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## **The Influence of Human Attentional State and Familiarity on Bengal Tiger (*Panthera Tigris Tigris*) and Siberian Tiger (*Panthera Tigris Altaica*) Sociability**

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**THE INFLUENCE OF HUMAN ATTENTIONAL STATE AND FAMILIARITY  
ON BENGAL TIGER (*PANTHERA TIGRIS TIGRIS*) AND SIBERIAN  
TIGER (*PANTHERA TIGRIS ALTAICA*) SOCIABILITY**

A Master's Thesis

Presented to

The Graduate College of

Missouri State University

In Partial Fulfillment

Of the Requirements for the Degree

Master of Science, Applied Behavior Analysis

By

Jenna Dail

July 2021

# **THE INFLUENCE OF HUMAN ATTENTIONAL STATE AND FAMILIARITY ON BENGAL TIGER (*PANTHERA TIGRIS TIGRIS*) AND SIBERIAN TIGER (*PANTHERA TIGRIS ALTAICA*) SOCIABILITY**

Psychology

Missouri State University, July 2021

Master of Science

Jenna Dail

## **ABSTRACT**

An essential element of an animal's character is sociability (Gosling and John, 1999) and is demonstrated through interactions with other individuals (Capitanio, 2002). Although it is common to believe that domestic cats are not social beings, they have exhibited variance in solitary and social behavior, contingent on their environment and history (Turner, 2014). Much of the research on sociable behavior of animals has been completed between conspecifics. There has been less examination into interspecies sociability, and that which has been reviewed is focused on the relationship between humans and the domestic dog (Udell et al., 2010). Following this work, Vitale and Udell (2019) examined the effect of human attentional state and familiarity on the sociable behavior of domestic cats. The authors found that cats spent significantly more time in proximity with attentive humans and meowed more with unfamiliar, inattentive humans. A wide range of sociable behaviors were observed, suggesting that individual behavior differences should be considered in the study of cat social behavior. The current study was designed to replicate and extend this work using Bengal tigers (*panthera tigris tigris*) and Siberian tigers (*panthera tigris altaica*). Human attention was found to influence the behavior of tigers. This impact, as well as the comparison of human familiarity, continue to be an interesting component of feline sociable behavior.

**KEYWORDS:** social behavior, sociability, tiger, attention, inattention, human-tiger bond

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In the interest of academic freedom and the principle of free speech, approval of this thesis indicates the format is acceptable and meets the academic criteria for the discipline as determined by the faculty that constitute the thesis committee. The content and views expressed in this thesis are those of the student-scholar and are not endorsed by Missouri State University, its Graduate College, or its employees.

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I dedicate this thesis to Carol Rakestraw, the most sociable tiger I have ever known.

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## INTRODUCTION

The study of animal behavior is something humans have been doing for as long as there have been humans and animals. As hunter-gatherers, early humans would have benefited in at least two ways by understanding the behavior of the animals that shared their environment. This knowledge would have helped them obtain prey and it would have helped them avoid predation. There is reason to believe that early humans participated in the natural study of behavior exhibited by animals, as found in review of cave paintings and other archeological indications.

To truly understand animal behavior, Tinbergen (1963/2010) asserted that four questions should be addressed, which include immediate causation, ontogeny, evolution, and function. The antecedents and consequences of the behavior of interest, as well as its motivational operations and processes refer to causation. That is, how does it work? Ontogeny involves individual behavior development across an organism's lifespan. Further, this behavior is facilitated through intricate relationships between environmental and genetic components. Here the question is, how did it develop during the lifetime of the individual? The modification of behavioral procedures across generations, which impact the process of speciation is referred to as evolution. That is, how did it evolve over the history of the species? Finally, function involves queries into the contributions behavior provides to the association between the environment and an organism. Here the question is, what is it for? E. O. Wilson classified the first two questions (causation and ontogeny) as those of proximate causation (Wilson, 1998). What interactions that occur between endogenous and exogenous variables produce the behavior of interest? The latter two questions were determined to suggest ultimate causation (Cohn and MacPhail, 1996). How is the continued

development of a species furthered by these behavior occurrences and why do they initially take place (Bateson and Laland, 2013)?

## **Sociability**

The ability to work with others likely conferred an evolutionary advantage as it seems to be a broadly shared trait of most animals. Humans are obviously social animals, but so are our closest relatives (chimpanzees, bonobos, gorillas, etc.), as well as many other species. In fact, “man’s best friend” is supremely social and that sociability likely sealed the close dog-human relationship over time. Some animals are commonly considered less social, like cats, and yet we have kept them close as well. This affinity is usually explained in terms of the utility of cats with respect to grain stores in early human settlements, but the role of dogs was equally practical at that time.

Sociability is an essential facet of an animal’s character (Gosling and John, 1999) and is demonstrated through relations with others (Capitanio, 2002). It is identified by pursuing another individual’s attention and maintaining physical closeness with them. Sociability is one element of a broader aspect of personality known as extraversion. Extraversion includes engagement in sociable and outgoing behaviors, showing boldness, and maintaining high activity levels (Gosling and John, 1999).

Much of the research on sociable behavior of animals has been completed between conspecifics. There has been less examination into interspecies sociability, and that which has been reviewed is focused on the relationship between humans and the domestic dog (Udell et al., 2010). Vitale Shreve and Udell (2015) recently reviewed behavioral factors that influence a cat’s

personality, sociability, and sensitivity to cues. The successful domestication of cats into human homes can be attributed to such sociable components.

A study by Vitale Shreve, et al. (2017) with pet cats and shelter cats conducted free operant preference assessments of their preference for social interaction, scents, toys, or food. Results of their research varied; however, engagement with a human was discovered to be the most preferred stimuli for most of the cats tested, proceeded by food. Another study by Vitale and Udell (2019) focused entirely on pet and shelter cat sociability. The influence of human attentional state, population, and human familiarity were controlled, to which they found that shelter cats spent more time in proximity (and meowed more) to the inattentive, unfamiliar human while familiarity of the human did not affect sociable behavior of pet cats.

### **Shaping Sociability**

There are several aspects to consider in the development of animal sociability. Ontogenic factors such as a human's attentional state when engaging with a cat, the cat's population, and the familiarity status of the human with whom a cat is interacting with. For example, domestic dogs (Udell et al., 2010), domestic pigs (Nawroth et al., 2013), and some non-human primates (Defolie et al., 2015) have demonstrated an ability to differentiate a human's attentive versus inattentive state and consequently alter their behavior. Sociable behavior is influenced by attentional state. Research completed by Barrera et al. (2010) controlled for attentional state of a human and demonstrated that dogs spent significantly more time in proximity to humans actively providing them with attention over those who were not. Ito et al. (2016) showed cats to approach a human engaging in overt prompts requesting their proximity. Further, a study by Mertens and

Turner (1988) found cats exhibited an increased frequency of social behaviors behavior with the human who was unfamiliar and attentive over a unfamiliar inattentive human.

Former life experience has an influence on sociability. For instance, Barrera et al. (2010) administered sociability examinations with pet and shelter dogs and discovered that shelter dogs spent significantly more time in proximity to an inattentive, unfamiliar human than did pet dogs. Other research has shown this, which demonstrated that a reduced chance of human interaction, or the lack of a familiar attached individual, may result in an increase in sociable behavior toward humans, including those they are not familiar with (Gácsi et al., 2001). This differs somewhat with Collard (1967), whose research demonstrated that cats exposed to fewer novel humans engaged in more social contact with a familiar human.

The social interactions between conspecific mammals are impacted by familiarity. Individuals who are more familiar with one another display more allied behaviors toward the other than aggressive behaviors (Curtis et al., 2003; Faerevik et al., 2007; Ancillotto and Russo, 2014). Interspecies relationships have been studied and demonstrate that cats can distinguish humans who are unfamiliar and familiar and frequently exhibited a preference toward a human who was familiar to them (Collard, 1967; Edwards et al., 2007; Galvan and Vonk, 2016). However, not all studies have indicated a cat's preference of a human who is familiar to them (Podberscek et al., 1991; Potter and Mills, 2015). Saito and Shinozuka (2013) showed that cats were significantly more likely to respond to the voice of their owner in comparison to that of stranger's. When presented with an individual with whom a cat was attached to, their blood pressure significantly increased, as demonstrated with humans (Baun et al., 1984) and dogs (Astrup et al., 1979). This finding may suggest enthusiasm for engagement or anticipation of reinforcement based on past relationship with the individual (Slingerland et al., 2008).

Vitale and Udell (2019) completed a study to examine the impact of human attentional state and familiarity status, in addition to population (pet versus shelter), on the sociability of domestic cats. As dependent measurements of sociable behavior, they collected the duration of time a cat spent in proximity to and in contact with a human in addition to the frequency of the meows they emitted. They demonstrated that human attentional state impacted the behavior of cats, as both populations spent significantly more time in proximity to a human providing attention to them. The shelter cats were shown to spend significantly more time in proximity to an unfamiliar human who was not providing attention to them than did pet cats. Shelter cats, in comparison to pet cats, meowed at least one time within periods of being presented with an inattentive, unfamiliar human. The familiarity status of a human was not shown to significantly affect the sociable behavior of pet cats. The authors observed cats to exhibit a wide range of sociable behavior, indicating that variation between individuals should be included in the exploration of cat sociability.

### **The Current Study**

The current study was designed to replicate systematically the study by Vitale and Udell (2019) using adult Bengal (*Panthera tigris tigris*) and Siberian (*panthera tigris altaica*) tigers at an animal sanctuary in southwest Missouri. It is hypothesized that attention will have an influence on behavior, as shown with shelter cats in the original study. Small revisions to the methodology were made to facilitate the safety of the human observers and the tigers. The change in species and the revisions to the methodology allowed for additional measures of sociability. For example, head-rubbing and quick glances in the observer's direction.

## METHOD

The protocol for the current study, 2020-01, was granted on March 6, 2020. An addendum was also given approval on August 17, 2021, which added Jenna Dail as personnel. The research compliance documentation, including the completed Animal Care and Use Application and Addendum, can be reviewed in Appendix A.

### Subjects

Eight Bengal tigers (*panthera tigris tigris*) and one Siberian tiger (*panthera tigris altaica*) were tested in the main area of their enclosures. Tigers ranged in age from 7 to 21 years old (mean = 13.11, SD = 4.93) and were mixed sex, with 5 neutered males and 4 spayed females. Tigers lived both singly and in multi-cat enclosures at the National Tiger Sanctuary in Saddlebrook, MO. The tigers were on an intermittent fasting schedule. Meaning, over a 7-day period, tigers were given a decreased portion of their typical meals for one 24-hour time frame. Tigers had only received about a half-dozen raw chicken wings on days when data collection took place.

### Setting

The sizes of the enclosures varied, ranging from approximately 350 to 700 square meters. Each area provided adequate space for the tiger to explore and sit away from the human observer. The average temperature during data collection was 67° Fahrenheit, with an average humidity of 47.5%.

## **Materials**

A video camera (Sony Handycam, HDR-SR11) was attached to a tripod at a height of 1.5 meters and positioned 1.8 meters from the fence to record each procedure in its entirety. Two fluorescent poles (height = 1 meter) were placed approximately 4.5 meters apart in front of the enclosure to aid in video analysis.

## **Dependent Measures**

Unlike in the original study, humans cannot be in physical contact with the subjects in this analysis. Thus, approximately 4.5 meters across the front fence line of each enclosure was measured and marked with ground posts to specify the target observation area when measuring proximity to a human. The frequency of prusten (chuffs/vocalizations) was counted in place of meows. Chuff vocalizations (or prusten) are communicative behavior exhibited by tigers, as well as leopards and jaguars (Rose et al., 2017). As opportunities for contact were removed due to safety, additional sociable behaviors, including glances and head or body rubbing on the fence, were measured to supplement interaction possibilities between the tiger and human.

## **Procedure**

Two protocols were utilized to control for contrasting levels of human attentional state. A limited attention protocol was employed (similar to Barrera et al., 2010) within the inattention phase. During the attention phases, a free interaction protocol was applied to assist in establishing the tiger's typical expectation for attentive engagements with humans (Ellis et al., 2015).

Two human observers participated as either a stranger to the tiger or a familiar caretaker. The sex of the unfamiliar and familiar human was held constant, aged between approximately

20-30 years old. Both humans implemented the two attending protocols, described in more detail below, in immediate succession. The inattentive protocol was consistently executed first, to gain baseline data, then followed with the attentive protocol. The observations were counterbalanced in terms of the test order beginning with the unfamiliar human versus familiar. Regardless of which participant was initially employed, all protocols occurred back-to-back, making the entire procedure for each tiger last a total of 8 minutes. All cats were tested with the procedure once.

### **Inattention Protocol**

After the recording and measurement equipment was positioned at the fence line, the human observer approached the fence to stand in the middle, approximately, of the target area in view of the camera. They emitted a simple greeting (i.e., “Hi Harry”) then remained quiet, ignoring the tiger, and looking at the ground for 2 minutes.

### **Attention Protocol**

The human observer then interacted freely with the tiger over a 2-minute interval. The human attempted to initiate an interaction with the tiger by engaging in a variety of relatively standardized verbal and gestural behaviors, which included calling them by name, whistling, tapping the ground, tapping the fence, patting their own leg, and speaking to them in a friendly tone and volume of voice (e.g., “Come see me”, “Come over here”, “Are you tired/hungry/happy today?”, etc.). Although human interaction behaviors were regulated, participants were given a choice in what order to utilize behaviors along with selecting their own talking topics. Their goal was to gain the cat’s attention and maintain their physical closeness for as much of the two



minutes as possible. If the tiger moved away, the person continued to attract the tiger's attention; however, the person did not move from their initial position.

## **Data Analysis**

Behaviors were coded manually by watching the video recording of each 8-minute procedure multiple times and reporting specified occurrences with paper and pencil within 1-minute intertrial intervals (ITI). Duration data was collected by documenting then calculating between the initial and final time stamps a tiger was located within a specified area. Proximity to the human (4.5 meters across the front fence line and within two of their own footsteps away) was coded. Frequency data was collected by tallying occurrences of target behaviors. Glances from the tiger toward the human, chuffs or other vocalizations emitted by the tiger, and a tiger touching some part of their body (e.g., head, paw, or side) to the front fence line, in the specified area, were coded. These behaviors were eventually grouped and referred to as sociable behaviors throughout the current study. A task list defining each sociable behavior more precisely is provided in Appendix B. This was used to train participants in coding behavior with accuracy and consistency. To calculate inter-observer reliability for duration and frequency data, at least 85% of videos were double-coded. An inter-observer reliability score of 89% was calculated for time spent in proximity and an agreement score of 97% was calculated for the total frequency of all sociable behaviors.

Descriptive statistics were analyzed in GraphPad Prism, version 9.1.2 for MacOS (GraphPad Software, San Diego, California USA, [www.graphpad.com](http://www.graphpad.com)) for each data set, consisting of a phase (unfamiliar or familiar), protocol (inattention or attention), or a variation of both to account for all groupings analyzed throughout the study. The descriptive statistics are

outlined in Tables 1 and 2, separated by duration and frequency data, respectively. A Shapiro-Wilk analysis was also completed on all data sets to determine their normality. A mix of normal and non-normal data are represented, which is shown in Table 3. Data sets were also separated to account for the counterbalanced test orders. The descriptive statistics and normality results, via a Shapiro-Wilk analysis, are outlined in Tables 4 and 5, respectively. Using GraphPad Prism, average differences were analyzed on non-normal data, mixed data, and one normal data set with a two-tailed Mann-Whitney  $U$  test. A two-tailed independent  $t$  test was completed on one grouping of normal data for the analysis of time in proximity to a human, between unfamiliar and familiar phases within the attentive protocol. A two-tailed Fisher's exact test was used to examine individual differences for the duration of time in proximity and frequency of sociable behavior(s). To do this, 2 X 2 contingency tables were created in GraphPad Prism and analyzed by grouping tigers by a percentage of the 2-min interval they spent in proximity to a human (0-49%, 50-100%), as shown within the duration data, and by whether a tiger engaged in at least one sociable behavior during the 2-min session (0, 1) using the frequency data. All statistical tests had an alpha level of ( $p < .05$ ).

## RESULTS

ITI data was used to create a line graph, using an ABAB experimental design format. Using GraphPad Prism, Mann-Whitney  $U$  tests were completed to compare inattentive baselines and attentive treatment phases. There were no statistically significant results found, which is outlined in Table 6. The duration and frequency data are shown in Figures 1 and 2, respectively. Each figure depicts two graphs to account for both test orders. The occurrences of specified sociable behaviors collected and coded as frequency data were eventually combined and utilized as such in all corresponding analyses described below. Figure 3 depicts average frequency for each behavior during inattentive and attentive protocols and within the unfamiliar and familiar phases before being grouped together. Table 7 provides descriptive statistics for each sociable behavior throughout all trials.

### Attentional State

The influence of human attentional state, determined by the implementation of either the inattentive or attentive protocol, on the average sociable behavior of tigers was examined. Figure 4 shows that attentional state did not have a significant influence on the average proportion of time tigers spent in proximity to the human participant, determined by the Mann-Whitney  $U$  test (see Appendix C),  $U(18) = 131$ ,  $Z = -0.96498$ ,  $p = .3307$ . Individual data confirms this by showing no significant differences for tigers receiving the inattention versus attention protocol, in terms of remaining in proximity to a human, (Fisher's Exact Test,  $p > .9999$ ). Using Mann-Whitney  $U$  tests, the current study also explored the effect of attentional state within phases, unfamiliar and familiar, shown in Figure 5. During intervals wherein an unfamiliar human was present, there was no significant difference between them implementing the inattentive versus

attentive protocol,  $U(9) = 29$ ,  $Z = -0.97132$ ,  $p = .33204$ . This is also true for the familiar phase, as no statistical significance was found between the utilization of the inattentive protocol in comparison to the attentive protocol,  $U(9) = 39.50$ ,  $Z = -0.04415$ ,  $p = 0.9681$ . The descriptive statistics for each group comparison of duration of time in proximity data, considering attentional state, can be found in Table 2. The individual data, which separated tigers between groups of 0-49% and 50-100% of time in proximity to a human, confirms these results, as there were no significant differences found between the unfamiliar phase, (Fisher's Exact Test,  $p = .6372$ ), or familiar phase, (Fisher's Exact Test,  $p > .9999$ ), when examining attentional state.

However, a Mann-Whitney  $U$  test (see Appendix C) showed human attentional state significantly influenced the frequency of sociable behaviors tigers engaged in, which can be seen in Figure 6. Tigers being actively attended to, on average, showed more engagement with the participating human than tigers whose attention was disregarded by a human,  $U(18) = 98.5$ ,  $Z = -1.99323$ ,  $p = .0466$ . Individual data differs from this finding, as results showed attentional state did not have an influence on whether or not tigers engaged in at least one sociable behavior, (Fisher's Exact Test,  $p = .4018$ ). Figure 7 shows differences within the unfamiliar and familiar phases, which were analyzed in regard to human attentional state using Mann-Whitney tests. Tigers undergoing an attentive protocol during the unfamiliar phase engaged in significantly more sociable behaviors than those receiving the inattentive protocol,  $U(9) = 17$ ,  $Z = -2.03$ ,  $p = .0424$ . Attentional state, however, did not influence tigers participating in the familiar phase. There were no significant difference in the average sociable behavior frequencies of tigers either receiving attention or not from a consistent caretaker,  $U(9) = 32.5$ ,  $Z = -0.66227$ ,  $p = .50926$ . The descriptive statistics for each group comparison of frequency of sociable behaviors in terms of attentional state are also shown in Table 2. Individual data, which grouped tigers by whether they

engaged in a sociable behavior at least once, provides a differing result for the unfamiliar phase, as individual cats were not influenced by an unfamiliar human's attention or lack thereof, (Fisher's Exact Test,  $p = .0824$ ). Similar to the group differences, individual data shows that tigers in the familiar phase were not affected by the attentional state of a human, (Fisher's Exact Test,  $p > .9999$ ).

## **Familiarity**

Multiple analyses showed that there was no significant influence of the participating human's familiarity toward the tiger, whether a stranger (unfamiliar) or consistent caretaker (familiar), on sociable behavior occurrences. A Mann-Whitney  $U$  test (see Appendix C) demonstrated that familiarity of the participating human, in general, did not influence the average duration of time a tiger spent in proximity to a human,  $U(18) = 131$ ,  $Z = -0.96498$ ,  $p = .33706$ , as shown in Figure 8. Individual data, which defined tigers by whether they spent 0-49% or 50-100% of their time in proximity to the human, is consistent with these findings, showing no significant difference on the average proportion of time tigers spent in proximity to either an unfamiliar or familiar human, (Fisher's Exact Test,  $p = .5051$ ). By using the Mann-Whitney test, closer examinations were completed on familiarity differences by protocol (inattentive or attentive) with results shown in Figure 9. During intervals wherein the inattentive protocol was executed, there was no significance difference in the average time a tiger spent in proximity to a human, regardless of their familiarity,  $U(9) = 28$ ,  $Z = -1.05963$ ,  $p = .28914$ . This is consistent within the attentive protocol execution as well,  $U(9) = 37$ ,  $Z = -0.26491$ ,  $p = .79486$ . Individual data demonstrated no significant differences between those spending at least 50% of their time in

proximity to a human and those who did not within the inattentive (Fisher's Exact Test,  $p = .3469$ ) and attentive (Fisher's Exact Test,  $p > .9999$ ) protocols.

It was also shown with a Mann-Whitney test (see Appendix C) that familiarity, in general, did not significantly impact the frequency of sociable behaviors demonstrated by tigers,  $U(18) = 154.5$ ,  $Z = -0.22147$ ,  $p = .82588$ , depicted in Figure 10. The individual data suggested similar results, as human familiarity did not have an influence on whether or not tigers engaged in at least one sociable behavior, (Fisher's Exact Test,  $p > .9999$ ). Again, the influence of familiarity was examined more specifically within both the inattentive and attentive protocols, as shown in Figure 11. A Mann-Whitney  $U$  test demonstrated that familiarity did not affect the frequency of sociable behaviors tigers demonstrated during inattentive protocol intervals, as no significant difference was found during periods wherein a familiar human was present versus an unfamiliar human,  $U(9) = 29$ ,  $Z = -0.97132$ ,  $p = .33204$ . Similarly, an independent  $t$  test provided evidence that no significant differences occurred during intervals of the attentive protocol being employed by either an unfamiliar or familiar human,  $t(16) = 0.2452$ ,  $p = .8094$ . These findings for the inattentive and attentive protocols are consistent with their respective individual data. It was shown that regardless of the familiarity of the human participant implementing specified protocol strategies, there was no influence in whether or not a tiger engaged in at least one sociable behavior per session within inattentive, (Fisher's Exact Test,  $p = .2941$ ) and attentive, (Fisher's Exact Test,  $p = .4706$ ) intervals.

## DISCUSSION

The social behavior of cats was influenced by human attentional state. The frequency of social behaviors increased with contingent attention when the human was unfamiliar to the tiger relative to inattention. If the human was novel (unfamiliar), the tiger responded to the attention that the human provided but did not respond as frequently when no attention was forthcoming. When the human was familiar to the tiger, the relative frequency of social behaviors was similar regardless of attention or inattention. When the human was familiar (not novel), the tigers tended to engage in social behavior at similar frequencies in response to both attention and inattention.

It was expected for tigers to adjust their behavior consequently to human attentional state, as studies have shown felines can acknowledge and react to human prompting (for a review see Vitale Shreve and Udell, 2015). When the human was novel, the tigers responded to the attention they were given but responded less when no attention was given. The tigers had no history with the unfamiliar human, so social reinforcers were immediately salient, and then they quickly adjusted their behavior when the unfamiliar human did not give them attention. In this study, ignoring was the first condition for both familiar and unfamiliar humans. So, their first exposure to the novel human was no attention, to which they responded in kind. As soon as the novel person provided some social reinforcers, the tigers adjusted quickly and paid more attention to the unfamiliar person. The interaction with the familiar person involved a shared history that the unfamiliar person did not provide. In this case, the tigers tended to display various social behaviors when being ignored and when being attended to. Their shared history may explain persistence when being ignored in this condition.

This study supports the research completed by Vitale and Udell (2019), as they did not discover significant differences when comparing the average sociable behaviors exhibited with an unfamiliar human versus an owner. As some research has shown individual data suggests a strong predilection for their owners (Edwards et al., 2007), others do not demonstrate preference of a familiar human over an unfamiliar (Potter and Mills, 2015). Within the current study, although not significant, a portion of the individual data presented the possibility for preference. This was made apparent as some tigers exhibited all occurrences of their sociable behavior throughout the procedure solely in the presence of either the unfamiliar or familiar human.

The tigers in this study corresponded to the shelter cats in the earlier study. In general, the capacity to respond to social cues of a human has likely influenced cats' achievement as a domesticated animal within human homes, not dissimilar from the domestication of the dog (Udell et al., 2010). The tigers in this study also spent most of the early life in human homes, or in close contact with humans. Once they arrived at the sanctuary, their living conditions closely resembled the shelter cats in the previous study. Humans continued to provide their meals, but otherwise, human interaction ceased.

Moreover, the impact of novel situations or objects on an animal's behavior has been demonstrated, as some animals exhibited an increased likelihood to advance toward and explore novel objects, such as a human not familiar to them (Waiblinger et al., 2003), through a study completed with colony cats (Podberscek et al., 1991). The previous study by Vitale and Udell (2019) did not vary test order, as the unfamiliar human consistently executed the attention protocols first, while the familiar human went second. This was completed to allow for population (pet versus shelter) comparison, which potentially presented a factor within the



influence of familiarity. The current research counterbalanced the order of humans to control for the novelty of initial unfamiliar human influences on cat social behavior.

To summarize, this study demonstrated that tigers differentiate between varying levels of human attentional state and consequently adjust their behavior. This demonstrates they are responsive to social signaling from humans and are likely to exhibit increased sociable behaviors when an attentive human is present. Much like domestic cats do. This finding is more surprising when considering the animals involved. In general, tigers have had a different history with humans than domestic cats have had. Domestication of the domestic cat occurred nine thousand years ago (Driscoll et al., 2007) but tigers have remained in the wild. In general, tigers are not domestic cats, but these specific tigers have spent their lives in close contact with humans. Much of the contact was quite aversive (thus, they were eventually surrendered to the authorities), but familiar with humans just the same. It would be difficult to test, but it is possible that wild tigers would respond differently than tigers raised by humans in captivity. For the tigers in the current study, a lifetime with humans resulted in behavior similar to that of domestic cats. The question then becomes one of phylogeny or ontogeny. The phylogenetic histories of domestic cats and tigers are divergent, but the ontogenetic histories of captive tigers and domestic cats are very similar.

Potter and Mills (2015) proposed the existence of a continuum, on which the sociability of cats is found, with the possibility of independence being favored. A wide variety of individual social behavior is present within domestic cat and tiger populations; however, a bias toward independence was not observed. The mean proportion of time a tiger spent in proximity to an unfamiliar human was 0.35 in the inattentive and 0.60 in the attentive phase. Previous studies investigating individual cat preference for a variety of incentives showed that most cats (50%)

indicated their most preferred stimuli to be human engagement, as food (37%) was the following favored item; this was proceeded by the toy category (11%), while scent was the least preferred stimuli (2%) (Vitale Shreve et al., 2017). It is reasonable to infer, as considerable variation is demonstrated within the social behavior of cats, individual experiences, such as environment and past history, influence the development of their sociability.

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**Table 1**

Descriptive statistics of attentional state and familiarity condition variations for duration data

Phase, protocol, or variation of	<i>n</i>	Mean	SD	SEM	Median	Skewness	Kurtosis
Unfamiliar	18	0.47	0.46	0.11	0.44	0.14	-1.97
Familiar	18	0.63	0.47	0.11	1.00	-0.58	-1.77
Inattentive	18	0.50	0.47	0.11	0.53	-0.02	-2.07
Attentive	18	0.60	0.46	0.11	0.97	-0.41	-1.90
Unfamiliar Inattentive	9	0.35	0.43	0.14	0.08	0.76	-2.13
Unfamiliar Attentive	9	0.60	0.48	0.16	0.93	-0.44	-2.17
Familiar Inattentive	9	0.65	0.49	0.16	1.00	-0.82	-1.73
Familiar Attentive	9	0.61	0.47	0.16	1.00	-0.46	-2.14

*Note.* This table summarizes the descriptive statistics for each condition variation analyzed over the course of this study. Data set includes measurements for average frequency of sociable behaviors.

**Table 2**

Descriptive statistics of attentional state and familiarity condition variations for frequency data

Phase, protocol, or variation thereof	n	Mean	SD	SEM	Median	Skewness	Kurtosis
Unfamiliar	18	3.11	3.55	0.84	2.00	1.72	2.90
Familiar	18	3.89	4.30	1.01	2.00	1.10	-0.18
Inattentive	18	1.94	2.10	0.49	1.50	1.58	2.97
Attentive	18	5.06	4.67	1.10	3.50	0.64	-1.16
Unfamiliar Inattentive	9	1.44	1.74	0.58	1.00	1.13	0.78
Unfamiliar Attentive	9	4.78	4.18	1.39	4.00	1.21	0.57
Familiar Inattentive	9	2.44	2.40	0.80	2.00	1.75	3.51
Familiar Attentive	9	5.33	5.36	1.79	3.00	0.38	-1.93

*Note.* This table shows the descriptive statistics for each condition variation analyzed throughout the study. Data set includes the measurement of the average duration of time tiger spent in proximity to human.

**Table 3**

Shapiro-Wilk analysis results for attentional state and familiarity condition variations

Phase, protocol, or variation thereof	Average Duration of Time in Proximity	
	<i>p</i> value	Normality
Unfamiliar	.0005	Non-normal
Familiar	<.0001	Non-normal
Inattentive	.0002	Non-normal
Attentive	.0001	Non-normal
Unfamiliar Inattentive	.0118	Non-normal
Unfamiliar Attentive	.0040	Non-normal
Familiar Inattentive	.0005	Non-normal
Familiar Attentive	.0032	Non-normal
	Average Frequency of Sociable Behaviors	
	<i>p</i> value	Normality
Unfamiliar	.0014	Non-normal
Familiar	.0019	Non-normal
Inattentive	.0049	Non-normal
Attentive	.0120	Non-normal
Unfamiliar Inattentive	.0535	Normal
Unfamiliar Attentive	.0664	Normal
Familiar Inattentive	.0388	Non-normal
Familiar Attentive	.0561	Normal

*Note.* This table provides the results of the Shapiro-Wilks test for each phase, protocol, or variation thereof and reports the normality of both the duration and frequency data collected.



**Table 4**

Descriptive statistics of counterbalanced test order groupings

Test order and Condition Variation	Average Duration of Time in Proximity				
	<i>n</i>	Mean	<i>SD</i>	<i>SEM</i>	Median
Unfamiliar First					
Unfamiliar Inattentive	5	0.4120	0.4232	0.1892	0.4900
Unfamiliar Attentive	5	0.5940	0.5250	0.2348	0.9300
Familiar Inattentive	5	0.8000	0.4472	0.2000	1.0000
Familiar Attentive	5	0.6000	0.5477	0.2449	1.0000
Familiar First					
Familiar Inattentive	4	0.4625	0.5375	0.2688	0.4250
Familiar Attentive	4	0.6275	0.4461	0.2231	0.7000
Unfamiliar Inattentive	4	0.2700	0.4881	0.2441	0.0400
Unfamiliar Attentive	4	0.5975	0.4913	0.2456	0.6950
Test order and Condition Variation	Average Frequency of Sociable Behaviors				
	<i>n</i>	Mean	<i>SD</i>	<i>SEM</i>	Median
Unfamiliar First					
Unfamiliar Inattentive	5	1.2000	1.3040	0.5831	1.0000
Unfamiliar Attentive	5	4.2000	5.0700	2.2670	2.0000
Familiar Inattentive	5	3.6000	2.7020	1.2080	3.0000
Familiar Attentive	5	3.8000	4.7640	2.1310	1.0000
Familiar First					
Familiar Inattentive	4	0.7500	0.5000	0.2500	1.0000
Familiar Attentive	4	7.2500	6.1310	3.0650	7.5000
Unfamiliar Inattentive	4	1.7500	2.3630	1.1810	1.0000
Unfamiliar Attentive	4	5.5000	3.3170	1.6580	5.0000

*Note.* This table provides the descriptive statistics for the data groupings separated by the counterbalanced test orders. It is represented as either duration or frequency data.

**Table 5**

Shapiro-Wilk analysis results for counterbalanced test order groupings

Test order and Condition Variation	Average Duration of Time in Proximity	
	<i>p</i> value	Normality
Unfamiliar First		
Unfamiliar Inattentive	.4014	Normal
Unfamiliar Attentive	.0187	Non-normal
Familiar Inattentive	.0001	Non-normal
Familiar Attentive	.0065	Non-normal
Familiar First		
Familiar Inattentive	.0888	Normal
Familiar Attentive	.2190	Normal
Unfamiliar Inattentive	.0080	Non-normal
Unfamiliar Attentive	.2572	Normal
Test order and Condition Variation	Average Frequency of Sociable Behaviors	
	<i>p</i> value	Normality
Unfamiliar First		
Unfamiliar Inattentive	.4211	Normal
Unfamiliar Attentive	.0221	Non-normal
Familiar Inattentive	.4272	Normal
Familiar Attentive	.0202	Non-normal
Familiar First		
Familiar Inattentive	.0012	Non-normal
Familiar Attentive	.2017	Normal
Unfamiliar Inattentive	.2200	Normal
Unfamiliar Attentive	.5150	Normal

*Note.* This table provides the normality results via a Shapiro Wilks test for the data groupings separated by the counterbalanced test orders. It is represented as either duration or frequency data.

**Table 6**

Statistical significance comparison across test orders

Phase, protocol, or variation thereof	Duration		
	<i>p</i> value	Confidence Intervals	Mann Whitney U
Unfamiliar First			
UA v UI	.5714	96.83% CI [-0.5700, 1.000]	9.0
FA v FI	>.9999	96.83% CI [-1.000, 1.000]	10.0
Familiar First			
FA v FI	.4000	97.14% CI [-0.8900, 1.000]	5.0
UA v UI	.4857	97.14% CI [-1.000, 1.000]	5.0
	Frequency		
	<i>p</i> value	Confidence Intervals	Mann Whitney U
Unfamiliar First			
UA v UI	.246	96.83% CI [-1.000, 12.00]	6.5
FA v FI	.7381	96.83% CI [-7.000, 7.000]	10.5
Familiar First			
FA v FI	.1143	97.14% CI [0.000, 13.00]	1.5
UA v UI	.2000	97.14% CI [-3.000, 10.00]	2.5

*Note.* This table shows the statistical analysis results for duration of time in proximity and frequency of sociable behaviors between a baseline of inattention (I) and treatment of attention (A), for unfamiliar (U) or familiar (F) human beginning the procedure first.

**Table 7**

Descriptive statistics for the frequency of specific sociable behaviors

Protocol and behavior	Unfamiliar Phase							
	<i>n</i>	Mean	<i>SD</i>	Median	Min	Max	Skewness	Kurtosis
Inattentive								
Glances	9	0.56	0.53	1.00	0	1	-0.27	-2.57
Chuffs/vocalizations	9	0.56	0.73	0.00	0	2	-1.01	0.19
Fence touches	9	0.33	0.71	0.00	0	2	2.12	4.00
Attentive								
Glances	9	2.22	1.39	2	1	5	1.28	0.83
Chuffs/vocalizations	9	1.67	2.44	0	0	7	1.55	1.96
Fence touches	9	0.89	1.36	0	0	4	1.77	4.00
Familiar Phase								
Inattentive								
Glances	9	1.11	1.27	1.00	0	4	1.63	3.15
Chuffs/vocalizations	9	0.56	0.73	0.00	0	2	1.01	0.19
Fence touches	9	0.67	2.00	0.00	0	6	3.00	9.00
Attentive								
Glances	9	2.22	1.92	2.00	0	5	0.41	-1.18
Chuffs/vocalizations	9	1.89	2.31	1.00	0	6	0.95	-0.54
Fence touches	9	1.22	1.48	0.00	0	3	0.41	-2.26

*Note.* This table shows the descriptive statistics for each sociable behavior collected and documented as frequency data. It is split between two phases of human familiarity and shows the attentional state protocol implemented by the human participant within each phase.

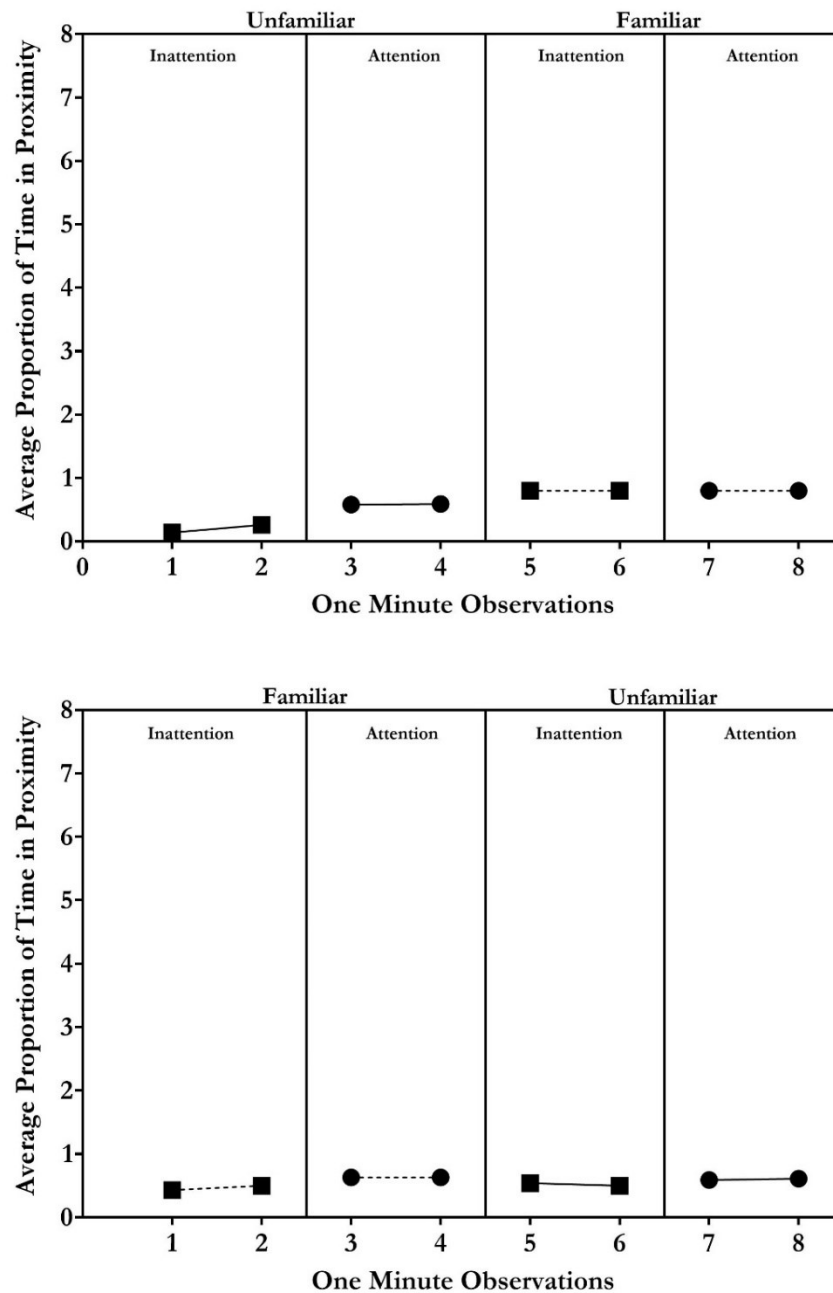


Figure 1. Average duration of time in proximity shown in counterbalanced test order

*Note.* This figure shows the average duration of time spent in proximity to human and accounts for the counterbalanced test orders, showing which human participant began implementing attentional state protocols first. Each procedure began with the inattentional protocol.

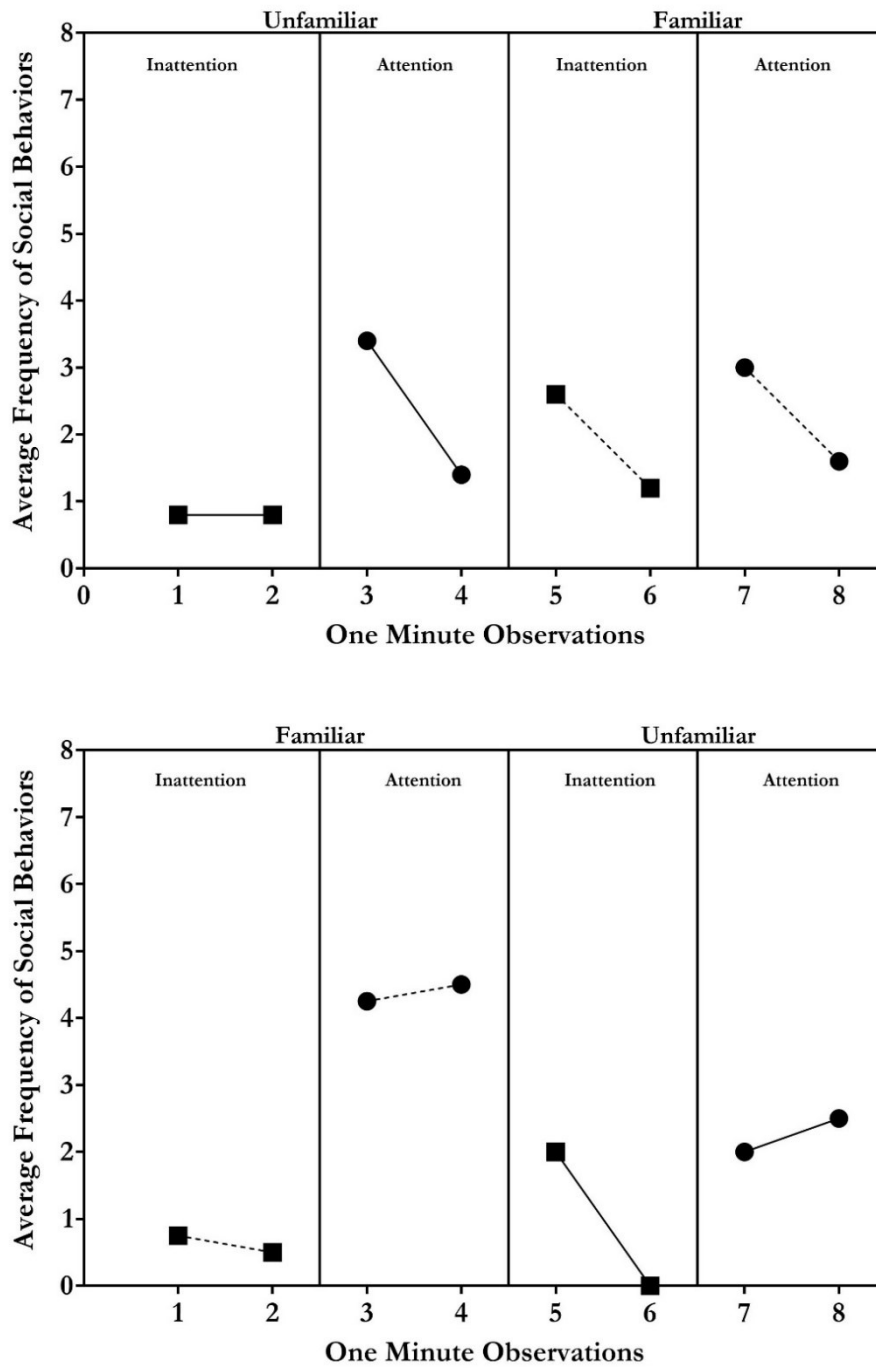


Figure 2. Average frequency of social behaviors shown in counterbalanced test order

*Note.* This figure shows the average frequency of social behavior and accounts for the counterbalanced test orders, showing which human participant began implementing attentional state protocols first. The inattentive protocol was utilized at the start of each procedure.

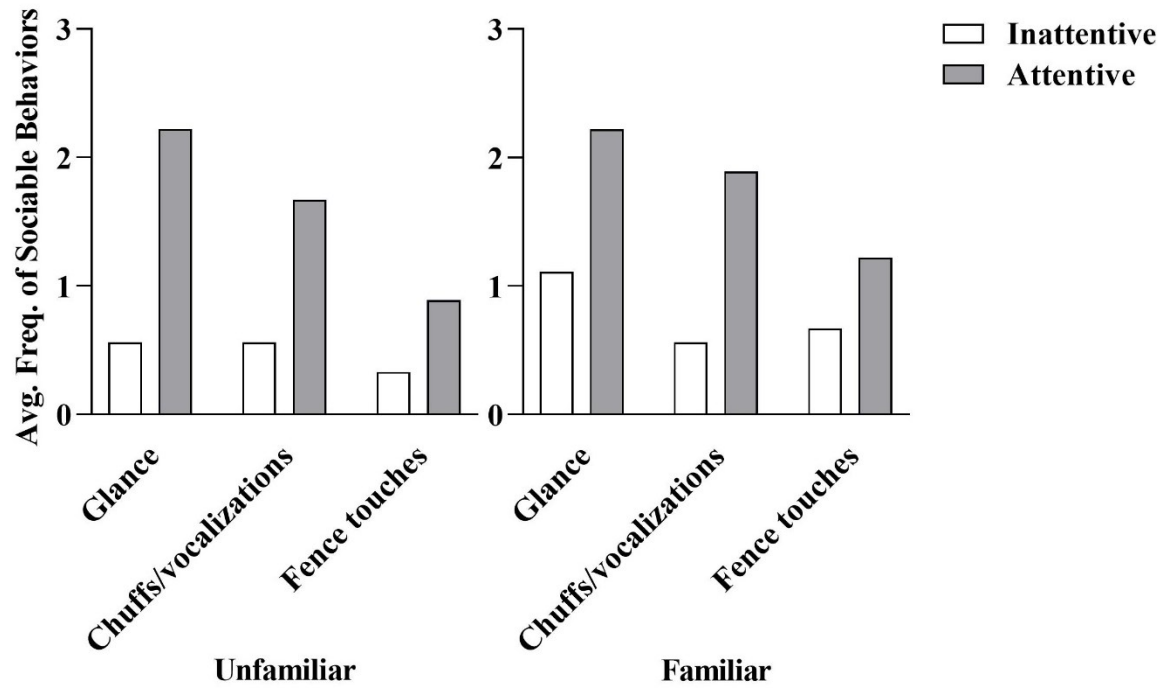


Figure 3. Average frequency of sociable behaviors across multiple condition variations

*Note.* This figure depicts average frequencies for each specified sociable behavior under the influence of two human attentional states, inattentive (white bars) and attentive (gray bars), and within two human familiarity phases, unfamiliar and familiar.

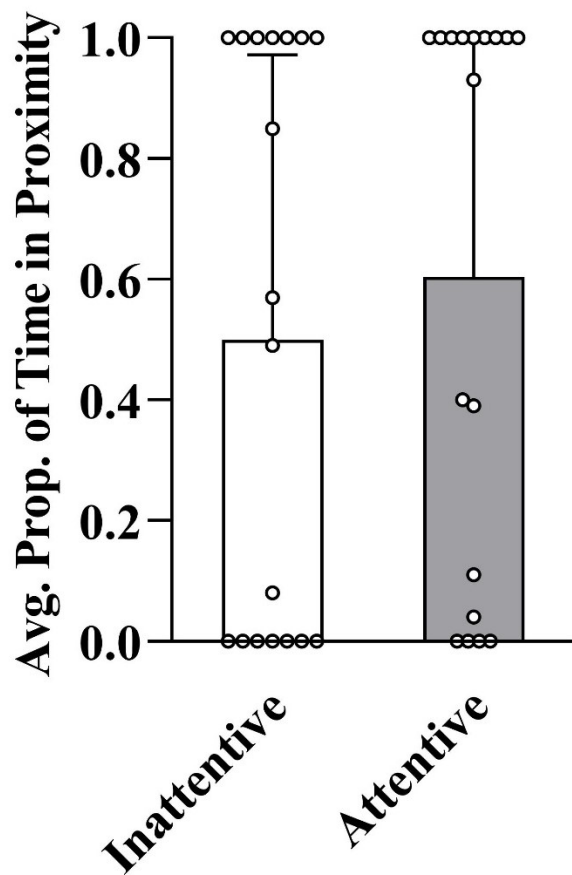


Figure 4. Average proportion of time spent in proximity comparing human attentional state

*Note.* This figure depicts the average proportion of a 2-min session the tiger spent in proximity to a human, specifically comparing the influence on behavior when implementing the inattentive (white bar) versus attentive (gray bar) protocol. Each dot represents an individual tiger.



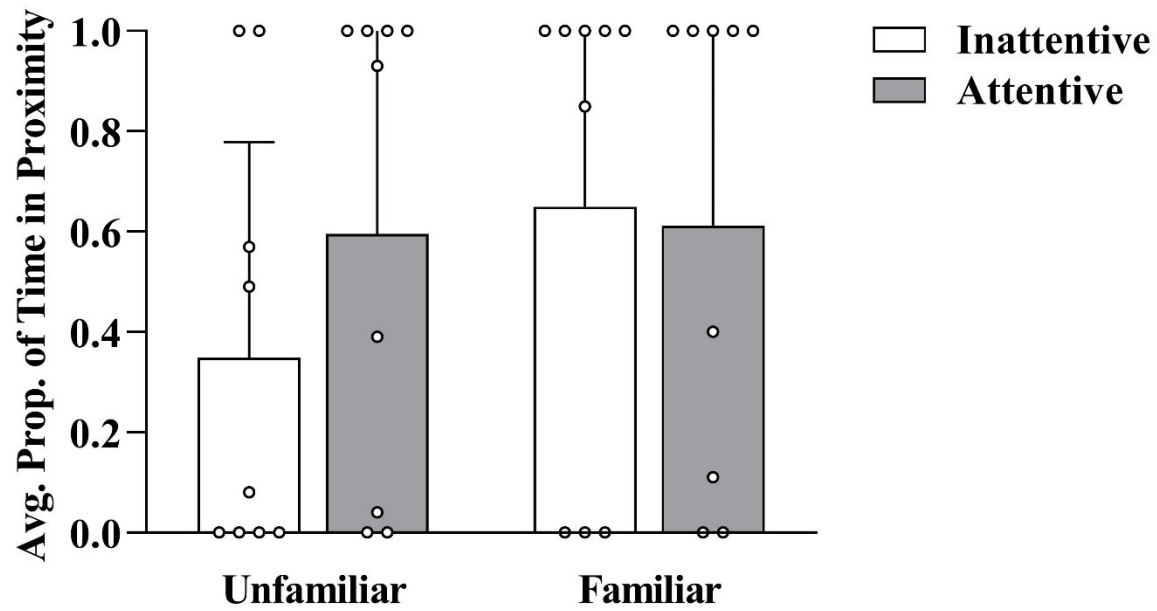


Figure 5. Average proportion of time in proximity comparing attentional state within varying familiarity conditions

*Note.* This figure depicts the average proportion of a 2-min session the tiger spent in proximity to a human. The comparison in behavior between inattentive (white bars) and attentive (gray bars) attentional states are both depicted within the unfamiliar and familiar states of human familiarity. Each dot represents an individual tiger.

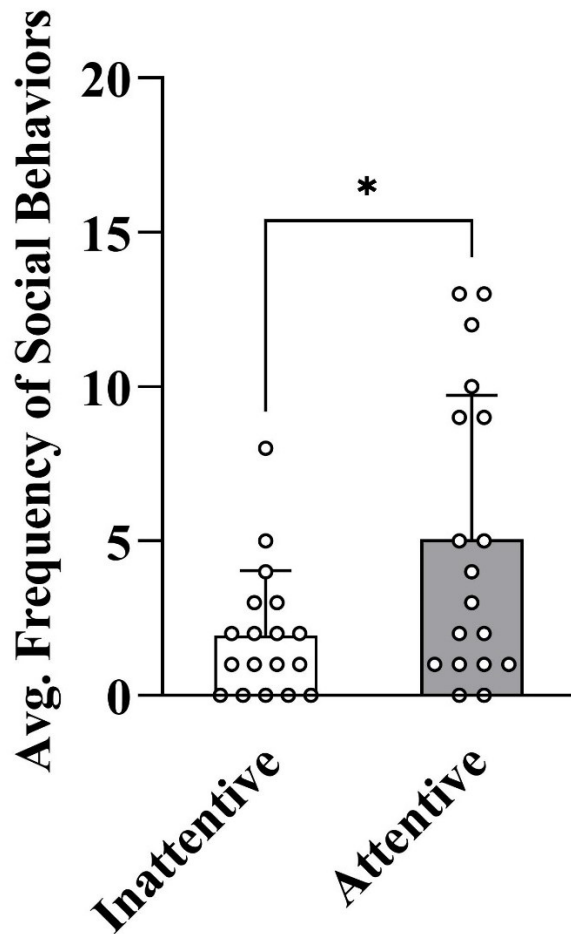


Figure 6. Average frequency of sociable behaviors comparing human attentional state

*Note.* This figure depicts the average frequency of sociable behaviors a tiger exhibited during a 2-min session, specifically comparing the influence on behavior when implementing the inattentive (white bar) versus attentive (gray bar) protocol. Each dot represents an individual tiger. \*  $P < 0.05$ .

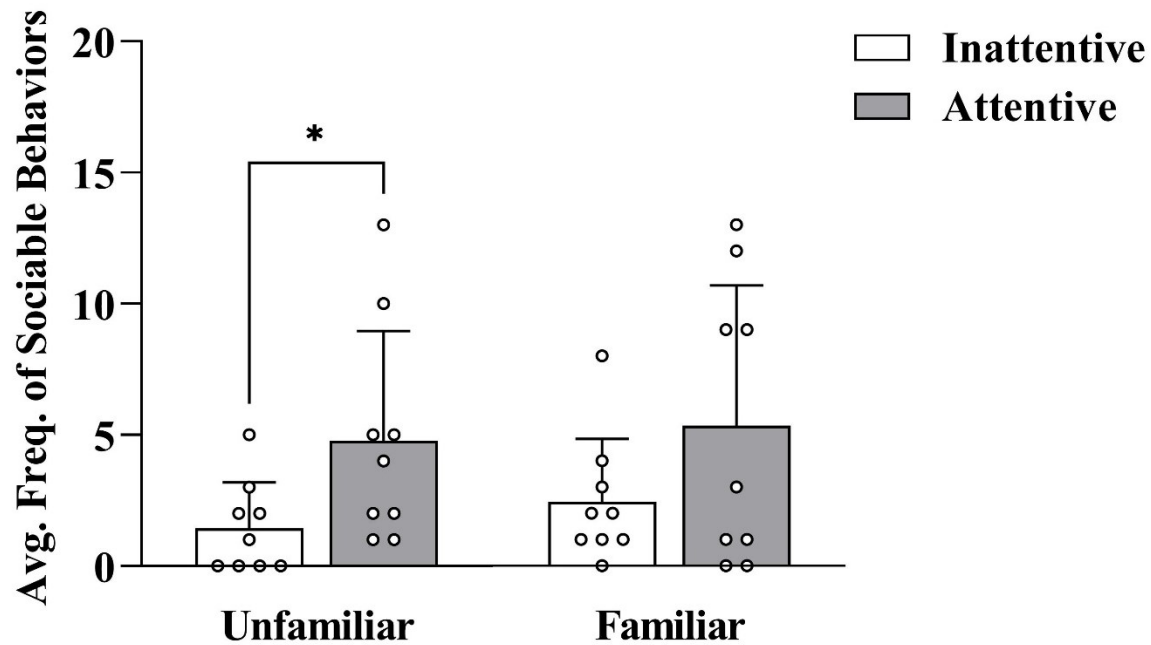


Figure 7. Average frequency of sociable behaviors comparing attentional state within varying familiarity conditions

*Note.* This figure depicts the average frequency of sociable behaviors a tiger exhibited during a 2-min session. The comparison in behavior between inattentive (white bars) and attentive (gray bars) attentional states are both depicted within the unfamiliar and familiar states of human familiarity. Each dot represents an individual tiger. \*  $P < 0.05$ .

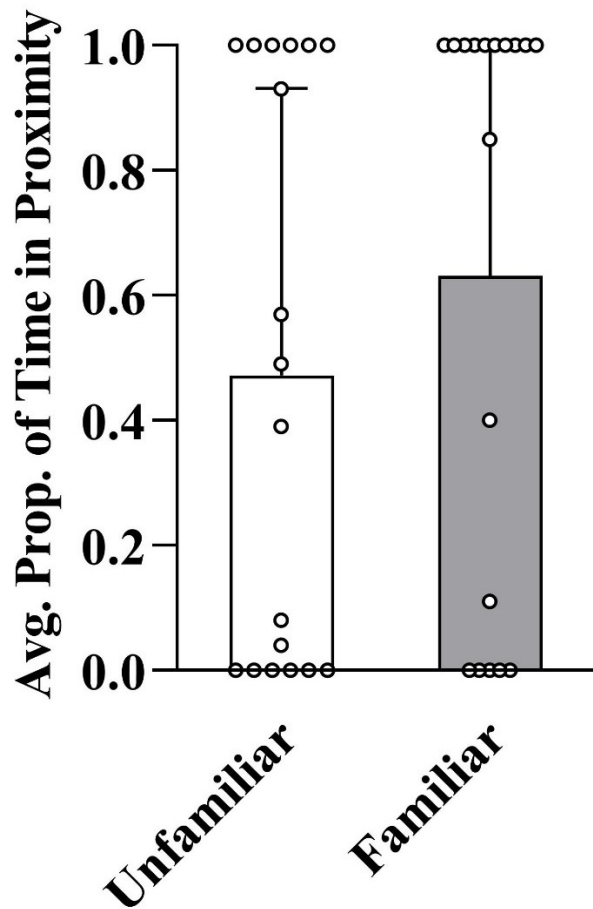


Figure 8. Average proportion of time in proximity comparing human familiarity

*Note.* This figure depicts the average proportion of a 2-min session the tiger spent in proximity to a human, specifically comparing the influence of human familiarity, unfamiliar (white bar) versus familiar (gray bar) on behavior. Each dot represents an individual tiger.

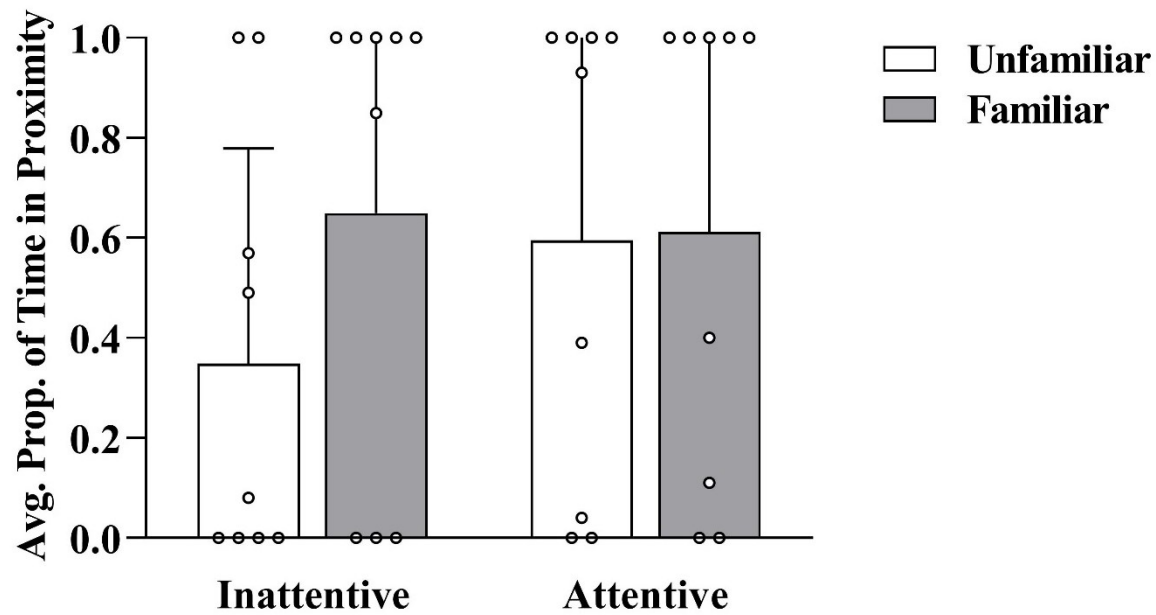


Figure 9. Average proportion of time in proximity comparing familiarity within varying attentional state conditions

*Note.* This figure depicts the average proportion of a 2-min session the tiger spent in proximity to a human. The comparison between behavior occurring for two forms of human familiarity, unfamiliar (white bars) and familiar (gray bars), is depicted across varying intervals of human attentional state. Each dot represents an individual tiger.

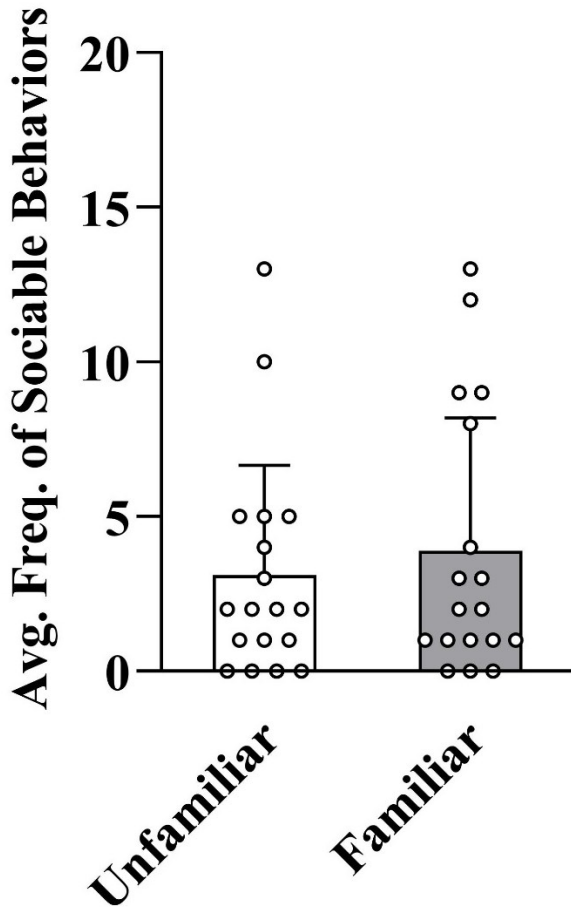


Figure 10. Average frequency of sociable behaviors comparing human familiarity

*Note.* This figure depicts the average frequency of sociable behaviors a tiger exhibited during a 2-min session, specifically comparing the influence of human familiarity, unfamiliar (white bar) and familiar (gray bar) on behavior. Each dot represents an individual tiger.

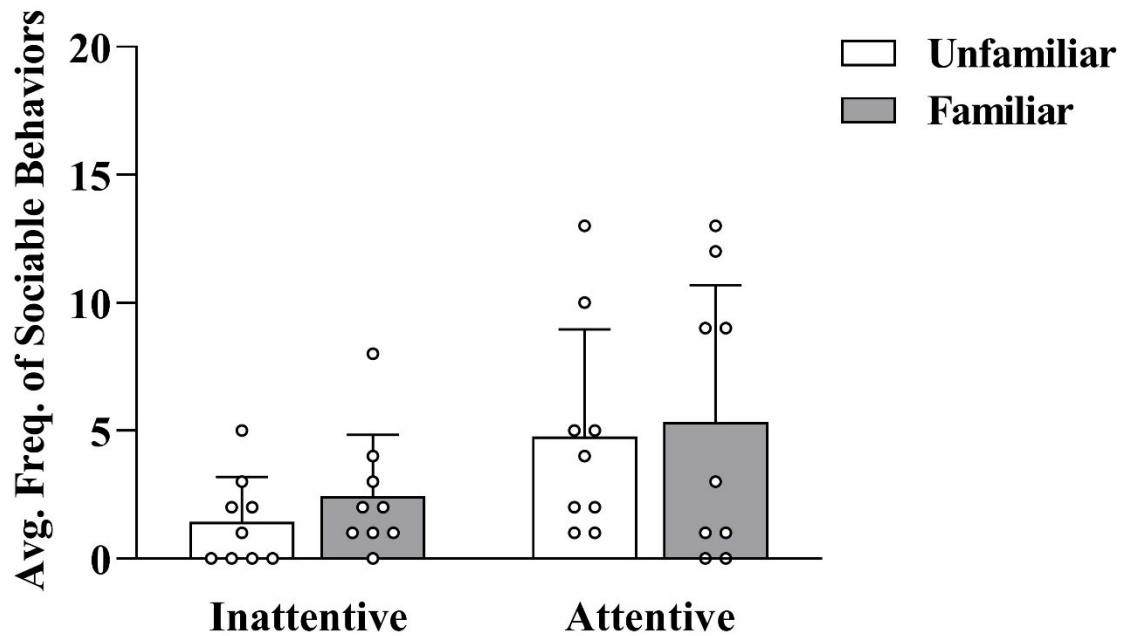


Figure 11. Average frequency of sociable behaviors comparing familiarity within varying attentional state conditions

*Note.* This figure depicts the average frequency of sociable behaviors a tiger exhibited during a 2-min session. The comparison between behavior occurring for two forms of human familiarity, unfamiliar (white bars) and familiar (gray bars), is depicted across varying intervals of human attentional state. Each dot represents an individual tiger.

## APPENDICES

### Appendix A. Animal Care and Use Application and Addendum



#### ANIMAL CARE & USE APPLICATION

INSTITUTIONAL ANIMAL CARE & USE COMMITTEE

v. July 2019

All Animal Care & Use Applications should be submitted electronically to [IACUC@missouristate.edu](mailto:IACUC@missouristate.edu).

<b>A. Investigator Information</b>		
Principal Investigator: Michael Clayton	Department: Psychology	Office Address: 409 Hill Hall
Office Phone: x3783	Emergency Phone: x3783	Email: mclayton@missouristate.edu
<b>B. Project Information</b>		
Project Title: The Effects of Human Attentional State and Familiarity on Tiger ( <i>Panthera tigris</i> ) Sociability		
<b>Protocol Action:</b> <input checked="" type="checkbox"/> New Proposal <input type="checkbox"/> Pilot Study <input type="checkbox"/> Renewal (due to protocol expiration) <input type="checkbox"/> Review for Exemption	<b>Protocol Type:</b> <input checked="" type="checkbox"/> Research <input type="checkbox"/> Teaching	<b>Protocol Class:</b> <input type="checkbox"/> Agricultural <input checked="" type="checkbox"/> Behavioral <input type="checkbox"/> Biomedical <input type="checkbox"/> Wildlife/Conservation
Is this project externally funded and/or do you anticipate future funding? <input type="checkbox"/> Yes <input checked="" type="checkbox"/> No If Yes, what is the name of the Funding Agency and grant number/title?		
<b>C. Previous Approved Protocol</b>		
For work that is similar to a previously approved protocol, provide the original protocol number and approval date. On the remainder of the forms, indicate changes to the originally approved protocol in bold font.		
Original Protocol Number: 2020-01	Approval Date: 03/06/2020	
<b>D. Investigator Assurances</b>		
1. The information provided herein is accurate and, to the best of my knowledge, conforms to all applicable University, PHS, and USDA policies on the use of animals in research and teaching. 2. I affirm that all procedures involving vertebrate animals will be performed only by personnel trained in the humane care, handling, and use of animals and that all personnel will abide by the recommendations of the University's Occupational Health and Safety Program. 3. I agree not to proceed with any portion of this project until I receive written approval from the Missouri State University Institutional Animal Care and Use Committee. 4. I agree any changes in the procedures contained in this protocol will be promptly forwarded to the IACUC for review and approval prior to performing any revised procedures. 5. I agree to provide proper, current documentation (e.g., licenses, permits and additional approval forms), when applicable, to the Office of Research Compliance throughout the course of this project. 6. I agree to allow inspection of my research facilities by members of the IACUC, including the veterinarian, and to comply promptly if informed of any violations of the Missouri State University's Animal Care and Use Policy. 7. I have taken into consideration the three "Rs" (replacement, reduction, and refinement) for my study and provided adequate justification for the animal model chosen, animal numbers requested, and procedures to reduce pain and distress. 8. I have conducted a literature search to ensure that I am not unnecessarily duplicating previous experiments.		
Michael Clayton	January 23, 2020	
Signature of Principal Investigator	Date	



## ANIMAL CARE & USE PROTOCOL

Read all sections for instructions. Answer all questions or answer N/A if the question does not apply. Complete electronically, handwritten versions will not be accepted. Submit electronically to [IACUC@missouristate.edu](mailto:IACUC@missouristate.edu).

### Section 1. Personnel Information

List all individuals, including the PI, performing manipulations or working with animals. Indicate each individual's role (PI, graduate assistant, undergraduate student, etc.) in the position column. Training should indicate both online training modules and lab specific procedures. Experience should indicate length of involvement (months, years, etc.) in the relevant area of research.

Name	Title/Position	Degrees	*Training/Experience
Michael Clayton	PI	Ph.D.	one year previous experience
Caroline Lundy	undergrad asst	n/a	n/a
Lauren Krieg	undergrad asst	n/a	n/a

\* All personnel must take the Online Animal Care & Use Training as well as enroll in the Occupation Health and Safety Program prior to animal related activities.

### Section 2. Project Description

#### 2.1 Nontechnical Summary

Provide in terms comprehensible to a nonscientist (abstracts or methods section of grant proposals are not acceptable):

- The project's goals & objectives:  
Determine how social tigers are with familiar/unfamiliar humans
- The project's benefit to society, education, or animals:  
Showing that tigers "like" people and are "happy" when they are near them would be good for public perceptions and increase support for conservation efforts
- A summary of the experimental design/teaching plan:  

**Setting**  
Each enclosure will have a 2 m radius semi-circle marked with water-soluble, non-toxic white paint. Although the size of each enclosure varies, all enclosures were large enough for the cat(s) to explore and sit outside the semi-circle away from the human observer. A video camera (Sony HandyCam, HDR-SR11) will be attached to a tripod and placed outside the two fence lines to videotape interactions.

**Interaction protocol**  
Two interaction protocols will be utilized to reflect two different levels of human attentional state (see Figure 1). For the inattentive phase, a limited attention protocol was used (similar to Vitale and Uddell, 2019) and for the attentive phase, a free interaction protocol was used to help match the cat's normal expectation for attentive interactions (Ellis et al., 2015). The human was either a stranger to the cat (Experiment 1) or a familiar caretaker (Experiment 2). The stranger will be an undergraduate student at Missouri State University and the familiar human will be a volunteer at the National Tiger Sanctuary.

**Inattentive phase**  
To begin, the human will stand at the outer fence line facing the semicircle on this inside of the enclosure. The human will not talk or make eye contact, ignoring the cat(s) and looking at the ground. If the cat(s) enters the semicircle (at least two paws inside the semicircle), the person will continue to ignore the cat(s) for the duration of this phase (2 min).

**Attentive phase**  
During this phase, the human can now interact freely with the cat by calling the cat by name and trying to make contact with it (keep the cat's attention and keep the cat within the semicircle) for as much of the 2 minutes as possible.

**Experiment 1**

<p>All cats will be tested with the procedure one time incorporating two phases (Inattention and Attention). The sex of the of the unfamiliar human will be held constant and will always be a White female aged between 20-30 years old.</p> <p><b>Experiment 2</b></p> <p>Immediately following Experiment 1, cats will be exposed to two additional phases using a familiar human. All cats will be tested with the procedure one time. The sex of the of the familiar human will be held constant and will always be a White female aged between 20-30 years old.</p> <p><b>Data analysis</b></p> <p>Behaviors will be coded from video using the program JWatcher, 1.0 (Blumstein et al., 2000). We will code the proportion of the time the cat spends in proximity to the human (inside the semicircle/within 2 m) and the frequency of vocalizations ("chuffs"). Chuffing is a non-threatening vocalization displayed by tigers. Captive tigers will exchange chuffs with their human caretakers in a way to express a greeting or excitement.</p> <p>To calculate interobserver reliability, 85% of videos will be double-coded. An 8% range of tolerance will be applied for the time spent in proximity with the human.</p>	<p>D. What is the project duration dates (start and end) and the disposition of the animals at the end of the project?</p> <p>We hope to start in early February and be done by the end of March</p>
<p><b>2.2 Justification</b></p> <p><i>This section should indicate consideration of the "three Rs."</i></p> <ul style="list-style-type: none"> <li>• <b>Replacement</b>- replacing the use of animals with non-animal techniques (i.e. computer models, in vitro assays, or cell culture)</li> <li>• <b>Reduction</b>- reducing the number of animals used (i.e. limiting group sizes, sharing tissues, or performing experiments simultaneously)</li> <li>• <b>Refinement</b>- changing experiments or procedures to reduce pain and distress in animals (new anesthetics/analgesics or surgical procedures)</li> </ul> <p>Briefly describe the following.</p>	
<p>A. Why each species was chosen.</p> <p>The tigers were chosen as a "convenience sample." We have access to them and the support of the sanctuary</p>	<p>B. Why the number of animals requested is warranted. (Why the proposed number of animals is sufficient, but not excessive for achieving valid results)</p> <p>We want to test as many tigers as we can to increase confidence in our statistics.</p>

2.3 Literature Search			
Searches should provide justification of the three "Rs" and be conducted within 2 months of protocol submission.			
Date of Search	Keywords	Resources Used	Years Covered
3/21/2020	domestic cat, human-cat bond	PSYC Info	1990-2017
3/21/2020	cat, human-cat bond, sociability	PSYC Info	1990-2019
3/22/2020	bengal tigers, preference assessments	unpublished thesis	2019
Results: Summarize how the searches indicate the necessity of an animal model, lack of duplication, the need to repeat previous studies.			
Replacement is unnecessary. We are interested in the sociability of live captive Bengal tigers			
Reduction is unnecessary. We are not doing anything invasive and there are only a limited # of tigers available			
Refinement is unnecessary. We will not be causing distress or discomfort to the animals.			
Section 3: Animal Use			
3.1 Animal Sources			
Provide number and source for each species used.			
Species	Common Name	Approximate Number	Source
panthera tigris	Bengal tiger	7	National Tiger Sanctuary
3.2 Animal Facilities			
Identify buildings and room numbers where species will be housed and procedures performed.			
Species	Housing	Procedure Area	
panthera tigris	large outdoor enclosures	National Tiger Sanctuary	
3.3 Animal Husbandry			
Describe how animals will be maintained including feeding, cage or housing conditions, and laboratory environment.			
National Tiger Sanctuary will handle all aspects of the animals' well-being			
3.4 Animal Procedures			
Check all that apply. Please fill out the appropriate addendum if one is indicated.			
<input checked="" type="checkbox"/> Behavioral Studies <input type="checkbox"/> Blood Sampling/Tissue Collection <input type="checkbox"/> Capture of Wild Animals (Addendum 5) <input type="checkbox"/> Death as an Endpoint <input type="checkbox"/> Field Observation Only <input type="checkbox"/> Food Restriction/Special Diet <input type="checkbox"/> Injections <input type="checkbox"/> Long Term Restraint		<input type="checkbox"/> Marking, Microchip, Tattoo <input type="checkbox"/> Non-Standard Housing <input type="checkbox"/> Non-Standard Husbandry <input type="checkbox"/> Physiological Studies <input type="checkbox"/> Sleep Deprivation <input type="checkbox"/> Student Project Involving Animals (Addendum 4) <input type="checkbox"/> Surgery* (Addendum 3) <input type="checkbox"/> Use of Hazardous Material(s) (Fill out 3.4 A)	
A. List hazardous materials to be used on this project. Materials can be harmful to humans, animals, or both. It is the PI's responsibility to have, up-to-date Material Safety Data Sheets (MSDS) for materials included on this form. Some materials (e.g., radioactive materials, rDNA and biohazardous materials) require additional institutional approval; contact the Office of Research Compliance for more information.			
Material(s): n/a			

\*Disruption of any integumentary surface is considered surgery except when: hypodermic needle, biopsy needle, ear punch, or a tail snip is performed. Addendum 4 details major, minor, multiple, survival and non-survival surgeries.



3.5 Pain or Distress					
Please complete the following table indicating the number of animals of each species used for each pain category. Pain categories are based on USDA criteria. USDA Categories:					
<ul style="list-style-type: none"> <li>Category B: Animals are those that are being bred, conditioned, or held for use in teaching, testing, experiments, research, or surgery but have not yet been used for such purposes, however minor.</li> <li>Category C: Animals are those that are subjected to procedures that involve no pain or distress, or procedures that would not require the use of pain-relieving drugs. (i.e. animal behavior or routine injections and blood samples)</li> <li>Category D: Animals are those subjected to potentially painful procedures for which anesthetics, analgesics, or tranquilizers will be used. (i.e. surgery with appropriate anesthesia and postoperative analgesia)</li> <li>Category E*: Animals are those subjected to painful or stressful procedures without the use of anesthetics, analgesics, or tranquilizers. (i.e. lethal dose studies or pain studies that do not allow pain-relieving agents)</li> </ul>					
Species	Category B	Category C	Category D	Category E*	
panthera tigris		7			
* Provide scientific justification for this pain category. (include criteria for moribundity and euthanasia)					
We are simply observing the animals from behind two (2) fence lines. There is no opportunity for us to impose any duress on the animals.					
3.6 Pain Alleviation					
Complete the table below.					
Species	Agent	Dose (mg/kg body weight)	Route	Frequency & Duration	Purpose

## ADDENDUM 2 PROTOCOL AMENDMENT REQUEST

This form is intended to be filled out if changes to ongoing, previously approved research or teaching activities involving animals are needed. Examples of changes include: title, funding, expiration date, animal species and numbers, procedures, personnel, etc. Protocol Amendment Requests should be submitted electronically to [IACUC@missouristate.edu](mailto:IACUC@missouristate.edu).

<b>Office Use Only</b>	Protocol Number: 2020-01 Date Filed: 8/3/2020	Approved: 8/17/2020 Date of IACUC/DMR Review: 8/17/2020	
<b>A. Investigator Information</b>			
Principal Investigator: Michael Clayton	Department: Psychology	Office Address: 409 Hill Hall	
Office Phone: 417-836-3783	Emergency Phone:	Email: mclayton@missouristate.edu	
<b>B. Project Information</b>			
Original Approval Date: April 6, 2020	Current Expiration Date: April 6, 2023		
Project Title: The Effects of Human Attentional State and Familiarity on Tiger ( <i>Panthera tigris</i> ) Sociability			
<b>C. Amendment(s)</b>			
Check the appropriate box(es) indicating the requested amendment(s) and provide information as needed.			
<b>TITLE</b>			
<input type="checkbox"/> Change Title To:			
<b>FUNDING</b>			
<input type="checkbox"/> Add Funding <input type="checkbox"/> Remove Funding			
Funding Source:			
<b>EXPIRATION DATE</b>			
<input type="checkbox"/> Change Project Expiration Date To:			
<b>PERSONNEL</b>			
<input checked="" type="checkbox"/> Add personnel			
Name	Title/Position	Degrees	Training/Experience
Jenna Dail	grad student	BA	<del>XXXXX</del> Complete
<input type="checkbox"/> Remove personnel			
Name		Title/Position	
<b>ANIMAL INFORMATION</b>			
<input type="checkbox"/> Remove animal species previously approved:			
<input type="checkbox"/> Add animal species not previously approved ( <i>Fill in the rest of ANIMAL INFORMATION section</i> )			
<input type="checkbox"/> Request additional animals previously approved ( <i>Fill in the rest of ANIMAL INFORMATION section</i> )			
Species	Common Name	Approximate Number	Source
<b>Justification for additional animals or species:</b>			

Pain or Distress				
Species	Category B	Category C	Category D	Category E*
*Justification for Pain Category E:				
PROCEDURE(S)				
<input type="checkbox"/> Add Procedure <input type="checkbox"/> Remove Procedure <input type="checkbox"/> Modify Procedure				
Please describe, in detail, the procedural change(s) requested. The following topics should be included, as applicable: necessity of change, species involved, number of animals involved, treatments, drugs, euthanasia, etc.				
USE OF HAZARDOUS MATERIAL(S)				
List hazardous materials to be used on this project. Materials can be harmful to humans, animals, or both. It is the PI's responsibility to have, up-to-date Material Safety Data Sheets (MSDS) for materials included on this form. Some materials (e.g., radioactive materials, rDNA and biohazardous materials) require additional institutional approval; contact the Office of Research Compliance for more information.				
Material(s):				
D. Literature Search				
Searches should provide justification of the <u>three "R's"</u> . A new search using the same keywords from the original protocol may be sufficient. If you completed the PROCEDURE(S) or ANIMAL INFORMATION section(s), please complete a literature search.				
Date of Search	Keywords	Resources Used	Years Covered	
Results: Summarize how the search indicates necessity of an animal model, lack of duplication or the need to repeat previous studies.				

Michael Clayton  
Principal Investigator

August 3, 2020  
Date

## Appendix B. Task List for Coding Sociable Behaviors

- In Proximity
  - Tiger is within specified 4.5-meter area, using the designated ground stake markers on either side of human, as a guide.
  - Tiger must also be within two of their own footsteps away from the front fence line of their enclosure to be considered ‘in proximity’.
    - A step is considered the lifting and forward placement of one foot.
  - To document intervals of duration:
    - Record arrival time stamp of video recording once tiger enters specified proximity.
    - Record departure time stamp once tiger leaves specified area.
      - Calculate and record seconds between arrival and departure time stamps to gain one interval of time.
    - Record new occurrence if tiger is outside of proximity for 2 consecutive seconds, then re-enters.
    - Record beginning and/or end time stamp(s) of video recording if tiger begins session in proximity and/or ends session in proximity.
- Glance
  - Tiger overtly moves their head or eye line to approximately align in some variation (horizontally or vertically) with current position of human’s head.
  - Behavior occurrences should be tallied.
  - Record new occurrence if tiger desist behavior completely for 2 consecutive seconds, then reengages, following the above definition.
- Chuff/vocalization
  - Tiger produces an audible noise, referred to as a chuff, or other vocalization including sub roar, long call, coughing growl, or bark. To accurately document, examples for each should be listened to on The Prusten Project’s website, (Ferlemann, 2018) before collecting data.
  - Behavior occurrences should be tallied.
  - Record new occurrence if tiger desist behavior completely for 2 consecutive seconds, then reengages, following the above definition.
- Fence Touch
  - Tiger rubs against fence with their head or another body part (e.g., side or paw).
  - Behavior occurrences should be tallied.
  - Record new occurrence if tiger desist behavior completely for 2 consecutive seconds, then reengages, following the above definition.



## **Appendix C. Statistical Analysis Footnote**

Given that data represents repeated measures, by combining the tigers attentional and familiarity data for analysis, results in doubling the n size for the statistical tests. This could have resulted in a confound in the part of the analyses given the data represent repeated measures for attentional and familiarity observations. However, given the visual inspection of the data (see Figures 4, 6, 8, and 10), those interpretation and findings appear in concordance. It is possible to create summary scores across observations and repeat the data analyses using the Wilcoxon vs the Mann Whitney U. Again, given the visual observations and results of independent group comparison, it is the contention of the author that the stated findings would be represented.