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Treatment Analysis of a Captive Male Jaguar (Panthera onca)

Megan Colleen Morris
University of North Florida

Treatment Analysis of a Captive Male Jaguar (*Panthera onca*)

Megan Colleen Morris

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Megan Colleen Morris

University of North Florida
The Graduate School

This is to certify that the thesis prepared:

By: Megan C. Morris
Entitled: A Treatment Analysis of a Captive Male Jaguar (*Panthera onca*)

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Signed by the final examining committee

____ Date: _________________
Chair of Thesis Committee

____ Date: _________________
Second Reader, Thesis Committee

____ Date: _________________
Third Reader, Thesis Committee

Approved by

____ Date: _________________
Dr. Lori Lange
Chair, Department of Psychology

____ Date: _________________
Dr. George Rainbolt
Dean, College of Arts and Science

____ Date: _________________
Dr. Len Roberson
Dean of the Graduate School

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Abstract

Large carnivores in human care have been reported to engage in stereotypic behaviors. Such behavior is thought to be correlated with high stress levels, in part, due to captive environments limiting opportunities for functional consequences and environmental stimuli. Moreover, there are several arguments stating that stereotypic behaviors can be indicative of poor welfare, as they can often have severe negative emotional and physical effects on the animal. The first portion of this study included a five-phase treatment analysis which evaluated whether environmental manipulations decrease the frequency of stereotypic behaviors including pacing, over-grooming and tail-sucking exhibited by a single male jaguar housed at Jacksonville Zoo and Gardens. The aim of the present study was to evaluate possible environmental variables that were reported by staff as likely variables maintaining or promoting stereotypic behavior. Data collection occurred during a 10-week evaluation and followed an ABCAD reversal design. Both behavioral data and fecal glucocorticoid metabolite levels were analyzed. The second portion of this analysis included a multi-institutional survey to assess the prevalence of the stereotypic behaviors exhibited by zoo-housed jaguars in North American AZA-accredited institutions. Results from the behavioral assessment revealed a decrease in stereotypic behaviors with the implementation of treatment conditions. Hormone analyses revealed that stereotypic pacing is not presently correlated with higher stress levels for this animal. Finally, survey results revealed that a significant portion of the North American jaguar population engages in stereotypic behaviors. Further analyses are necessary to identify potential patterns or environmental predictors for the development of stereotypic behaviors.

Treatment Analysis of a Captive Male Jaguar (*Panthera onca*)

The jaguar (*Panthera onca*) is the third largest species of felid (Johnson & Ivy, 2016) and the largest cat species of the Americas (Nowell & Jackson, 1996). Historically, the jaguar was known to range from the southern United States to as far as the Argentinean Patagonia and throughout Central and South America (Eizirek et al., 2001). However, a 2002 survey indicated that jaguars can be found in only 46% of its original range (Sanderson et al., 2002). Jaguars are currently listed as "near threatened" by the International Union of Conservation of Nature Redlist of threatened species (Caso et al., 2008). In 1973, jaguars were listed in Appendix I of the Convention on International Trade of Endangered Species of Wild Fauna and Flora (CITES, 2017), meaning they are of the most endangered animals listed by this organization. As a species listed in Appendix I, jaguars have the highest level of regulation and protection from over-exploitation (CITES, 2017). Wild jaguar populations have declined due to habitat fragmentation, deforestation, poaching, and human conflict. Frequent attacks on livestock has led to persecution by local ranchers and increased species vulnerability. Zoo-housed animals are often considered ambassadors for their wild counterparts. For this reason, the management and welfare of zoo-housed jaguars is of increasing importance.

Wild jaguars currently subsist in fragmented populations (Eizirek et al., 2001). While the jaguar typically associates with bodies of water, habitats range in both terrain and elevation. This species has been known to inhabit rainforests, thorn scrub woodland, dry forests, savannahs, and areas heavily covered by swamps (Caso et al., 2008; Nowell & Jackson, 1996). Although they typically reside in lowlands of below 1,000 meters, jaguars inhabit a range of elevations and can occupy expanses as high as 3,000 meters (Brown & Lopez Gonzalez, 2001; Caso et al., 2008.) Their broad range in habitat is perhaps why there have previously been eight distinguished subspecies of jaguar. Today, however, the jaguar is considered monotypic despite variations in body size, skull shape, and diets (Eizirek et al., 2001; Pocock, 1939). Due to these variations, information about wild groups or individuals cannot be recognized as absolute for the species, and there is still much to learn about the physical and behavioral needs of both wild and captive populations (Johnson & Ivy, 2016).

Although the population was unstable until the 1930s, jaguars have been housed in zoological institutions since 1885 (Mechak, Johnson, & Goff, 2016). A 2016 analysis of the Association of Zoos and Aquariums (AZA) captive jaguar population reported 109 animals (52 males, 57 females) currently managed in human care. This population is distributed among 49 AZA-accredited institutions (Johnson & Ivy, 2016). Jaguars are considered a premier cat species in terms of exhibition in zoological institutions (Johnson & Ivy, 2016). They are also considered a conservation icon in that efforts to educate the public about current threats to wild populations and their environment have also extended to conservation efforts for other species throughout the jaguar's home range (Johnson & Ivy, 2016). As such, the jaguar is of particular importance as an ambassador species.

Missions of Animal Care Facilities

Whereas new animal welfare initiatives are on the rise in zoos and aquariums, much of what is known about animal welfare science stems from the agricultural industry (Melfi, 2009). Unsurprisingly, zoo animals differ from domesticated animals in terms of their psychological and physiological needs (Carlstead & Shepherdson, 2000). Unlike farm animals, animals housed in zoos and aquariums are bred in conjunction with the fundamental conservation mission held by many modern zoos: to maintain genetic diversity with the goal of reintroducing individuals into the wild. As such, zoo-housed animals are bred to behave similarly to their wild counterparts rather than adapting to captive conditions (Carlstead & Shepherdson, 2000). Inability to cope with captive environments can lead to unsuccessful breeding attempts and the exhibition of species-atypical behavior (DeCaluwe, Wielebnowski, Howard, Pelican, & Ottinger, 2013; Wingfield et al., 1998).

In addition to a conservation-based mission, Maple (2016) discusses a recent shift in zoological missions: conservation first, animal welfare second. Maple further discusses the necessary shift toward the adoption of an empirical approach to zoological management. Essentially, zoological institutions with an empirical approach strive to make management and husbandry decisions based on systematic observations and concrete data rather than intuition of the animal care staff. Since the shift towards a conservation focus in zoological institutions in the past few decades (Anderson, 2003), efforts have been made to incorporate science into management practices and conservation initiatives (Hopper, 2017).

Empirical Goals of Management and Conservation of Species in Zoos

Behavioral observation and recording provides an empirical approach that allows for the quantification of behavioral events (Powell, Martindale, & Kulp, 1975). Though informal observations made by animal care staff are helpful in basic husbandry practices, formal behavioral observations are essential in assessing captive animal welfare (Jauhiainen & Korhonen, 2005). As zoological institutions shift towards an empirical approach, quantification of behavior will be a necessary characteristic of zoo management (Crockett & Ha, 2010). In addition to providing information about an individual animal's current state of welfare, data can increase knowledge on the basic biology of the species in question and transfer to in situ conservation efforts (Crockett & Ha, 2010). According to Broom (2010), observation of behavior provides a means to interpret physiological measures and is of value in welfare assessments. For example, just as abnormal repetitive behaviors and stereotypies can be an indicator of poor welfare, many behaviors can be indicative of good welfare (e.g., play, engaging in positive reinforcement training) (Melfi & Hosey, 2011). However, welfare must be measured on an individual level in addition to focusing on groups or species. Maple (2014) asserts the necessity of elevating animal welfare standards to include defining what it means to thrive for each and every animal housed in human care. Maple further describes thriving in similar terms to Webster's (2008) characterization of animal welfare, which he defines as "living a natural life, being fit and healthy, and being happy" (Maple, 2014; Webster, 2008). Behavioral observations will aid researchers in answering questions to help provide animals opportunities to reach a level of thriving and obtain the optimum level of wellness.

A new empirical approach to zoo management has also given rise to the use of applied behavior analysis in zoos and aquariums. Applied behavior analysis (ABA) is the science of "systematically applying interventions based upon the principles of learning theory to improve socially significant behaviors to a meaningful degree" (Baer, Wolf, & Risley, 1968). Best known for its use in treating humans with developmental disabilities, ABA has recently been become more prevalent in the study of non-human animal behavior, specifically, animals housed in captive environments (Maple & Segura, 2014). In recent decades, zoos have seen significant growth in research opportunities, as such research offers chances to answer questions regarding breeding, management, biological, and behavioral questions of zoo-housed animals (Hosey, 1997; Macdonald & Hofer, 2011). Zoo animal research often focuses on the relationship between animals, their behavior, and their environment making ABA an important tool for ensuring the psychological well-being of zoo-housed animals (Maple & Segura, 2014). Data collected from formal behavioral observations and analyses can assist in developing treatment conditions aimed at reducing problem behaviors or produce alternative species-appropriate behaviors (Farmer-Dougnan, 2014).

Due to the recent emphasis on defining thriving for individual animals rather than a population or species as a whole, single subject research design is also gaining traction across zoos and aquariums. Single subject research is a principle of ABA that applies behavior-change strategies to individuals or small-n populations, rather than focusing on large groups with the goal of generalizing across populations. (Alligood, Dorey, Mehrkam, & Leighty, 2017). There is an inherent logistical challenge when conducting research on zoo animal populations as a result of generally small sample sizes (Kuhar, 2006). As such, inferential statistics are not often feasible for use in zoo research (Saudargas & Drummer, 1996). Multi-institutional research studies have typically been used to address this issue, with methods that include the use of cross-species analyses (Alligood et al., 2017; Clubb & Mason, 2004; Shepherdson, Carlstead, & Wielebnowski, 2004).

Though there is a long history of single subject design in many areas of the psychological sciences (e.g., Kazdin, 2003; Lenz, 2015), single subject research design has seen much opposition (Alligood et al., 2017; Bloomsmith, Marr, Maple, 2007). Critics question the ability to derive accurate information using inferential statistics with small-n studies due to possible inadequate statistical power necessary to make inferences about the reliability of behavioral observations based on statistical significance (Alligood et al., 2017) and inability to infer external validity or generalize across populations (Swaisgood & Shepherdson, 2005). However, Kazdin (2003) discusses methods to alleviate these concerns by highlighting experimental control using repeated measures, continuous measurement, baseline assessments, and comparisons within and across conditions or phases. The goal of single subject research design is to acquire evidence of behavior change as it relates to environmental states or events. In addition to using single subject research design in assessing general questions regarding animal well-being, many experts detail the value of using such methods to evaluate enrichment (e.g., Alligood et al., 2017; Swaisgood & Shepherdson, 2005). The practice of environmental enrichment is common in animal husbandry protocols and will be discussed more in depth later.

Functional behavior assessments (FBA) are commonly used in ABA to evaluate problem behavior in both humans and non-human animals. Friedman (2007) describes a FBA as the initial means of assessing “what’s going on and why,” which she argues is essential for developing intervention and treatment plans. FBAs have previously been used in schools in the form of both indirect and direct measures. Indirect measures typically involve interviews or rating scales, whereas direct measurement require observation of the behavior being measured (Borgmeier, Loman, Hara, & Rodriguez, 2015; Scott & Kamps, 2007; Lewis, Mitchell, Harvey, Green, & McKenzie, 2015). This information is used to evaluate conditions under which the behavior in question occurs and what conditions are hypothesized as maintaining the behavior (Friedman, 2007). FBAs involve four steps as described by Friedman (2007); describing the problem behavior; identifying environmental conditions under which the problem occurs and does not occur; understanding the purpose behind the behavior, including consequences that maintain the behavior; developing a treatment or intervention plan to eliminate or redirect the behavior. Because ABA combines experimental design and behavioral observations, it is effective in providing a framework for addressing problem behaviors in captive animals (Forthman & Ogden, 1992).

Problem Behaviors in Zoo-Housed Animals

Problem behavior, such as stereotypic and abnormal repetitive behaviors (ARBs), in zoo-housed animals is not uncommon and has been documented even in the earliest zoological institutions (Melfie, 2009). Stereotypic behavior is understood as a repeated behavior pattern with no obvious function or goal (Carlstead & Seidensticker, 1991; Lyons, Young, & Deag, 1997). More recently, studies suggest these ARBs/stereotypic behaviors represent aversive conditions and can be indicative of poor welfare (e.g., Broom, 1983; Mason, Clubb, Latham, & Vickery, 2007; Shyne, 2006). For example, Mason and colleagues suggest that stereotypic behavior is a broad term that not only encompasses repetitive behavior, but repetitive behavior that is induced by frustration, repeated attempts to cope, or C.N.S. dysfunction (Mason et al., 2007). Such behavior is also thought to be correlated with high stress levels due, in part, to captive environments limiting opportunities for functional consequences and exposure to environmental stimuli (Lyons et al., 1997). For example, stereotypic behaviors can develop when an animal is motivated yet unable to perform a particular behavior (Mason, 1991). Wooster (1997) describes notable contributors to the establishment of stereotypic behaviors to include structured husbandry routines, proximity to conspecifics, impoverished environment, and visitor densities, among other environmental factors associated with captivity.

Large carnivores are particularly known to engage in stereotypic behaviors. Some argue that this is principally due to limitations associated with feeding behaviors (McPhee, 2002) and inability to range (Kroshko et al., 2016). Historically, carnivores vary greatly in their ability to cope with captive conditions. Kroshko and colleagues discussed great variation in the prevalence of stereotypic behaviors and breeding capacity of carnivores, and they suggested that these negative responses stem from stress (Kroshko et al., 2016). Furthermore, Mason and colleagues proposed intelligence as a risk factor for stress-related behaviors, suggesting that species with a higher intelligence level when compared to less intelligent species were less likely to adjust appropriately to captivity and had an increased risk of stereotypic behaviors due to boredom and frustration (Mason et al., 2013).

Problem behaviors in felids typically include pacing, head-swinging or bobbing, excessive grooming, and self-mutilation (especially tail-sucking or chewing), and long periods of inactivity (Clubb & Mason, 2007; Markowitz, Aday, & Gavazzi, 1995; Tudge, 1991). Wild felids spend significant amounts of time exhibiting appetitive behaviors including locating, capturing, killing, and consuming prey (Bashaw Bloomsmith, Marr, & Maple, 2003; Lindburg, 1988). With scheduled meals and strict diets, captive conditions can limit opportunities to engage in such appetitive behaviors (Bashaw et al., 2003). Historically, problem behaviors and other welfare concerns have been dealt with by implementing training and enrichment programs as well as naturalistic exhibit design (Law, Macdonald, & Reid, 1997; McPhee, 2002).

Pacing is the most common stereotypic behavior seen in captive carnivores (Mason et al., 2007) and perhaps the most recognized stereotypic behavior by the general public (Clubb & Vickery, 2006). Pacing has not only been shown to have negative emotional effects on individuals engaging in this behavior, but there are also negative physical effects of pacing including hair loss, abrasions, and sores (Morris, 1964). Experts suggest this behavior arises from hunting limitations (Clubb & Mason, 2007) or unsuccessful attempts at escape (Clubb & Vickery, 2006). Some studies have also shown that pacing heightens just before scheduled feedings (Clubb & Vickery, 2006; Kroshko et al., 2016). Furthermore, bouts coinciding with feeding times were longer in duration than other pacing bouts such as those associated with keeper activity (Mallapur & Chellum, 2002). Thus, pacing has been considered a redirection of appetitive behaviors unable to be performed in captivity (Carlstead & Shepherdson, 1991). New environmental enrichment aimed at promoting hunting behaviors have typically been used to reduce instances of stereotypic pacing (Mallapur & Cheullum, 2002), but this behavior remains prevalent in many zoo-housed carnivores.

Self-injurious behavior (SIB), including tail sucking or biting, is also a well-documented stereotypic behavior (Bloomsmith et al., 2007; Wielebnowski, Fletchall, Carlstead, Busso, & Brown, 2002). In a 2006 study, Hope and Deem assessed the morbidity and mortality of captive jaguars from 1982-2002 and found reported cases of tail sucking in all age classes of jaguars, including juveniles, adults, and geriatrics. Furthermore, mortality resulting from diseases of the integumentary system were present in greater than 20% of cases included in the study (Hope & Deem, 2006). Tail sucking can lead to alopecia and inflamed tail lesions thereby increasing opportunities for infection and decreasing disease resistance (Wielebnowski, et al., 2002). SIBs are thought to emerge as a coping mechanism and are indicative of a limiting or stressful environment (Carlstead, 1998). Interestingly, when compared to individuals who did not self-mutilate, clouded leopards engaging in tail sucking had higher corticoid concentrations, (Wielebnowski, et al., 2002). Given the potentially dangerous nature of SIBs, it is important that care givers determine possible environmental triggers and devise a treatment plan to decrease or eliminate these behaviors.

Over-grooming, or excessive licking, is also considered a stereotypic behavior (Tudge, 1991), though the parameters that constitute grooming as "excessive" have been debated (Mason et al., 2007). According to Novak and colleagues, a routine behavior can become a pathological behavior if its "frequency of occurrence disrupts basic biological functions or if it replaces other species-typical behaviors" (Novak, Meyer, Lutz & Tiefenbacher, 2006). Some researchers suggest that in such situations, grooming becomes a coping behavior and is, therefore, indicative of stress and poor welfare (Beisner & Isbell, 2009). In a 2009 study looking at factors influencing hair loss in captive rhesus macaques (*Macaca mulatta*), researchers suggested that over-grooming may arise as a result of redirected foraging behavior. Consequently, animals motivated but unable to perform foraging behaviors redirected species-typical motor patterns associated with foraging to grooming behaviors (Beisner & Isbell, 2009). Though the threshold for grooming to be considered excessive should be defined on both a species and individual level, exposed skin and lesions resulting from over-grooming prompt welfare concerns.

Stress-Hormone Analysis in Zoo-Housed Animals

Many problem behaviors are thought to stem from the stress of captive environments (Carlstead, 1998). Stress, as defined by Selye, is a "response to any stimulus that threatens or appears to threaten homeostasis of an individual (Selye, 1976). Attempts to adapt to this homeostatic imbalance involve changes behavior, activation of the adrenal medulla, and glucocorticoid release including corticosterone and cortisol (Dantzer & Mormède, 1983; Metrione & Harder, 2011; Moberg, 1991). In the past, stress has been measured using behavioral observations or post-mortem evaluations (von der Ohe & Servheen, 2002). Although experts agree that measuring stress cannot be limited to a single biomedical or behavioral measure (Mason & Mendl, 1993; Mostl & Palme, 2002; Wielebnowski, 2002;), measuring glucocorticoids using fecal samples offers a non-invasive approach to quantifying stress levels in non-human animals (von der Ohe & Servheen, 2002). Glucocorticoids can be measured using samples of blood, urine, saliva and feces (Beerda, Schilder, Janssen, & Mol, 1996; Möller, Wendt, & Waldmeier, 1991; von der Ohe & Servheen, 2002). As stress can have impacts on reproductive success, the prevalence of stereotypic behaviors, the development and decrease in disease resistance, and mortality (Carlstead & Shepherdson, 2000; Metrione & Harder, 2011;

von der Ohe & Servheen, 2002), it is important to routinely monitor each animal for signs of stress.

Purpose of the Present Study

Although stereotypic behaviors have been studied extensively, such behaviors continue to be a concern among zoo-housed animals. As zoos and aquariums continually strive to provide optimal care for their charges, it is necessary to implement evidence-based approaches to address problem behavior and reach a level of thriving for all individuals in their care. The goal of the present study was to empirically evaluate environmental components thought to be contributing to the engagement of stereotypic behaviors by 1.0 jaguar “Saban” housed at the Jacksonville Zoo and Gardens (JZG). The study subject had a history of engaging in stereotypic behaviors including pacing and tail sucking. In the past, both tail sucking and pacing have led to exposed skin and lesions. These behaviors were also believed to be correlated with high stress levels and therefore, indicative of poor welfare.

Assessments of environmental variables were accomplished by implementing principle components of applied behavior analysis including single subject and reversal design research methods. Careful consideration was taken when devising the methodology with the goal of making it easily adoptable by other zoological institutions. Too often animal caregivers employ the “kitchen sink” method to addressing problem behavior which can result in inconsistent or inaccurate conclusions regarding the effectiveness of treatments (Swaisgood & Shepherdson, 2005). Consistent with decreasing species-atypical behaviors, investigators hypothesized that species-typical behaviors, such as manipulation, investigative and patrolling behaviors would increase with the implementation of treatments. Fecal glucocorticoid levels were assessed in conjunction with analyzing any behavioral changes associated with the environmental manipulations to 1) determine if stereotypic behavior was at all correlated with stress levels and 2) measure any changes in cortisol levels across conditions.

The second portion of the present study aimed to evaluate the frequency of stereotypic pacing, tail-sucking, and over-grooming in jaguars housed in human care. This evaluation was created in the form of a survey distributed to AZA accredited institutions across North America. The goal of this report was two-fold. One goal was to determine the prevalence of stereotypic behaviors exhibited by jaguars in AZA institutions. The second goal was to determine if any environmental or ontogenetic factors were predictor variables among those individuals known to engage in stereotypic behaviors. Determining commonalities in individuals engaging in stereotypic behaviors could be used to inform husbandry and management practices and improve the welfare of the animals in our care.

Method

Overview

Jacksonville Zoo and Gardens (JZG) is home to five jaguars: 2 males and 3 females. Pacing is prevalent in all jaguars housed at the Jacksonville Zoo. Tail-sucking has been observed in both male jaguars; however, one individual has only been observed to engage in this behavior during the colder weather months (November-February). The study subject, Saban, reportedly exhibits this behavior throughout the year, with some extreme bouts leading to open wounds and alopecia. Tail-sucking is perhaps most hazardous of the stereotypic behaviors being exhibited. In the past, this behavior has led to abrasions and lesions, opening up the possibility of infections and reduced physical welfare.

Preliminary Data Collection

Initial attempts to identify environmental triggers for the tail-sucking behavior included constructing a daily log to check four times daily for evidence of observed tail-sucking for the two males. Data were collected on a Microsoft Excel spreadsheet (Table 1). Keeper checks were scheduled for (1) 7:00am-7:45am, (2) 12:15pm-1:00pm, (3) 2:30pm-3:30pm and (4) 4:45pm-5:30pm. Both feeding times and times of the checks were recorded. Though keeper checks were conducted for both male cats, the subject of this study had a different diet than the other male jaguar. The subject was fed four times daily and his diet consisted of Toronto Zoo Feline Diet, Chunk meat and Capelin (*Mallotus villosus*). Feeding times were generally around the same time every day with the first feeding occurring between 7:00AM and 8:00AM, the second feeding between 9:00AM and 10:00AM, the third feeding between 12:00PM and 1:00PM, and the final feeding usually occurred between 4:00PM and 5:00PM. Due to the subject's history of exhibiting stereotypic behaviors, he did not receive a "fast day" and was fed the mentioned diet seven days a week. Keepers also reported the location of each cat at each check and coded "N" for no evidence or observation of tail-sucking, "E" for evidence of stereotypic tail-sucking and "O" for observed tail-sucking. Criteria for "evidence" of tail-sucking included a visibly wet tail or a dried white ring around the tail, which was indicative of dried saliva.

Animal care staff were also concerned this behavior was occurring after hours, when staff was not present. Animal Care Staff set up a GoPro camera to record behavior after hours. This footage revealed occasional stereotypic tail-sucking, but it also revealed long bouts of grooming and excessive stereotypic pacing. Following this discovery, the wellness lab at Jacksonville zoo and gardens began to formulate a methodology for the treatment for pacing and over-grooming in addition to stereotypic tail-sucking.

Study Subjects

A single male jaguar housed at Jacksonville Zoo and Gardens was the subject of this treatment analysis. "Saban", a 5-year-old, in-tact jaguar, was born January 26, 2013 at Brevard Zoo, where he was mother-reared. In April 2014, he was transported to Jacksonville Zoo and Gardens. Records from the subject's previous zoo revealed that the study subject had a history of engaging in stereotypic behaviors prior to being transported to Jacksonville Zoo and Gardens. Additionally, the subject's biological father had been documented to exhibit stereotypic tail-sucking.

A 4-year old male jaguar "Kahn" housed at the Birmingham Zoo, Inc. served as the control subject for the fecal glucocorticoid analysis. This jaguar was born at Jacksonville Zoo and Gardens in 2013. In February of 2017, he was transferred to Birmingham Zoo, Inc. in accordance with the Species Survival Plan breeding and transfer plan.

Housing

The subject is housed in the *Range of the Jaguar* exhibit at the Jacksonville Zoo and Gardens in Jacksonville, Florida. Jacksonville Zoo and Gardens is home to 2.3 Jaguars ranging from 5-14 years of age. All jaguars are housed separately. *Range of the Jaguar* consists of two exhibits: "Plaza" and "Temple." Both exhibits contain rockwork, natural trees, and water features. The off-exhibit holding consists of four outdoor holding yards and attached indoor dens. A series of chutes connect each holding yard to the interior dens. Dens consists of three concrete block walls and a metal mesh wall. These dens contain a shelf approximately 4-5 feet high. Dens are separated by solid shift doors. The cats, therefore, do not have visual access to jaguars housed in adjacent dens. They do, however, have visual access to jaguars housed across the keeper hallway (See Figure 1). Per management protocol, the study subject is not housed next to the other resident male jaguar "Tuco." Animal care staff previously attributed stress-related behaviors, including stereotypic pacing and frequent vocalizations exhibited by both the subject and Tuco to proximity to each other, as both are sexually mature males. A black shower curtain is drawn to block visual contact between these two cats if housed in a manner where they have visual contact across the interior keeper hallway. With two exhibits, two of the jaguars are typically locked on exhibit during visitor hours (approximately 9:00 AM-5:30 PM,) while all other animals remain in the off-exhibit holding areas. Overnight, jaguars are rotated, so two different jaguars are given access to the exhibits. Overnight jaguars are also given access to an indoor den and the connecting chute.

The subject's caretakers previously attributed the exhibition of stereotypic behaviors to the presence of zoo visitors. For this reason, the subject typically remains off-exhibit during the zoo visitor hours and is given access an exhibit overnight (approximately 5:30pm-9:00am). The exhibit the subject is given access to rotates nightly.

With the exception of phase 2, the subject will continue to remain off-exhibit during zoo hours. While the subject is housed off-exhibit he will either have access to an outdoor holding yard and either 1 or 2 indoor dens and the connecting chutes, or he will have access to 3 adjacent dens. The subject will have routine access to food and water. Routine environmental enrichment will be given throughout the duration of the study. Phase 3 will involve manipulating environmental enrichment. The parameters of this phase will be discussed later.

Functional Behavior Assessment

To determine environmental variables to manipulate for the intervention, each jaguar keeper was asked a series of questions pertaining to stereotypic tail-sucking and pacing (Appendix 1). Questions relating to jaguar behaviors, management practices, and environmental stimuli were included in the questionnaire. For example, keepers were asked questions about the subject's behavior budget before and after instances of stereotypic pacing and stereotypic tail-sucking. Four primary members of the animal care staff were interviewed individually. Interviews were conducted via phone.

Data Collection

This behavioral assessment included a ten-week study, consisting of five phases. Three stereotypic behaviors (pacing, tail-sucking, and over-grooming) were the focal behaviors of this study (Table 2). Both during and after zoo operation hours data were collected at predetermined times. Therefore, camera footage was used to observe and code behaviors. Cameras were installed in each of the off-exhibit holding dens, outdoor holding yards, chutes leading to the exhibits, and the exhibits. 13 HikVision network dome cameras (Model: DS-2CD2132F-IWS) were used in the exhibits and chutes leading to the exhibits. These cameras were equipped with 2.8mm fixed lenses, night-vision capability, and were waterproof and vandal resistant. Cameras allowed for a wireless range of ~165 feet which permitted visual coverage for all areas of the exhibits with minimal blind spots. The Plaza exhibit was outfitted with five cameras and the Temple exhibit was outfitted with four cameras. Cameras were strategically placed to reduce the probability of "out of views." CAT5 ethernet cables connected each camera to a Network Video Recording device (NVR) located in the jaguar night-house. Cameras were not equipped with sound functions. The indoor holding dens and outdoor holding yards were installed with QSEE 720P HD Bullet cameras and were connected to a DVR located in the night-house. These Bullet cameras were located closer to the DVR, they did not require wireless capability and were connected using AV cords. Cameras allowed for behavioral observations after zoo hours when animal care staff was not present. The jaguar night house and exhibits are currently installed with a total of 29 cameras.

Once permanent cameras were installed in jaguar holding areas and the two exhibits, investigators reviewed footage to determine general activity budgets for the study subject. Three consecutive days of continuous footage were reviewed to determine periods of activity vs. inactivity. These data would help determine observation session times for the study. Session times were also determined based on keeper availability to deliver enrichment during the second manipulation phase.

Behavioral observations were recorded four days a week on Monday-Thursday using the camera footage. The focal subject was observed for six 30-minute sessions at predetermined times. Observations took place at 0:30-1:00AM, 6:30AM-7:00AM, 9:00-9:30AM, 12-12:30PM, 15:45-16:15PM and 18:00-18:30PM. After each session time, the footage for the locations the subject had access to were downloaded onto an external hard drive and reviewed. Behavior was recorded using partial-interval time sampling and whole-interval time sampling methods (Powell, 1977). Each behavior was predetermined as whole-interval or partial-interval. Thus, designated partial-interval behaviors will be recorded if they occur during any portion of the interval, wheras whole-interval behaviors will be recorded only if the behavior occurs for the entire duration of the interval (Powell, 1977). Intervals were 10 seconds in length. The investigator paused the video footage to record behaviors exhibited in each interval. All behavior observations were recorded using a Microsoft Excel spreadsheet. All predefined behaviors can be found in Appendix 2.

Behavioral Assessment Phases

Data collection occurred in five phases and followed an ABCAD reversal design (Table 3). Each phase was two weeks in length. Data collection occurred from 8/21/17-11/4/17.

Phase "A" served as the baseline phase. All management and husbandry practices remained constant throughout the two-week baseline period. Baseline consisted of rotation of proximate jaguars each night, normal diet preparation/delivery and standard enrichment administration. Routine enrichment protocol consisted of following a monthly calendar that indicated which enrichment would be delivered on that day. Enrichment delivery time varied. As part of current management protocol, a curtain was drawn down the middle of the keeper hallway to block visual contact between the two adult male jaguars in opposite dens during this phase.

Phase "B" was the first manipulation. This phase consisted of manipulating the study subject's housing designations. Keepers previously attributed the study subject's engagement in stereotypic behaviors to stress from visitor densities. As a result, the study subject was deemed a "night cat" and his daily routine consisted of spending the day time visitor hours in off exhibit holding and having one of the exhibits with access to one or more holding dens overnight. The other male jaguar followed the opposite schedule and was on exhibit during the day and housed in holding overnight. Investigators wanted to test this visitor density theory by reversing the housing designations for the study subject. For phase B, the study subject would be housed on exhibit during the day and off-exhibit overnight. Unfortunately, this phase was disrupted by the path of Hurricane Irma, and therefore occurred for 2 non-consecutive weeks. In the day prior and two days following the impact of Irma, the study subject remained off-exhibit with access to two holding dens and a covered chute for safety reasons. For the remainder of the week, phase 2 resumed without behavioral data collection or fecal sampling.

Phase "C" consisted of manipulating environmental enrichment provided for the subject. Specifically, enrichment was provided four times per day with at least one enrichment item provided overnight. Several new enrichment items were proposed for approval for this study, meaning the subject would have access to novel enrichment items in addition to the increase in frequency of enrichment delivery. Enrichment was provided 45-minutes to an hour prior to any observation session. Enrichment was provided between 8-8:30am, 10:45-11:30am, 2:30-3:30pm, and 4:30-5:15pm. A zipline was also installed prior to conducting the study. The zipline was anchored to two metal poles, which are part of the exhibit frame. The zipline was constructed using ¼-inch stainless steel aircraft cable, stainless steel cable clamps, thimbles and firehose. The zipline allowed for items to be attached to the cable using a stainless-steel pully connected to stainless steel chain, which was covered in firehose for safety reasons. All enrichment was approved by management and veterinary staff prior to its use in this study. New enrichment items provided to the study subject followed current new-item observation and evaluation protocols, which requires a minimum of 15 minutes of direct observation for the first two times the animal is presented with the item (See Appendix 3 for more detail regarding the enrichment evaluation protocol). Additionally, the investigator kept a log of enrichment items provided with specific details about the item itself, the location of access, date given, and the location of the item within the exhibit or holding area.

The next phase of the study consisted of a repeat of the baseline phase. During this period, all husbandry and management practices will return to normal. This return to baseline aimed to demonstrate experimental control and eliminate confounding effects. Data from phases B and C were analyzed during the second baseline phase to determine which manipulation phase resulted in the least amount of stereotypic behaviors.

The final phase, Phase "D" was a repeat of the condition which evoked the least amount of stereotypic behaviors. Based on data analyses, the condition which evoked the least amount of stereotypic behaviors was determined to be phase B (i.e., the on-exhibit phase). As such, this phase repeated during the final two weeks of the study.

Fecal Glucocorticoid Analysis

Preliminary fecal glucocorticoid analysis.

Prior to beginning the 10-week study, whole fecal samples were collected from the study subject for one week. For four consecutive days, every fecal passed by the study subject was collected for analysis. Samples were dated and stored at -20°C within two hours of collection (Washburn & Millspaugh, 2002). The fecal collection protocol can be found in Appendix 4. At the end of the week, the fecal samples were taken to South-East Zoo Alliance for Reproduction and Conservation (SEZARC) laboratory located at the University of North Florida in Jacksonville, Florida. Brown and colleagues found variation in the distribution of steroid metabolites within felid samples (Brown, Wasser, Wildt, & Graham, 1994). For this reason, the collected samples were partitioned into four pieces to ensure ≤20% variation of steroid distribution within samples. Determined ≤20% variation would allow for animal care staff to collect only a portion of a fecal sample rather than whole fecal samples. Both the preliminary fecal glucocorticoid analysis and within-study fecal glucocorticoid analysis used the same extraction process. This is done to save on shipping costs and keeper labor.

Within-study fecal glucocorticoid analysis.

Fecal cortisol levels were also assessed throughout the five phases of the behavioral assessment. Animal care staff was asked to collect fecal samples three times weekly from the study subject. As such a total of 30 fecal samples were collected. They were dated and stored at -20°C within two hours of collection. At the end of the study, fecal samples were taken to the SEZARC laboratory located at the University of North Florida.

1.0 Jaguar "Kahn" housed at the Birmingham Zoo, Inc. served as the control subject for the fecal glucocorticoid analysis. An internal research request form was completed and submitted to the Birmingham Zoo Inc. steering committee for review. The sample collection was reviewed and approved on May 11, 2017. Birmingham was asked to follow the same collection protocol for their jaguar. Keepers were asked to collect 3 samples per week for 10 weeks resulting in 30 total fecal samples. They were also asked to document any stereotypic behaviors exhibited by the study subject. Fecal collection for this jaguar occurred from 5/22/17-7/30/17. Following the 10-week collection period, samples were sent to the SEZARC lab located at the University of North Florida for analysis.

Extraction process.

Fecal samples were freeze-dried, homogenized and crushed. Using water that had been boiled (90-100^{o}C) for 20 minutes, dry fecal matter was extracted in (0.19±0.01g) in 5ml of ethanol (90% v/v). Plates used for the enzyme immunoassay (EIA) were pre-coated with 10 µg/mL Goat anti-rabbit IgG (Mesa-Cruz, Brown, & Kelly, 2014). and stored in a Ziploc bag with desiccant at 4^{o}C until analysis. This cortisol EIA also utilized a cortisol-horseradish peroxidase (HRP) and an antiserum.

Necessary standards, controls, and antibody samples were prepared according to the SEZARC protocol. Control preparation consisted of combining 6mL of Cortisol horseradish peroxidase ligand (1:100,000) in assay buffer with 6 µL stock and 6 mL assay buffer. Sample preparation consisted of combining 6mL of Cortisol antibody (1:25,000) in assay buffer with 20.4 µL stock and 6 mL assay buffer. 50µl of the standards, controls, and samples were loaded into the pre-coated plates. Within 1 minute, 50 µl of horseradish peroxidase ligand and 50 µl of antibody were added to each well. The plate was then covered and placed on a shaker for 1 hour.

After shaking, the plates were washed four times with 300 µl of a wash buffer. Then, 100 µl of ABTS was created and added to the wells and the plate was resealed. The plate was then read at 450 nm incubated at room temperature. The EIA protocol can be found in Appendix 5.

Data Analysis

Behavioral assessment phases.

First, overall behavior budgets were calculated while excluding any out-of-view scores. The mean percent of intervals spent engaging in each behavior was calculated for each day and then compiled for each phase (Table 4) Data from phases A, B, and C were analyzed to determine which manipulation phase would be repeated following the return to baseline. This analysis occurred during the second baseline period. To determine which phase to repeat, the mean percent of intervals spent engaging in aberrant behavior (tail-sucking, pacing, and/or over-grooming) was assessed. Mean percent of time spent engaging in each of the predetermined behaviors and mean percent of aberrant behavior was calculated for each the remaining two phases following the data collection period. Using SPSS statistical software version 24.0, descriptive statistics were performed and used to calculate the mean, standard deviation, skewness, and kurtosis for each of the 5 phases. Descriptive statistics were performed to represent the percent of intervals in which pacing occurred as a function of phase and day within the phase. The mean percent of intervals spent engaging in aberrant behavior was used to identify behavior change across baseline and treatment conditions and to determine if treatments were effective in reducing aberrant behavior. Mean percentages across phases were also used to determine if the pattern of behavior change followed that of a standard reversal design.

Investigators were also interested if treatment conditions had an effect on the frequency of object manipulation and enrichment manipulation when compared to baseline phases. It was expected that enrichment manipulation would increase during the enrichment manipulation phase. However, we were also interested in testing whether or not space allocation evoked changes in frequency of object/enrichment manipulation

Fecal glucocorticoid analysis.

First, overall mean FGM concentrations (ng/g) were calculated for both the study subject and the control subject. Descriptive statistics were performed and standard deviations were calculated and used to determine the existence of any outliers. Fecal samples were then compared to corresponding behavioral data, accounting for the 12-48hr excretion lag time observed in felid species (Naidenko et al., 2011; Terio, Citino, & Brown, 1999; Wielebnowski et al., 2002). Mean fecal cortisol levels were then calculated for each of the five phases and compared mean percent of aberrant behavior per phase using Pearson's correlation. Concentrations of hormones are stated as nanogram cortisol mass per gram of feces (ng/g).

Behavioral assessment of zoo-housed jaguars.

Surveys were closed from the public on January 12, 2018. The list of participating institutions (Question 2 on the survey) was exported into a Microsoft Excel spreadsheet. The most recent studbook published on the AZA website was used to identify animals and collect any missing information from the survey responses. In some instances, institutions listed juvenile animals that had not yet been given a regional studbook number. Juvenile animals not yet given a studbook number were omitted from the analysis. As the aim of the survey analysis was primarily one of exploratory nature, descriptive data were compiled to determine the percentage of the population that has been observed engaging in stereotypic behavior. Descriptive data were then compiled to compare keeper perception of the environmental conditions and possible motivations surrounding stereotypic behaviors. These exploratory analyses were conducted for each of the 3 focal stereotypic behaviors.

Results

Behavioral Assessment Phases

Within the subject's overall behavior budget, the treatment phase with the lowest percent of intervals spent engaging in aberrant behaviors was used to determine which manipulation phase would be repeated for the final phase of the study. This analysis was conducted during the second baseline period (phase 4). Following the completion of the data collection period, descriptive statistics were performed to determine the mean, standard deviation, skewness, and kurtosis of aberrant behavior frequencies between phases (Table 4).

As seen in Table 4, the percent of aberrant behavior did not remain constant between phases. Recall that we first hypothesized that our treatment conditions would result in lower frequencies of aberrant behavior when compared to the baseline conditions. The mean percent of intervals in which pacing occurred during the initial baseline period was 25.0. As we were also interested in which of the manipulation phases resulted in lower aberrant behaviors, the mean frequencies between phases 1 and 2 were compared. Both treatment conditions resulted in lower mean frequencies of aberrant behavior than the initial baseline period. Phase 2 resulted in the lowest mean percent of intervals of aberrant behavior with a mean of 15.4, and phase 3 had the next lowest mean percent of aberrant behavior at 20.8. As such, it was determined that the second manipulation, the on-exhibit phase, would be repeated for the final phase of the study.

These findings support our first hypothesis that aberrant behavior will decrease as a result of implementation of treatment conditions. Also consistent with our first hypothesis, the percent of intervals in which pacing occurred increased during the return to baseline; That is, the mean percent of intervals in which pacing occurred was 35.9 in phase 4. Although the mean percent of aberrant behavior for phase 5 (27.5) was higher than phases 1, 2, and 3, aberrant behavior decreased as expected from the second baseline phase to the reinstatement of the treatment condition (phase 5).

Interestingly, the standard deviations from phases 1,2,3, and 4 revealed fairly equal variability of aberrant behavior across days within phases with a range of 13.3 to 17.7. This was not the case, however, for the final phase of the study which resulted in much lower variability with a standard deviation of 4.7. As the standard deviations were calculated as a function of days within phases, the lower standard deviation from the mean in phase 5 indicates that the percent of aberrant behavior was more consistent across days than it was in the other phases of the study.

Investigators were also interested in any potential changes in the frequency of manipulation behaviors with the implementation of different treatment conditions. Manipulation behaviors included any behavior which involved investigating or interacting with (1) an object fixed in the environment (e.g., log, tree) or (2) an environmental enrichment item provided to the study subject. It was expected that an increase in enrichment manipulation behaviors would occur during the enrichment condition phase. However, investigators were interested in exploring whether object manipulation would increase as a result of additional enrichment and if the on-exhibit phase impacted manipulation behaviors when compared to baseline. As expected, the frequency of enrichment manipulation behaviors increased during phase 3 of the study. The frequency of enrichment manipulation behaviors was also considerably higher for the on-exhibit, or phase B, of the study when compared to the two baseline phases. Interestingly, this pattern was not replicated during the repeat of the on-exhibit phase (Figure 2). The frequency of manipulation of fixed objects in the environment remained steady with little variation across phases.

Fecal Glucocorticoid Analysis

Pre-trial glucocorticoid of the Jacksonville Zoo and Gardens jaguar.

Prior to the data collection period, animal care staff was asked to collect every fecal passed by the study subject over a period of four days. These samples would be used to determine the variation of fecal cortisol metabolites within samples. Jaguar care staff was able to collect fecal samples on 3 consecutive days. On the first and third day, keepers were able to collect one fecal sample. On the second day, keeper staff was able to collect 4 fecal samples from the study subject.

Cortisol extracted from the fecal samples collected in the pre-trial collection period ranged from 36.92744 ng/g to 136.3226 ng/g. The inter-assay coefficient of variation (CV) was used to determine the variability in cortisol levels in the fractions (separated portions) created from each sample. Cortisol levels between fractions of fecal samples ranged from 36.92744 to 242.212. The coefficient of variation was first calculated using the fecal cortisol levels (ng/g) across the 3 days in which samples were collected. Next, the coefficient of variation was calculated for the 4 fecal samples within one day. The enzyme-immunoassay revealed a coefficient of variation of 14.3% for cortisol levels across days. This variation in the fecal cortisol metabolites between fractions of fecal samples was lower than the accepted variation between plates (≤20% variation). The variation in FGM across the day was no higher than the accepted between-plate variation with a CV of 18.9% (FDA, 2001). These results indicated that there was no need for keepers to collect whole fecal samples and that the samples did not have to be collected within a certain time-frame each day.

Within-study fecal glucocorticoid analysis.

Animal care staff was asked to collect 3 fecal samples per week for the duration of this study. When possible, they were asked to collect fresh samples. Keepers were able to collect 28 fecal samples for the study subject over a 10-week period. A total of 26 fecal samples were collected from a control subject housed at Birmingham Zoo. In-study fecal cortisol levels from the study subject ranged from 18.46758ng/g to 73.68175 ng/g with a mean of 35.9333 ng/g (Figure 3). With scores ranging from 51.3943 to 349.8638 ng/g with a mean of 136.1049 ng/g, the control subject had consistently higher cortisol levels than did the study subject, (Figure 4).

Standard deviations were calculated to determine the existence of any outliers. Outliers were defined as any sample that resulted in less than or greater than two standard deviations from the mean (Brown, Bellem, Fouraker, Wildt, & Roth, 2001). Data revealed one outlier for both the study subject and the control subject. The study subject's outlier was a spike in FGM and occurred in the final phase of the study.

Mean fecal glucocorticoid levels for the study subject were compared across phases (Table 5). Both manipulation phases resulted in the lowest FGM levels. The second phase of the study resulted in the lowest mean FGM level with a mean of 32.8866 ng/g and the third phase resulted in a mean of 33.2241. Interestingly, the repeat of phase 2 resulted in the 2nd highest FCM level.

Pearson's correlation was used to assess the relationship between the percent of aberrant behavior and the subject's fecal cortisol levels (Table 5). There was a moderate but unreliable positive relationship between percent of aberrant behavior and fecal glucocorticoid levels per phase (r = +.22, p = .717).

Behavioral Assessment of Zoo-Housed Felids

A survey was administered to AZA accredited institutions to assess the prevalence of stereotypic behaviors in zoo-housed jaguars across North America. According to the 2016 regional studbook, the North American population jaguar population consists of 80 males and 72 females at 63 institutions. Although two of those institutions are members of the Species Survival Plan for the North American jaguar population, these two institutions are not located in North America, and therefore responses from them were not solicited. Of the 63 institutions listed in the regional studbook, 16 were not accredited by the Association of Zoos and Aquariums. Responses were received from 37 institutions, 36 of which are AZA accredited and one that was not. This provided a 58.7% response rate. Of those responses, one did not consent to the use of his answers in broader research purposes and, therefore, his responses were not included in the data analysis.

It was determined that 51.3% of the North American jaguar population is housed at the 37 institutions from which we received survey responses. Survey results provided detailed information concerning 78 individual jaguars: 37 male and 41 female. Of the 78 individuals included in the survey responses, we discovered that 62% engage in one or more of the 3 focal stereotypic behaviors (pacing, tail-sucking, excessive grooming) addressed in our survey.

Data were compared for respondents who indicated that one or more jaguar engaged in stereotypic behaviors. Of the 36 responses received, 24 indicated that one or more jaguars engages in stereotypic pacing, meaning that 67% of the North American jaguar population has been observed pacing. Investigators found that pacing is observed at various time frames. Of the 24 respondents who reported observing pacing, 25% indicated that this behavior is most frequently seen in the early morning (before 10AM), 20.8% reported seeing this behavior in the late morning, 25% reported seeing this behavior most frequently in the early afternoon, and 29.2% reported this behavior in the late afternoon (after 3pm).

Respondents were also asked if they believed the animal engaging in stereotypic pacing was attempting to gain or avoid a stimulus. Of the 24 respondents who noted observing an animal engaging in stereotypic pacing, only 8.3% indicated that he/she thought the animal was attempting to gain something by engaging in stereotypic pacing. By comparison, 37% indicated that the animal was not attempting to gain something, and 54.2% were not sure if the animal was attempting to gain something by engaging in stereotypic behaviors.

Conversely, 12.5% of respondents reported thinking the animal was trying to avoid something by engaging in stereotypic behaviors. Almost half (45%) of respondents did not think the animal was attempting to avoid something, 29.2% were not sure, and 12.5 percent selected other for this question. Respondents who selected other indicated that the animal was "potentially attempting to avoid vet staff, but the behavior persisted once vet staff had been removed from the environment." Another respondent indicated that in some instances the animal was trying to avoid something by engaging in stereotypic pacing. One respondent who selected "other" for this question said that at times, the animal was moving away from the direct viewing area (i.e., attempting to avoid the public).

In one of our survey questions respondents were asked to describe environmental conditions prior to observing an animal engaging in stereotypic pacing. As this was an open-ended question, we were hoping to identify some pattern that might allow us to identify different environmental variables that potentially elicit this behavior. The responses received for this question varied greatly. Some respondents (25%) noted that they were unable to identify any pattern in environmental conditions. A number of respondents (42%) indicated that large crowds, loud noises, construction, and the presence of maintenance technicians were potential triggers for this behavior. One respondent indicated that one of the animals in her care was more likely to engage in stereotypic pacing on fast days or when veterinarians were present. Interestingly, four respondents (17%) cited cold or inclement weather to be a trigger.

Finally, respondents were asked if any physical or mental health deficits had been observed as the result of stereotypic pacing. Nearly half of respondents (43.3%) noted never seeing any physical or mental health deficits as a result of pacing. One the other hand, some respondents (10%) indicated that hair loss was observed as a result of the pacing behavior. Three respondents (10%) also indicated that he or she had seen open wounds or raw skin as a direct result of stereotypic pacing. There were also 5 respondents (20%) who indicated seeing cracked and bloodied paws from excessive stereotypic pacing. Respondents (24%)who selected the "other" option for this question revealed that he or she had seen rocks or other environmental debris embedded in the paws of pacing animals and cracked or worn down paw pads as a direct result of stereotypic pacing.

The next set of questions referred to any animal that had been observed engaging in stereotypic tail-sucking in the past or at present. Nine respondents (25%) indicated that one or more jaguars housed at his/her institution engages in stereotypic tail-sucking. Of these responses, more than half (55.6%) indicated that this behavior is most frequently observed in the late afternoon (after 3pm). The next most frequently reported time period for observed stereotypic tail-sucking was the late morning (22.2%). An equal number of respondents (11.1%, early morning; 11.1%, early afternoon) indicated that the behavior is observed most frequently in the early morning and the early afternoon.

When asked if their animal was attempting to gain something by engaging in stereotypic tail-sucking, 11.1% answered affirmatively. However, 33.3% said the animal was not attempting to gain something by engaging in this behavior, 44% said they were unsure, and 11.1% (i.e. one respondent) selected "other" in response to this question. This respondent noted that veterinarian staff at his institution suggested that endorphin release may be triggered by destructive tail-chewing/licking (injury).

When asked if the animal engaging in stereotypic tail-sucking was attempting to avoid something, no respondent indicated that the animal was, in fact, attempting to avoid something. Conversely, 66.6% indicated that the animal was not attempting to avoid something, and 33.3% noted that they were unsure whether or not the animal was attempting to avoid something by engaging in stereotypic tail-sucking.

Respondents were also asked to describe environmental conditions immediately before instances of stereotypic tail-sucking. Of the nine who reported caring for an animal that engages in this behavior, 2 indicated that cold weather seemed to be a trigger. Three of the respondents indicated that they were not able to identify a pattern in environmental conditions prior to observing this behavior. One respondent indicated that visitor densities seems to be correlated with the behavior and one individual indicated that being locked in holding for exhibit work/maintenance seemed to be a trigger of this behavior. One individual said that she saw the behavior immediately before eating or enrichment manipulation and suggested that this behavior might be elicited by anticipation of food or other tangible objects such as enrichment items.

When asked if any physical or mental health deficits correlated with the stereotypic behavior, 42.6% reported observing hair loss as a result of tail-sucking. Furthermore, 30.8% reported seeing open wounds from aggressive or repeated bouts of stereotypic tail-sucking.

However, 15.4% reported seeing no physical or mental deficits correlated with stereotypic tail-sucking. The respondent who answered “other” indicated that the animal no longer engages in the behavior, but in the past reported seeing hair loss and open wounds as a result of tail-sucking.

The final set of questions in the survey referred to excessive grooming behaviors. Only 10% (4 respondents) of participating respondents indicated that one or more individuals housed at their institution engages in excessive grooming. Of these 4 responses, half (50%) reported that the behavior is most frequently observed in the late afternoon. Of the remaining respondents, 1 reported observing the behavior in the early morning and the other reported seeing it most frequently in the early afternoon

All four of these respondents indicated that they did not believe the animals reported to engage in excessive grooming were attempting to gain something by doing so. Similarly, 100% of respondents indicated that they did not believe the animal was attempting to avoid something by engaging in excessive grooming behaviors.

When respondents were asked to describe environmental conditions prior to an animal engaging in excessive grooming, only one respondent was able to identify a possible environmental trigger. This respondent indicated that the animal in her care tested positive for season allergies that elicited an increase in grooming behaviors. All other respondents indicated that they were unable to identify potential environmental triggers for this behavior.

Finally, respondents were asked to describe any mental or physical health deficits resulting from excessive grooming. All four respondents indicated that hair loss was a physical deficit observed in animals who engaged in this behavior. One of the four respondents (20%) indicated that he has seen open wounds as a result of this behavior.

Discussion

Behavioral Assessment Phases

Overall, comparisons of the 3-focal aberrant behaviors between phases followed the expected pattern of a typical reversal design. In a standard reversal design, one would expect to see behavior change during intervention phases, followed by the reoccurrence of aberrant behaviors upon return to baseline or discontinuation of a treatment (Kazdin, 2011). Moreover, one would expect to observe the same behavior change again upon the reinstatement of a condition (Kazdin, 2011). This pattern of behavior was evident in this study, as we saw higher levels of aberrant behavior for baseline conditions when compared to treatment conditions. Data revealed a reduction in the percent of intervals spent engaging in aberrant behavior from the initial baseline period to the first treatment. There was an observed increase in percent of aberrant behavior from the phase 2 (manipulation of exhibit access) to phase 3 (enrichment manipulation); however, the percent at which the subject engaged in aberrant behavior during phase 3 was still considerably lower than that which was observed in the initial baseline period. These results indicate that while both treatments were effective in reducing aberrant behavior, the treatment involving a manipulation in daytime exhibit access was more effective at reducing aberrant behavior than the enrichment manipulation treatment. Upon the return to baseline for phase 4, we observed and increase in aberrant behavior, as expected. Phase 4 produced the highest percent of aberrant behavior within the subject's overall behavior budget compared to the other phases of the study. Finally, aberrant behavior percentages decreased following the recurrence of treatment 1 (manipulation of exhibit access) for phase 5. Although the percent of aberrant behavior in the final phase was higher than the initial baseline, these data still follow the pattern of expected behavior change within a reversal design.

Although we saw a decrease in aberrant behavior from phase 4 (baseline) to phase 5 (repeat of manipulation of exhibit access), phase 5 resulted in a higher mean percent of aberrant behavior than phases 1(baseline), 2 (exhibit manipulation), and 3(enrichment manipulation). Whereas this pattern of behavior change still follows the typical results from a reversal design, we expected to see more similar behavior budgets between phases 2 and 5. It is possible that the higher percentage of aberrant behaviors observed in phases 4 and 5 resulted from a phenomenon known as the rebound effect, whereby an increase in responses is elicited following the suppression of a behavior (Comings, 1990; Himle & Woods, 2005; Himle, Woods, Piacentini, & Walkup, 2006). The idea of a rebound effect may possibly account for the slightly higher aberrant behavior percentage observed in phase 4 when compared to phase 1 as well as the differences observed between phase 2 and phase 5. On the contrary, it is possible that these results indicate that there may be other external variables maintaining the pacing behavior. Other possible maintaining variables might include season, proximity to conspecifics, or an underlying psychopathological mechanism.

In addition to an increase in stereotypic pacing between phase 2 (on-exhibit manipulation) and phase 5 (repeat of on-exhibit manipulation), we also observed stereotypic tail-sucking in phase 5 for the first time in this study. Animal care staff now theorize that stereotypic tail-sucking may be triggered by some factor associated with colder temperatures. This was a limiting factor in this analysis, as the last phase of the study occurred during the transition to colder temperatures. We did see a decrease in temperatures during this phase of the study, which took place in the last week of October and the first week in November. The hypothesis that stereotypic tail-sucking is in some way related to colder temperatures requires additional testing.

As previously mentioned, prior to this study the subject was a deemed a "night cat," as animal care staff attributed the high frequency of aberrant behaviors to stress induced by high visitor densities. Baseline phases followed this husbandry protocol, which for this animal consisted of providing him access to the exhibit overnight and housing him in off-exhibit holding areas during zoo visitor hours. The decrease in aberrant behavior during the on-exhibit phase confirmed that stereotypic behaviors exhibited by the study subject are not maintained by visitor densities.

In addition to decreasing aberrant behaviors through treatment conditions, we also hypothesized that such treatments would increase manipulation and other species-typical behaviors. First recognized by Yerkes (1925), environmental enrichment involves providing environmental stimuli to achieve psychological and physiological well-being and enhance the quality of care provided to animals housed in captivity (Shepherdson, Mellen, & Hutchins, 2012). Environmental enrichment has only recently gained traction in the last few decades (Mellen & MacPhee, 2001). Today, however, environmental enrichment is part of standard husbandry protocols for many zoological institutions (Association of Zoos and Aquariums) and is at the forefront of improving animal welfare (Maple & Perdue, 2013). Furthermore, enrichment has been shown to reduce stereotypic behaviors (Carlstead & Shepherdson, 1994). For example, one study found that introducing inexpensive enrichment items to the environment decreased stereotypic pacing, escape behaviors, and coprophagy in captive greater rhea (*Rhea Americana*) (De Azevedo, Lima, Cipreste, Young, & Rodrigues, 2013). New environmental enrichment aimed at promoting hunting behaviors has been used to reduce instances of stereotypic pacing and other problem behaviors (Mallapur & Cheullum, 2002).

During phase 3, this study subject received novel enrichment items more frequently than the standard husbandry protocol in addition to receiving enrichment overnight. Enrichment items provided were designed to evoke species-typical behaviors such as investigative behaviors, exploratory behaviors, and behaviors associated with hunting. As expected, the frequency of enrichment manipulation behaviors increased during the enrichment phase when compared to all other phases. Interestingly, the total frequency of enrichment manipulation was also considerably higher for the second phase of the study (on-exhibit condition). However, enrichment manipulation was very low for the repeat of the on-exhibit condition for phase 5 of the study. During the exhibit manipulation phase, routine enrichment per management protocol was provided for the study subject. Routine enrichment for these jaguars usually involved following a monthly enrichment calendar whereby one specific enrichment item was provided to each animal per day. However, enrichment can come in many forms including sensory (e.g., perfumes, audio playbacks), feeding (e.g., puzzle feeders), novel additions to the environment (e.g., fallen driftwood, exhibit modifications) and social interactions (e.g., positive reinforcement training) (Melfi, 2013). It is possible that the exhibit manipulation was enriching in itself. By providing a more complex environment and, in turn, increasing behavioral choices, animals have opportunities to engage in species-typical behaviors and improve their welfare (Shepherdson, 2003). While it is possible that daytime exhibit access and access to all the sensory stimuli (guest noises, smells, etc.) was enriching for the subject, we would have expected to see the same pattern in enrichment manipulation behaviors in phase 5 that we saw in phase 2.

As indicated by the results of this study, environmental enrichment did not eliminate stereotypic behaviors; however, the presence of novel enrichment and increased access to enriching stimuli did result in a decrease in stereotypic behaviors for the study subject. When it is utilized correctly and evaluated regularly, enrichment can be an effective tool in decreasing species-atypical behaviors while simultaneously increasing species-typical behaviors, thereby improving animal welfare. Still, once environmental enrichment strategies are employed, there is an increased necessity of systematically evaluating enrichment to ensure its effectiveness in targeting species-appropriate behavior on the individual level. Furthermore, Mellen and Sevenich-MacPhee (2001) suggest a single definition of enrichment should not exist. Instead, enrichment should be redefined and continuously evaluated on the species level as well as the individual level.

Fecal Glucocorticoid Analysis

In general, the study subject maintained relatively low cortisol levels throughout the 10-week study. We hypothesized that the treatment conditions would reduce cortisol levels when compared to baseline conditions. However, even baseline cortisol levels did not indicate that the subject was experiencing high stress levels. Moreover, the analysis of fecal cortisol levels revealed no significant difference in cortisol levels between phases. This was true even for the phases which revealed significantly elevated aberrant behavior frequencies. These results suggest that perhaps pacing is not correlated with higher stress levels for this particular animal.

Previous research indicates that stereotypic behaviors are often associated with high stress levels and are seen as an indicator of poor welfare and an inability for an animal to cope with its environment (e.g., Mason, 1991). Conversely, it has been reported that stereotypic behaviors are not necessarily correlated with poor welfare or high stress levels (Mason & Lantham, 2004) and, in some instances, have been proven to be negatively correlated with increased stereotypies (Mason & Lantham, 2004; Redbo, 1993; Vestergaard, Skadhauge, & Lawson, 1997). It has been argued that over time, stereotypies can become independent of the stimulus (e.g., stressful event) that originally elicited them (Hinde, 1970; Levy, 1944; Mason, 1991). As such, it is possible that a stereotypic behavior that was originally elicited by a stressful event no longer occurs in conjunction with a high-arousal situation. Instead, some argue that repetitively engaging in stereotypies becomes an automatic process described as “central control,” whereby minimal or no cognitive processing is necessary to perform this behavior (Fentress 1973, 1976; Martiniuk, 1976; Mason & Lantham, 2004). In essence, this behavior becomes somewhat of a habit engrained and readily available in the animal’s behavioral repertoire. Moreover, Toates (2001) suggests that these behaviors can then be elicited by a range of stimuli regardless of whether or not those stimuli are stressful. For this study subject, the pacing behavior may have been originally elicited by a stressful event; however, measures of his fecal cortisol levels revealed that this behavior was not correlated with stress. Therefore, we can conclude that while the treatments were effective in reducing stereotypic behavior, they were not effective in reducing baseline stress levels.

The control subject’s cortisol levels were consistently higher than Saban’s cortisol levels, with the exception of Saban’s one outlier which occurred in the final phase of this study. In addition to the fact that this animal is housed at a member institution for SEZARC, the control subject was chosen based on his age, sex, and history of relatively low engagement in stereotypic behaviors. However, it was later revealed that though infrequently, this individual also engaged in stereotypic tail-sucking. This revelation did not make this animal the ideal candidate to serve as a control subject to indicate baseline cortisol levels for a young, male jaguar.

The data revealed one outlier for both the control subject and the study subject. Interestingly, the outliers for both individuals corresponded with a stereotypic a tail-sucking bout. Though investigators cannot infer that stereotypic tail-sucking is in some way correlated with elevated fecal cortisol levels with only one data point per animal, past studies have indicated such a relationship. For example, researchers found that clouded leopards who engaged in self-mutilation behaviors (tail plucking, chewing) had significantly higher fecal cortisol concentrations when compared to individuals who did not engage in self-mutilation behaviors (Wielebnowski et al., 2002). Further testing of cortisol levels and behavioral sampling is necessary to identify if a relationship exists between cortisol levels and stereotypic tail-sucking.

Behavioral Assessment of Zoo-Housed Jaguars

The survey administered to AZA accredited institutions in North America was distributed with the goal of exploring the prevalence of stereotypic behaviors among zoo-housed jaguars. As previously mentioned, stereotypic or abnormal behaviors are often associated with poor welfare. Although there is evidence that both supports and disputes this statement, the general public commonly perceives stereotypic behaviors as indicators of poor welfare, lack of a stimulating environment, and “unhappy” or “unhealthy” animals. For example, in a 2012 study, researchers found that zoo guests who witnessed tigers pacing thought the level of care the animals were receiving was suboptimal (Miller, 2012). Furthermore, Miller found that zoo guests who observed frequent pacing behaviors were less likely to support zoos in the future and also less likely to support in-situ conservation efforts made by zoological institutions. In this regard, stereotypic behaviors can have a negative impact on the fulfillment of many zoos’ missions. Still, stereotypic behaviors are present in many zoo-housed animals and this is particularly true for large carnivores.

Although we only received responses from a little over half (58.7%) of institutions included in the regional studbook and members of the jaguar SSP, we found that 62% of the jaguars in this population engage in stereotypic behaviors. Furthermore, these stereotypic behaviors are exhibited by animals housed at 37 different zoological institutions across North America. Although the circumstances surrounding the development of stereotypic behaviors likely differs between individual animals, the presence of stereotypic behaviors increases the likelihood that zoo guests will develop negative opinions regarding the level and quality of care provided to these animals.

Our survey results revealed that pacing was the most common stereotypic behavior exhibited by zoo-housed jaguars (67%). Participants who indicated caring for an animal who engages in stereotypic behaviors were later asked a series of questions regarding the environmental conditions surrounding pacing bouts. No clear trend was revealed regarding the time of day animals were observed engaging in stereotypic pacing. Instead, about 20-29% of (roughly a quarter) of respondents indicated observing behavior most frequently during one of the specified time frames (early morning, late morning, early afternoon, late afternoon). These results imply that pacing is observed at all times of the day and, as such, time of day is not a predicting variable for pacing. More data is necessary to confirm if a relationship exists between time of day and frequency of pacing.

Stereotypic behaviors are thought to develop as a result of many factors including attempts to re-direct a behavior that is unable to be performed (Hinde, 1970; Mason, 1991) or inability to escape an aversive stimulus (Duncan, & Wood-Gush, 1972; Hinde, 1962; Mason, 1991). With this in mind, survey participants were asked if they thought animals were either trying to gain something (tangible object, primary reinforcer, keeper attention) or avoid something (aversive stimuli) by engaging in stereotypic pacing. As the majority of respondents were not sure of the motivations behind their animals' behavior, it is possible that keepers considered pacing to be motivated by some other external or internal variable or a simply a behavior of habit. It is also possible that animal care staff is unable to identify a motivating operation for the pacing behavior in their animals. By comparison, many respondents said their animal was not attempting to gain anything by engaging in stereotypic pacing and only a small portion of respondents attributed their animals' engagement in pacing behaviors to an attempt to gain something.

In response to the question about animals attempting to avoid something by engaging in stereotypic pacing, the majority of zookeepers believe that pacing is not a stress response elicited by an aversive stimulus. The perceived absence of an aversive stimulus can be viewed from an animal welfare standpoint as a positive in that pacing is not necessarily an indicator of poor welfare. However, an environment lacking aversive stimuli does not necessarily mean the environment is of optimal quality, as it is still possible that the environment is lacking components that the animal finds reinforcing (i.e. the animal would be attempting to gain something by engaging in pacing). As with the previous question regarding whether or not the animal was attempting to gain something by engaging in stereotypic behavior, it is also possible that zookeepers thought the behavior was being maintained by something other than the motivation to gain or avoid something. Future research would likely include open-ended question structure to allow for more detailed responses.

The majority of survey questions were structured using a multiple choice format. In one question, participants were asked in an open-ended format to describe environmental conditions prior to observed pacing bouts. There was no clear trend in responses, as the majority of respondents noted that they were unable to identify any specific pattern in environmental conditions or variables that routinely elicit the pacing behavior. Respondents' inability to identify specific patterns in the environment that could elicit alone suggests that there is still much to learn about stereotypic behaviors both on the species and individual level. If keepers are unable to recognize triggers for problem behaviors, it will be difficult to identify methods for treatment and the likelihood that these behaviors will persist or worsen is increased. Interestingly, those who did provide information regarding environmental conditions prior to observed pacing bouts attributed the behavior to conditions that would likely be interpreted as aversive (high visitor densities, presence of maintenance technicians, construction, and veterinarians). In these instances, naming these aversive stimuli would imply the animal was, in fact, engaging in stereotypic pacing to avoid said aversive conditions. In regard to visitor densities as a potential trigger of pacing, previous research has shown that higher visitor densities can increase stereotypic behaviors (Sellinger & Ha, 2005). However, recall that the current study found the opposite to be true. Pacing decreased when the animal was given exhibit access during daytime hours when visitors are typically present as compared to baseline conditions when the animal had exhibit access overnight (no visitors). As previously mentioned, the "kitchen sink" method to problem solving is too easily adopted. As such, empirical testing of hypotheses surrounding environmental triggers of any behavior is necessary to confirm whether or not a particular variable elicits a response or if alternative variables need to be explored.

Because physical ailments or visible injuries can be alarming for zoo patrons and prompt questions regarding standards of care provided to the animals, our survey respondents were asked about such injuries as they related to stereotypic pacing. Many respondents reported observing no mental or physical ailments as a direct result of pacing. However, the remaining respondents indicated observing physical deficits including hair loss, open wounds/raw skin, and cracked or bloodied paw pads. One respondent even reported observing environmental debris including pebbles and rocks embedded in an animal's paw pads. No respondents indicated observing any mental health deficits as a result of pacing. Assessing an animal's psychological well-being is a difficult task, as it is not something we can easily quantify or directly measure. Nevertheless, considering an animal's psychological well-being is necessary to ensure we provide the best possible welfare for the animals in our care.

Although fewer survey respondents reported housing animals who engage in stereotypic tail-sucking and over-grooming, those who did so also noted that these behaviors seem to occur most frequently in the late afternoon. This perhaps implies that time of day is a possible predictor these behaviors. Further research is necessary to identify variables associated with the late afternoon which could possibly tail-sucking and/or over-grooming. For example, it is possible the behavior is most frequently observed at this time because it is typically the time of day animals receive their evening diet or the time of day care staff close up and leave for the night.

Concerning the motives for tail-sucking, most respondents were unsure if the animals in their care are attempting to gain something. However, one respondent indicated that the facility's veterinary staff theorized that the tail-sucking behavior to be triggered by an endorphin release, thus suggesting the behavior is reinforced automatically rather than by an external stimulus. This belief is consistent with the idea that stereotypies can themselves be reinforcing. For example, Mason and Lantham (2004) describe how stereotypies can have reinforcing properties or even become "do-it-yourself enrichments," providing opportunities for mental and physical stimulation. Even so, tail-sucking can have extreme negative impacts on the health and well-being and there have been reports of fatalities due to tail-sucking leading to hair loss, open wounds, and eventually infection (Hope & Deem, 2006). These reports are similar those described in the answers to the survey question detailing observed physical and mental health deficits of tail-sucking, whereby over 73% reported observing either hair loss or open wounds as a direct result of tail-sucking. Similar to stereotypic pacing, no respondents reported observing any mental health deficits as a result of stereotypic tail-sucking.

The majority of respondents did not believe their animal was attempting to avoid something by engaging in stereotypic tail-sucking. This finding indicates that the animal care staff does not believe the environment contains something aversive. These staff members instead believe the behavior is elicited or maintained by some other variable.

Interestingly, two respondents indicated that colder temperatures could be a tail-sucking trigger when asked to describe environmental conditions prior to observing tail-sucking bouts. This is consistent with the theory currently held by the Jacksonville Zoo and Gardens jaguar care staff who believe tail-sucking to be a seasonal behavior most often exhibited in the colder months. Previous research indicates that seasonal variation can impact the prevalence of stereotypic behavior (Carlseatd & Seidensticker, 1991). As jaguars are native to South and Central America, it is possible that jaguars housed in institutions in areas with colder temperatures may find such cold aversive. Although most respondents in our survey did not believe their animal was attempting to avoid something by engaging in stereotypic tail-sucking, it is also possible that the care staff did not recognize the cold as an aversive stimulus. Stereotypic tail-sucking should be further investigated to confirm a relationship between tail-sucking frequency and seasonal variation.

All participants who reported caring for an individual who engages in over-grooming thought their animal was neither attempting to gain nor avoid something by doing so. Further research and more detailed questioning is necessary to discover whether keeper staff believe the behavior is maintained by some other underlying environmental variable. Additionally, only one of the four respondents who reported caring for an animal who engages in over-grooming behaviors was able to identify environmental conditions prior to bouts of over-grooming. This person identified seasonal allergies as a trigger for over-grooming and indicated that this behavior was corrected with allergy medications. All other respondents reported being unaware of any environmental conditions that may be correlated with over-grooming bouts. Hair loss was reportedly observed by all respondents who reported caring for an animal who engages in over-grooming. As previously mentioned, the threshold for considering grooming behaviors excessive is ambiguous. It is possible that for the participants in this study, observed hair loss is considered the threshold for excessive grooming. This would explain the fact that 100% reported observing hair loss and a direct result of over-grooming. Additionally, one respondent reported observing open wounds as a result of over-grooming behaviors. Like tail-sucking, these physical ailments can open up the possibility of infection and decreased physical and mental welfare. Identifying potential triggers and determining if stereotypic behaviors are performed as a stress response or a habitual response is necessary to mitigating harmful behaviors that can lead to reduced welfare.

Limitations and Future Directions

The original goal of the present study was to identify environmental variables contributing to stereotypic tail-sucking. While we were pleased that the study subject did not regularly engage in tail-sucking, it meant we were not able to effectively identify environmental triggers for this behavior in the context of this study. We found that the treatments were effective in reducing stereotypic pacing. Still, we are interested in determining if the colder temperatures are, in fact, correlated with tail-sucking bouts as the animal care staff believes. Future directions would include a continuation of baseline data collection to reassess possible treatments for the tail-sucking behavior. Identifying what specifically about the winter months elicits this behavior might generalize to other jaguars who reportedly exhibit this behavior more frequently or exclusively in the winter months.

A major limitation of this study was the fact that the control subject for the fecal cortisol analysis began engaging in the stereotypic tail-sucking behavior during the data collection period. This did not make him the ideal control candidate to provide a comparison between cortisol levels for an animal who engages in stereotypic behaviors versus one who does not. Future directions would include additional control subjects with a known history of no or infrequent stereotypic behaviors. Fecal collection from additional individuals would also be beneficial in identifying a baseline representation of cortisol levels for adult, male jaguars housed in zoos. Consistent with the idead that stereotypic tail-sucking is in some way associated with seasonal variation, it would be beneficial to continue collecting fecal samples to (1) identify if a relationship exists between tail-sucking and cortisol levels and (2) to identify if fecal cortisol levels are affected by seasonal variation.

Finally, the survey served to explore possible patterns in the stereotypic behaviors exhibit by zoo-housed jaguars. Although surveys can certainly be beneficial for collecting pertinent information, there is also limitations involved in self-report. For example, respondents may be less inclined to answer truthfully. As stereotypic behaviors are commonly associated with poor animal welfare, zoo personal tend to be sensitive in discussing this topic or all together admitting the presence of stereotypies in animals in their care. Still, it would be beneficial to explore additional variables that may predict stereotypic behaviors in zoo-housed jaguars. Additionally, as stereotypic behaviors are prevalent in many species of felid, exploring the prevalence of stereotypic behaviors, the history surrounding them, and possible contributing factors would be beneficial to the zoo community at large. Such information has the potential to better inform management practices and improve the welfare and wellness of felids housed in human care.

Conclusion

In summary, the results of the present study suggest that environmental manipulations including exhibit access and increased frequency/novelty of enrichment were effective in reducing stereotypic pacing behaviors exhibited by a captive male jaguar. The reversal design was effective in showing the behavior change between baseline and treatment conditions. This design is easily adoptable and can be used to assess a number of welfare and wellness evaluations including preference assessments, problem behavior, and enrichment effectiveness. A fecal glucocorticoid analysis did not show a significant correlation between the percent of pacing behavior and cortisol levels for this particular jaguar, indicating that pacing is not a response to stressful environmental stimuli. However, cortisol levels may be positively correlated with the frequency/percent of stereotypic tail-sucking. Further analyses are necessary to confirm this relationship. Finally, a significant portion of the North American jaguar population reportedly engages in stereotypic behaviors. Many of these stereotypic behaviors have unknown triggers. It is possible that environmental or ontogenetic characteristics exist that could help identify predictor variables for the development of these behaviors. Identifying a pattern in individuals who engage in these behaviors could inform management practices and methods for mitigating stereotypic behaviors.

Table 1.
Example of the data recording sheet used in the preliminary data collection period which included daily "checks" for evidence or observed tail-sucking for 2.0 jaguars "Tuco" and "Saban."

DATE	3/13/2017	PLAZA CAT	Am √: N/A	Day: T	Pm: N/A	TEMPLE CAT		Am √: S	Day: X	Pm: S	
		TUCO						SABAN			
FEEDINGS		1:00	8:58					1:00	7:39		
		2:00	4:48					2:00	10:18		
		3:00						3:00	12:16		
		4 (PM):						4 (PM):	4:43		
	Keeper AM routine		AB				Keeper PM routine		AB		
CHECKS	Initials	TIME	LOCATION	CODE	VISIBILITY		Initials	TIME	LOCATION	CODE	VISIBILITY
7:00-7:45	AB	7:38	den 3 shel	N			AB	7:25	TE3	N	Y
12:15-1:00	VS	12:41	PE39	N	Y		AB	12:15	den 1	N	
2:30-3:30	MR	2:45	PE39	N	Y		AB	2:35	HY1	N	
4:45-5:30	AB	4:45	den 3	N			AB	5:15	den 5	E	
	Comments						Security worked on plaza exhibit overnight.				
	Brian from security added more camera wires from 11:00-1:00										

Table 2.
Condensed ethogram of the three focal stereotypic behaviors.

Behavior	*Definition*
Pacing	Repetitive, apparently functionless movement, usually on a fixed route within the enclosure (after Odberg, 1978).
Tail-Sucking	Repetitive sucking, chewing, biting or raking teeth along the tail
Over-grooming	Determined during data analysis as ≧75% of sessions

Table 3.
Overview of the behavioral assessment phases.

Phase	Treatment
A	Baseline
B	Manipulation of exhibit access, whereby the study subject had exhibit access during zoo operation hours and was housed in off-exhibit holding overnight
C	Manipulation of enrichment delivery time and frequency
A	Baseline
D	Repeat of the condition which evoked the lowest percent of stereotypic behaviors

Table 4.
Percent of intervals in which pacing occurred as a function of phases and days within phases.

Day	Phase A (1)	Phase B (2)	Phase C (3)	Phase A (4)	Phase B (5)
1	28.6	16.3	44.0	32.7	31.4
2	4.0	0.9	19.7	34.2	20.7
3	27.9	30.0	4.3	27.3	23.5
4	3.5	15.8	0.0	63.3	23.8
5	15.1	38.5	20.5	46.1	34.4
6	57.7	12.8	17.4	18.4	28.5
7	33.9	7.9	42.6	47.7	26.5
8	29.5	0.6	17.6	17.1	31.3
Mean	25.0	15.4	20.8	35.9	27.5
Standard Deviation	17.69	13.30	15.77	15.76	4.7
Skewness	+.53	+.72	+.46	+.53	+.01

Table 5.

Mean fecal glucocorticoid levels and mean percent of aberrant behaviors for the study subject across phases.

Phase	Fecal Glucocorticoid Average ng/g	Mean Percent of Aberrant behavior
A	41.8922	25.0
B	32.8866	15.3
C	33.2241	20.8
A	34.1048	35.9
B	38.2359	27.5

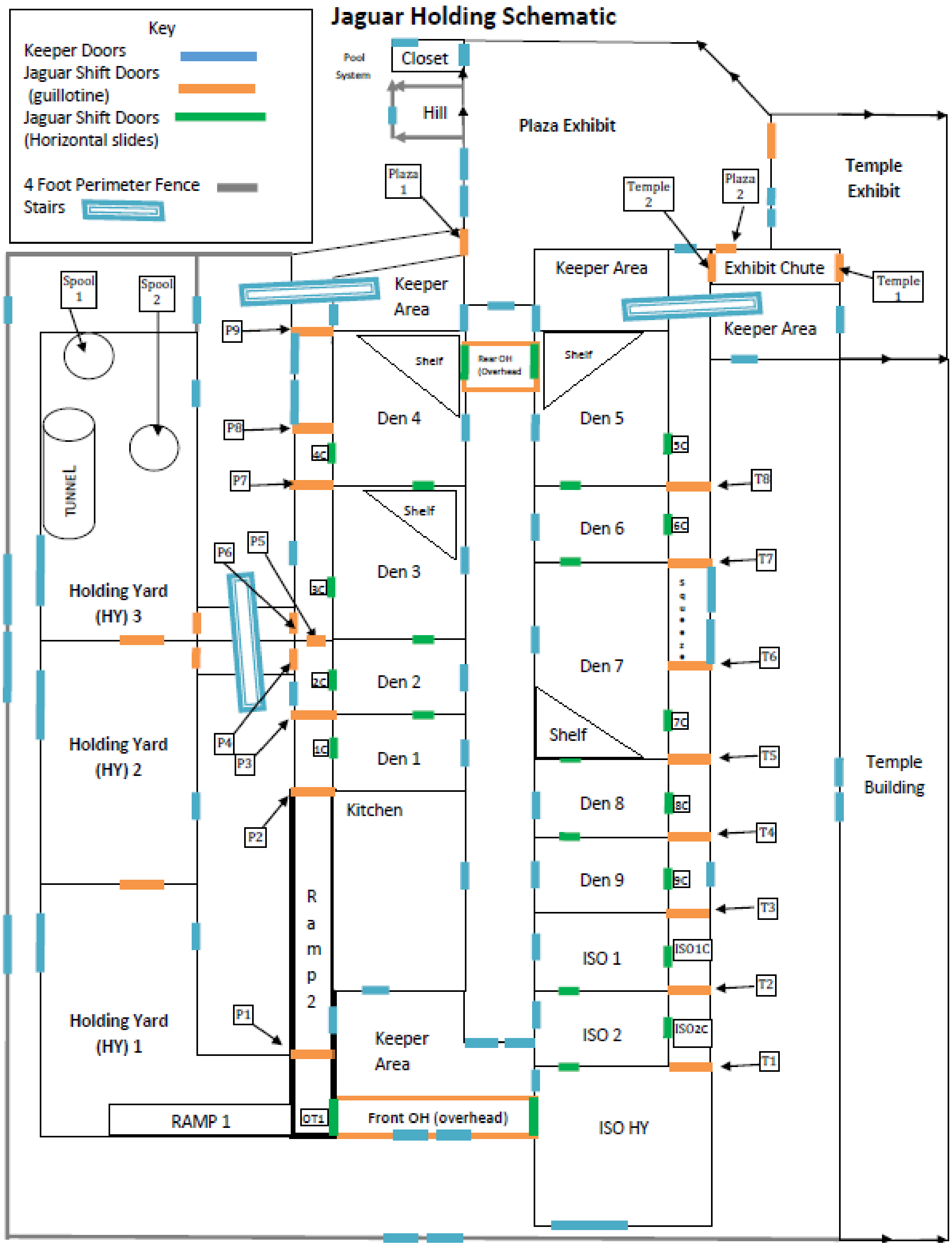

Fig1. Schematic of the jaguar holding facilities.

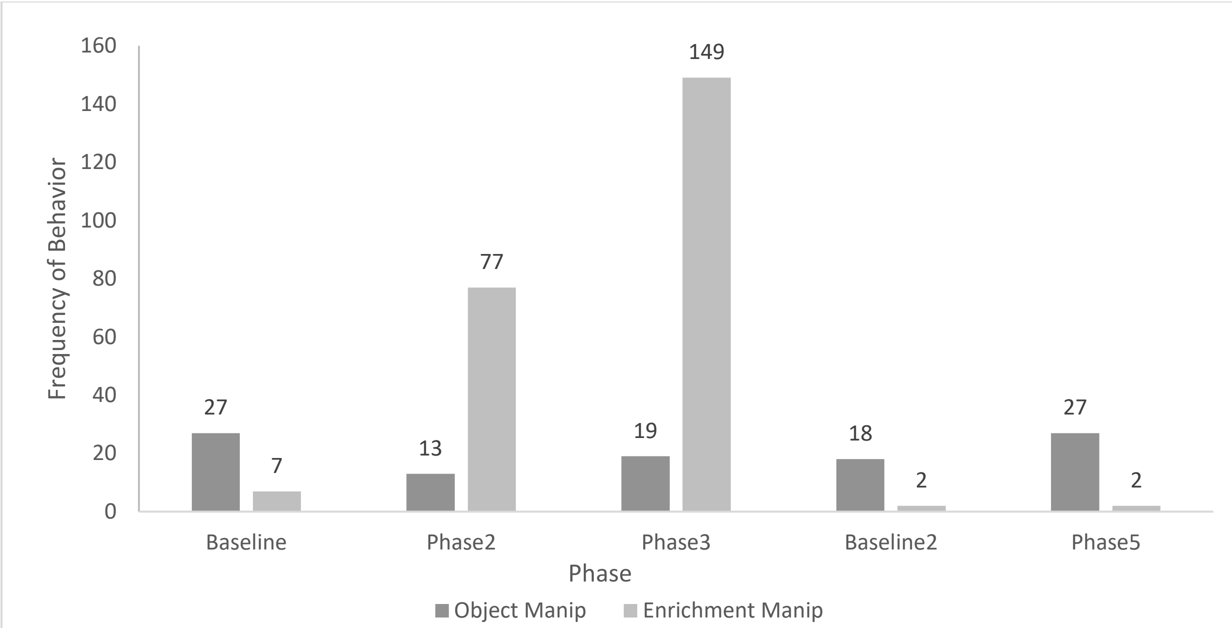

Fig 2. Frequency of manipulation of object fixed in the environment and enrichment items provided during baseline and treatment conditions

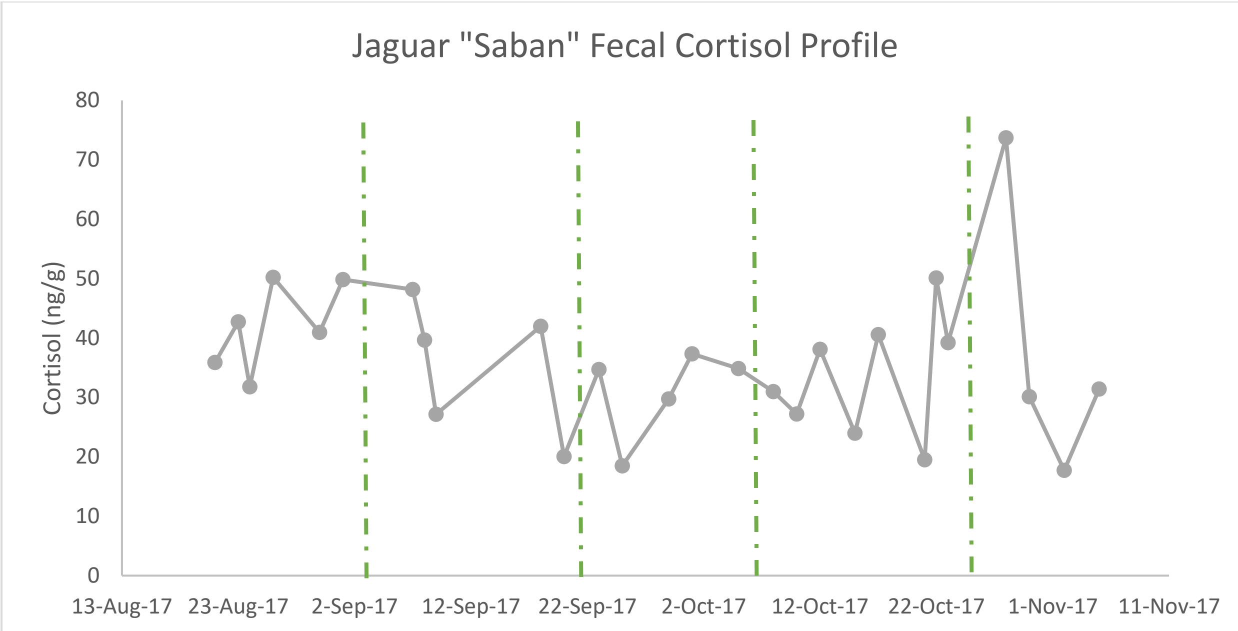

Fig 3. 1.0 Jaguar "Saban's" fecal cortisol profile throughout the 10-week study. Vertical lines represent the five different phases in the 10-week study.

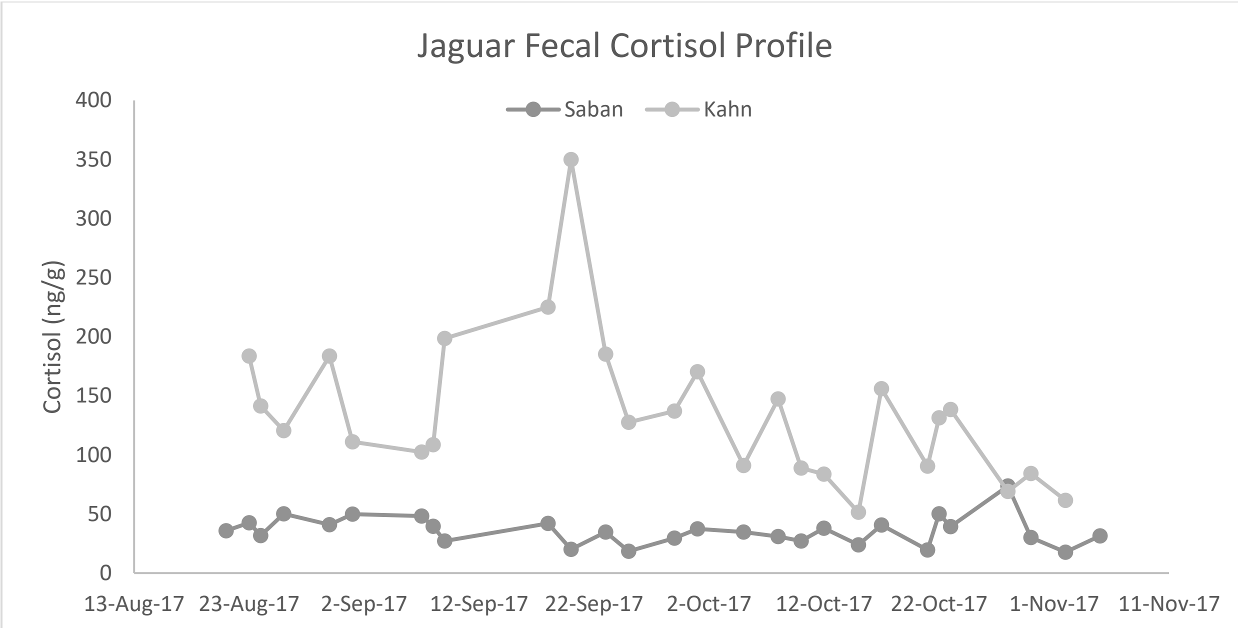

Fig 4. A comparison of cortisol levels (ng/g) between the study subject and the control subject "Kahn."

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Appendix 1

Keeper Interview Questions

1. In your experience, what time of day does Saban exhibit the most stereotypic behaviors?
2. Explain in detail what occurs 30mins prior to Saban exhibiting stereotypic pacing?
3. Explain in detail what occurs 30mins prior to Saban exhibiting stereotypic Tail-sucking?
4. Explain in detail what your actions are immediately following instances where you observe stereotypic pacing?
5. Explain in detail what your actions immediately following instances where you observe stereotypic tail-sucking
6. During what time of the day have you observed Saban being most vocal?
7. What enrichment items would you like to provide to the cats if there was no budget?
8. To your knowledge, when did stereotypic tail-sucking first occur (season, year)
9. To your knowledge, when did stereotypic pacing first occur? (season, year)
10. In your opinion, under what circumstances does tail-sucking occur?
11. In your opinion, under what circumstances does tail-sucking NOT occur?
12. In your opinion, under what circumstances does pacing occur?
13. In your opinion, under what circumstances does pacing NOT occur?
14. In your opinion, what does Saban avoid by exhibiting stereotypic pacing, e.g. presence of people, demands/requests (training sessions), sensory stimulation (loud noises, in view of other jaguars)
15. In your opinion, what does Saban avoid by exhibiting tail-sucking?, e.g. presence of people, demands/requests (training sessions), sensory stimulation (loud noises, in view of other jaguars)
16. In your opinion, what does Saban gain by exhibiting tail-sucking?, e.g. presence of people, demands/requests (training sessions), sensory stimulation (loud noises, in view of other jaguars)
17. In your opinion, what does Saban gain by exhibiting pacing?, e.g. presence of people, demands/requests (training sessions), sensory stimulation (loud noises, in view of other jaguars)
18. What steps has keeper staff taken in order to address the problem behaviors?
19. In the past, have you noticed varying levels of intensity of pacing (i.e. faster pacing, more aggressive tail-sucking)
20. In the past, have you noticed varying levels of intensity of stereotypic tail-sucking (i.e. more aggressive tail-sucking)
21. In your opinion, what conditions are most likely to precipitate or set off the behavior of pacing?
22. In your opinion, what conditions are most likely to precipitate or set off the behavior of tail-sucking?
23. To your knowledge, are there any indications that a stereotypic behavior is about to start?
24. To your knowledge, are there any indications that a stereotypic behavior is about to start?
25. What behaviors would you like to see instead of pacing?
26. What behaviors would you like to see instead of tail-sucking?

27. If you could make one meaningful change to current management protocols would it be?
28. If you could make one meaningful change to keeper practices what would it be?
29. If you could identify two distinct variables triggering pacing what would they be?
30. If you could identify two distinct variables triggering tail-sucking what would they be?

Do you have anything else you would like to share that might contribute to creating an effective behavioral intervention plan?

Appendix 2

Ethogram

Aberrant behavior

Stereotypic movement

(P) Pacing

Repetitive, apparently functionless movement, usually on a fixed route within the enclosure (after Odberg, 1978).

(TS**)** Tail-Sucking

Repetitive licking or chewing of the tail

(EG) Excessive Grooming

Determined in data analysis as greater than or equal to 75% of a single session.

Information Gathering and Learning

Attentional Behavior

(A) Alert

Animal disengages from all other activities with eyes open and aware of surroundings.

(I) Investigate

Animal is actively interested in any tangible part of its environment. Can include sniffing objects or looking at something intently. To be considered intently looking at something, animal must be within 3 meters of the object of intent. Does not include physical manipulation of an object.

Maintenance

(U/D) Urinating/Defecating

Any projection of bodily fluids (except scent-marking), includes vomiting.

(G) Grooming

Animal licking or scratching itself.

Static Poses

(I) Inactive

Animal is not engaging in any of the predetermined behaviors. Eyes can be open or closed

Nutrient acquisition

Ingest

(F) Feeding

Eating, chewing, or licking edible substances.

(D) Drinking

Ingesting water

Manipulate

(O) Object Manipulation

Interacts with or manipulates something fixed in the environment (e.g. log, bush). Does not include water play

(E) Enrichment Manipulation

Interacts with, investigates, or manipulates an enrichment item (e.g., toy)

Play

(LP) Lone Play

Engaging in playful activities alone. Can include chasing the tail, pouncing.

Owner advertisement behavior

(SM) Scent marking
Animal releases spray from posterior toward an object.
(V) Vocalize
Animal makes noise with the mouth.

Behaviors without Functional Category

(Lo) Locomote
Generalized movement within the enclosure such as walking or climbing.
(R) Rolling
Animal on one side and completely rotates to the other side while laying down.
(S) Swimming
Any activity when the animal is in the water.
(O) Other
A category which included many important behavior patterns rarely seen during scan sampling,
(Out) Out of View
Animal is not in view of the investigator or the view is obstructed by an object fixed in the environment.

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Appendix 3

Jacksonville Zoo and Garden's New Enrichment Approval and Observation Form

Jacksonville Zoo and Gardens
Behavior Husbandry Program
Enrichment Proposal Form (EPF)

Date: 12/6/16 Submitted by: Megan Morris Division: Wellness Area:

Enrichment name:

This sheet refers to:

☐ Specific species

☐ Specific individual

☐ Specific enclosure:

☐ General for order:

This enrichment will be given:

☐ On exhibit/During hours

☐ On exhibit/After hours

☐ Off exhibit/During hours

☐ Off exhibit/After hours

What is the goal of this enrichment idea?

☐ Increase activity and/or visibility

☐ Mental stimulation

☐ Encourage natural behavior: Describe –

☐ Discourage undesired behavior: Describe – Stereotypic pacing and tail-sucking

☐ Other: Describe –

Enrichment idea details:

Describe details, including materials required. –

What is the estimated cost? –

What frequency/duration will this item be given? – This enrichment device will be used specifically for the jaguar stereotypic behavior study. However, it can be used in the regular enrichment rotation after the study.

What is the estimated time involved (build, set up) –

Are there any safety concerns? –

Other important information –

Area Supervisor

Initials: ☐ Approved ☐ Not Approved ☐ Approved with changes – Describe:

Curator:

Initials: ☐ Approved ☐ Not Approved ☐ Approved with changes – Describe:

Veterinarian:

Initials: ☐ Approved ☐ Not Approved ☐ Approved with changes – Describe:

Deputy Director of Animal Care and Conservation:

Initials: ☐ Approved ☐ Not Approved ☐ Approved with changes – Describe:

Picture or Photo Attached?

☐ **Yes** ☐ **No**

Jacksonville Zoo and Gardens

Behavior Husbandry Program

Enrichment Evaluation Form

This form is to be used when a new enrichment item is presented to animals for the first two times. Conduct direct observations for at least 15 minutes each time. Transfer observations to computerized form and e-mail to supervisor upon completion. Submit original hard copy with monthly enrichment logs. For any established enrichment initiatives this form will be used at the supervisor's discretion. Initiatives that are not "items" will not be given indirect scores.

Enrichment Response Codes

Direct Observation	**Indirect Observation**
-1 = undesired or unsafe interaction (notify supervisor)	-1 = conditions indicate undesired or unsafe interaction
0 = no interaction	0 = conditions indicate no direct interaction with enrichment
1 = fear; avoidance	1 = conditions indicate direct interaction with the enrichment
2 = brief interaction or investigation; aggression	
3 = species-appropriate interaction, but not goal behavior	
4 = appropriate display of goal behavior	

Direct observations are based on visual monitoring of animal interaction with the enrichment. Indirect observations are based on condition or location of the enrichment after some time has passed without direct observation.

Enrichment Item:

Comments:

Presentation #1	
Observer:	
Date:	

Time:	
Species:	
Animals:	
Enrichment response code:	Direct: __ ___ Indirect: _ ____
Response:	Individual: ☐ Group: ☐
Ad lib observations:	

Presentation #2	
Observer:	
Date:	
Time:	
Species:	
Animals:	
Enrichment response code:	Direct: _ __ Indirect: _ ____
Response:	Individual: ☐ Group: ☐
Ad lib observations:	

Appendix 4

Fecal collection protocol

SEZARC

South-East Zoo Alliance for Reproduction & Conservation

Fecal Collection Protocol for Hormone Analysis

- A minimum of 3 morning fecal samples per week should be collected from the study subject (Mondays, Wednesdays and Saturdays).
- Collect 2-3 grams of fresh fecal material *More is not always better & adds to shipping costs, our freezer space & our garbage.*
- Sample should be fresh with as little urine, dirt, straw, & debris included as possible.
- Place sample into an airtight, leakproof, polyethylene (Whirl-Pak) bag, preferably a smaller 3×5" or 3×7" size.
- Label with minimum of animal ID & date using a black permanent marker.
- Samples should be placed in a freezer as soon as possible and within an hour of collection. A cooler with ice pack can be used when unable to get to a freezer within a reasonable period of time or on hot days.
- The recording of any aggressive or abnormal behaviors occurring throughout the study will be greatly appreciated & will assist with interpretation of the hormone assays. Please note the date and individuals involved.

Appendix 5

Cortisol (R4866) Assay

- Pre-coated, Pre-blocked plates should be stored at 4°C in a ziplock bag with desiccant.
- This assay uses plates pre-coated with 10 µg/mL Goat anti-rabbit IgG (Arbor Assays)

PREPARATION

Step 1: Take pre-coated plate and ASSAY BUFFER out of the fridge to warm to room temperature.
Step 2: Prepare all sample dilutions in Assay Buffer.
Step 3: Remove Standard Stock (1000 pg/well), Controls, and Cortisol Antibody from the freezer to thaw
Step 4: Make Standards, standard values are 1000, 500, 250, 125, 62.5, 31.2, 15.6, 7.8, 3.9 & 0.0 pg/well, by diluting standard serially 2-fold using 200 µL standard stock (1000 pg/well) &200 µL assay buffer. The stock standard at 1000 pg/well will be the top standard
Step 5: Prepare 6 mL of Cortisol (Cort) **-HRP (horseradish peroxidase ligand) at 1:100,000** in ASSAY BUFFER (1:100 HRP stock stored at 4 °C). Add 6 µL stock (1:100) to 6.0 mL assay buffer. Vortex gently.
Step 6: Prepare 6 mL of Cortisol (Cort) –**Ab at 1:25,000** in ASSAY BUFFER (1:50 Ab stock stored at -20 °C). Add 20.4 µL antibody stock (1:85) to 6 mL assay buffer. Vortex gently.

LOADING

Step 7: Load your plate with 50 µl of your standards, controls and samples in duplicate within 8 minutes. Your blank wells should get 50 µl of assay buffer.
Step 8: CAREFULLY add 50 µl of HRP to each well in < 1 minute.
Step 9: CAREFULLY add 50 µl of Ab to each well in < 1 minute. **DO NOT add Ab to Blank Wells**.
Step 10: Briefly agitate the plate (by tapping the sides gently), cover fully with plate sealer, and place on plate shaker for 1 hour. Then, measure out Substrate buffer to warm to room temperature. **DO NOT ADD ABTS YET.**

SUBSTRATE

Step 11: Wash the plate 4 times with 300 µl 1X WASH buffer. Leave WASH buffer in the wells until ready to add substrate. When ready, dump and blot on paper towels.
Step 12: Prepare ABTS substrate immediately before use
Step 13: Combine 125 µL 40 mM ABTS, 40 µL 0.5M H_2O_2, & 12.5 mL substrate buffer. Mix well.
Step 14: Add 100 µL substrate to all wells using either repeater or multichannel pipette.
Step 15: Replace plate sealer.

STOP & READ

Step 16: Read the plate at 450 nm wavelength.
Step 17: Incubate at room temp until zero standards read about 1.000 OD

Appendix 6

Demographics

1. Your Name
2. Your Institution
3. Your position within your institution
4. Your email address
5. Please read carefully and select an option

   I consent to allowing my responses to be used for broader research purposes

   I do not want my responses to this survey to be used for broader research purposes

Collection and Housing

6. How many jaguars does your institution house? Please indicate the sexes of your individuals using common zoo notations (E.g. 1.2= one male, two females)
7. Please Provide the animal ID for all jaguars housed at your institution. The studbook number or local accession number are acceptable, but studbook number is preferred.The label (Jaguar 1, 2 etc.) associated with each individual will be used throughout the survey, so please be sure to remember which label you have given each animal in your collection.

Pacing

8. Do any of the individuals housed at your institution engage in * stereotypic pacing?

   Yes

   No

Other (please specify)

9. Please indicate which individuals engage in stereotypic pacing

   Jaguar 1

   Jaguar 2

   Jaguar 3

   Jaguar 4

   Jaguar 5

   Jaguar 6

   Jaguar 7

   Jaguar 8

10. What are your immediate actions when you see a jaguar engaging in stereotypic pacing?
11. In your experience, what time of day do your jaguars exhibit the most stereotypic pacing?

    Early morning (Before 10am)

    Late morning (10:00am-12:00pm)

    Early afternoon (12:00-3:00pm)

    Late afternoon (After 3pm)

12. Please describe environmental conditions immediately before pacing is observed (e.g. cold/hot weather,unfamiliar noise, etc.)

13. Is the animal gaining something by engaging in * stereotypic pacing?

    Yes

    No

    Unsure

    Other (please specify)

14. Is the individual attempting to avoid something by engaging in stereotypic pacing?

    Yes

    No

    Unsure

Other (please specify)

15. Have you observed any physical/mental health deficits as a result of stereotypic pacing? Please select all that apply

Hair loss
Open wounds
Bloody feet
I have not observed any physical/mental health deficits as a result of stereotypic pacing
Other (please specify)

16. What steps has keeper staff taken to address stereotypic pacing?

Tail-sucking

17. Do any of the individuals housed at your institution engage in stereotypic * tail-sucking?

Yes
No
Other (please specify)

18. Please indicate which individuals engage in stereotypic * tail-sucking?

Jaguar 1
Jaguar 2
Jaguar 3
Jaguar 4
Jaguar 5
Jaguar 6
Jaguar 7
Jaguar 8

19. What are your immediate actions when you see a jaguar engaging in stereotypic tail-sucking? (e.g. offer enrichment, shift to another area etc.)

20. In your experience, what time of day do your jaguars exhibit the most stereotypic tail-sucking?

Early morning (Before 10am)
Late morning (10:00am-12:00pm)
Early afternoon (12:00-3:00pm)
Late afternoon (After 3pm)

21. Please describe environmental conditions immediately before tail-sucking is observed (e.g. cold/hot weather, unfamiliar noises etc.)

22. Is the animal gaining something by engaging in stereotypic * tail-sucking?

Yes
No
Unsure
Other (please specify)

23. Is the individual attempting to avoid something by engaging in stereotypic tail-sucking?

Yes
No
Unsure
Other (please specify)

24. Have you observed any physical/mental health deficits as a result of stereotypic pacing? Please select all that apply

Hair loss
Open wounds

I have not observed any physical/mental health deficits as a result of stereotypic pacing
Other (please specify)

25. What steps has keeper staff taken to address stereotypic tail-sucking?

Excessive Grooming

26. Do any of the individuals housed at your institution engage in * excessive grooming?
    Yes
    No
    Other (please specify)
27. Please indicate which individuals engage in * excessive grooming?
    Jaguar 1
    Jaguar 2
    Jaguar 3
    Jaguar 4
    Jaguar 5
    Jaguar 6
    Jaguar 7
    Jaguar 8
28. What are your immediate actions when you see a jaguar engaging in excessive grooming? (E.g. offer enrichment, shift to another area etc.)

29. In your experience, what time of day do your jaguars exhibit the most excessive grooming?
    Early morning (Before 10am)
    Late morning (10:00am-12:00pm)
    Early afternoon (12:00-3:00pm)
    Late afternoon (After 3pm)
30. Please describe environmental conditions immediately before excessive grooming is observed (e.g. cold/hot weather, unfamiliar noises etc.)
31. Is the animal gaining something by engaging in * excessive grooming?
    Yes
    No
    Unsure
    Other (please specify)
32. Is the individual attempting to avoid something by engaging in excessive grooming?
    Yes
    No
    Unsure
    Other (please specify)
33. Have you observed any physical/mental health deficits as a result of excessive grooming? Please select all that apply
    Hair loss
    Open wounds
    I have not observed any physical/mental health deficits as a result of stereotypic pacing
    Other (please specify)
34. What steps has keeper staff taken to address excessive grooming?

Vita, Megan C. Morris

EDUCATION:

University of Illinois at Urbana-Champaign
- Bachelor of Science in Animal Science: Concentration in Companion Animal and Equine Science

University of Illinois Exchange Program: University of Western Australia
  Perth, Western Australia.

University of North Florida
- Masters of Science in General Psychology

PROFESSIONAL EXPERIENCE:

Animal Behavior Specialist, Jacksonville Zoo and Gardens	Jul 2017-Present
Keeper I Mammals, Zoo Atlanta	Apr 2015-Aug 2015
Seasonal Keeper: Mammals, Zoo Atlanta	Aug 2014- Apr 2015
Carnivore/Panda Keeper Intern, Zoo Atlanta	May 2013-Apr 2014
Champaign County Humane Society: Dog/Cat Handler	2011-May 2013

RESEARCH EXPERIENCE:

Jacksonville Zoo and Gardens Research Fellow	Aug 2015-Present
NSF New Biology Fellowship: Undergraduate Research and Mentoring	May 2012-Dec 2013
Summer Research Opportunities Program	May 2012-Aug 2012

CONFERENCES

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PUBLICATIONS

Tennant, K. S., Segura, V. D., **Morris, M. C.,** Snyder, K. D., Bocian, D., Maloney, D., & Maple, T. L. (2017). Achieving optimal welfare for the Nile hippopotamus (*Hippopotamus amphibius*) in North American zoos and aquariums. *Behavioural processes*.