Associations Between Anxiety and Attention in Laboratory-Housed Rhesus Macaques (Macaca mulatta)

Lauren E. Hobbs
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Associations Between Anxiety and Attention in Laboratory-Housed Rhesus Macaques

(Macaca mulatta)

A Thesis Presented

By

LAUREN E. HOBBS

Submitted to the Graduate School of the University of Massachusetts Amherst in partial fulfillment of the requirements for the degree of

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Neuroscience and Behavior Program
Associations Between Anxiety and Attention in Laboratory-Housed Rhesus Macaques
(Macaca mulatta)

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I would first like to thank my advisor, Melinda Novak, for her continuous guidance and support ever since I joined the lab back in 2013. Through the encouraging atmosphere that she instills within our lab, I found a home, a true love for research, and unwavering support from a wonderful group of dedicated students. I would also like to thank my committee members, Jerrold Meyer and Christina Metevier for their extremely honest and helpful suggestions for this project.

I wish to express my appreciation to all of my wonderful lab members who helped me get this project off the ground. Through the planning and experimental phase, you all helped me in a significant way, and I am truly grateful for your insightful ideas and feedback.

Finally, I would like to thank the wonderful monkeys in our lab – even those who were too stubborn to participate in my project. Without these intelligent creatures I would not be where I am today, and I am extremely grateful for their lifelong commitment to research and science.
ABSTRACT

ASSOCIATIONS BETWEEN ANXIETY AND ATTENTION IN LABORATORY-HOUSED RhesUS MacaQUes (MACACA MULATTA)

May 2015

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Previous studies completed with humans have revealed insight into the effects of anxiety on attention tasks such as the dot-probe task, but there is little information about such effects on non-human primates. This study aimed to assess whether anxiety or anxious behaviors would impact rhesus macaque performance on a three stimuli paradigm similar to the dot-probe task. Utilizing images of conspecifics (strong threat, mild threat, and neutral), eight monkeys were video recorded completing a task that required them to slide two doors, which held these images, to the side to obtain a treat. We hypothesized that behavioral phenotype (high or low anxiety) would affect attention on this modified dot-probe task. Additionally, we predicted that time spent looking at mildly threatening stimuli would be positively correlated with high levels of anxious behaviors (e.g., scratching, yawning, pacing, self-biting) and cortisol concentrations over a four month period. We also predicted that a higher percentage of the mildly threatening stimuli as a first choice would be positively correlated with high levels of anxious behaviors and cortisol concentrations. However, anxious behaviors and cortisol concentrations did not affect performance on this task. Interestingly, a sex difference was found for the mild threat stimuli, with females taking significantly more time to complete the task when presented
with the mild stimuli ($p = 0.01$), and also looking at the mild stimuli longer than males ($p = 0.03$). These data suggest that males and females interpret ambiguous facial expressions differently, possibly indicating the significance of attention in female dominance hierarchies in macaque social groups.
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CHAPTER 1
INTRODUCTION

1.1 Background Information

Anxiety disorders are cognitively debilitating and negatively affect millions of people worldwide. The prevalence of these disorders affects thousands of children and adolescents (Schaffer et al. 1996), and also play a role throughout the lifespan for many adults (Blazer, 2003; Schutzer & Graves, 2004). In the United States alone, 25% of adults report a lifetime prevalence of an anxiety disorder, making it the most common mental disorder (Kessler et al. 2005). The detrimental effects of anxiety disorders encompass both psychological and physical health, and can also be a financial burden, as the U.S. reported spending more than $42 billion on these disorders (Greenberg et al. 1999).

As a result of their increasing prevalence in society, humans have been the main focus of experimental research regarding the complex relationship between anxiety, emotion, and cognition. Analyses of basic cognitive processing experiments have shown that humans are biased when processing emotionally relevant information. It is now widely accepted that anxious individuals attend more to threatening stimuli than non-threatening stimuli (Mineka & Sutton, 1992). Initially, this bias was tested with the Stroop task, which requires participants to name the color of threat and non-threat words. Anxious individuals were significantly slower to name the color of threat words. (Mathews & MacLeod, 1985; McNally et al. 1990; Foa et al. 1991). However, questions were raised as to whether the Stroop task was an adequate measure of attentional bias. Thus, other visual attention task paradigms have been developed. MacLeod, Mathews, and Tata (1986) used an early version of the dot-probe task, where two words (a threat
related and non-threat related word) appeared on a computer monitor one on top of the other. Participants were asked to read the top word aloud, but sometimes a probe would randomly appear in place of one of the words, and they were required to respond using the mouse. In this study, the experimenters predicted that faster reaction times would occur when the probe took place of the word they were asked to pay attention to, or read aloud. The study demonstrated that clinically anxious participants had faster reaction times when responding to the probe that replaced the threat word, as opposed to the non-threat word. Non-anxious participants had faster reaction times when responding to the probe that replaced the non-threat word. MacLeod et al. concluded that clinically anxious participants are more inclined to attend to threat related words, while non-anxious participants attend to non-threat related words.

With MacLeod’s dot-probe task as a foundational model, scientists were now able to investigate further the relationship between anxiety and attentional biases. Bradley et al. (1998) found that high-trait anxious groups spent more time attending to faces with threatening expressions when compared to non-anxious individuals. Around the same time, Bradley et al. (1999) demonstrated that participants with generalized anxiety disorder (GAD) had faster response times to threatening faces on a dot-probe task, and Mogg et al. (2000) showed that participants with GAD had faster initial eye movements toward threatening faces as opposed to non-threatening faces. It has been demonstrated more recently that individuals with panic disorder displayed enhanced attention to fearful faces, but not happy faces on a dot-probe task (Reinecke et al., 2011). These findings have been extended to post-traumatic stress disorder (Fani et al., 2012) and to individuals with high social anxiety (Yu et al., 2014). All of the above findings support the notion
that different types of anxious individuals have increased vigilance for threatening stimuli, are attending to the locus of threat, and are biased in their performance on these tasks.

While some experimental paradigms demonstrate that anxious individuals have faster response times to threatening stimuli and others report enhanced attention that result in slower response times to threatening stimuli, it is important to note that these differences are prominent competing theories in the literature. The vigilance-avoidance model suggests that anxious individuals are more inclined to orient quickly to a threatening stimulus and then actively avoid it, while the attention-maintenance model suggests that an anxious individual will have difficulty disengaging from a threatening stimulus once it has been attended to (Weierich et al., 2008). Attentional control has been found to a mediating factor in both of these theories, and it’s been shown to vary widely with both anxious and non-anxious individuals (Matthews & Wells, 2000). The degree to which an individual can mediate bottom-up processing of emotional stimuli by actively limiting the influence of these distractors, directly affects one’s attentional control (Eysenck et al., 2007). Research regarding regulating mechanisms of attentional biases is ongoing, with the goal of gaining a better understanding of this higher-order processing that occurs in the brain.

With substantial support for the theory that attentional bias is a feature of anxiety disorders, recent research focuses on the specific underlying mechanisms that promote this propensity for anxious individuals to allocate attention to threat. Studies implicating specific brain regions have shown that the amygdala is associated with a strong attention to threat (El Khoury-Malhame et al. 2011). It is also been shown that increased grey
matter volume in the anterior cingulate cortex is correlated with increased attention to threat (Carlson et al. 2011), but it still remains unclear why specific variation in the brain relates to individual differences on attention tasks. While analyzing these brain structures can be informative, many researchers strive to pinpoint the cause of individual differences by relying on less invasive techniques. Many suspect ecological and evolutionary implications, that is, there is a biological imperative for species to successfully identify potential threats in order to survive and reproduce.

Some scientists have taken this theory one step further, suggesting that there is an intensity threshold that threatening stimuli must exceed in order for the organism to actively attend to it (Mathews & MacLeod, 2005). This model suggests that individual differences are crucial and will determine if an organism is going to actively attend to or avoid a particular stimulus. This model was proposed by Mathews & Mackintosh (1998) as well as Mogg & Bradley (1998), and was one of the first theories to propose that moderately threatening stimuli should be introduced in experimental procedures to assess how low and high-trait anxious individuals react to low-level threatening stimuli. A probe task study by Wilson & MacLeod (2003) investigated all three levels of stimuli with high and low-trait-anxious individuals, and found that all participants had slower reaction times to neutral probe images, indicating avoidance. They also found that all participants had faster reaction times to threatening probe images, indicating vigilance. Most importantly, they found that only high-trait-anxious individuals had fast reaction times to moderately threatening probe images. For the moderate threat level, the transition from avoidance to vigilance occurred only with the high-trait-anxious individuals, indicating
that these participants might have a lower threshold for their ability to actively attend to threatening stimuli.

1.2 Background Information: Non-human Primates

From an evolutionary standpoint, the theory that human beings attend to potentially threatening stimuli in order to survive should also hold true with our closest relatives. However, research regarding attentional biases is a relatively recent focus in the field of cognitive testing with non-human primates. Fear conditioning studies conducted by Cook & Mineka (1989, 1990) found that rhesus monkeys were more likely to develop a fear of snakes as opposed to a fear of flowers. A study conducted more recently with Japanese macaques showed that these monkeys have a bias for threatening stimuli, as they responded faster to images of snakes than images of flowers in a visual search task (Shibasaki & Kawai, 2009). Furthermore, male rhesus macaques were also found to have an attentional bias to threatening faces of conspecifics in a dot-probe task (King et al. 2012). To further investigate attentional bias to social threat in a natural setting, Mandalaywala et al. (2014) compared infant rhesus macaques’ response to threatening and non-threatening faces of conspecifics. The three-month-old free-ranging infants did not display a bias, whereas the nine-month-old infants showed significant increased attention to threatening stimuli. Despite the fact that attentional biases for threatening stimuli have been reported in these few studies, non-human primate cognition research has yet to assess if differences arise between high-trait and low-trait anxious subjects on similar testing paradigms.

In order to assess the possible differences in performance on an attention task between high and low-trait anxious rhesus monkeys, their standard behavioral conditions
must first be observed. Behavioral observations will be used initially to assess anxious behavior in their standard home pen environment, since you cannot simply administer a questionnaire to a rhesus monkey. Observing an individual monkey and recording their daily behaviors gives researchers the opportunity to look back over a given amount of time, and calculate average rates of behavior and behavioral change. These behavioral rates are indicative of their standard behavioral responses in a home cage setting, thereby giving researchers insight to what is considered normal for each individual monkey. In captivity, behavioral pathology can indeed vary across different individuals, but overall, they are a resilient species that are able to thrive in a laboratory setting (Novak, 2003).

Regardless of their adaptive nature, abnormal patterns of behavior can arise in the form of stereotypic behavior, which are repetitive actions that do not serve any biological purpose (Ridley & Baker, 1982). Stereotypies can manifest in two different forms: whole body motions such as pacing or rocking, and self-directed motions such as digit-sucking or eye/ear covering (Novak, 2003). Most stereotypies are not severe; however, if these behaviors interfere with biological functions, or interfere with the animal’s overall wellbeing, then interventions may be required (Bayne & Novak, 1998). These behaviors, in addition to scratching and yawning can be considered indicators of an anxious temperament. Over a given time period, daily behavioral observations can be averaged together to demonstrate a reliable measure for anxious and non-anxious behaviors in every individual monkey.

1.3 Advantages of Our Study

With such little information regarding attentional biases in non-human primates, the need to further explore this area of research is evident. To our knowledge, no
researcher has designed an experiment to determine if anxious tendencies in rhesus monkeys relate to performance on an attention task. Thus, the present study was designed to assess how high-trait and low-trait anxious rhesus monkeys perform on a testing paradigm similar to the dot-probe test. Following the model described earlier by Mathews and Mackintosh (1998), the study examines the response of rhesus monkeys to three levels of social stimuli (neutral, mild threat, and strong threat) to determine if a threat bias exists using a modified dot-probe procedure. In addition to examining the relationship between hair cortisol concentrations and attention on this task, this study also has the advantage of utilizing daily behavioral observations for each monkey in the study. The monkeys at the UMass Primate Laboratory are observed 5 days per week for most weeks throughout the year, resulting in a comprehensive data set that we can use to determine if average rate of behavior over the course of four months is related to performance on this task.

1.4 Hypotheses and Predictions

In this study we hypothesized that behavioral phenotype (high or low anxiety) would affect attention on this modified dot-probe task. Additionally, we predicted that time spent looking at mildly threatening stimuli would be positively correlated with high levels of anxious behaviors (e.g., scratching, yawning, pacing, self-biting) and cortisol concentrations over a four month period. We also predicted that a higher percentage of the mildly threatening stimuli as a first choice would be positively correlated with high levels of anxious behaviors and cortisol concentrations.
2.1 Subjects and Housing Conditions

This study was conducted at the UMass Primate Laboratory at the University of Massachusetts Amherst. Subjects are eight (3 female) rhesus macaques (*Macaca mulatta*) housed in four separate colony rooms (Subject ID numbers: ZA31, ZA01, V43, V42, ZA65, ZA56, ZA63, ZA54).

All colony rooms were on a 13 hour light-dark cycle, where lights come on at 7:00 AM and turn off at 8:00 PM. Subjects were given Lab Diet Monkey Chow twice per day (8:30 AM and 2:00 PM) and had ad libidum access to water. Every morning at the same time, the health and wellness of each animal was assessed and recorded. The animals also received a food treat in the mornings (e.g., nuts, fruits, vegetables, grains, and monkey dough.) The animals were also exposed to a daily enrichment program that they received on a rotational basis (e.g., ice cube treats, lunch bags, music, and television).

2.2 Testing Apparatus

The apparatus used in this study was a plastic grey rectangular box (24in x 6in x 11in) with two sliding doors attached to the front. Both doors had a Plexiglas covering which shielded the laminated images of conspecifics, as they were placed behind this covering. Behind each sliding door is a circular hole that leads to the other side of the apparatus, where treats were placed for the monkey (see Figure 1).
2.3 Procedure

Starting in June 2014, eight subjects began a series of two familiarization phases with the testing apparatus. Phase one allowed subjects access to the testing apparatus, with the doors completely open, with a treat on top of the apparatus and on the opposite side of both doors. Treats varied by subject preference, but usually included raisins or unshelled peanuts. Once subjects successfully met criterion by obtaining the treats behind both doors in three out of four consecutive trials, they were able to move on to the next step. Subjects had to meet the same criterion with both doors half open, and then finally fully closed. Subjects then began phase two, where the doors were completely closed, and images of flowers were placed behind the Plexiglas screen. Flower images were chosen as neutral stimuli to allow the subjects to become familiarized with an image appearing on the door.

The experimental sessions began in October 2014 with the eight subjects. One session consisted of six trials, and one trial consisted of a pair of images from two of the three stimuli groups: neutral, mild, and strong (Images are shown in Figure 8 in Appendix). Each stimuli group had 6 images each, and they also had simple facial feature requirements. The strong images had an open mouth with teeth showing and eyes staring straight ahead, the mild images were just staring straight ahead, and the neutral images had their heads turned to the side with the eyes not facing forward. Images were block randomized, and the location of the image (right or left door) was also randomized. To
start a session, images were placed in the appropriate doors, one treat was placed behind each door, and the apparatus was moved forward to the subject’s cage with both doors closed. Once the subject had opened the doors and obtained both treats, the apparatus was pulled away from the cage, and the images were removed and replaced with the next trial’s images. This procedure was then repeated for the next five trials. Each subject completed each condition (Neutral vs. Mild, Strong vs. Neutral, Mild vs. Strong) twice, giving a total of 36 trials per subject.

2.4 Behavioral Analysis

All sessions were video recorded using a GoPro camera that was attached to the apparatus via a clamp (see Figure 2). The videos were then analyzed and coded for which image was chosen first, how long the subject attended to each image, and how long the subject took to complete the task from each side of the apparatus. Completion time was measured from the start point, when the monkey first touched the door and the end point, when the treat touched their mouth (measured in seconds.)

Figure 2: Testing apparatus with GoPro camera attached.
2.5 Modified Frequency Data Collection

Daily behavioral data were collected twice daily on every weekday from September through the end of December 2014. Observers used a modified frequency sampling procedure to record the presence of 32 behavior categories in 15-second intervals for a 5-minute sampling period between the hours of 9:00-10:00AM and 4:00-5:00PM. Inter-observer reliability between all lab members was calculated over all categories by percent agreement scores and averaged over 90%. The relevant behaviors for this particular study are listed in Table 1 below. Additionally, certain behaviors were combined to create comprehensive categories that represent different behavioral phenomena. These categories included threat (vocalization, initiate threat, initiate aggress, cageshake, initiate fear grimace, self mouth), anxiety (yawn, crooktail, locomotion, active stereotypy/pace, self directed stereotypy, scratch), and passive (visual explore).

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<td>Crooktail</td>
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<td>Initiate Aggress</td>
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<td>Scratch</td>
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<td>Visual Explore</td>
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Table 1: Species typical behaviors relevant for analysis

2.6 Hair Cortisol Collections and Analyses

Hair samples were collected during routine health exams during the winter of 2014 and 2015. A small patch of hair was shaved from the back of the neck from each of
the monkeys. Hair samples were then washed twice in isopropanol to remove any excess cortisol from the outside of the hair shaft. The hair was then completed dried, and ground into a fine powder to increase the surface area for extraction. Cortisol from the interior of the hair shaft was then extracted into methanol, the methanol was evaporated, and then the extract is reconstituted in assay buffer. The extracted cortisol is then analyzed with a commercially available enzyme immunoassay (EIA) kit. The readout from the EIA kit is then converted to pg cortisol per mg powered hair weight (Meyer et al., 2014)
CHAPTER 3
RESULTS

3.1 Modified Dot-Probe Task Performance Correlations

Performance on the modified dot-probe task was split into two categories: time to completion and looking time. Every subject’s score for each of the three stimuli levels was averaged, resulting in a mean subject score for neutral, mild, and strong. A strong positive correlation was found between average time to completion for neutral and strong stimuli across subjects ($r = 0.87, p < 0.03$, Means: Time Neutral = 15.48, Time Mild = 15.52, Time Strong = 16.48) (see Figure 3).

Figure 3: Time to completion (measured in seconds) for the neutral and strong stimuli.
A strong positive correlation was also found between average looking time for neutral and strong stimuli across subjects ($r = 0.92$, $p < 0.01$, Means: Time Neutral = 11.98, Time Mild = 11.59, Time Strong = 11.40) (see Figure 4).

![Figure 4: Looking time (measured in seconds) for the neutral and strong stimuli.](image)

Pearson correlations were used to analyze the relationship between Time Neutral, Time Mild, and Time Strong, but found no other significant results. Additionally, Pearson correlations were used to analyze the relationship between Look Neutral, Look Mild, Look Strong, but found no other significant results.
3.1.1 Modified Dot-Probe Task Performance Correlations – Hair Cortisol

There was no significant relationship between time to completion for the neutral stimuli and hair cortisol concentrations from 2014, but there was a negative correlation \((r = -0.63, p < 0.58)\) (see Figure 5).

![Figure 5: Time to completion (measured in seconds) for the neutral stimuli vs. hair cortisol concentrations from Winter 2014 (pg/mg).](image)

There was no significant relationship between time to completion or looking time for the three stimuli when compared to hair cortisol concentrations from 2014 or 2015.
3.2 Modified Dot-Probe Task Performance Analysis of Variance

An analysis of variance was performed across the three stimuli levels for time to completion with sex as the between subject variable. There was a significant difference in how long it took females and males to complete the task across the three stimuli levels \((F(2,12) = 6.2, p = 0.01)\) Females took significantly longer to complete the task when presented with mild stimuli (Means: Time Neutral = 16.11, Time Mild = 21.45, Time Strong = 17.75), as opposed to males who took significantly less time to complete the task when presented with mild stimuli (Means: Time Neutral = 15.1, Time Mild = 11.97, Time Strong = 15.71) (see Figure 6).

![Time to Completion](image)

Figure 6: Average time to completion between males and females for each of the three stimuli levels.
An analysis of variance was performed across the three stimuli levels for looking time with sex as the between subject variable. There was a significant difference in how females and males looked at each of the three stimuli levels (F(2,12) = 4.84, p = 0.03). Females looked significantly longer when presented with mild stimuli (Means: Look Neutral = 11.7, Look Mild = 13.75, Look Strong = 11.56), as opposed to males who looked significantly less when presented with mild stimuli (Means: Look Neutral = 12.15, Look Mild = 10.3, Look Strong = 11.3) (see Figure 7).

Figure 7: Average looking time between males and females for each of the three stimuli levels.
CHAPTER 4
DISCUSSION

4.1 Discussion

This study examined the relationship between anxiety and attention on a modified dot probe task. More specifically, we investigated whether high and low-trait anxious rhesus monkeys (measured through hair cortisol concentrations and average rates of anxious and non-anxious behaviors over four months) differed in their looking times, time to completion, and first image choice for a three stimuli dot probe paradigm. When analyzing the task performance data to assess whether completion time or looking time correlated with average rates of behavior or cortisol concentrations, we did not find any significant correlations. Additionally, we did not find any significant correlations between the subject’s first image choice and the average rates of behavior or hair cortisol concentrations.

When comparing the relationship between mean completion times in each of the three stimuli levels, we found a significant positive correlation between neutral and strong images. We also found a significant positive correlation between looking times for neutral and strong images. While this dual finding is not surprising, it’s a good measure of the paradigm’s effectiveness. When a subject looks longer at either the neutral or strong images, the longer it takes them to obtain the treat and complete the task. This clearly demonstrates that the subjects are processing the stimuli correctly. This finding also suggests that the eight monkeys interpreted the strong and neutral stimuli’s communicative significance at the same rate.
Interestingly, we found a sex by stimuli level interaction when we completed an analysis of variance for both time to completion and looking time. More specifically, we found a significant difference in the amount of time it took males and females to complete the task for the mild stimuli, with females taking significantly longer than males to complete the task. Additionally, we found a significant difference in the amount of time males and females looked at the mild stimuli with females looking significantly longer than males when presented with mild stimuli. In an attempt to explain these differences, we completed additional analyses of variance to determine if males and females differed in their average rates of anxious, threatening, or passive behaviors. We also looked to see if males and females differed in their hair cortisol concentrations from both time points (Winter of 2014 and 2015). However, none of these ANOVAS resulted in significant findings.

When we analyzed the data for which image was the subject’s first choice when completing the task, and we did not find an overall significant stimulus bias. To further investigate this sex difference we found for time to completion and looking time with the mild stimuli, we also looked to see if males or females had a bias for their first image choice when completing the task. Unfortunately, we did not find any significant bias in their first image choice.

As these results are the first of their kind in the non-human primate literature, it is important to note that a sex dependent threat-related attentional bias has also been reported in attention paradigms completed with humans. Some behavioral studies have shown that women are better at recognizing emotional facial expressions than men, (Du & Martinez, 2011) and when presented with threatening stimuli of human faces, men are
more likely than women to avoid these threatening images (Tan et al., 2011). Gender was also found to be an important factor in an fMRI study that explored processing of emotional pictures. The authors of this study reported that women showed higher activity in the primary and secondary visual cortex when shown unpleasant pictures when compared to pleasant pictures, whereas men had the opposite reaction (Lang et al., 1998). In evaluating our results, it is difficult to make comparisons between the findings in the human and non-human primate literature, because humans and rhesus monkeys interpret sources of threat in different ways. Humans that participate in these types of experiments know that they are not in any real danger when presented with threatening stimuli, but the same cannot be said for the monkeys in this task. Although our monkeys are used to completing cognitive and enrichment tasks almost daily, they do not usually see images of unfamiliar monkeys. When comparing this task to a real interaction between two rhesus monkeys, it is important to understand how macaques normally communicate. In terms of species typical communicative behavior, rhesus monkeys have a variety of facial expressions that they learn to use and respond to. In general, the most common facial expression among rhesus monkeys is the silent bared teeth face, which is important for communication between individuals of different social ranks- with the less dominant individual utilizing this expression the most (Maestripieri, 1999). This facial expression happens to be most similar to our strong threat stimuli group, which was generally looked at as frequently as the neutral stimuli group in both sexes. Since our monkeys looked at both the strong and neutral images at relatively the same rate, and took a similar amount of time to complete the task when presented with these two stimuli, we can presume that they are able to process these two stimuli at the same rate. The strong stimuli depicting
the silent bared teeth face might be an unambiguous form of communication to them, so they simply take a look at the photo and then proceed to complete the task. The neutral stimuli presented to them depicts a monkey whose face is turned to the side with no direct eye contact, so they also might simply take a look at the photo, realize that there is no ambiguous facial information being presented to them, and move on to the task at hand.

With the mild threat stimuli however, the facial expression may be a bit more ambiguous, as the images depict monkeys that are staring straight ahead with no open mouth. This direct eye contact may be interpreted differently in males and females, as the data clearly shows that females are spending more time looking at the mild images and taking longer to complete the task when presented with the mild images. Considering the fact that the majority of the stimuli images were of male monkeys, our three female subjects might have spent more time attempting to interpret this ambiguous stare from the males in order to assess their potential dominance rank, their sexual maturity, or their potential to be a threat. Since rhesus monkey social groups are generally characterized by large matrilineal dominance hierarchies with males wandering between these large groups (Melnick et al., 1984), females may be more inclined to be concerned with the potential threat of an unfamiliar male, especially since males have large canine teeth that can be used as a weapon. Since females do not have these canines at their disposal for protection, they might be more inclined to pay attention to unfamiliar males to assess potential threats.

While our hypotheses mainly focused on anxiety, anxious behaviors, and their possible affects on attention task performance, our data did not support our predictions. With such a small sample size for this particular experiment, we knew that power was going to be an issue when analyzing our data. However, even with only eight subjects we
were able to identify a sex difference in how the mild stimuli were interpreted by the monkeys (represented by time to completion and looking time). To further investigate the potential effects of anxiety on an attention task similar to the one used in this experiment, future studies should utilize larger stimuli so the monkeys can see the images more clearly, and be less likely to ignore the stimuli presented to them. In order to substantiate the mild stimuli sex difference that was found, future studies should use an equal number of male and female monkey faces in each stimuli level, while also tracking which image was used in each trial. This would help to further explain our finding that females were more likely to spend more time completing the task, and spend more time looking at the mild stimuli than males. Additionally, it could potentially clarify whether or not this difference was simply due to the fact that the majority of our stimuli were of male monkey faces.
APPENDIX

FIGURES AND GRAPHS

Figure 1: Testing Apparatus

Figure 2: Testing apparatus with GoPro camera attached
Figure 3: Time to completion (measured in seconds) for the neutral and strong stimuli.
Figure 4: Looking time (measured in seconds) for the neutral and strong stimuli.

Figure 5: Time to completion (measured in seconds) for the neutral stimuli vs. hair cortisol concentrations from Winter 2014 (pg/mg)
Figure 6: Average time to completion between males and females for each of the three stimuli levels.

Figure 7: Average looking time between males and females for each of the three stimuli levels.
Figure 8: Stimulus photos (Top row = Strong, Middle row = Mild, Bottom row = Neutral)
BIBLIOGRAPHY


