American Journal of Primatology 70:490–494 (2008)

BRIEF REPORT

The Effects of Observer Presence on the Behavior of *Cebus capucinus* in Costa Rica

KATHARINE M. JACK[1*], BRYAN B. LENZ[1], ERIN HEALAN[1], SARA RUDMAN[1], VALÉRIE A.M. SCHOOF[1], AND LINDA FEDIGAN[2]
[1]*Department of Anthropology, Tulane University, New Orleans, Louisiana*
[2]*Department of Anthropology, University of Calgary, Calgary, Alberta, Canada*

We report on the responses of *Cebus capucinus* in the Santa Rosa Sector of the Area de Conservación Guanacaste, Costa Rica, to the presence of observers over a 4-week period. Study groups were habituated to different degrees: (1) Cerco de Piedra (CP): continuous observations began in 1984; (2) Exclosure (EX): focus of an 18-month study on males from 1998 to 1999; and (3) NBH: never studied/followed but the group frequently encounters researchers. We collected three types of data: group scans (group state was coded as calm or agitated at observer presence), focal animal data (observer-directed behaviors were recorded), and fecal cortisol levels. The two less-habituated groups (NBH and EX) differed significantly from the habituated group (CP) in their behavioral and cortisol responses, and they showed an increase in habituation over the study period (agitation and cortisol levels both dropped). Individuals in NBH also decreased their responses to observers during focal follows; however, at the end of the study the responses of the two less-habituated groups (NBH and EX) remained elevated in comparison to the habituated group (CP), suggesting the need for further habituation. Unlike capuchin groups that rarely encounter humans, NBH and EX never fled from observers and they rarely emitted observer-directed alarm calls. We suggest that the permanence of habituation and the ability to habituate animals passively through a neutral human presence are both important considerations for researchers conducting studies in areas where animal safety from poachers, etc. cannot be guaranteed. Am. J. Primatol. 70:490–494, 2008.

Key words: habituation; *Cebus*; passive habituation; fecal cortisol; stress; observer effects

INTRODUCTION

Habituation, defined as the reduction in the attention directed toward human observers following repeated neutral contacts [Williamson & Feistner, 2003], is the first step in most primatological field research. Despite its importance and long-standing interest in the topic [e.g. Wrangham, 1974], we know little about the process of habituation and its short- and long-term effects on the animals that we study. These types of data can provide researchers with tips on habituation methodology, improve estimates of habituation time [Williamson & Feistner, 2003], and have potential conservation applications [e.g. eco-tourism; Blom et al., 2004; Werdenich et al., 2003]. Habituation research is also important because of the potential negative effects that habituation can have on wild primates. For example, habituation can increase the susceptibility of primates to disease by bringing them into close proximity to humans [Engel et al., 2006; Williamson & Feistner, 2003] or induce stress responses that may act as immunosuppressants [Woodford et al., 2002]. Habituation also increases the potential for poaching, a problem that can intensify when researchers are not present [Dupain et al., 2000; Plumptre et al., 2001]. To contribute to the literature addressing habituation, we examined the effects of observer presence on three groups of wild white-faced capuchins (*Cebus capucinus*) in Costa Rica.

Each of the study groups was previously habituated to different degrees: (1) Cerco de Piedra (CP) was completely habituated. Behavioral studies of this group began in 1984 and continue to the present with observers present year round. (2) Exclosure (EX) was somewhat habituated as this group was the focus of an 18-month behavioral study on males from

Contract grant sponsors: Tulane University; Newcomb College at Tulane University; Stone Center for Latin American Studies at Tulane University; NSERC and the Canada Research Chairs Program.

*Correspondence to: Katharine M. Jack, Department of Anthropology, Tulane University, 1326 Audubon Street, New Orleans, LA 70118, USA. E-mail: kjack@tulane.edu

Received 21 May 2007; revised 2 November 2007; revision accepted 5 November 2007

DOI 10.1002/ajp.20512
Published online 12 December 2007 in Wiley InterScience (www.interscience.wiley.com).

1998 to 1999 [Jack, 2001]. (3) North Bosque Humedo (NBH) was the least habituated of the three groups. Although NBH had not previously been studied or followed by researchers, they and EX frequently encounter researchers working on other projects in their home ranges (i.e. ongoing research on neighboring capuchins groups by Fedigan and Jack, *Ateles geoffroyi* by F. Aureli, and various research projects by D. Janzen). We predict that these varying levels of previous exposure to observers will lead to differences in observer-directed responses by individuals residing in the three groups: individuals in CP should display the lowest levels of observer-directed behaviors, NBH should display the highest, and EX should be intermediate. We also predict that the observer-directed responses of individuals in EX and NBH will decrease with repeated exposure to observers, whereas CP will show no change. Additionally, stress levels, measured through fecal cortisol [Whitten et al., 1998], are predicted to decrease for NBH and EX over the study and fluctuate little for CP.

METHODS

The study took place in the Santa Rosa Sector of the Area de Conservación Guanacaste, Costa Rica, where L.M.F. began her observations of *C. capucinus* in 1983 [for additional information, see Fedigan & Jack, 2001]. Data were collected over a 4-week period between May 25 and June 23, 2005. Capuchins reside in multimale, multifemale groups characterized by female philopatry and male dispersal. Males reside in many different groups throughout their lives, and our groups experience complete changeovers in the adult male membership approximately every 4 years [Fedigan & Jack, 2004]. During the study period CP was composed of 17 individuals (three adult males, four adult females, and ten immatures), EX contained seven individuals (one adult male, three adult females, and three immatures), and NBH was the largest group with a minimum of 21 individuals (six adult males, five adult females, 10+ immatures; all adults were identified).

Over the habituation period, we logged 25.5, 50.5, and 60 contact hours with CP, EX, and NBH, respectively. We spent less time with our habituated group (CP) than with the other two groups because our desire was to habituate EX and NBH as new study groups. For several of our analyses we divided the study period into two phases: phase 1 represents the first half of our contact time with each group and phase 2 represents the second half of our contact time. Three types of data were collected: (1) group scans, (2) focal animal, and (3) fecal cortisol.

(1) Instantaneous group scans were conducted at 5-min intervals during which we recorded whether the group was calm or agitated; the group was coded as agitated if at least one individual directed aggression toward observers (i.e. threat faces, threat displays, or alarm calls). We computed the proportion of scans scored as agitated for a given group and day and then used an arcsine square root transformation of these proportions. We conducted a covariance analysis using R's glm procedure with normal errors to test if this proportion differed between groups, changed with time, and whether the slope of the relationship with time differed among groups.
(2) Ten-minute focal animal samples (131 in phase 1 and 111 in phase 2) were collected during which we recorded all instances of observer-directed behaviors (look, watch, threat, and alarm call). All focal animals were adults and could be individually identified; we sampled six in CP (three females and three males), all four adults in EX (three females and one male), and nine in NBH (four females and five males). For each individual we computed the log-transformed ratio of the number of observer-directed behaviors per hour in phase 2 of the study to those in phase 1; the log of the ratio of before/after data is one of the acceptable ways of dealing with measures of relative change [Crawley, 2005; p 514]. We conducted a two-way analysis of variance using R's glm procedure with normal errors, where we examined whether the ratio was a function of group, sex, and the interaction between group and sex. We partitioned the 2 degrees of freedom of the group effect into two planned orthogonal contrasts. The first compared NBH to EX, the two groups that we expected to be relatively similar. The second combined NBH and EX and compared them to CP.
(3) Fecal cortisol levels. A total of 62 fecal samples were used in these analyses, including 23 (phase 1 = nine, phase 2 = 14) from seven individuals in CP (three females and four males), 15 (phase 1 = eight, phase 2 = seven) from the four adults in EX (three females and one male), and 24 (phase 1 = 11, phase 2 = 13) from six individuals in NBH (one female and five males). With the exception of two samples collected on our first day of contact with NBH and EX, fecal samples were collected following 1 or 2 consecutive days of exposure to observers and thereby reflect post-contact cortisol levels. Samples were collected opportunistically from known individuals and stored with an ice pack in an insulated bag until they could be frozen at the field station. Initial field extraction of cortisol, using solid phase extraction cartridges, followed Strier and Ziegler [1997]. Samples were shipped to the Wisconsin National Primate Research Center for analysis. For each individual included in this

portion of the study, we calculated average fecal cortisol for phases 1 and 2. Because cortisol levels are known to vary due to sex, reproductive status [Keay et al., 2006], and dominance rank [Jack & Schoof, unpublished data], we computed the log of the ratio between the cortisol measures in phase 2 of the study to those in phase 1 for each individual. Data were analyzed in the same way as the focal animal data described above. All research conducted during this study complied with protocols approved by Tulane University's IACUC and adhered to the legal requirements of Costa Rica.

RESULTS

Scan Data

There were differences among groups in the slopes of the relationship between the proportion of scans coded as agitated and time ($F_{2,20} = 3.58$, $P = 0.046$). The proportion of scans coded as agitated decreased markedly through time for NBH, slightly for EX, and increased slightly for CP (Fig. 1). There were also significant main effects of the groups ($F_{2,20} = 18.37$, $P < 0.001$), with NBH more likely to be agitated than EX ($t = -2.803$, df = 20, $P = 0.011$), and both NBH and EX more likely to be agitated than CP ($t = -3.179$, df = 20, $P = 0.005$).

Focal Data

There was no effect of sex on the rates of observer-directed behaviors during phases 1 and 2 ($F_{1,15} = 1.07$, $P = 0.316$), but the effect of group was significant ($F_{2,15} = 3.89$, $P = 0.043$). Planned comparisons showed that while individuals in NBH and EX did not differ significantly in their response to observers in phases 1 or 2 ($t = 0.933$, df = 15, $P = 0.336$), collectively their responses did differ from those of CP ($t = 2.215$, df = 15, $P = 0.042$). Overall, NBH displayed a decrease in their rate of observer-directed behaviors from phases 1 to 2, EX showed little change, and CP experienced an increase (Fig. 2).

Cortisol Data

The sexes did not differ in their log-transformed ratio of phases 2 to 1 cortisol levels ($F_{1,13} = 0.034$, $P = 0.855$) but there was a strong group effect ($F_{2,13} = 4.26$, $P = 0.038$). Planned comparisons showed no difference in the direction of change in cortisol levels for individuals in NBH and EX ($t = -0.393$, df = 13, $P = 0.701$), as levels declined from phases 1 to 2 for both groups. However, individuals in CP did differ from those in NBH and EX combined ($t = 2.92$, df = 13, $P = 0.012$) as they showed an increase in average cortisol levels from phases 1 to 2 (Fig. 3).

DISCUSSION

During our park-wide censuses [e.g. Fedigan & Jack, 2001], we encounter groups of capuchins that only rarely meet humans. These are truly unhabituated groups—they respond to our presence with alarm calls, extreme vigilance, and immediate flight. On the basis of these experiences, we did not anticipate that our task of habituating the new study groups, NBH and EX, would proceed as smoothly as it did. Although these new groups, NBH in particular, responded aggressively to our presence during the early part of the study, they never fled at our approach, and observer-directed alarm calls were only recorded twice (once for each new group) throughout the study. We suggest that neutral exposure to humans likely facilitated the habituation of the new study groups. Although NBH has never been the focus of a behavioral study and EX has not been studied since 1999, both of these groups range in areas that are heavily utilized by researchers in

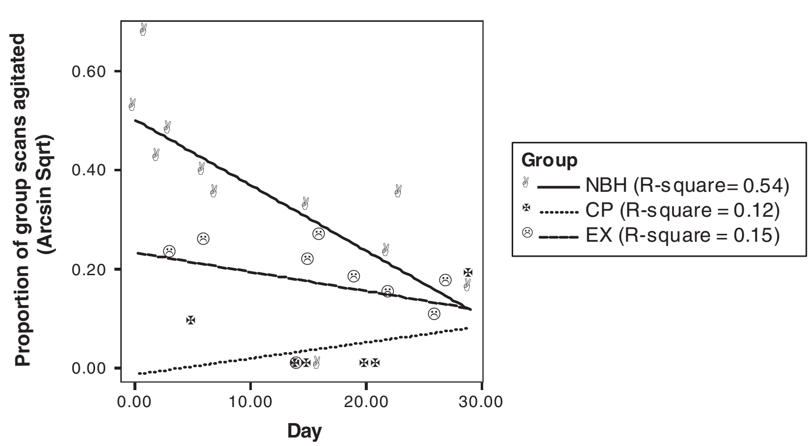

Fig. 1. Linear regression of the relationship between the proportion of group scans coded as agitated and habituation time for each of the three study groups.

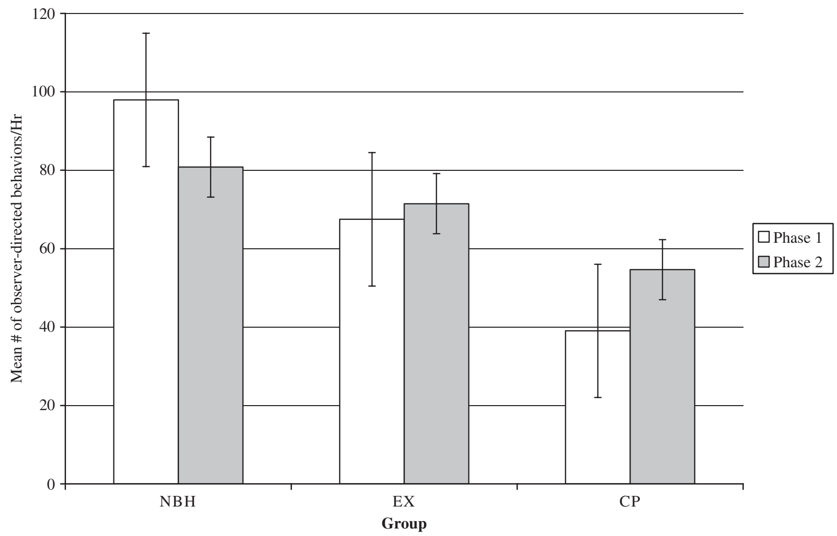

Fig. 2. Comparison of the mean rate/hour of observer-directed behaviors (look, watch, threat, alarm call) between phases 1 and 2 of the study.

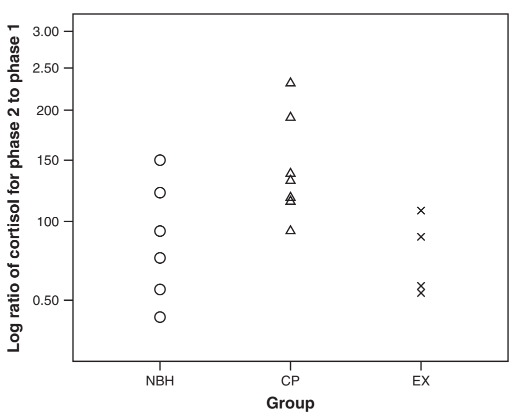

Fig. 3. Log ratio of individual cortisol responses from phases 2 to 1 for three groups of *Cebus capucinus*.

Santa Rosa. We further suggest that the presence of previously habituated individuals in both NBH and EX may have acted as a buffer to speed up the process of habituation in these new study groups [Vercauteren Drubbel et al., 1998; Williamson & Feistner, 2003]. The three EX adult females were present in the group when it was studied in 1998 and 1999 [Jack, 2001]. Although the sole resident male was new to the group and appeared unhabituated, the relatively low levels of observer responses by the EX females may have acted to facilitate the male's acceptance of our presence. Likewise, in NBH there was an old adult female that we suspect was a member of a small group also included in the 1998–1999 study and an older male who appeared to have been previously habituated, perhaps as a member of one of our long-term study groups. These observations illustrate the relative permanence of habituation—the familiar females in NBH and EX had not been intensively followed for 6 years. Our finding that repeated exposure to humans who are not actually studying/following the monkeys may be serving to *passively habituate* animals and the apparent permanence of habituation are both extremely important considerations for researchers embarking on projects in areas where primates are, or will be, at risk owing to human activity.

Our predictions that the three groups would differ in their behavioral and stress responses and that the responses of the two less-habituated groups would decrease over the study period were supported in this study. In particular, the data collected during group scans showed that after only 4 weeks of repeated neutral exposure to observers the frequency of observer-directed aggression by our less-habituated groups (NBH and EX) was similar to that of our long-term study group (CP) (Fig. 1). However, in terms of observer-directed behaviors recorded during focal animal sampling (Fig. 2), only NBH showed a decrease over the habituation period and the frequency of these behaviors (mainly in the form of looks and watches during phase 2) remained higher for both NBH and EX in comparison to CP. Consequently, 2 additional months were spent with NBH and EX before they were habituated enough to include them in our behavioral studies.

Woodford et al. [2002] suggest that the process of habituation is likely to induce stress in the target animals, potentially leading to decreased reproductive success or disease owing to immunosuppression. However, the extent to which researcher presence is inducing stress, and whether or not this decreases as habituation continues, has not been previously tested in wild primates [for review, see Keay et al., 2006]. The cortisol levels of individuals in NBH and EX, our least habituated groups, decreased over the study period indicating that observer presence does cause some stress to animals in the early phases of habituation. However, because these groups had been passively habituated before the start of our study, it is possible that stress responses would be more intense in truly unhabituated groups with no prior, or negative, exposure to humans.

Although our long-term study group, CP, displayed much lower levels of observer-directed responses on all measures, the fact that they are still responding to our presence after more than 20 years of observation is of interest. As primatologists, we are aware that we influence certain aspects of the lives of the animals we study, such as susceptibility to predation [Isbell & Young, 1993] or the outcome of inter-group aggression when unhabituated groups are encountered [Jack, personal observation]; however, we often fail to consider the more subtle effects that our presence may have. Although the habituation process appears to induce both behavioral and stress responses in *C. capucinus*, these responses show a marked decrease over a short period of time in the groups studied. Nonetheless, our finding that observer-directed behaviors were not completely extinguished even in our most habituated study group indicates that we can never become a truly neutral presence to our study animals.

ACKNOWLEDGMENTS

This research was made possible through start-up funds to K.M.J. from Tulane University, grants to S.R., E.H. and K.M.J. from Newcomb College at Tulane University, and grants to K.M.J. and B.L. from the Stone Center for Latin American Studies at Tulane University. L.M.F. is supported by NSERC and the Canada Research Chairs Program. We are thankful to the National Park system of Costa Rica for permitting us to conduct long-term research on the Santa Rosa capuchins, and we are particularly grateful to Roger Blanco Segura (Research Director for the Area de Conservación Guanacaste) for his ongoing support of our research. Special thanks to John Addicott for statistical consulting and to Toni Ziegler and Dan Wittwer at the WNPRC for cortisol analyses. The comments and suggestions of two anonymous reviewers greatly improved this manuscript. All research reported here was conducted in compliance with Tulane University's IACUC and the legal requirements of Costa Rica.

REFERENCES

Blom A, Cipolletta C, Brunsting AMH, Prins HHT. 2004. Behavioral responses of gorillas to habituation in the Dzanga-Ndoki National Park, Central African Republic. Int J Prim 25:179–196.

Crawley MJ. 2005. Statistics: an introduction using R. New York: Wiley. 342p.

Dupain J, Van Krunkelsven E, Van Elsacker L, Verheyen RF. 2000. Current status of the bonobo (*Pan paniscus*) in the proposed Lamoko reserve (Democratic Republic of Congo). Biol Conserv 94:265–272.

Engel G, Hungerford LL, Jones-Engel L, Travis D, Eberle R, Fuentes A, Grant R, Kyes R, Schillaci M. 2006. Risk assessment: a model for predicting cross-species transmission of simian foamy virus from macaques (*M. fascicularis*) to humans at a monkey temple. Am J Primatol 68:934–948.

Fedigan LM, Jack K. 2001. Neotropical primates in a regenerating Costa Rican dry forest: a comparison of howler and capuchin population patterns. Int J Primatol 22: 689–713.

Fedigan LM, Jack K. 2004. The demographic and reproductive context of male replacements in *Cebus capucinus*. Behavior 141:755–775.

Isbell LA, Young TP. 1993. Human presence reduces leopard predation in a free-ranging population of vervet monkeys (*Cercopithecus aethiops*). Anim Behav 45:1233–1235.

Jack KM. 2001. Life history of male white-faced capuchins (*Cebus capucinus*). Ph.D. dissertation. University of Alberta.

Keay JM, Singh J, Gaunt MC, Kaur T. 2006. Fecal glucocorticoids and their metabolites as indicators of stress in various mammalian species: a literature review. J Zoo Wildl Med 37:234–244.

Plumptre, AJ, Masozera M, Vedder A. 2001. The impact of civil war on the conservation of protected areas in Rwanda. Washington, DC: Biodiversity Support Program.

Strier KB, Ziegler TE. 1997. Behavioral and endocrine characteristics of the reproductive cycle in wild northern muriquis, *Brachyteles arachnoides*. Am J Primatol 42:299–310.

Vercauteren Drubbel R, Gautier JP, Fleury MC, Martinez B. 1998. Difficulties of observing forest dwelling primates and anti-predator strategies. Congress Int Primatol Soc, Abstracts 17:141.

Werdenich D, Dupain J, Arnheim E, Julve C, Deblauwe I, Van Elsaker L. 2003. Reactions of chimpanzees and gorillas to human observers in a non-protected area in south-eastern Cameroon. Folia Primatol 74:97–100.

Whitten PL, Stavisky R, Aureli F, Russell E. 1998. Responses of fecal cortisol to stress in captive chimpanzees (*Pan troglodytes*). Am J Primatol 44:57–69.

Williamson EA, Feistner ATC. 2003. Habituating primates: processes, techniques, variables and ethics. In: Setchell JM, Curtis DJ, editors. Field and laboratory methods in primatology: a practical guide. Cambridge: Cambridge University Press. p 25–39.

Wrangham RW. 1974. Artificial feeding of chimpanzees and baboons in their natural habitat. Anim Behav 22:83–93.

Woodford MH, Butynski TM, Karesh WB. 2002. Habituating the great apes: the disease risks. Oryx 36:153–160.