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Research Article

Determinants of Grooming Behavior in Captive Langurs under Human Disturbance: A Case Study from the Central Zoo, Kathmandu, Nepal

Shailendra Sharma, Smriti Shrestha, Naresh Pandey and Laxman Khanal*

Central Department of Zoology, Institute of Science and Technology, Tribhuvan University, Kathmandu, Nepal.

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LK conceptualized the study. SS performed the behavioral sampling. SS, SS and NP analyzed data and prepared the manuscript. LK supervised the research and finalized the manuscript. All authors approved the manuscript for submission.

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Activity budget, Captive primates, Ethogram, Focal animal sampling, Social behavior

Abstract | Social behavior among primates is crucial to their health and survival. Captive primates under human disturbance are often stressed, which disrupts their social interactions. Therefore, identifying the factors affecting social behavior is important for managing animals in captivity. This study investigated whether visitors' number and disturbance affect the grooming frequency and duration of the langurs (*Semnopithecus* sp.) housed in Central Zoo, Kathmandu, Nepal. Further, it tested if the effects were biased to the age and sex of the animals. Behavioral data were collected through the focal animal sampling method for 101 hours, and human disturbances were categorized into four levels based on the number and behavior of visitors. Out of the entire activity budget, resting (49.27%) constituted the highest proportion, followed by feeding (22.93%) and grooming (12.59%). There was a negative association between the grooming behavior and the number of visitors; however, no significant difference was observed in grooming proportion before and after visits. Higher levels of disturbance had a significant negative effect on the grooming frequency and time investment. The non-lactating adult female delivered higher grooming to the troop members, while the sub-adult male received more grooming than others. We conclude that the nature of disturbance rather than the number of visitors affects social bonding among the captive langurs.

Novelty Statement | This study categorized the zoo visitors' disturbance into different levels and tested its effects on the grooming behavior of captive langurs against age/sex categories.

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Introduction

Sociality measures the degree to which animals interact and form long-term or transient associations (Ward and Webster, 2016). Social interactions develop when the net benefits of close associations with group members exceed the costs (Krause and Ruxton, 2002). Such interactions are of various types, such as playing, huddling, sexual behavior, affiliative behavior (grooming), aggressive behavior, etc. (Kaplan *et al.*, 1991; Khan, 2013). Socio-ecological and hormonal factors are the major drivers of sex-difference in social behavior performance among a wide range of animal species (Meredith, 2013). Social behavior (e.g., grooming) is performed for strong bonding among the group members (social bonding hypothesis) (Arlet *et al.*, 2015), cope with stress (social stress hypothesis) (Marty *et al.*, 2019), and as an indication of willingness to interact (immediate investment hypothesis) (Allanic *et al.*, 2020).

Corresponding author: Laxman Khanal
lkhanal@cdztu.edu.np

Besides these, social interactions are also performed for safety from predators, collaborative hunting, reproductive success, reduction of parasitic abundance among the members of the society, etc. (Alexander, 1974; Robinson *et al.*, 2008). Therefore, social interactions have important roles in the health, survival and reproduction of group-living animals.

Among social animals, primates are best known to exhibit social interactions that act as a glue for binding members of the troop together (Minhas *et al.*, 2010). Social bonds in primates are often expressed in the form of grooming, with females engaged more in social grooming or social interaction than males (Mitchell and Tokunaga, 1976; Kulic *et al.*, 2015). Auto-grooming helps in thermoregulation, elimination of dirt and parasites, and self-soothing, whereas allo-grooming plays an essential role in animal hygiene and group cohesion (Grueter *et al.*, 2013). The social rank of the animal, proximity to other individuals, and climatic factors (i.e., temperature, humidity, etc.) cause variations in grooming behavior in primates (Borries, 1992; Borries *et al.*, 1994; Kaburu *et al.*, 2019; Marty *et al.*, 2019; Li *et al.*, 2020; Caselli *et al.*, 2021). The level of social interaction among primates differs between wild and captive conditions as the latter sets many limitations in activities. Maintaining natural environmental conditions inside the captive setting is crucial for the normal development of the animals, but controlling every aspect of such natural environments within limited spaces is difficult (Lutz and Novak, 2005). Animals start to develop abnormal behavior to cope with the adverse situation inside the captive setting and as a result, the expression of social behavior is reduced significantly (McPhee and Carlstead, 2010). Lack of natural surroundings, multiple mates, social set up and appropriate substrate, routine husbandry, limited spaces, and scheduled diet are the major reasons for stressful situations and alteration of social behavior among captive animals (Morgan and Tromborg, 2007; Khan, 2013). Crowding among captive primates due to high population density contributes to overexpression of aggressive behavior, which diminish social interactions (Nieuwenhuijsen and De Waal, 1982). Additionally, visitors' activity level outside predicts visitor-directed aggression by the zoo monkeys more than the size of the crowd (Woods *et al.*, 2019).

Human disturbances along with potential food availability affects the pattern of grooming behavior in primates (Jasso *et al.*, 2020). A wide range of potentially impactful activities are performed by humans, directly or indirectly affecting a variety of non-human primates' behavior (including social behavior) in captivity or in the wild (Marty *et al.*, 2019). Positive human interaction with primates works as environmental enrichment inside the captivity which promote social behavior performance and reduce abnormal behavior triggered by high human pressure (Baker, 2004). Some studies in captive setting reported the number of visitors significantly affect the performance of social behavior and aggressive behavior. For example, in the presence of visitors, social behavior in captive primates is reduced by almost half whereas aggressive behavior is increased rapidly (Chamove *et al.*, 1988; Wells, 2005). Other behaviors like social support, avoidance, aggression, displacement, self-scratching and social affiliation are performed by monkeys to avoid tourist pressure (Maréchal *et al.*, 2011; Marechal *et al.*, 2016).

Langurs are colobine monkeys that are widely kept in zoos and are one of the major attractions to visitors (Little and Sommer, 2002; Sha *et al.*, 2014). Hanuman/Himalayan langurs (*Semnopithecus* sp.) are shy, folivores and canopy dwellers living in multimale-multifemale social groups in the wild (Kavana *et al.*, 2015; Khanal *et al.*, 2018; Dasgupta *et al.*, 2021). Behavioral studies in langurs have been carried out in the wild but are rarely studied in captive conditions. Grooming constitutes one of the major behavioral activities of langurs in their natural habitats (Ahsan and Khan, 2007). However, it has never been documented under the ecological limitations imposed by the captive conditions. Therefore, this study aimed to assess the proportion of the grooming behavior in the overall time budget and the impacts of visitor's number and disturbance levels on the grooming behavior. Using the focal animal sampling method (Altmann, 1974), it tested for the age-sex dependent variation in grooming behavior of captive langurs.

Materials and Methods

Study animals and housing condition

Behavioral observation was carried out on a cohort of six langurs (Table 1) housed in the Central Zoo, Kathmandu, Nepal. The cohort constituted two adult females rescued from the wild and two males born inside the enclosure. A subadult male in the cohort is the child of the non-lactating female and an infant (approximately 3–4 months of age) is the child of lactating female (personal communication with Lina Chalise, Conservation Education and Information Officer, Central Zoo, Lalitpur, Nepal). The 10×20 m^2 enclosure housing animals was made of iron wired mesh on three sides and a concrete walled room on one side. Physical enrichment of the cage included fallen logs and hanging ropes. On two sides of the cage, the langurs were exposed to visitors who could feed and tease them. The zoo management feeds the animals three times a day (morning, afternoon and evening), and water is provided *ad libitum*.

Behavioral data collection

Behavioral observations were done between 7:00 AM and 3:00 PM from 14 to 30 March, 2022. Captive langurs were observed three hours in the morning during the

complete absence of visitors (7:00 AM – 10 AM) (total 50.5 hours) and three hours after 10:00 AM during the presence of visitors (total 50.5 hours). Behavioral data on a total of 101 observational hours, i.e., 202 focal observations of each 30 minutes, were collected over the entire study period (for LF, NLF, SAM_5, AM_3 and AM_1, 40, 41, 40, 39 and 42 focal samples were collected, respectively). Each animal was sampled for at least two hours each sampling day (one hour before 10 AM and one hour after 10 AM). Behavioral sampling was done by using 30 minutes focal animal sampling method (Altmann, 1974). Number of visitors near to the cage and their behavior towards langurs (level of disturbance) were recorded by instantaneous scan sampling method for 30 seconds at the interval of every 10 minutes during focal animal sampling (Altmann, 1974). Each grooming bout (duration and frequency) along with other predefined behavioral states and events (Table 2) performed by the focal animal were recorded (starting and ending time) using a stopwatch. Groomer and groomee's identities were recorded carefully for each grooming act. If the focal animal was out of sight for more than 10 minutes, behavioral observation was terminated. The behavior of the infant was fully dependent on its mother and other members of the cohort and was excluded from the observation.

Table 1: Age/sex status of the study animals with individual description.

S.	Code	Description
1	AM_1	Older male, male with little dark gray coat color
2	LF	Lactating female, older female
3	AM_3	Middle-aged adult male, male with light gray coat color
4	NLF	Middle-aged adult female, non-lactating
5	SAM_5	Subadult male, having somewhat white coat color
6	Infant	Infant frequent contact with the mother's (Lac) nipple

Level of disturbance

During the behavioral sampling, human disturbances on the langurs were identified based on the visitors' behavior around the cage. The disturbances were categorized into four levels- no disturbance (before 10 AM, under complete absence of visitors), low disturbance, medium disturbance and high disturbance based on their potential impacts on the study animals (Table 3).

Table 2: Ethogram of state and event behaviors of langurs used in the study.

S. No.	Behavior	Behavior description
1	Grooming	Auto-grooming (GRA), cleaning or manipulating own hair or skin
		Grooming received (GRR), cleaning or manipulating the hair or skin by another group member
		Grooming delivered (GRD), cleaning or manipulating the hair or skin of another individual
2	Hurdling	The posture of sitting with another individual in a hugging position anywhere inside the cage for a long time (>15 seconds)
3	Resting	Sitting or lying down without engaging in other activities
4	Moving	Covering some distance by the animal purposively or in response to another member inside the cage
5	Feeding/ Foraging	State of the animal searching or consuming palatable item inside the cage for >10 seconds
6	Play	Teasing, pulling, pushing and grabbing activities in between two animals or state of playing with other non-palatable material (modifier) inside the cage
7	Abnormal behavior	Self-biting behavior, hanging behavior, self-injurious behavior
8	Swinging	Swing on the rope or any other rigid rod for >5 seconds
9	Begging	State of animal sitting near the fence in presence of visitors and one or both hands out of fence for begging food or sitting in such a way that animal looks towards visitors for >30 seconds
10	Aggression	Behavioral sequence including at least one of the following: facial expressions, bite, fight, scream, chase, vocal threat
11	Mating	Mate with any of the members inside the cage
12	Lactation	State of a mother breastfeeding her infant
13	Other	Any behavior which is not mentioned above considered as 'other' behavioral activities

Table 3: Disturbance levels and their description.

S. No.	Disturbance level	Description
1	No disturbance	Visitors' absence (before 10:00 AM)
2	Low disturbance	Visitors watch the study animals but do nothing
3	Medium disturbance	Visitors teased the study animals but were not responded by the animals
4	High disturbance	Visitors teased and fed the study animals

Statistical analysis

The association between the number of visitors and the duration and frequency of grooming was tested using Karl Pearson's correlation coefficient. Analysis of variance (ANOVA) was performed to check the mean difference of both types of grooming (grooming receiving and grooming delivery) among the different age/sex status. Multiple linear regression was done to predict grooming behavior based on different age/sex status; age/sex status was five level categorical variables (LF, AM_1, AM_3, SAM_5 and NLF). For analysis of both types of grooming behavior i.e., grooming receiving and delivery were treated as response variables whereas in both models, age/sex status (categorical variable) was treated as an explanatory variable. After running multiple linear regression, assumptions were checked in order to validate the results of the model. Normality of the residuals was checked using Q-Q plots and homoscedasticity was checked by residuals vs fitted scatter plot. All the analyses were performed using R version 3.6.1 (R Core Team, 2021).

Results and Discussion

Activity budget of the captive langurs

Out of 12 behavioral activities recorded, resting constituted the highest proportion (49.27%), followed by feeding/foraging (22.93%) (Figure 1). Grooming constituted the third-most prevalent behavioral state (12.59%) which did not vary significantly before and after 10.00 AM. The highest proportion of the activity time budget on resting and feeding might be related to easy food availability. As emphasized by Alam *et al.* (2014), resting comprised the highest proportion of the overall activity time budget (41.04%) followed by feeding (33.75%) and grooming (11.73%) in langurs in forest patches under varied human disturbance. High resting and foraging in langurs even in the wild, might be attributed to a habitat condition with enough resources (Minhas *et al.*, 2010; Alam *et al.*, 2014). Resting also varies based on the temperature; animals rest more at high and low temperatures than during moderate temperatures and it constitutes the highest proportion of the activity budget in langurs (Li *et al.*, 2020). White-headed langurs also invest the highest proportion of diurnal time on resting (50%), followed by feeding (13%), moving (18%) and social behavior (19%) (Li and Rogers, 2004). During this study, we observed a marked decrease in resting, lactation, playing, and huddling behaviors after 10.00 AM when visitors were allowed inside the zoo, whereas the proportion of feeding/foraging and begging increased (Figure 1). Feeding/foraging ($r = 0.228$, $P < 0.05$), begging ($r = 0.251$, $P < 0.05$), moving ($r = 0.161$, $P > 0.05$), and swinging ($r = 0.066$, $P > 0.05$) were positively associated with the number of visitors, whereas, resting behavior ($r = -0.5823$, $P < 0.05$) was negatively associated. All other behaviors had a statistically non-significant negative association with the number of visitors. As disturbance increases in langur's habitat, foraging behavior is increased in proportion, but resting and social behaviors are decreased (Yang *et al.*, 2005, 2007). Comparable results were obtained from this study where resting decreased and foraging increased proportionately when the number of visitors and the intensity of disturbance increased inside the zoo.

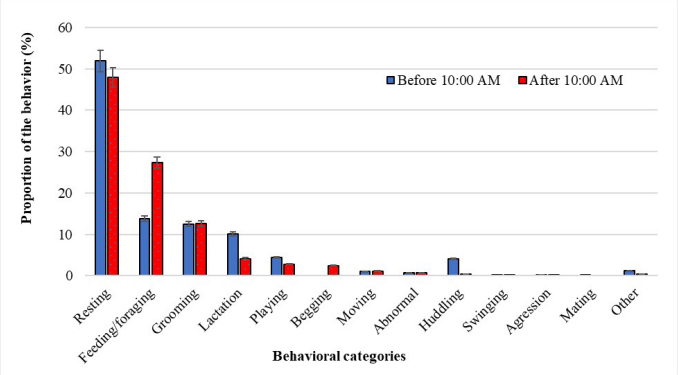

Figure 1: Overall activity proportion of captive langurs before and after 10:00 AM.

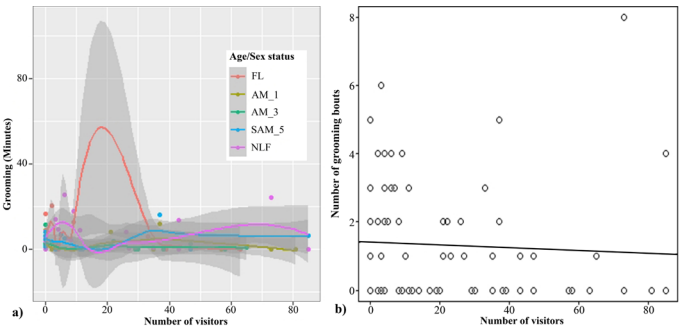

Figure 2: Grooming behavior by langurs in response to the number of visitors near the cage; (a) Age/sex-wise variation in time invested on grooming, (b) number of grooming bouts by the langurs (FL- lactating female, NLF- non lactating female, AM_1- adult male_1, AM_3- adult male_3, SAM_5- sub adult male_5).

Impact of visitors on grooming behavior

There was a negative non-significant association between the number of visitors with grooming bouts ($r = -0.031$, $P > 0.05$) and grooming time budget ($r = -0.051$, $P > 0.05$) of the langurs (Figure 2a and 2b).

Visitors have a meaningful impact on primates' grooming behavior and stress physiology in zoos (Chamove *et al.*, 1988; Davis *et al.*, 2005). Grooming delivery was the highest under the low human disturbance condition (Figure 3). During high disturbance, visitors provided supplement food to the study animals that in turn negatively affected the grooming behavior. The involvement of animals in other behaviors limit the time for performing social activities under high disturbance in captive settings. The results overlap with the time constraints hypothesis, i.e., under high human disturbance study animals engage in other activities (feeding/ foraging,

begging behavior) reducing the time investment on social activities (grooming, huddling, playing behavior) (Marty *et al.*, 2019). Beyond feeding, animals groomed more during high disturbance than during low or no disturbance, which may be due to attempts to avoid aggression by high-ranking animals and relieve stress through grooming (Lee *et al.*, 2021). Under the medium disturbance condition, langurs watched the visitor's majority of the time.

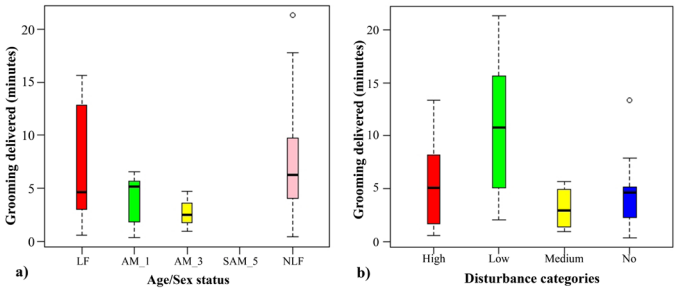

Figure 3: Boxplot representing mean time of grooming delivered based on- (a) age/sex status of the animal, and (b) level of disturbance from visitors.

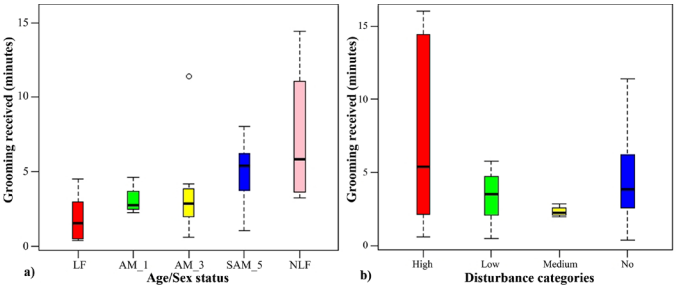

Figure 4: Boxplots representing mean time of grooming received based on- (a) age/sex status of the animal, and (b) level of disturbance from visitors.

High disturbance creates a situation in which the animals are fed or forced to interact with the source of the disturbance. All individuals inside the cage did not engage in such interactions equally. Adult males interacted more frequently with the visitors than the females. Most of the grooming was received under a high level of disturbance (Figure 4). Females and subadults were engaged in grooming behavior under high disturbance levels. Subadult males and non-lactating females received more grooming than any other age-sex category. It could be speculated that there was a high level of grooming reciprocity under high human disturbance than the low and no human disturbance.

Age-sex variation in grooming behavior

Results from the ANOVA showed that sex/age status was a significantly associated variable for grooming delivered ($F= 8.724$, $P < 0.001$) and grooming received ($F= 3.903$, $P < 0.01$) in captive langurs. The number of visitors did not show a significant association with grooming. Multiple linear regression models predicted that adult and subadult males showed a negative association with the grooming delivery while non-lactating female showed a significant positive association with it ($\beta \pm SE= 2.956 \pm 1.211$, $t = 2.441$, $P < 0.05$). Subadult male was positively associated with grooming receiving behavior ($\beta \pm SE= 2.9473 \pm 0.9268$, $t = 3.18$, $P < 0.05$). Other age and sex categories did not show a distinct pattern of grooming delivery or receipt.

Non-lactating females had a high tendency to engage in grooming delivery behavior whereas sub-adult male had a similar overtone with the grooming received behavior. A comparable result was presented by McKenna (1977) and Borries *et al.* (1994), who concluded adult females are more frequently engaged in grooming than other age groups among captive langurs whereas males are not much involved in the grooming behaviors. Lactating females spent most of their time breastfeeding and resting, which might be the probable reason for less participation in grooming than adult non-lactating females. In primate communities, adult females coordinate with the other members of the society for food and mate, which could be the reason for more grooming behavior than other age/sex groups and might support the social bonding hypothesis (Arlet *et al.*, 2015). Akinyi *et al.* (2013) concluded that age, sex and dominance rank significantly affect the quality of grooming received in the baboons; younger and higher-ranking individuals were groomed more frequently than older ones, and females were generally groomed more often than males in wild baboon troops. Adult females and adult males are groomed more in the langur troop, immature females spent significantly more time on grooming than the immature males, and immature females spent more time grooming all age-sex classes inside the troop (Nikolei and Borries, 1997). Male langurs have a different tendency to groom at different stages of their life. Grooming behavior peaks during and immediately following puberty; this period corresponds to the establishment of the coalition bonds (Ram *et al.*, 2011). Inside the langur troop, low-ranking females are frequently engaged in grooming with the high-ranking female members. A similar pattern was also observed inside the all-male band in which high-ranked males were frequently groomed by the low-ranked males (Minhas *et al.*, 2010). Under high-quality habitat, feeding time is lesser than in low-quality habitat, and adults invest more time in grooming than infants and juveniles (Li and Rogers, 2004). Cat Ba langurs also exhibit age class differences in social behavior as adults are engaged for longer duration in social interactions than the young members of the troop (Hendershott *et al.*, 2017). Under the captive conditions of the central zoo, subadult males and non-lactating adult females were engaged in grooming for higher duration because of the disturbance imposed by the visitors.

Conclusions and Recommendations

Captive langurs spent most of their time resting, followed by feeding and grooming. The number of visitors did not have a significant effect on grooming behavior inside the captivity. However, the intensity of disturbance had a negative association. Sex/age status was one of the major determinants of grooming behavior. The non-lactating adult female showed a significantly higher association with grooming delivery, and the subadult male showed a strong and significant association with the grooming receipt inside the captivity. Social behavior in captive langurs is under a strong effect of visitors' disturbance, hence, the zoo management should take initiatives to minimize such interference.

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Conflict of interest

The authors have declared no conflict of interest.

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