the novel object test and predator test were also correlated (Spearman ϱ=0.4302, p=0.0125*).

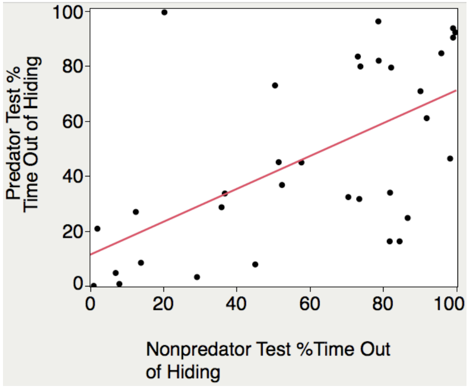

Fig. 5. Correlation between the percentage of time spent out of hiding in the predator versus non-predator tests on *Dascyllus trimaculatus* in Mo'orea, French Polynesia.

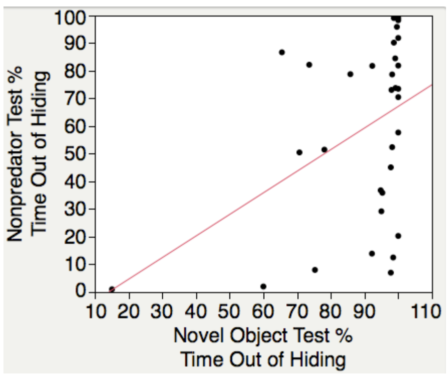

Fig. 6. Correlation between the percentage of time spent out of hiding in the non-predator versus novel object tests on *Dascyllus trimaculatus* in Mo'orea, French Polynesia.

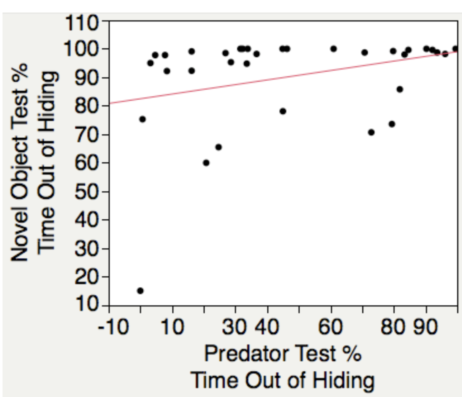

Fig. 7. Correlation between the percentage of time spent out of hiding in the novel object versus predator tests on *Dascyllus trimaculatus* in Mo'orea, French Polynesia.

The consistency between behavior in the three tests are also shown using correlation tests (predator/non-predator: RSquare=0.354, t Ratio=4.12, p=0.0003*; non-predator/novel object: RSquare=0.177, t Ratio=2.58, p=0.0149; novel object/predator: RSquare=0.095, t Ratio=1.81, p=0.0806).

Effect of group size

Based on the limited data obtained, the number of fish in each group living around the anemone also had an effect on boldness. Average boldness increased with group size on every test except for the novel object test (Fig. 8, 9, 10) . For example, average boldness increased by about 40% in the predator test when the group size increased from two fish to six fish.

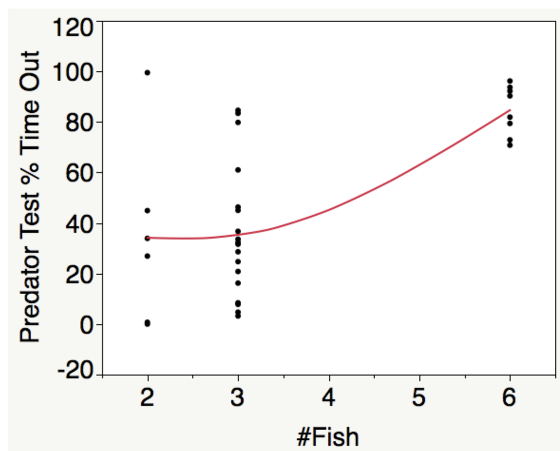

Fig. 8. The effect of group size on the percentage of time spent out of hiding by *Dascyllus trimaculatus* during the predator test in Mo'orea, French Polynesia.

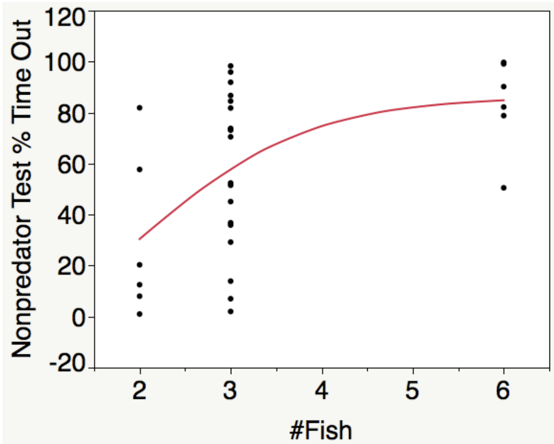

Fig. 9. The effect of group size on the percentage of time spent out of hiding by *Dascyllus trimaculatus* during the non-predator test in Mo'orea, French Polynesia.

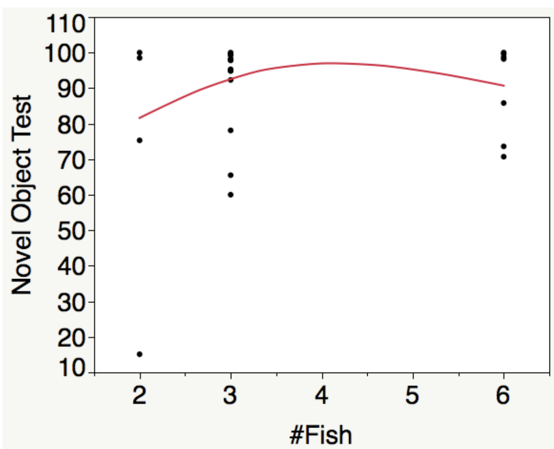

Fig. 10. The effect of group size on the percentage of time spent out of hiding by *Dascyllus trimaculatus* during the novel object test in Mo'orea, French Polynesia.

Discussion

Significance of behavioral syndromes

Based on the statistical analysis, both between and within individual behavioral consistency exists in *Dascyllus trimaculatus*, indicating that a behavioral syndrome is present. The existence of behavioral syndromes is significant because they provide insight into the evolutionary history of the three-spot *Dascyllus*. More time spent feeding and more time spent hiding both have positive effects on fitness (Sih *et al.* 2004a). Feeding gives *Dascyllus trimaculatus* the energy it needs to survive and reproduce, but requires a higher level of boldness because hiding helps to protect it from predation. These two behaviors, therefore, are negatively correlated. The three-spot *Dascyllus* cannot hide while feeding, so it has to make trade offs. Variations in the type of behavior an individual exhibits also shows variations in the level of risk an individual will take for potential gains in resources. It is essential for an individual to make these trade offs. In some situations a high level of boldness is more favorable, in others, like the example of predation, it is not (Sih *et al.* 2004b). With this reasoning, there is no perfect route to fitness so behavioral syndromes may be an essential evolutionary tool to protect the population of a species as a whole through various situations (Dzieweczynski *et al.* 2011).

Applications in behavioral biology

Though several studies have addressed this question in other species, behavioral syndromes have not been studied in *Dascyllus trimaculatus*. It is, however, an important goal to obtain a more complete understanding of three-spot *Dascyllus* behavior and the evolution of behavioral syndromes. Individuals that were more bold when the predator model was present were also more bold when the non-predator model and novel object were present. The consistent behavioral differences across contexts indicate that a behavioral syndrome is present in juvenile three-spot *Dascyllus*. Behavioral syndromes provide an evolutionary advantage because they allow a population to quickly adapt to dynamic selective pressures. The existence of this behavioral syndrome indicates limited (less than optimal) behavioral plasticity and behavioral carryovers across contexts, instead of optimal plasticity in each isolated situation (Sih *et al.* 2004a).

Applications in Conservation Biology

The existence of behavioral syndromes is also significant for the field of conservation biology. The presence of these individual variations offers evidence that not only is genetic diversity important for the survival of a population, but behavioral diversity is well. Further research that examines the links between genetics and behavioral types could be beneficial for future conservation projects (Adriaenssens & Johnsson 2010). These implications also apply on an ecosystem level. Coral reefs are threatened on a global scale, and interactions between species have huge effects on the health of coral reefs. Though this study did not address this issue, variations in behavior may result in varying interactions between species, so it is important to consider when examining ecosystems as well.

Issues with the Novel Object Test

The predation test was found to be highly repeatable and the non-predator test was slightly repeatable. This shows that each individual behaved consistently, again indicating the existence of a behavioral syndrome. The novel object test, however, had very low repeatability. There are many possible reasons for this. The novel object did not have a large effect on the fishes' behavior. Because of this, there was a much smaller range in the percent time spent out of hiding. This may have led to low repeatability because variations in a couple seconds had a much larger effect on the differences between individuals. If that is the case, the way boldness was quantified also could have created a bias during the novel object test. More time spent out of hiding was considered to be a higher level of boldness, but since the novel object had such a small effect on behavior, some of the time spent hiding may have simply been normal patterns of movement rather than an attempt to avoid a perceived threat.

Furthermore, the novel object may not have actually been considered an unknown by the fish. There were many locations along the reef that were flagged for research, and these flags were very similar and size and shape to the novel object. If it were possible to continue this study, there are some changes that could be made to avoid this issue. Rather than using a shape for the novel object test, a novel fish model could be used. The behavior of the three-spot *Dascyllus* would most likely be more affected by this unknown potential risk.

Future Research

Though there was limited data, the results also show that group size affects the level of boldness in a fish. This may be because though individual variations in behavior exist, there is still a level of plasticity in behavior. In a previous study on rainbow trout, fish were documented changing their behavior after observing the behavior of their conspecifics (Frost *et al*. 2007). Individuals that typically would have been more shy may have realized that it was safe to come out of hiding when the more bold individuals were not harmed while they were exposed, and changed their behavior based on this information. This is a topic that should be further addressed by future research. It is important to note, however, that although average boldness levels increased in larger groups, even within groups of different sizes there were still variations between individuals.

Based on field observations, the size of an individual did not seem to affect the level of boldness. There were some both large and small fish were seen at both ends of the bold/shy continuum. This, however, is also something that should be studied further.

It would also be interesting to see how varying other social factors affect the behavior of these fish. One possible way to study this is to first test behavior of individuals within a group, then isolate individuals to see how behavior changes once a fish is alone.

Additional research should also be done to determine the specific selective pressures that shape and maintain behavioral syndromes. Dynamic environmental conditions change the selective pressures that favor various behaviors over the course of an individual's lifetime (Adriaenssens and Johnsson 2010). Conducting a long term study on both the juvenile and adult *Dascyllus trimaculatus* could provide insight into how behavioral syndromes are maintained.

Acknowledgments

I would like to thank my professors Dr. George Roderick, Dr. Jere Lipps, Dr. Pat Kirch, and Dr. Vince Resh; and my GSIs Bier Kraichak, Jenny Hofmeister, and Virginia Emery for their continuous support throughout this project. It would not have been possible without their help. I would also like to thank the entire IB158 class for their support, specifically Jennifer Bates for her help creating the models used in this project; as well as Allie Shea, Bryan Frossard, and Gwen Hubner for their assistance in the field. Lastly, I would like to thank my family for all the love and support they have given me.

Literature Cited

Adriaenssens, B. and J.I. Johnsson. 2010. Shy trout grow faster: exploring links between personality and fitness-related traits in the wild. Behavioral Ecology **22:** 135-143.

Bernardi, G., S.J. Holbrook and R.J. Schmitt. 2001. Gene flow at three spatial scales in a coral reef fish, the three-spot dascyllus, *Dascyllus trimaculatus*. Marine Biology **138**: 457-465.

Brown, C., F. Jones and V. Braithwaite. 2005. In situ examination of boldness-shyness traits in the tropical poeciliid, *Brachyraphis episcopi*. Animal Behavior **70:** 1003-1009.

Dzieweczynski, T.L. and J.A. Crovo. 2011. Shyness and boldness differences across contexts in juvenile three-spined stickleback *Gasterosteus aculeatus* from an anadromous population. Journal of Fish Biology **79:** 776-788.

Frost, A.J., A. Winrow-Giffen, P.J. Ashley, and L.U. Sneddon. 2007. Plasticity in animal personality traits: does prior experience alter the degree of boldness? Proceedings of the Royal Society **274:** 333-339.

Holbrook, S.J. and R.J. Schmitt. 2002. Competition for shelter space causes density-dependent predation mortality in damselfishes. Ecology **83**: 2855-2868.

Holbrook, S.J. and R.J. Schmitt. 2005. Growth, reproduction and survival of a tropical sea anemone (Actiniaria): benefits of hosting anemonefish. Coral Reefs **24:** 67-73

JMP, Version 9. SAS Institute Inc., Cary, NC, 1989-2011.

Lessells, C.M. and P.T. Boag. 1987. Unrepeatable repeatabilities: a common mistake. The Auk **104**: 116-121.

Randall, J.E.. 2005. Reef and Shore Fishes of the South Pacific: New Caledonia to Tahiti and the Pitcairn Islands. University of Hawai'i Press, Honolulu, Hawai'i.

Réale, D., S.M. Reader, D. Sol, P.T. McDougall, N.J. Dingemanse. 2007. Integrating animal temperament within ecology and evolution. Biological Reviews **82:** 291-318.

Scharnweber, K., M. Plath and M. Tobler. 2011. Examination of boldness traits in sexual and asexual mollies (*Poecilia latipinna*, P. formosa). Acta Ethologica 14: 77-83.

Sih, A., A. Bell and J.C. Johnson. 2004. Behavioral syndromes: an ecological and evolutionary overview. Trends in Ecology and Evolution **19:** 372-378.

Sih, A., A.M. Bell, J.C. Johnson and R.E. Ziemba. 2004. Behavioral syndromes: and integrative overview. The Quarterly Review of Biology **79:** 241-277.

Smith, B.R. 2008. Individuality in animals: Consistent behavioral differences from an adaptive and conservation perspective. ProQuest, Ann Arbor, Michigan.