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## **The Effects of Olfactory Stimuli on the Behavior of the Far Eastern/Amur Leopard (*Panthera pardus orientalis*) in Captivity**

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The University of Southern Mississippi

THE EFFECTS OF OLFACTORY STIMULI ON THE BEHAVIOR OF THE FAR  
EASTERN/AMUR LEOPARD (*PANTHERA PARDUS ORIENTALIS*) IN CAPTIVITY

by

Marks Stimson McWhorter

A Thesis

Submitted to the Graduate School  
of The University of Southern Mississippi  
in Partial Fulfillment of the Requirements  
for the Degree of Master of Science

Approved:

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Director

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Dean of the Graduate School

May 2014

## ABSTRACT

### THE EFFECTS OF OLFACTORY STIMULI ON THE BEHAVIOR OF THE FAR EASTERN/AMUR LEOPARD (*PANTHERA PARDUS ORIENTALIS*) IN CAPTIVITY

by Marks Stimson McWhorter

May 2014

Two Amur leopards from the Jackson Zoological Park were observed under continuous surveillance while on exhibit over 21 days to better understand the activity budget of leopards in captivity, and what effects olfactory stimuli may have on those activity patterns. Behaviors were observed through surveillance systems located around their enclosure and scored using an ethogram, and proportions of behaviors shown were calculated for each day, and by 15-minute increments throughout the day. Over the course of this study, leopards received three olfactory stimuli on separate occasions, where we then compared behaviors before stimuli presented to behaviors expressed during and after enrichment was presented.

Overall, leopards were active on average around two hours, 30 minutes each day, with pacing accounting for approximately 30 minutes to one hour of that time. Leopards interacted with stimuli; however, their use was extremely short-lived, primarily only the first two hours of the first day, with use dropping between 85% and 90% from the first to second day. Animals were also selecting certain areas of their exhibit, using 51% of their exhibit 83% of the time. Ultimately, some stimuli may have positive effects on modifying captive leopards, but those effects may not last to a second day. Zoos should continually modify stimuli and account for almost immediate habituation for an enrichment program to be effective.

## ACKNOWLEDGMENTS

I would like to take this opportunity to thank everyone who has assisted in this project. First, this thesis would not have been possible without the continual advice and counsel of my graduate committee, Dr. Carl P. Qualls, Dr. Stan Kuczaj, and Dr. Aimée K. Thomas. Each member of this committee created a unique perspective that, I believe, made this study different from most seen in this field. Without their guidance, this thesis would not have ended in what I believe are valuable results to the zoo field.

I would also like to thank the Jackson Zoo and every staff member who assisted in the creation and implementation of this study. The zoo's animal management team (Beth Poff, Director; Dave Wetzel, Assistant Curator; Willie Bennett, Animal Curator; and Donna Todd, Veterinary Technician) provided valuable insight during the creation of our prospectus and approved our study to observe two of their Amur leopards. They were excited to listen to the results of our research and ultimately modified their exhibit, taking into account some of our findings to further improve the welfare of their animals. Two of the zoo's leopard keepers, Jonathan Fields and Jessica Howard, were also extremely helpful with providing time to set up camera equipment and place olfactory stimuli on exhibit.

Lastly, I would like to thank my friends and family for their support throughout the pursuit of my master's degree, including Jennifer Lamb, Kayla DiIorio, Sean Kochtitzky, Kelly Breland, Jay Eubanks, and Melissa Gutierrez. This study culminated from a passion in science education and conservation, established by a friend and colleague, Percy King, who first sparked my interest in science at the Jackson Zoo years ago. To him and all others, I say thank you for your support.

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## CHAPTER I

## INTRODUCTION AND OBJECTIVES

*The Amur Leopard; An Endangered Subspecies*

The Far Eastern/Amur is one of nine subspecies currently recognized within the species *Panthera pardus* and has the lowest genetic diversity of all subspecies (Uphyrkina et al., 2001). The conservation status of this subspecies is of critical concern, listed as endangered through the International Union for Conservation of Nature and Natural Resources (IUCN) in 1994, and critically endangered since 1996 (Jackson & Nowell, 2008). With the wild population listed as declining (Jackson & Nowell, 2008), from 25-40 individuals in 2002 (Uphyrkina, Miquelle, Quigley, Driscoll, & O'Brien, 2002) to 25-35 individuals in 2011 (Hebblewhite, Miquelle, Murzin, Aramilev, & Pikunov, 2011), the survival of this subspecies is currently relying on organizations through captive efforts. While it is currently established that one founder leading to the current population may not have been of the same subspecies, the genetic diversity of captive Amur leopards is more diverse than the wild population (Uphyrkina et al., 2002). Further, current wild populations have also become so fragmented that inbreeding could further deteriorate their genetic diversity (Uphyrkina & O'Brien, 2003). With habitat loss and poaching a constant threat (Miquelle et al., 2010), and genetic diversity strongest within captive programs, zoological institutions have the strongest position to ensure that the Amur subspecies can survive for further generations.

*Environmental Enrichment in a 21<sup>st</sup> Century Zoo*

Several studies have been conducted to better prepare programs for reintroduction efforts (e.g. Hebblewhite et al., 2011; Miquelle et al., 2010; Uphyrkina & O'Brien, 2003),

but until further reintroduction programs are established, zoological institutions are responsible for maintaining both the genetic and behavioral diversity that Amur leopards would likely have in the wild. Zoos have now become increasingly aware of the behavioral needs of animals under their care, with the number and diversity of environmental enrichment programs increasing dramatically over the past several decades (Shepherdson, 2003). These programs ultimately seek to better the welfare of captive animals. While environmental enrichment has been defined in several ways, one of the first books written on the subject describes it as "...an animal husbandry principle that seeks to enhance the quality of captive animal care by identifying and providing the environmental stimuli necessary for optimal psychological and physiological well-being" (Shepherdson, Mellen, & Hutchins, 1998). Enrichment provides zoo staff the opportunity, on a species-specific basis, to increase the behavioral diversity of their animals by incorporating the needs of an animal in a captive environment into their exhibit, and that need is paramount in the 21<sup>st</sup>-century zoo (Shepherdson, 2003).

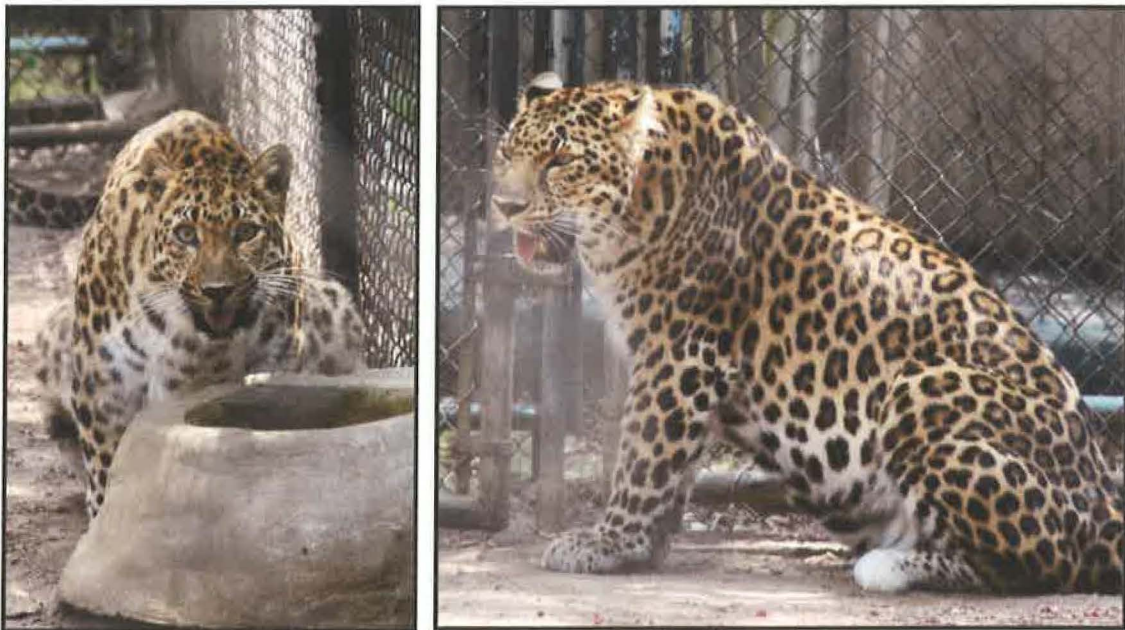
The increased awareness of environmental enrichment has brought about hundreds of studies in the past decade, many of which focused on assessing behavioral changes. Zoos are now monitoring the behaviors of their collection in a more comprehensive way, understanding that monitoring an animal's behavior can aid in detecting health issues and recognizing when abnormal behaviors appear (Watters, Margulis, & Atsalis, 2009). There were 374 studies of environmental enrichment published between 1984 and 2004, through popular zoo journals, that quantified the effectiveness of stimuli by measuring animal behavior (de Azevedo, Cipreste, & Young, 2007). With the advent of environmental enrichment, several studies covering a variety

of taxa have not only focused on promoting species-typical behaviors but also on decreasing or eliminating atypical or “stereotypic” behaviors (e.g. Miller, Kuczaj, & Herzing, 2011; Quirke, O’Riodran, & Zuur, 2012; Rees, 2009; Smith & Litchfield, 2010; Swaisgood et al., 2001; Therrien, Gaster, Cunningham-Smith, & Manire, 2007; Vickery & Mason, 2004). A review of the current literature by Shyne (2006) found that 90% of the 63 studies reviewed saw a reduction in observed stereotypic behaviors after enrichment was given. Behaviors that were identified as stereotypical ranged from pacing observed in several species, to swaying, head swinging, regurgitation, and hair plucking. While pacing is the most observed atypical behavior among the carnivore order, hair plucking, or over-grooming, and self-biting have also been observed. The term stereotypic behavior has been used inconsistently to cover various behaviors among taxa, resulting in a vague understanding as to what behaviors fall under a stereotypic category (Mason, Clubb, Latham, & Vickery, 2007). The term stereotypic behavior can also create further problems, given that the causes of a given atypical behavior could vary from another atypical behavior of the same individual. Further discussion into the causes of such behaviors and difficulties in a catchall category of stereotypies are addressed in Mason et al. (2007), and for the purpose of this study, any undesired behaviors observed will be categorized as abnormal repetitive behaviors (ARB). By describing this behavior in further detail, I hope to provide a better understanding of the atypical behaviors of captive animals.

#### *The Focal Animal at the Jackson Zoological Park*

The focal animal for this study is the Far Eastern/Amur Leopard (*Panthera pardus orientalis*). Two individuals (Figure 1), one male and one female, were chosen at

the Jackson Zoological Park (JZP), an Association of Zoos and Aquariums (AZA) institution in Jackson, Mississippi. Nikolai, the zoo's male was born 30 June 2002 at Erie Zoological Gardens, Erie, Pennsylvania, and Katya, the zoo's female was born 15 July 2000 at the Pittsburgh Zoo & PPG Aquarium, Pittsburgh, Pennsylvania. Both individuals were transferred to JZP in November 2004 and have been housed together since arriving. Both individuals were transferred to a new exhibit before this study was conducted and released into their new exhibit for the first time the day before this study began. Leopards were not being relocated for this study; instead, zoo personnel were shifting individuals to a larger exhibit, which provided us with the opportunity to conduct this study.



*Figure 1.* Two adult Amur leopards observed at the Jackson Zoological Park for this study. Katya, the zoo's female, is shown on the left, and Nikolai, the zoo's male, is shown on the right.

### Objectives

The objectives of this study were to (a) determine an approximate activity budget for captive leopards (b) determine if species-typical and atypical behaviors correlate to time of day (c) evaluate the effectiveness of olfactory stimuli on the behaviors exhibited by captive Amur Leopards at JZP (d) determine if different olfactory stimulus treatments have equal effect in modifying a leopard's behaviors, and (e) evaluate exhibit use by leopards in a new enclosure. Using the methodology described below (i.e., comparing behaviors for each enrichment treatment to a pre-enrichment period), this study statistically evaluates the effect of each olfactory stimulus (scent) on the behavior of captive individuals. Further, by statistically comparing the changes in behavior (from non-stimulus control periods) among treatments, this study will also determine which of the stimuli given have the greatest effect on modifying behaviors.

CHAPTER II  
ACTIVITY BUDGET OF TWO CAPTIVE AMUR LEOPARDS  
AT THE JACKSON ZOOLOGICAL PARK

Introduction

Activity budgets for species-typical behaviors vary across species; however, most studies suggest that resting is the dominant behavior for felines (e.g. Mallapur & Chellam, 2002 with Indian leopards; Sulser, Steck, & Baur, 2008 with snow leopards; Weller & Bennett, 2001 with ocelots; & White, Houser, Fuller, Taylor, & Elliott, 2003 with Siberian and Sumatran tigers). Mallapur and Chellam (2002) also note that the highest concentration of pacing occurred while keepers were near the exhibit, in late morning and late afternoon before feeding. Leopards were significantly more active shortly before feeding time at the end of the day, likely due to the predictability of feeding schedules in captivity. Weller and Bennett (2001) also showed that little resting and a higher concentration of pacing occurs shortly before feeding time in the afternoon. While the captive ocelots had these two peaks of activity, times for these peaks varied slightly among individuals (Weller & Bennett, 2001).

To further understand the activity budgets of captive leopards, we installed video surveillance equipment to record daily activity. While several studies were conducted by having observers directly watch focal animals throughout the day (Burgener, Gusset, & Schmid, 2008; Jenny & Schmid, 2002; Powell, 1995; Skibieli, Trevino, & Naugher, 2007; Yu et al., 2009) we used surveillance equipment both to avoid any potential acclimation time needed for leopards towards the observer and so the observer could identify behaviors exhibited by leopards in further detail from recorded video. This also gave us a

greater advantage in recognizing any potential correlations between expressed behaviors and time of day. Given that keepers' schedules vary by day, this also gave us the opportunity to compare behaviors by day of leopards from the moment they were released into the exhibit, as opposed to time of day (i.e., compare hour 1 on exhibit instead of 0800 – 0900).

While some studies have shown that visitors can potentially modify an animal's behavior (see Fernandez, Tamborski, Pickens, & Timberlake, 2009 for a review), this study did not film or collect data on visitors. However, this study took place over a time period where no major events occurred, meaning no extreme rise in visitor attendance. Two studies (Hosey, 2008; Margulis, Hoyos, & Anderson, 2003) also suggest that visitor interaction may not have a large impact on captive felines, or at least that those effects do not necessarily modify their behavior. This review also discusses potential behavioral effects due to keeper interaction. Keepers work in close proximity with captive animals on a daily basis, and since research was done in close proximity with animals on exhibit, keepers that assisted in this study followed a normal schedule consistent with their previous routines, and no other personnel were present over the course of the study.

### Methodology

Leopards were observed for 21 days while on exhibit. Video cameras were placed around the perimeter of the exhibit, and video was digitally recorded from 0827 to 1627. Given the continuously changing schedules of keepers, leopards were shifted into the outdoor exhibit between 0830 and 1000 and shifted into the night enclosure between 1600 and 1630. Video was stored on a digital video recorder and behaviors were scored using an ethogram (Table 1) modified from previous studies (Margulis et al., 2003;

Powell, 1995; Yu et al., 2009). While other studies characterized behaviors during only limited time periods, using various sampling methods described by Altmann (1974), (Macri & Patterson-Kane, 2011; Mallapur & Chellam, 2002; Powell, Carlstead, Tarou, Brown, & Monfort, 2006; Sulser et al., 2008) or a combination of sampling and continuous observation (Yu et al., 2009), we scored all behaviors in view of surveillance equipment during the entire time the animals were on exhibit each day. While more time and labor intensive, this method of continuous observation provided us with more available data for the two leopards observed during this study.

Both individuals had resting locations on exhibit that were out of view of our surveillance system. While the female rested in areas that were typically in view, our male almost always rested on a section below a platform that was not visible. Given that this location was large enough that any active movement would have facilitated a view on camera, but small enough that we could not determine if he was resting, sleeping or grooming, these occurrences were scored as out of view. With a lack of data for the male's resting durations, resting durations were not determined.

Table 1

*Ethogram for Amur Leopards at Jackson Zoological Park*

Behavior	Description
<b>Active Behaviors</b>	Individual moves from one location to another or interacts with exhibit features
Exploratory	Individual moves limbs at multiple paces in a forward direction while searching; no shorter than 30 seconds
Self Play	Individual moves in an excited manner by interacting with a part of its exhibit that is not enrichment being studied or its own body (feet, tail etc)
Claw Sharpen	Individual moves paws, with claws extended, in a repetitive motion against a surface



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Scent Mark	Individual raises tail and sprays an object, rubs cheeks against a surface, or scratches with claws not in a repetitive motion
Interact w/foilage	Individual plays, chews, or manipulates foliage and/or trees in some way for the purpose of using foliage as enrichment
Interact w/enrichment	Individual interacts with enrichment item being studied
<b>Abnormal Repetitive Behavior</b>	
Pacing	Individual acts in a repetitive manner expressed as stress; must cover repeated area at least three times in sequence
<b>Rest</b>	Individual stays in one location without any other action
Relax	Individual lays on stomach or side while awake
Sleep	Individual lays on stomach or side with eyes closed and no movement for an extended period
<b>Affiliative Behavior</b>	Individual interacts with another individual of the same species without expressing stereotypical behavior
Huddle/sleep	Individual rests against or in close proximity to an individual of the same species
Groom	Individual licks with the apparent intent to clean another member of the same species
Play	Individual interacts with another individual of the same species while moving in an excited manner
Aggression (Display)	Individual shows display postures and/or threats to an individual of the same species without contact
Aggression (Physical)	Individual shows display postures and/or threats to an individual of the same species with contact
Mate	Individual attempts to or does mate with another individual on exhibit
Other	Individual interacts with another individual in some way not categorized above
<b>Self Groom</b>	Individual licks with the apparent intent to clean itself
<b>Out of view</b>	Individual is not visible through video surveillance
<b>Urinate or Defecate</b>	Individual excretes waste
<b>Drink</b>	Individual intakes water
<b>Eat</b>	Individual acquires, chews, and swallows food
<b>Aggression</b>	Individuals show the intent through display to cause harm to an individual not of the same species

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Data were analyzed by calculating duration of all behaviors observed (states) and frequency of behavior expressed (events) for appropriate behaviors. Activity levels for each behavior were first calculated by averaging duration of behaviors observed for each day over the 21-day study. Considering that video was recorded for approximately 8 hours each day, days were divided into thirty-three, 15-minute increments (i.e., 0815-0830, ... 1615-1630), and values were calculated for each behavior as the proportion of time that behavior was exhibited over the 15-minute increment the individual was in view. Wilcoxon Rank-Sum tests were used to compare durations of behaviors between individuals, considering that the assumptions for parametric tests (i.e., normal distributions or equal variances) were not met (Zar, 2010).

In addition to daily activity, we also analyzed differences in proportion of behavior throughout the day. We divided the amount of time leopards were on exhibit into thirty-three, 15-minute increments (i.e., 0815-0830...) and compared proportions of each behavior expressed among each time frame using Kruskal-Wallis tests. This analysis can determine if any behaviors are occurring at a specific time of day. We also compared differences among behaviors for the first three hours animals were released into their outdoor enclosure. Considering that both animals were not released on exhibit at the same time each day, we compared the first twelve 15-minute increments leopards were on exhibit (i.e., 00-15 min, 15-30 min...). This analysis determined the most prevalent behaviors expressed when first on exhibit, and any potential changes in behavior over the first three hours.

## Results

### *Daily Activity Patterns of Captive Leopards*

Leopards were observed for 168 hours of continuous observation, divided into 21 days, 8 hours per day, and activity budgets were created by day and are summarized in Table 2. Resting behaviors (relaxing, sleeping, and self grooming) were not compared between individuals due to the male's primary resting location being not visible to the surveillance system. Active behaviors, including pacing, accounted for approximately 2 hours, 31 minutes per day for the male, and 2 hours, 23 minutes for the female. There was no significant difference in the proportion of time exhibiting exploratory behavior between leopards (32.82% male, 30.97% female,  $Z = 0.553$ ,  $P = 0.580$ ) or interacting with foliage (3.23% male, 1.24 % female,  $Z = 1.488$ ,  $P = 0.137$ ). There was however, a significant difference in the proportion of time spent scent marking (1.48% male, 0.58% female,  $Z = 2.528$ ,  $P = 0.012$ ) and claw sharpening (0.04% male, 0.09% female,  $Z = 2.171$ ,  $P = 0.030$ ).

Table 2

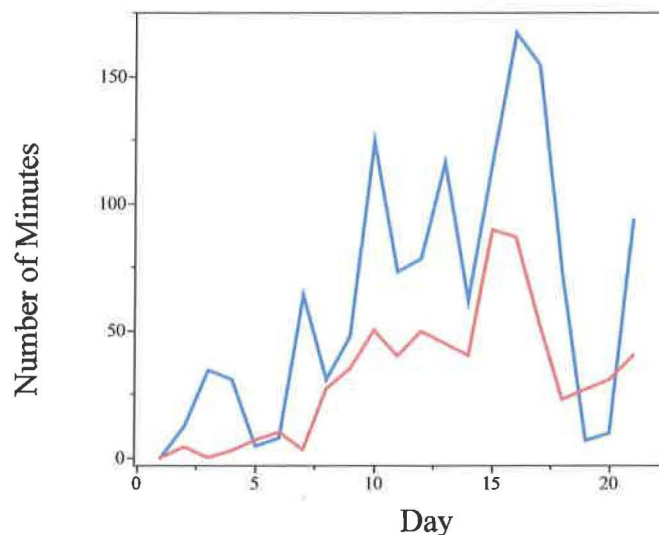
*Mean duration of behaviors (in minutes) per day of male and female leopards and standard deviation.*

Behavior	Male	Female	Behavior	Male	Female
<b>Active Behaviors</b>			<b>Rest</b>	N/A	199.14 ± 93.67
Exploratory	57.08 ± 17.16	98.30 ± 31.55	Relax	N/A	168.89 ± 70.59
Self play	9.99 ± 5.53	**	Sleep	N/A	25.05 ± 38.39
Claw sharpen	0.09 ± 0.18	0.34 ± 0.37	<b>Affiliative Behavior</b>	1.55 ± 1.03	
Scent mark	2.23 ± 1.40	1.71 ± 1.46	Huddle/sleep	0	
Interact w/foilage	7.51 ± 5.87	4.50 ± 3.27	Groom	0	
Interact w/enrichment	10.56 ± 13.07	3.64 ± 4.60	Play	0.83 ± 0.99	
<b>Abnormal Repetitive Behavior</b>			Aggression	0.72 ± 0.71	
Pacing	62.01 ± 50.93	31.96 ± 26.15	Mate	0	
<b>Self Groom</b>	N/A	11.72 ± 10.90			

Note: self play behavior not quantified for female. Also see discussion on abnormal repetitive behaviors below.

*Abnormal Repetitive Behaviors: Daily Activity and The Relationship between Behavior and Time of Day*

The most abnormal repetitive behavior recognized was pacing, observed on average 62.01 and 31.95 minutes a day, for the male and female, respectively. Pacing was observed in both individuals; however, this behavior was recorded significantly more in the male (17.97% male, 7.29% female,  $Z = 5.640$ ,  $P < 0.001$ ). It is also important to note that there was extreme day-to-day variability in pacing over the 21-day study (Figure 2). Further variability is also shown in the number of occurrences of pacing per day (range per day, Nikolai: 0-35; Katya: 0-45). While these ranges differed between individuals, there was no significant difference in the number of times the two individuals paced per day (12.71 times per day, Nikolai; 12.62 times per day, Katya;  $Z = -0.252$ ,  $P = 0.801$ ). In total, Nikolai was recorded pacing 257 times, at an average of 5.07 minutes per pacing bout and Katya was recorded pacing 262 times, at an average duration of 2.56 minutes.



*Figure 2.* Total number of minutes Nikolai (male; in blue) and Katya (female; in red) spent pacing by day over the 21-day study. Range between 0 and 166.8 minutes for Nikolai and between 0 and 89.45 minutes for Katya.

With respect to time of day, there was no statistically significant difference in pacing among all 15-minute increments (Figure 3) for Nikolai ( $H = 44.254$ ,  $df = 31$ ,  $P = 0.058$ ); however, there was a difference among times for Katya ( $H = 76.229$ ,  $df = 32$ ,  $P < 0.001$ ). Post-hoc analysis indicated that two 15-minute increments (1430-1445 & 1515-1530) had the largest mean proportion of pacing exhibited and had significantly larger proportions than all other 15-minute increments (1030-1045 & 1000-1015).

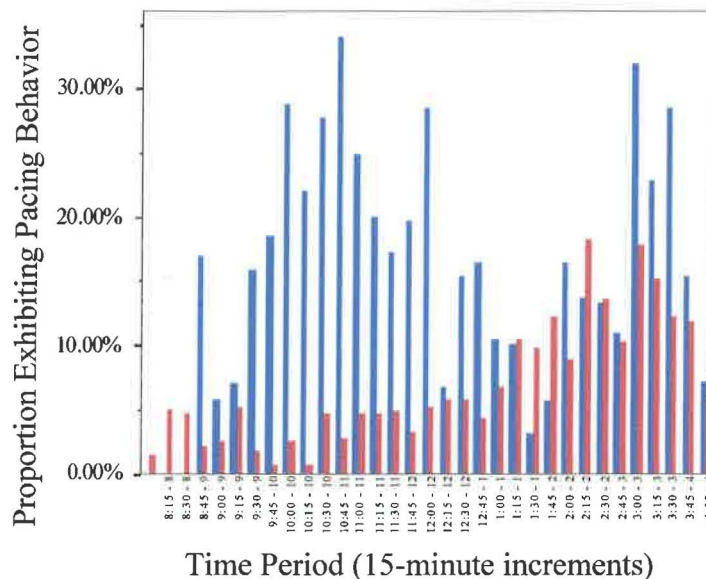
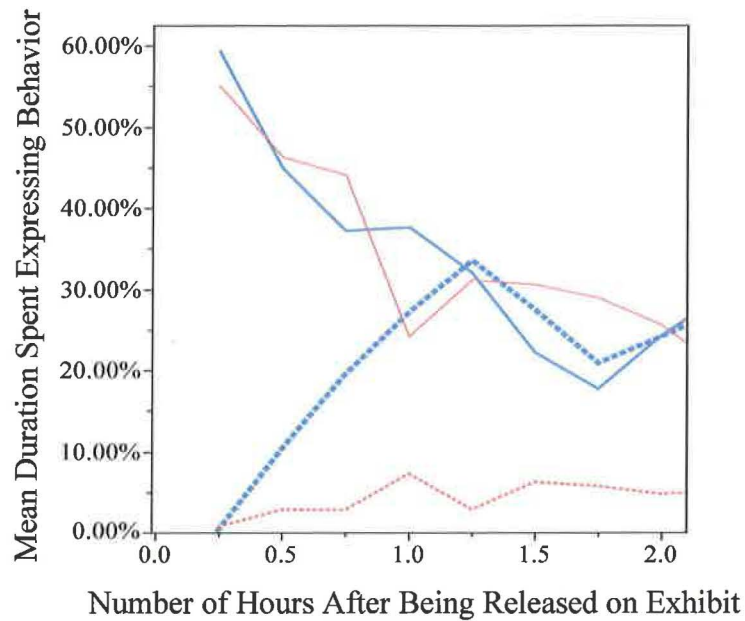


Figure 3. Proportion of time leopards paced while on exhibit. Nikolai (male) shown in blue and Katya (female) shown in red. Each bar represents the mean proportion of pacing exhibited per 15-minute increment from 0815 to 1630.

When accounting for both Nikolai and Katya's release time, there was a significant difference between two of the twelve 15-minute increments (0-15 minutes,  $mean = 0.003\%$ ; 75-90 minutes,  $mean = 33.5\%$ ;  $H = 26.368$ ,  $df = 11$ ,  $P = 0.0057$ ). As shown in Figure 4, pacing steadily increased for Nikolai over the first 90 minutes on exhibit, as proportion of exploratory behavior also decreased. There was no significant

difference in the proportion of time spent pacing among any of the twelve 15-minute increments for Katya ( $H = 13.721$ ,  $df = 11$ ,  $P = 0.2488$ ).



*Figure 4.* Proportion of time leopards spent pacing (dotted lines) and exploring (solid lines) for the first two hours while on exhibit. Nikolai (male) shown in blue and Katya (female) shown in red.

#### *Species-Typical Behaviors and their Relationship to Time of Day*

The most expressed active behavior when released on exhibit was exploratory behavior. Accounting for release time, there was a significant difference between the first 15 minutes and the four increments between 90 and 150 minutes ( $H = 29.758$ ,  $df = 11$ ,  $P = 0.0017$ ) for Nikolai (Figure 4). Specifically, the mean proportion of exploratory behavior during the first 15 minutes on exhibit was 59.23%, significantly greater than during the four consecutive increments between 90 and 150 minutes (22.13%, 17.67%, 24.22%, and 29.37%, respectively). Similarly, there was a significant difference between the first 15 minutes and five increments between 60 to 75 minutes and 120 and 180 minutes ( $H = 25.028$ ,  $df = 11$ ,  $P = 0.0090$ ) for Katya (Figure 4). The mean proportion of

exploratory behavior during the first 15 minutes on exhibit was 54.99%, significantly greater than the five increments listed earlier (24.12%, 25.54%, 19.93%, 25.56%, and 22.08%, respectively).

### Discussion

Leopards were on exhibit 7 hours, 56.5 minutes and 7 hours, 47.21 minutes on average per day, for Nikolai and Katya, respectively. The most dominant behavior expressed during this time was resting, with active behaviors (i.e., species-typical and abnormal repetitive behaviors) exhibited on average 31.67% for Nikolai and 32.90% for Katya. Exploratory and pacing behaviors were the most observed of these active behaviors, with pacing accounting for 13% and 6.84% of daily activity for Nikolai and Katya, respectively. This daily amount of pacing is similar to leopards observed at four zoos in India (Mallapur & Chellam, 2002), who spent 2-11% of daily activity in stereotypic pacing. Observed proportions of exploratory behavior accounted for 11.97% (Nikolai) and 21.04% (Katya), also similar to Mallapur and Chellam (2002), with the frequency of active behaviors varying between 6.22% and 14.9% among leopards at four zoos in India. Frequencies of observed active behaviors were significantly different between individuals, with Nikolai having a greater proportion of active behavior ( $Z = 9.318, P < 0.001$ ). This is contradictory to one study with captive ocelots (Weller & Bennett, 2001), which observed one male, compared to five females, having the lowest proportions of active behaviors; however, this difference is likely due to the Nikolai's increased frequency of pacing, which was significantly greater than Katya's.

Substantial standard deviations around the mean durations of these behaviors (Table 2) illustrate that there is considerable day-to-day variability among individuals

with respect to frequency of typical and atypical behaviors. While we observed days where frequency of pacing exceeded one hour (11 days, Nikolai; two days, Katya), we also should note that there was one day when Nikolai was not observed pacing, and two days where Katya exhibited no pacing behavior. This variability could suggest that modifying daily variables (e.g., introducing enrichment in an appropriate manner or modifying an animal's feeding schedule) could sufficiently reduce observed ARBs.

No other previously recognized ARBs most commonly seen in felines were observed over the course of this study, including over-grooming and self-biting, which have been identified among several species in the carnivore order (Mason et al., 2007). One behavior, however, was observed over the course of this study that could not be categorized within our ethogram, and seemed repetitive in nature. Katya, our female, was observed routinely pausing, quickly shifting weight to her hind-limbs, and standing perpendicular to the ground, then rotating her neck and head in a swift, counter-clockwise motion, fully rolling her neck from the right shoulder, left shoulder, and back to its original position. This behavior only lasted for approximately two seconds, and she never appeared to be staring at a single object before the behavior was exhibited (i.e., never following a running squirrel or bird). This was not similar to her normal stretching behaviors, or observed play behaviors. We observed this at several locations on exhibit, and at various times throughout the day. No specific behaviors routinely followed this, or preceded it. At first glance, this was categorized as a type of self-play; however, once we more thoroughly reviewed the behavior, we categorized it as an ARB. Considering that this was not reviewed until midway into data analysis, we could not statistically evaluate frequencies of this behavior over the course of the study. To my knowledge, no other



studies have described such a behavior expressed in felines, and considering we could not identify its origin or purpose, we categorized it as a type of ARB.

Frequencies of atypical behaviors observed could partially be due to a predictable feeding schedule, which has been shown to increase rates of pacing (Lyons, Young, & Deag, 1997; Moreira et al., 2007; Quirke & O’Riordan, 2011; Quirke et al., 2012). Modifying food presentation through the use of enrichment has been shown to alter behavior, decreasing repetitive pacing and increasing frequency of feeding behavior (Bashaw et al., 2003). In this study, Katya was more likely to pace in the afternoon, shortly before zoo staff arrived, and while not statistically significant with respect to Nikolai, both individuals showed increased rates of pacing shortly before zoo staff arrived to transfer individuals into their night-house and feed (Figure 3). Nikolai’s pacing levels are similar to those of leopards observed by Mallapur and Chellam (2002); however, Katya was significantly more likely to pace in the late afternoon (specifically between 1430-1445 & 1515-1530). Nikolai also seemed to exhibit higher frequencies of pacing in late morning, approximately 90 minutes after being released. When accounting for release time (Figure 4), Nikolai showed his highest frequency of exploratory behavior, and additionally, lowest frequency of pacing, immediately after being released. As time progressed his proportion of exploratory behavior steadily decreased, while his frequency of pacing increased, reaching its maximum frequency between 75-90 minutes. If Nikolai’s pacing levels are at their highest within this time frame, it could then be hypothesized that introducing stimuli approximately 90 minutes after being released or modifying an animal’s schedule around that time could reduce proportions of exhibited ARBs.

This component of the study assessed frequencies of behavior displayed by two individuals, one male and one female, housed in a single exhibit. It is important to note that, while we found significant differences between Nikolai (male) and Katya (female), we are not suggesting that these differences are solely based on gender. Several studies have also evaluated behavioral differences between gender and among age groups, including daily activity, groups vs. solitary individuals, or differences in response to stimuli (Jenny & Schmid, 2002; Powell, 1995; Weller & Bennett, 2001). It is our hope that as future data are collected in addition to previous literature and a larger sample size is compared, these data will be able to assist in identifying the myriad differences recognized among individuals in our captive population.

## CHAPTER III

OLFACTORY STIMULI AND THEIR EFFECTS ON THE BEHAVIOR OF AMUR  
LEOPARDS (*PANTHERA PARDUS ORIENTALIS*) IN CAPTIVITY

## Introduction

Studies of environmental enrichment have recently increased dramatically among the carnivore order, given that multiple species within this group are frequently housed in zoological institutions. The most common ARB observed among carnivores is pacing (Mason et al., 2007), which is of principle concern in almost all studies of behavior in captive felids. Pacing decreased with a male and female pair of Amur tigers after implementing a feeding box (Jenny & Schmid, 2002), and pacing significantly decreased with a pair of tigers once a visual barrier was created to decrease the viewing of conspecifics (Miller, Bettinger, & Mellen, 2008). A study completed in 2008 evaluating the effectiveness of feeding boxes, however, found no significant difference in proportions of pacing when introducing feeding boxes with two snow leopards (Burgener, Gusset, & Schmid, 2008). While it is important to note that pacing does not directly correlate towards poor animal welfare (see Swaisgood & Shepherdson 2005 for review), its presence should at the least focus the attention of institutions to remove it.

Aside from reducing ARBs, feline studies have also shown that environmental enrichment can significantly increase species-typical behaviors. Incorporating feeding boxes into the feeding schedule of Amur tigers not only decreased pacing but also increased sleeping in females (Jenny & Schmid, 2002). Frozen treats significantly increased paw manipulation and licking/gnawing in adult lions, and paw manipulation, licking/gnawing, and sniffing in cubs on exhibit (Powell, 1995). Bones and frozen treats

increased activity levels on the day the enrichments were given in six species of felines (Skibieli et al., 2007). Enrichments relating towards olfaction have also been used frequently among felines. Scents of nutmeg increased exploratory behavior in Amur leopards (Yu et al., 2009) and catnip increased active behaviors in black-footed cats (Wells & Egli, 2004). While these enrichments have been frequently used to elicit desired behavioral responses, few studies have shown them to have lasting effects, with some enrichments losing efficacy between two and seven days (see Skibieli et al., 2007; Wells & Egli, 2004; & Yu et al., 2009 for further review).

Given the monetary constraints upon most nonprofit zoological institutions, enrichment items must be inexpensive and readily available in order to see widespread use. Therefore, three olfactory stimuli were chosen for this study that could all be easily accomplished by zoo staff without any funds dedicated towards enrichment. This could ensure that if stimuli have positive effects on leopards, then other facilities could use similar stimuli in an inexpensive manner. Since these stimuli have not been shown, experimentally, to have a positive effect on the welfare of captive leopards, they should not yet be qualified as enrichment, so these items will herein be referred to as “olfactory stimuli.” Stimuli chosen include hay with the scent of a Sumatran tiger (*Panthera tigris sumatrae*), hay with the scent of a pot-bellied pig (*Sus scrofa domesticus*), and hay without any scents added. Each stimulus was given one time to the individuals on exhibit and their behaviors were recorded, quantified, and compared to assess any potential differences before and after exposure to the olfactory stimuli.

## Methodology

Three olfactory stimuli were each given individually on randomly established days, once each, over the course of the study: hay with tiger scent, hay with pig scent, and hay (as a control for scents). Stimuli were only used once in this study considering that habituation would likely occur after stimuli are first encountered (Kuczaj et al., 2002). Hay with tiger scent was rubbed in urine from the night enclosure of JZP's Sumatran tiger (*Panthera tigris sumatrae*) exhibit. Three Sumatran tigers are housed at JZP, and all are male. Hay with pig scent was removed from the night enclosure of JZP's pot-bellied pig (*Sus scrofa domesticus*) exhibit after being used for bedding the previous night. Two pigs are housed at JZP, and both are female. Hay for both of these treatments was taken from the hay barn at JZP. To account for any potential scents from the barn or previous encounters, hay from the same barn but without any additional scent was used as a control. The amount of hay was the same for all treatments, and each olfactory stimulus was randomly placed within the leopard exhibit, where it remained for three days. An error occurred on day 22 and an enrichment item (cardboard box) was placed on exhibit the third day with the tiger scent stimulus, voiding any data collected that day, and ending the study early at 21 days. Each stimulus was randomly selected in order of use, and was presented to leopards at random intervals over the 21-day study (i.e., no stimulus, days 1-8; hay [pig scent], days 9-11; no stimulus, days 12-13; hay [control], days 14-16; no stimulus, days 17-19; hay [tiger], days 20-21). No other stimuli were provided to leopards throughout the 21-day study, and to avoid behavioral changes continuing to a following treatment, we created a 2-day minimum period without a stimulus between treatments, or at least 5 days from one treatment to another.

Use of stimuli was compared for each leopard by averaging total duration of behaviors two days before stimuli were presented and comparing that to the day stimuli were placed on exhibit. It should be noted that I only compared total duration of behaviors on the first day each stimulus was given to pre-stimulus data, given that the primary use of each stimulus only occurred on the first day (87.19% of use for Nikolai; 90.82% for Katya). Wilcoxon Rank-Sum tests were then used to compare the prevalence of individual behaviors two days before versus the day after the introduction of each olfactory stimulus, separately for each leopard. Pre-stimulus data were taken from the two days before stimuli were present to compare and average duration of behaviors before a stimulus to the frequency of behaviors observed while interacting with a stimulus.

### Results

Both leopards interacted with all stimuli on the first and second day they were present; however, use of stimuli was reduced on average by 85.2% and 88.9% by the second day for the male and female leopard, respectively. By the third day, no stimuli were investigated or interacted with by either individual. The total number of minutes leopards interacted with each of the stimuli is presented in Table 3.

Table 3

*Total Number of Minutes Leopards Spent Interacting with Olfactory Stimuli for Each Day Stimuli were Present on Exhibit.*

Olfactory Stimulus	Day 1		Day 2		Day 3	
	Male	Female	Male	Female	Male	Female
Hay	33.82	12.22	0.27	0.87	0.00	0.00
Hay - pig scent	15.17	7.48	0.33	0.33	0.00	0.00
Hay - tiger scent	24.67	6.73	10.22	1.47	-	-

The proportions of expressed active behaviors displayed by leopards over the course of the study (including exploration, pacing, interacting with foliage, and scent marking) were compared to determine effects of olfactory stimuli on the focal animals' behaviors (Table 4). Other active behaviors listed on the ethogram (drinking, claw sharpening, affiliative behaviors, urinating or defecating) were not compared because each of these behaviors was expressed in low proportions per day. Bonferroni corrections were not implemented because each Wilcoxon Rank-Sum test was independent of the others, and no overall conclusion was drawn on the basis of any single result (i.e., a significant result in one would not suggest an overall effectiveness of a stimulus). Pacing significantly decreased in Nikolai with the use of hay, but no other behavioral effects were found with this individual. Pig-scented hay increased exploratory behavior by 4.83% for Katya; however, this stimulus had no other significant behavioral effects. Hay with tiger scent increased interacting with foliage and scent marking for Katya, and hay alone had no significant effects on her behavior.

Table 4

*Proportion of Time Per Day Individuals Spent Expressing Active Behaviors both Before and After Stimuli were Given, with Statistical Significance of Each Behavior for Each Stimulus.*

Behavior	Gender	Stimuli	Mean (before)	Mean (after)	Z-Score	Significance
Exploration	Male	Hay, pig scent	30.05%	34.43%	0.523	0.601
"	"	Hay, tiger scent	37.78%	40.35%	1.041	0.298
"	"	Hay	38.41%	27.36%	-1.336	0.182
"	Female	Hay, pig scent	18.15%	22.98%	2.684	0.007 *
"	"	Hay, tiger scent	37.23%	40.79%	0.759	0.448
"	"	Hay	31.05%	42.33%	1.72	0.086

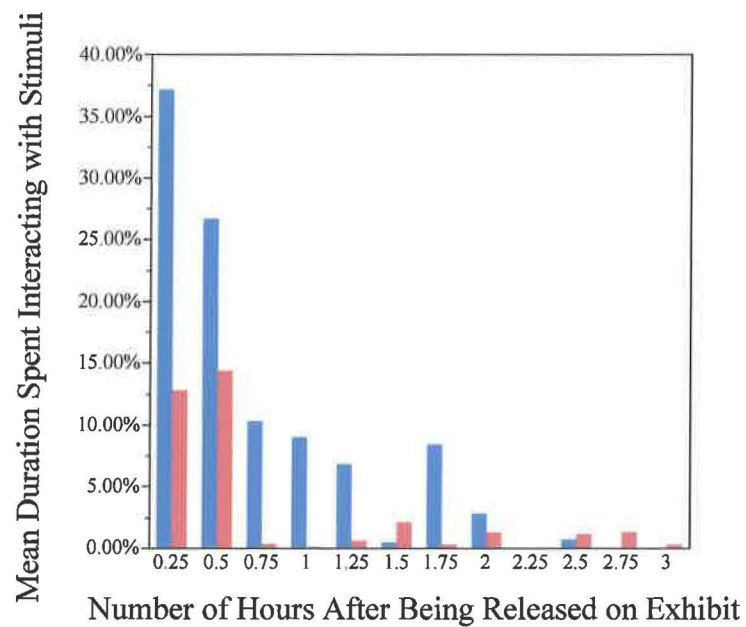
Table 4 (continued).

Pacing	Male	Hay, pig scent	14.32%	12.24%	-0.56	0.575
"	"	Hay, tiger scent	10.27%	4.54%	-0.721	0.471
"	"	Hay	28.71%	19.54%	-1.994	0.046 *
"	Female	Hay, pig scent	3.06%	7.25%	1.728	0.084
"	"	Hay, tiger scent	6.62%	9.25%	1.185	0.236
"	"	Hay	11.91%	10.69%	-0.759	0.448
Interacting with foliage	Male	Hay, pig scent	2.74%	1.25%	-1.253	0.21
"	"	Hay, tiger scent	3.85%	5.74%	1.942	0.052
"	"	Hay	1.78%	2.82%	1.953	0.051
"	Female	Hay, pig scent	1.10%	2.39%	1.728	0.084
"	"	Hay, tiger scent	2.19%	4.64%	2.024	0.043 *
"	"	Hay	1.03%	1.88%	1.35	0.177
Scent marking	Male	Hay, pig scent	2.81%	1.74%	-1.234	0.217
"	"	Hay, tiger scent	2.51%	0.40%	0.158	0.874
"	"	Hay	2.21%	2.04%	0.87	0.384
"	Female	Hay, pig scent	0.51%	1.98%	1.114	0.265
"	"	Hay, tiger scent	0.49%	2.12%	2.033	0.042 *
"	"	Hay	0.69%	0.85%	0.095	0.924

Asterisks indicate that there was a significant difference between the duration of that behavior expressed before versus after the stimulus was introduced.

Individuals interacted with olfactory stimuli primarily within the first three hours on exhibit (Figure 5). Combining each of the olfactory stimuli, Nikolai interacted with olfactory stimuli for a total of 73.66 minutes, 76.49% of that time was within the first two hours after he was released. Katya interacted with olfactory stimuli for only 26.43 minutes, with 81.13% of that being within the first three hours. The highest concentration of use for both individuals was within the first 30 minutes (45.49% total use, Nikolai; 53.47% total use, Katya).





*Figure 5.* Mean percentage of use of olfactory stimuli used per day for the first three hours leopards were released on exhibit. Nikolai (male) shown in blue and Katya (female) shown in red.

Considering that both individuals interacted with olfactory stimuli over the first three hours, I also compared the proportions of exhibited active behaviors before versus after stimuli, including only data collected within the first three hours animals were on exhibit. I found no significant difference between proportions of pacing before and after stimuli were given during those times for either individual, with any of the stimuli (Table 5).

Table 5

*Proportion of Time Individuals Spent Pacing Over the First Three Hours Released on Exhibit both Before and After Olfactory Stimuli were Given, with Significance Listed for Each Stimulus.*

Behavior	Gender	Stimuli	Mean (before)	Mean (after)	Z-Score	Significance
Pacing	Male	Hay, pig scent	17.97%	20.66%	0.212	0.832
"	"	Hay, tiger scent	19.98%	6.35%	-1.069	0.285
"	"	Hay	22.53%	30.01%	0.469	0.639
"	Female	Hay, pig scent	3.70%	1.15%	-0.698	0.486
"	"	Hay, tiger scent	3.83%	4.49%	0.977	0.328
"	"	Hay	5.12%	1.70%	-0.652	0.514

### Discussion

Leopards interacted with each of the olfactory stimuli in varying periods of time; however, use was primarily the first two hours on the first day (Table 3; Figure 5). Similar studies have also shown short-term use of enrichment (Skibieli, Trevino, & Naugher, 2007; Yu et al., 2009) yet to our knowledge, none have shown such a short time before apparent habituation. Skibieli and others (2007) observed decreased frequencies of pacing when using several spices as enrichment for felines; however, no significant difference in pacing was found a week after enrichment was provided. Yu et al. (2009) observed frequencies of behaviors exhibited by leopards after introducing olfactory enrichment and found negligible effects after four days of use. Habituation to novel stimuli has been observed among several taxa (Anderson, Arun, & Jensen, 2010; Kuczaj et al., 2002) and we expect that leopards are no exception; however, observing individuals rarely interacting with stimuli on the second day it was implemented suggests

that habituation to some types of stimuli could occur in extremely short durations. This habituation could be due to several reasons (e.g., type of stimulus, time stimulus was given, duration of stimulus on exhibit) and shows the variation that exists in observed habituation rates. To the contrary, a study by Swaisgood and others (2001) observed no habituation with respect to five enrichment items when given to giant pandas over 15 sessions in a 2.5-month period. These treatments, however, were only kept on exhibit for a single day, then provided again on a later date randomly within the 2.5-month period. We should also note that, with respect to this study, interaction with a single stimulus did not necessarily correlate to observed behavioral changes. Hay, acting as our control, had the most observed interaction time for both Nikolai and Katya. This stimulus reduced observed proportions of pacing behavior with respect to Nikolai, yet it was the only stimulus to have no behavioral effects on Katya.

No single stimulus presented to Nikolai promoted species-typical behavior, and only one treatment (hay as a control) decreased pacing. This control treatment was not the first treatment given to leopards, meaning that the novel effects of hay alone should not have had any behavioral effects. Nikolai interacted with this treatment longer than the others (Table 3), and given that no other scents were added, he could have been investigating to determine if any minute traces of scents were within the hay. Of the recent olfactory-related enrichment studies conducted with felines (Powell, 1995; Skibieli et al., 2007; Yu et al., 2009; Quirke & O’Riordan, 2011), none used hay as a medium for presenting stimuli. One brief report, however, suggested that straw could be used as a form of environmental enrichment for Wolf’s guenons, increasing observed frequencies of affiliative behavior (Fuller et al., 2010), and a burlap sack with straw as well as other

types of enrichment decreased pacing in giant pandas (Swaisgood et al., 2001). It is our hope that as future studies are conducted relating to environmental enrichment with felines, we will be able to more accurately determine what additional effects hay could have as a form of enrichment.

While no significant effects were observed with Nikolai in response to predator and prey-scented hays, these treatments had significant effects on modifying Katya's species-typical behavior. Exploration increased with pig-scented hay, and interacting with foliage as well as scent marking increased with tiger-scented hay. These results are similar to those of a recent study where exploratory behaviors and scent marking increased with the scents of tiger urine and roe deer feces on towels placed in a leopard exhibit (Yu et al., 2009). These treatments, however, did not modify frequencies of observed pacing, and the behavioral changes that did occur were short-lived. Such short-term use by the leopards of the chosen stimuli could suggest that a single olfactory stimulus presented may not promote the behavioral responses we desire. The use of olfactory stimuli in addition to a randomized feeding schedule and spatial variability with respect to feeding location reduced pacing and increased exploratory behavior in cheetahs (Quirke & O'Riordan, 2011), and multiple scents simultaneously used with felines reduced observed frequencies of pacing (Skibieli et al., 2007).

This study did not analyze differences in frequencies of species-typical and atypical behaviors off exhibit, which was shown to have increased proportions of pacing with lions (Bashaw et al., 2003). While we were not able to measure proportions of behaviors expressed at night, this study was able to further understand the daily behavior of captive leopards and how those behaviors can be affected by environmental

enrichment. Overall, these findings suggest that observed stimuli could be appropriate enrichment tools in promoting the welfare of captive leopards, but for a greater effect, olfactory stimuli should be simultaneously implemented with additional forms of enrichment and should only be used for short durations. If possible, zoos should provide intended enrichment stimuli to individuals for no longer than a few hours to a single day, minimizing the potential for habituation, and ensuring that the enrichment stimuli zoos have at their disposal are providing their greatest potential.

CHAPTER IV  
EXHIBIT USE BY CAPTIVE AMUR LEOPARDS  
IN A NEWLY RENOVATED EXHIBIT

Introduction

Over the past decades, zoos have been renovating their exhibits to look more natural, increasing the amount of soil and vegetation within their enclosures. Naturalistic exhibits not only increase visitor interest and attention (Davey, 2006), but they also act as their own form of environmental enrichment for the animals they house (Shepherdson, 2003). This transition has certainly introduced new concerns for zoo personnel, as larger, naturalistic exhibits tend to make observing individuals closely more difficult for zoo staff (Kawata, 2012). As zoos make these modifications, they have also begun to evaluate the use of exhibit space for several taxa within their collection (e.g. Lyons, et al., 1997; Leighty et al., 2010; Miller, Leighty et al., 2011; Blowers, Waterman, Kuhar, & Bettinger, 2012), and are now understanding that larger exhibits do not necessarily result in better welfare (Kawata, 2012). Larger exhibits typically result in mixed-species exhibits, which may have negative consequences for some species, may only be designed with the focal species in mind, and may not have a greater enrichment potential. A study by Lyons et al. (1997) showed that only half of a larger exhibit was used by felines, and raised surfaces and exhibit edges were the primary areas used by the individuals being housed. In this case, stereotypic pacing had no correlation to exhibit size, with approximately 79% of cats exhibiting pacing, regardless of available space. When increasing the size of an outdoor exhibit for captive spoonbills at Disney's Animal Kingdom®, keepers were soon faced with the problem of having wild migrants land, and

ultimately outcompete the captive animals for food (Kawata, 2012). Elephants at Disney's Animal Kingdom® have been shown to use exhibit space differently depending on individuals' social status within the group (Leighty et al., 2010), with the dominant individuals covering more land and possibly restricting use of the less dominant herd members.

These studies suggest that merely implementing larger exhibits for captive individuals does not necessarily equate to better welfare for the animals occupying them. The focus has now been placed on creating a habitat with characteristics similar to the habitat of specific species occupying it, or creating habitat that elicits behaviors that the animal would normally exhibit in the wild. Studies examining preference of exhibit features have been conducted with several species. Meller, Croney, and Shepherdson (2007) found that rubberized flooring in an Asian elephant exhibit increased patterns of behavior similar to their wild counterparts, even though individuals were not found to use this area more frequently after renovation. Renner and Lussier (2002) showed that providing climbing structures as environmental enrichment increased the use of other structures already in place in a spectacled bear exhibit.

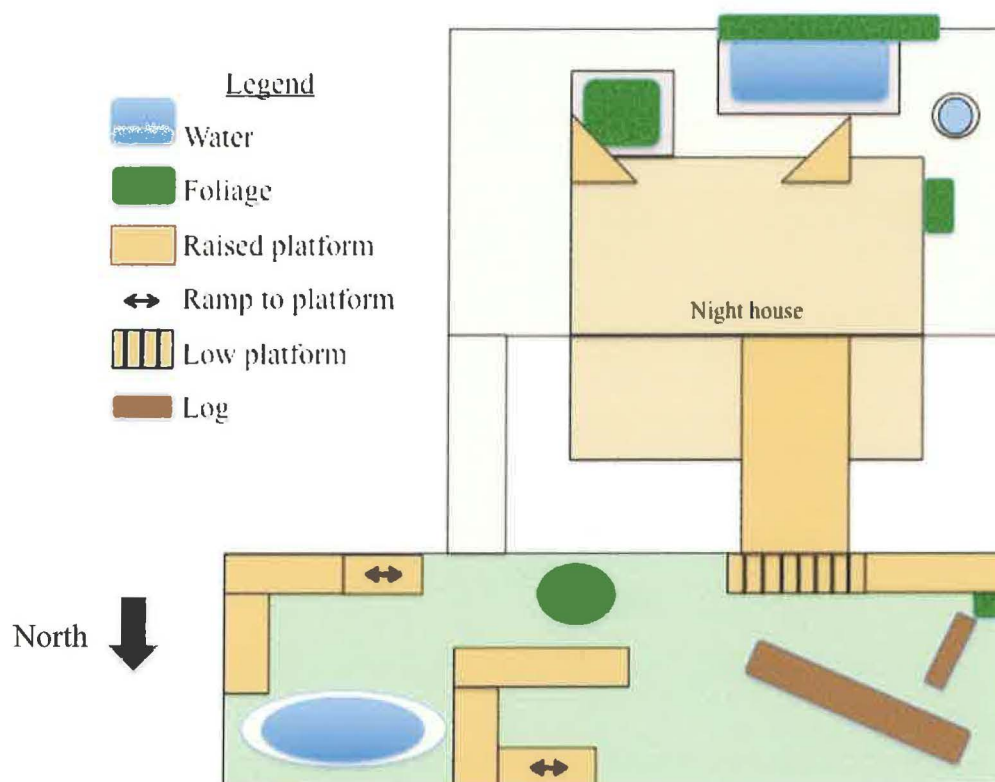
To further understand the potential choices made by leopards at JZP, we observed their two individuals while on exhibit and recorded which section of the exhibit was being most frequently used. Due to upcoming renovations, staff relocated the two leopards, just before this study, into two previously separated exhibits now connected by two catwalks. The two exhibits were approximately equal in size, but different in substrate (concrete vs. soil), number of resting platforms, water availability, visitor attendance, and vegetation. While we recognize that there are several differences

between the two exhibits that are now connected, preferential use of one section may help us better understand exhibit space selection among leopards.

### Methodology

Both leopards were transferred to a new exhibit in November 2011 and first given access on 8 November 2011. This exhibit (Figure 6) was previously two separate enclosures approximately equal in size, now joined by two catwalks. The total area of the combined exhibit is 177.3 m<sup>2</sup> (north section, 90.5 m<sup>2</sup>; south section, 68.1 m<sup>2</sup>; and 18.7 m<sup>2</sup> of connecting catwalks). The north section of the exhibit has four wooden platforms for climbing, one Crape Myrtle (*Lagerstroemia* sp.), one 14.5 m<sup>2</sup> pool approximately 1.5 m deep, fallen logs, small forbs, and a soil substrate with grass. The south section has two wooden platforms, a roof section of the night enclosure to climb on, one 13 m<sup>2</sup> shallow pool, one concrete water bowl, small forbs, and a concrete floor.





*Figure 6.* Aerial view rendering of the 177.3 m<sup>2</sup> Amur leopard exhibit at Jackson Zoological Park, Jackson, Mississippi. North section colored green to represent soil and grass, whereas the south section is colored gray to depict concrete flooring. Leopards had access to both sections by using two catwalks, one at ground level and the other above the night house.

As recorded video was being scored using our ethogram (Table 1), leopards were identified as either residing in the north or south section of the exhibit (i.e., we calculated duration of time spent in either section over the course of each day). Both catwalks, the corridors between the north and south sections, were considered as a part of the north section. The proportions of time spent in both north and south sections were then calculated for the thirty-three, 15-minute increments described in Chapter II. Data were also combined to determine overall proportion of time each leopard spent in the corresponding sides by day (i.e., combining data so that each day was a replicate). To

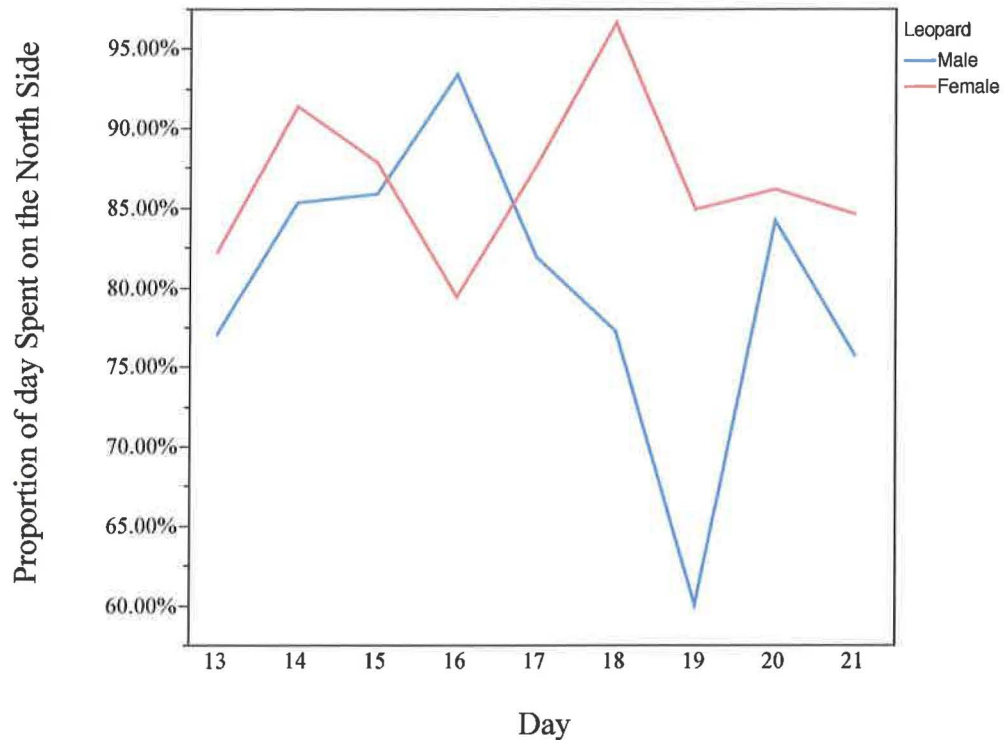
determine whether leopards spent a significantly different proportion of time on one side, we performed a Wilcoxon Rank Sum test for each leopard to determine if there was a significant difference between the proportions of time spent on each side per day.

Similarly, a Kruskal-Wallis test was used to analyze significant differences in usage of the north side, among times-of-day (with data pooled over all nine days). Statistical tests were conducted using *JMP 10* software (SAS Institute).

### Results

Leopards were recorded spending the majority of their time on the north section of the exhibit over the nine days we observed exhibit use ( $m$ : 79.95%  $\pm$  28.46% *SD*, Nikolai;  $m$ : 86.76%  $\pm$  16.73% *SD*, Katya). While there was considerable day-to-day variability (Figure 7), neither leopard spent a majority of his/her time on the south portion of the exhibit on any of the observed days. We found significant variability with respect to the amount of time Nikolai and Katya used the north side compared to the south side of their exhibit (Nikolai:  $H = 23.181$ ,  $df = 8$ ,  $P = 0.031$ ; Katya:  $H = 30.571$ ,  $df = 8$ ,  $P = 0.002$ ). The proportion of time Nikolai spent occupying the north section varied between 93.35% (Day 16) and 60.01% (Day 19), and a nonparametric Wilcoxon each pair post-hoc analysis showed that his lowest proportion on Day 19 was significantly lower than six other days (14, 15, 16, 17, 18, & 20) and that Day 16 was significantly higher than the remaining four days (13, 17, 19, & 21). Katya's variability was similar to Nikolai's day-by-day proportions, varying between 96.61% (Day 18) and 79.38% (Day 16); however, the days on which she used the north section proportionally less were different than Nikolai's (Days 13 and 16). The proportion of time she spent on the north section was

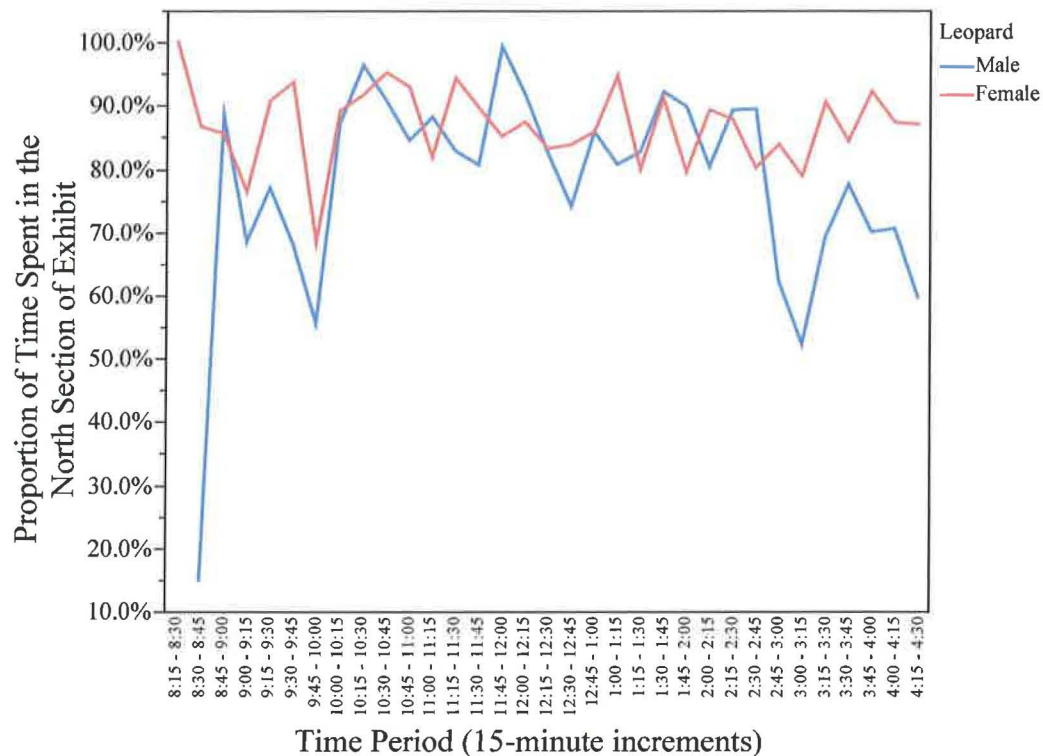
also significantly higher on Day 18 than all other days observed with the exception of one, Day 14.



*Figure 7.* Proportion of time leopards spent on the north section of the exhibit per day for the last nine days animals were observed on exhibit. Nikolai (male) shown in blue and Katya (female) shown in red.

With respect to time-of-day, there was no significant difference in proportion of Katya's exhibit use among the thirty-three, 15-minute increments ( $H = 37.045$ ,  $df = 32$ ,  $P = 0.2474$ ). A Kruskal-Wallis test determined that there was a significant difference in proportion of Nikolai's exhibit use among the thirty-three, 15-minute increments ( $H = 46.478$ ,  $df = 31$ ,  $P = 0.0366$ ), with the majority of time-increments in the late afternoon being proportionally spread between the north and south sides than values during the middle of the day (i.e., Nikolai remained on the north side for significantly longer periods of time during the middle of the day than the late afternoon). The lowest proportion of

time spent in the north section was 14.88% (0830–0845), but this proportion was observed only on one day, given that Nikolai was typically released shortly after Katya and not yet on exhibit during this time. All other proportions were cumulated over days where Nikolai was in view and varied between 52.22% and 99.26%.



*Figure 8.* Mean proportion of time leopards spent on the North section of the exhibit throughout the day divided into thirty-three 15-minute increments. Nikolai (male) shown in blue and Katya (female) shown in red.

## Discussion

Both leopards at JZP were selecting the north section of their enclosure more frequently than the south section. These sections have a multitude of differences between them, including but not limited to substrate, type of climbing structures, drinking-water locations, and foliage. This study did not quantify which objects (boardwalks, drinking-water locations, trees) or areas (central versus perimeter edges) were selected more by

both leopards; however, the choice of materials on either side is different, and leopards are selecting one area more frequently, using, on average, 48.96% of their exhibit only 16.65% of the time. Considering that space is always of principal concern in zoological institutions, these comparisons are vital in determining whether captive animals are appropriately using the facilities in which they are housed.

While both sides were similar in the amount of space available, the area used less frequently (south section) had features that would define it as more of a traditionally-styled exhibit, with the north section having more naturalistic design containing more foliage and platforms. Several studies have shown significant differences between these types of exhibits (e.g. Fàbregas, et al., 2009; Little and Sommer, 2002) noting that individuals have decreased proportions of stereotypical behaviors in naturalistic exhibits. Given that our individuals spent such little time on the south section, and that data collected for this project were not designed to specifically assess behavioral patterns in these areas, we were not able to quantitatively determine if species-typical or atypical behaviors occurred more frequently in specific areas. Both leopards, however, were recorded pacing in both sections as well as exploring and interacting with exhibit features. Nikolai, in particular, was most frequently observed pacing in one section of the north portion of the enclosure; however, several factors could have contributed to him pacing in this location (i.e., primarily pacing in the north section does not necessarily suggest that the north section is the cause of the activity). While our observations suggest that the naturalistic section of the leopards' enclosure was used more frequently, we must also recognize that classifying an exhibit as natural may not necessarily be more enriching to an individual (Kawata, 2011). In some cases, introducing items as part of

the exhibit that may be traditional could promote species-typical behaviors and may be less expensive for zoos to implement (i.e., bars as the physical barrier that could promote climbing behaviors in primates as opposed to a moat).

Proportions of time observed in the north section of the enclosure remained similar with respect to time-of-day, with a bimodal trend for Nikolai of movement between sections in the morning and afternoon (Figure 8). Activity periods with respect to pacing were also bimodal (Figure 3), occurring at roughly the same time each day, and similar patterns have been observed in smaller felids at other institutions (Moreira et al., 2007). Both individuals rested the majority of their day (Table 2) and typically rested in the north section of the enclosure, which could explain higher observed proportions of time spent there. Nikolai's increased use of the south section during the morning and afternoon could be due to the increased proportions of active behaviors also during those times.

The amount of space a captive animal is given should certainly be addressed when designing a new exhibit so that the exhibit is appropriate for the focal animal. Proportions of stereotypic behaviors expressed in captivity have been shown to relate to wild home ranges and the average daily amount of space a carnivore covers (Clubb & Mason, 2007); however, space should not be the only concern. For the quality and use of an animal's exhibit to be appropriately addressed, captive institutions should understand that individuals housed are selecting which areas of an exhibit to use, meaning that, like enrichment, this selection can be short-lived. This concept of quality is certainly not new, dating back one of the principal founders of our concept of an open, naturalistic exhibit: "The amount of space at the animal's disposal has had too much attention while

the quality of the space on the other hand has received too little” (Hediger, 1964).

Ultimately, if we are to ensure that the welfare of our captive animals is always maintained at its highest potential, we must understand that an effective enclosure design utilizing our knowledge of each individual’s wild and captive behaviors is paramount.

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