

Effect of behavioral enrichment on fecal corticosterone hormone levels in chimpanzees (*Pan troglodytes*) captive for conservation

Pawarat Jaidee¹, Tulyawat Sutthipat¹, Siriporn Umsook¹, Pitak Chainet¹, Kamphon Chomphuploi¹, Aratchaporn Meemey¹ and Nanthana Pothakam^{1*}

¹ Veterinary, Conservation and Research Section, Animal Management Division, Chiang Mai Night Safari, 33 Moo 12, Nong Kwai, Hang Dong District, Chiang Mai, 50230, Thailand

* Corresponding author: n.pothakam@gmail.com

Received: 30th April 2024, **Revised:** 16th June 2024, **Accepted:** 18th June 2024

Abstract - Chimpanzees (*Pan troglodytes*) are classified as an endangered species (IUCN), therefore their conservation is important. Raising chimpanzees in a captive environments different from the natural environment may cause them to exhibit fewer natural behaviors and affect stress. Behavior enrichment is another way to help maintain animals in captivity and relieve their stress. Corticosterone is a steroid stress hormone group, that is secreted when animals are in stressful conditions with decreased immune system function. The objective of this study was to examine the effect of behavior enrichment on corticosterone levels in chimpanzees captive for conservation by collecting the fecal material of 4 chimpanzees (2 male and 2 female) before, during and after behavior enrichment, then the fecal material was dried and hormones were extracted with 90 % ethanol. The solution was analyzed for level of corticosterone hormone by ELISA technique using completely randomized design. Results of the study on corticosterone levels in male chimpanzees found that during the period of before behavior enrichment, during enrichment and after enrichment period were not significant. However, there was a continuous decrease in corticosterone levels. Corticosterone levels in female chimpanzees found that during the behavior enrichment period were higher than after enrichment and before the enrichment period, the level was not

Citation: Jaidee, P., Sutthipat, T., Umsook, S., Chainet, P., Chomphuploi, K., Meemey, A., & Pothakam, N. (2024). Effect of behavioral enrichment on fecal corticosterone hormone levels in chimpanzees (*Pan troglodytes*) captive for conservation. *Food Agricultural Sciences and Technology*, 10(2), 84-94.

significant. The results of this study may be used as basic guideline information for designing behavior enrichment programs suitable for captive chimpanzees.

Keywords: Chimpanzee (*Pan troglodyte*), corticosterone, behavioral enrichment, captive

1. Introduction

Wild chimpanzees, have a potential lifespan about 60 years and experience an increase in levels of stress-related hormones as they age. This situation is similar to that of humans, as reported by NIA-supported researchers in the Proceedings of the National Academy of Sciences (Emery et al., 2020). These findings from a 20-year study of animals in their natural environment suggested that increases in stress hormones are a normal part of aging, rather than a consequence of other factors, such as the environment where the humans are studied or certain lifestyle factors such as poor nutrition or insufficient physical activity (Emery et al., 2020). Female chimpanzees are generally less gregarious than males but nevertheless are confronted with considerable intrasexual social competition (Newton-Fisher, 2006). The observations of captive female chimpanzees forming coalitions represent an inherent capacity rather than an artifact of confinement and close proximity to humans, the absence of reports of such behavior from wild populations suggests that it may occur only under appropriate ecological and demographic conditions. Chimpanzees show wide variation in many aspects of their behavior across communities and populations, which is produced or influenced by differences in ecology, demography, or local tradition (Whiten et al., 2001). Corticosterone is a steroid-based hormone of 21 carbons, classified among adrenal corticosteroids synthesized in the adrenal cortex.

Corticosterone, is the main glucocorticoid in rodents, birds, reptiles, and amphibians, controlling the metabolism and stress response (Yoshinao & Michael, 2021). In humans, corticosterone is mainly synthesized in the zona glomerulosa of the adrenal cortex; however, the activity of corticosterone is weak in humans. Corticosterone is important as an intermediate product for the production of aldosterone from pregnenolone in steroidogenesis. (Yoshinao & Michael, 2021). Numerous studies have demonstrated that the magnitude of the cortisol response marshaled by an individual depends upon both the physiological and the psychological aspects of the stressor that induces it (Miller & James, 2002). Psychological stressors have generally received more attention in the literature (Abbott et al., 2003). However, experimental evidence that both unpredictability and loss of control are associated with a heightened stress response led to an early expectation among researchers that in social animals, subordinate individuals should generally maintain higher levels of circulating glucocorticoids than dominants (Creel, 2001). Tests of this hypothesis in primates have been performed primarily on captive populations, producing mixed results. For some groups, this relationship normally holds true (Sapolsky, 1992). Wild animals raised in captivity in an environment that is different from their natural environment can exhibit different behaviors and display stress symptoms. When animals are deprived of the possibility to

perform species-specific behavior, they may show signs of suffering such as behavioral disorders, chronic stress, or other pathological conditions (Wurbel et al., 1996). Behavioral enrichment is a process of stimulating the five senses: eyes, ears, nose, tongue and touch (movement) so that animals can exhibit natural behavior and reduce stress, helping animals to achieve better mental and physical health. The behavioral enrichment format is divided into two types: 1. Food Enrichment (variety more than feed or feeding change method) 2. Non-Food Enrichment (make equipment from natural material or use toys not harmful to animals) to stimulating natural behavior in animals (Panisa, 2011). Behavior enrichment programs aims to provide chimpanzees with an enjoyable and fulfilling life. Billie et al. (2016) are dedicated to improving their overall well-being by encouraging cognitive and sensory stimulation, easing stress, reducing boredom, and promoting physical and mental health (In nature, chimpanzees spend most of their time foraging for food and exploring their habitat). However, in captivity, they have a lot more leisurely time. So Billie's team of volunteers and caregivers aim to originate fun and unique

ways to keep them entertained (Billie et al., 2016). The safety of the animals is paramount in all activities. In addition, the promotion of behavior is considered an indicator of the welfare of animals kept in a zoo (Young, 2003). The objective of the present study was to examine the effect of behavior enrichment on corticosterone levels in chimpanzees captive for conservation.

2. Materials and methods

2.1 Animals and housing

Subjects were four adult chimpanzees (two males and two females) from the Chiang Mai Night Safari (Table 1). The Chimpanzees had lived in captivity for about 17 years at Chiang Mai Night Safari. Chimpanzees were divided into two groups: two males and two females. Each group was housed in the same way. The enclosures they were held captive in ~9 x 12 m, and divided into two parts: an enclosed compound of 3 x 3 m, and an external "backyard" of 6 x 9 m. Inside the backyard there was a rope and tires, a timber bed for climbing and there was a bowl of water which chimpanzees could drink from at anytime.

Table 1. Data on Chimpanzees

No	Local ID	Microchip Number	Sex	Age (Year)
1	M40-001	900.012000102360	Female	33
2	M40-004	900.012000103046	Female	30
3	M40-002	900.012000103124	Male	33
4	M40-003	900.202011240342	Male	31

2.2 Behavior Enrichment Program

The behavior enrichment program designed for chimpanzees involved 15 activities,

each activity lasted approximately 30 min. Details about the enrichment program are shown in Table 2.

Table 2. Behavior enrichment programs and response times for chimpanzees

Date	Enrichment Program	Time to Response (min.)	Time to Enrichment (min.)	Behavior Response
Day 1	Coconut supplement diet	1-5	>10	They use their hands and teeth to gnaw off coconut shells and then eat coconut water and coconut meat. Some chimpanzees play with coconuts first and then eat coconut water and coconut meat.
Day 2	Scatter feeding substituting feeding pattern	1-5	6-10	Walking to collect feed pellets takes longer than the tray feeding method.
Day 3	Hand-fed substituting feeding pattern	1-5	>10	Get more familiar with the keeper.
Day 4	Hanging-fed substituting feeding pattern	1-5	>10	They stand and hang to eat feed pellets continuously.
Day 5	Grain scattered supplement diet	non	non	Not interested in grains.
Day 6	UHT milk supplement diet	1-5	1-5	They use their hands and teeth to unwrap a milk carton. After finishing, they returned the empty milk cartons to the keeper.
Day 7	Coconut supplement diet	1-5	>10	They use their hands and teeth to gnaw off coconut shells and then eat coconut water and coconut meat.
Day 8	Frozen fruit supplement diet	1-5	1-5	Enjoy frozen fruit Reduce aggressive behavior.
Day 9	Banana trunk and leaf supplement diet	1-5	>10	The first chimpanzee that got the banana tree played with it satisfied and then passed it on to other chimpanzees to play with. Some chimpanzees eat banana tree trunks.
Day 10	Change feeding time	1-5	>10	Chang’s feeding time from 2 times /day to 3 times/day affects chimpanzees not fighting and grabbing feed.

Table 2. Behavior enrichment programs and response times for chimpanzees (cont.)

Date	Enrichment Program	Time to Response (min.)	Time to Enrichment (min.)	Behavior Response
Day 11	Supplement frozen fruit dessert	1-5	>10	They use their hands to smash and find a place to sit and eat frozen fruit dessert.
Day 12	Add plastic ball	1-5	1-5	They run and pick up a ball to throw and play with.
Day 13	Add fecal other animal	non	non	Not interested in fecal material of other animals.
Day 14	Turn on Mozart's music	non	non	Not interested in Mozart's music.
Day 15	Add pile dead leaves	1-5	1-5	Walk to a pile of dead leaves, throw and tear play pile of dead leaves.

2.3 Fecal collection

Fecal samples were collected weekly before, during and after behavior enrichment during periods, when animals were individually housed for procedures required in other ongoing studies. Fecal samples were collected weekly in the morning between 09.00 to 12.00 h to avoid variation in the circadian rhythm of corticosterone secretion. Defecation was either observed or indicated to have occurred within the hour by the zoo keeper. Fecal boluses were broken open and approximately 50 g of material was taken from the middle and placed into zip-lock plastic bags labeled with the chimpanzees sex, time of collection and date. Samples were preserved at -20°C until fecal extraction and hormonal analysis.

2.4 Fecal extraction

Fecal samples were extracted as described by Brown et al., (2004). Frozen feces were thawed at room temperature (RT) and dried

in a conventional oven (60°C) for 24-48 h. Mixed powdered feces (0.1 g) were placed into glass tubes, 4.5 ml (18 x 180 cm) of EtOH and 0.5 ml of distilled water were added, and tubes were vortexed briefly. Samples were extracted by boiling in a water bath (90°C) for 20 min, with 90% EtOH added to keep the volume at 5 ml, and then centrifuged at 2,500 rpm for 20 min. The fecal extracts were combined, dried in a 90°C water bath, re-suspended in 5 ml of 90% EtOH, dried down again, and finally re-suspended in 1 ml of methanol into 1.5 ml microtube and were then stored at -20°C until analysis using enzyme-linked immunosorbent assay (ELISA).

2.5 Analysis by enzyme-linked immunosorbent assay (ELISA)

Concentrations of fecal glucocorticoid metabolites (fGCM) were measured in extracts diluted 1:3 in assay buffer (0.0137M Trizma base, 0.2 M Tris-HCl, 0.2 M NaCl, 0.2 M EDTA, 0.001% BSA, and 0.001% Tween 20; pH 7.5) using a double-antibody

EIA with a polyclonal rabbit anti-corticosterone antibody (CJM006, Coralie Munro, UC Davis, CA) previously used in Thailand. Samples and corticosterone standards (50 µl) were added to wells in duplicate followed by corticosterone-HRP (25 µl; 1:30,000) and anti-corticosterone antibody (25 µl, 1:100,000). Plates were incubated in the dark at RT for 2 h, followed by 100 µl of TMB solution and incubation for 20-35 min. Stop solution was added (50 µl, 1 N HCl) and absorbance measured at 450 nm (TECAN, Männedorf, Switzerland). Assay sensitivity was 0.12 ng/ml. Intra- (at 90% binding) and inter-assay coefficients of variation (CV, based on dose values) were <10% (all duplicates over 10% which were reanalyzed) and 12.28%, respectively. The level of the hormone corticosterone was determined to interpret the results in comparison with the behavior enrichment data.

2.6 Statistical analysis

All data were analyzed by using IBM SPSS statistic software version 25. Descriptive

data are presented as the mean ± standard deviation (SD) for corticosterone levels. Analyses compared means One-Way ANOVA and Duncan Post Hoc tests using 3 groups: before enrichment, during enrichment and after enrichment.

3. Results and discussion

3.1 Enrichment program

The enrichment program for chimpanzees consisted of 15 activities which were divided into two categories, Food and Non-food enrichment activities. Food enrichment activities were 11 activities that responded to 10 activities and non-response to 1 activity. Non-food enrichment activities were 4 activities that responded to 2 activities and non-response to 2 activities. The results of enrichment programs in chimpanzees were that for food enrichment, there was a 73.33% response to food enrichment 90.91% and non-food enrichment 26.67% response to non-food enrichment 50% (Figure 1).

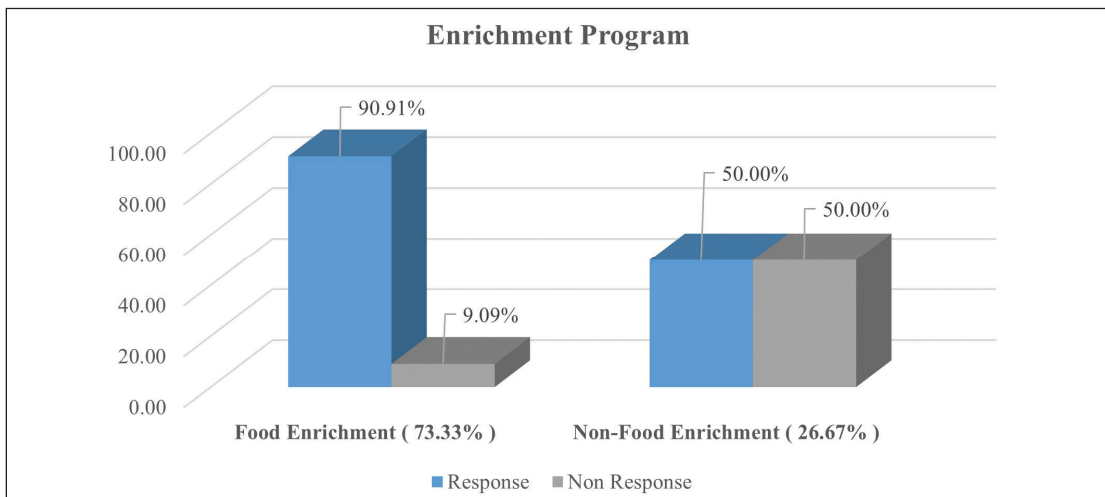


Figure 1. Results of chimpanzee responses to the enrichment program.

The positive results of this study generally corroborate the results of the few other published feeding enrichment studies. Chamove et al. (1982) reported reduced levels of aggression and abnormal behaviors and more foraging when woodchip litter seeded with food items was added to primate enclosures (Chamove et al., 1982).

3.2 Corticosterone in chimpanzees

It was found that corticosterone levels in male chimpanzees before, during and after the behavior enrichment period were not-significantly different ($P = 0.895$). However, there was a continuous decrease in corticosterone levels equal to 57.63 ± 24.93 ng/g feces, 54.51 ± 3.06 ng/g feces, and 52.25 ± 23.41 ng/g feces, respectively. Corticosterone levels in female chimpanzees during the behavior enrichment period were higher than after enrichment and before enrichment period was non-significantly ($P = 0.071$). Their values were 45.15 ± 10.16 ng/g feces, 36.04 ± 6.85 ng/g feces, and 45.05 ± 2.35 ng/g feces during, after and before enrichment respectively (Table 3). Under the report of the relation between the level of self-mutilation and the concentration of fecal metabolites of glucocorticoids in captive chimpanzees (*Pan troglodytes*). The influence of environmental on behavioral and endocrine variables of primates, have been increasingly studied by many authors, and it has been shown that abnormal behaviors associated with increased glucocorticoids may be directly related with the impairment of the animals' well-being. This study used 22 adult chimpanzees (*Pan troglodytes*), which were kept in captivity in three different institutions. Fecal samples were collected in triplicate and mean concentrations of glucocorticoid fecal

metabolites coincident with three different scores of self-mutilation were 34.65 ± 9.35 ng/g feces, 90.34 ± 29.08 ng/g feces and 138.82 ± 38.45 ng/g feces respectively, showing significant differences in this study (Cristiane et al., 2015). Past academic reports indicated that glucocorticoids play an especially important role in the regulation of aggression (Soma, 2006). Aggression between males typically heightens as they compete for reproductive opportunities with estrous females, and male cortisol levels frequently increase during such aggression in many primate species (Bergman et al., 2005; Setchell et al., 2005; Setchell et al. 2010; Fichtel et al., 2007). Also, research using fecal glucocorticoids to assess stress levels in captive river otters found that corticosterone levels in male otters were higher than in female otters (Rothschild et al., 2008). Environmental enrichment is commonly advocated as an effective method to improve the psychological wellbeing of nonhuman primates and other mammals in captivity (Markowitz & Shirley, 1987; Carlstead, 1996). As with psychological wellbeing, no widely accepted definition exists about what constitutes enriched environments. It is generally acknowledged, however, that such environments include both social and physical components (Erwin & Sackett, 1990; Poole, 1991). Furthermore, research on environmental enrichment of Brown Capuchins (*Cebus apella*) advocated that behavioral fecal cortisol measures associated with environmental enrichment are effective indicators of environmental enrichment is commonly advocated as an effective method to improve psychological well-being of nonhuman primates and other mammals in captivity, Boinski et al. (1999) found that across six conditions (preexperimental,

control, toy, box, box & toy, postexperimental) behavioral and fecal cortisol measures had significantly different within-subject differences across the six conditions. Research on daily and seasonal variation of basal and stress-induced corticosterone levels in captive starlings (*Sturnus vulgaris*) found changes in levels. Birds were bled four times during the daily cycle and during three different simulated seasons: under a

short-day photoperiod (mimicking winter), under a long-day photoperiod (mimicking summer) and it was found that handling and restraint elicited robust increases in corticosterone at all times of the day and during all three seasons. Levels were higher at night, during the bird’s inactive period, and decreased during the day (Romero & Remage-Healey, 2000).

Table 3. Effects of enrichment programs on corticosterone levels in chimpanzees

Chimpanzees	Corticosterone (ng/g feces)			SEM	P-value
	Before Enrichment	During Enrichment	After Enrichment		
Male	57.63±24.93	54.51±3.06	52.25±23.41	4.42	0.895
Female	36.04±6.85	45.15±10.16	45.05±2.35	1.90	0.071
SEM	2.79	4.15	3.99		
P-value	0.387	0.056	0.471		

SEM: Standard error of the mean in row comparison of data between corticosterone levels during period of before enrichment, during enrichment and after enrichment period.
Within a row, values with different non-significantly ($P < 0.05$).

From past reports, it is clear that older chimpanzees exhibited a significantly shallower rate of decline driven by higher levels of corticosterone at the end of the day. Males exhibited higher cortisol concentrations than females across the day. None of the interactions involving chimpanzee sex were significantly different ($P > 0.1$) (Emery et al., 2020). However, increases in fecal cortisol were observed in five of the six cases in which urinary cortisol was higher following stress. In the one case in which urinary cortisol declined following stress, a reduction also was observed in fecal cortisol, both the latter case and the one discordant case occurred in the same female (Whitten et al., 1998).

4. Conclusions

It was found that, in male chimpanzees found that before, during and after behavioura; enrichment, there were continuous decreases corticosterone levels. Enrichment programs in female chimpanzees found that during the enrichment period corticosterone levels were higher than after enrichment and before the enrichment period. Therefore, the results of this study may be used as basic guideline information for designing behavior enrichment programs suitable for captive chimpanzees.

Acknowledgement

Many thanks to the Director of Pinkanakorn Development Office (public organization), the Director of Chiang Mai Night Safari Office, the Director of the Department of Animal Management, Veterinary Medicine, Conservation and Research, and all the staff who made this study possible.

References

- Abbott, D. H., Kevern, E. B., Bercovitch, F. B., Shively, C. A., Mendoza, S. P., Saltzman, W., Snowdon, C. T., Ziegler, T. E., Banjevic, M., Garland, T., & Sapolsky, R. M. (2003). Are subordinates always stressed? A comparative analysis of rank differences in cortisol levels among primates. *Hormone & Behavior*, 43(1), 67-82. [https://doi.org/10.1016/S0018-06X\(02\)00037-5](https://doi.org/10.1016/S0018-06X(02)00037-5).
- Bergman, T. J., Beehner, J. C., Cheney, D. L., Seyfarth, R. M., & Whitten, P. L. (2005). Correlates of stress in free-ranging male chacma baboons, *Papio hamadryas ursinus*. *Animal Behaviour*, 70(3), 703-713. <http://doi:10.1016/j.anbehav.2004.12.017>.
- Billie, J., Adrienne, A., Rachael, R., Judy, G., Marsh, P., Elizabeth, B., & Kat, V. D. (2016). *Enrichment project*. Project Chimps. <https://projectchimps.org/chimpanzee-enrichment/>.
- Boinski, S., Swing, S. P., Gross, T. S., & Davis, J. K. (1999). Environmental enrichment of brown capuchins (*Cebus apella*): behavioral and plasma and fecal cortisol measures of effectiveness. *American Journal of Primatology*, 48(1), 49-68. [https://doi.org/10.1002/\(SICI\)1098-2345\(1999\)48:1<49::AID-AJP4>3.0.CO;2-6](https://doi.org/10.1002/(SICI)1098-2345(1999)48:1<49::AID-AJP4>3.0.CO;2-6).
- Brown, J. L., Walker, S. & Steinman, K. (2004). *Endocrine manual for the reproductive assessment of domestic and nondomestic species* (2^{ed}). Smithsonian institution.
- Carlstead, K. (1996). *Wild mammals in captivity: Effects of captivity on the behavior of wild mammals*. University of Chicago Press.
- Chamove, A. S., Anderson, J. R., Morgan Jones, S. C., Jones, S. P. (1982). Deep woodchip litter: Hygiene, feeding, and behavioral enhancement in eight primate species. *International Journal for The Study of Animal Problems*, 3, 308- 318.
- Creel, S. (2001). Social dominance and stress hormones. *Trend in Ecology & Evolution*, 16(9), 491-497. [https://doi.org/10.1016/S0169-5347\(01\)02227-3](https://doi.org/10.1016/S0169-5347(01)02227-3).

- Cristiane, S. P., Manuela, G. F. G. S., Danielle, A. L., Cecília, P., Adauto, N., Priscila V. F., Cláudio, A., Oliveira, D., & Marcelo, A. B. V. G. (2015). Relation between the level of self-mutilation and the concentration of fecal metabolites of glucocorticoids in captive chimpanzees (*Pan troglodytes*). *Pesquisa Veterinaria Brasileira*, 35(1), 62-66. <https://doi.org/10.1590/S0100-736X2015000100013>.
- Emery, T. M., Melissa, E. T., Stephanie, A. F., Andreas, B., Kris, H. S., Sarah P. G., Drew, K., Emily, O., Zarin, P. M., Richard, W. W., Martin, N. M. (2020). Wild chimpanzees exhibit humanlike aging of glucocorticoid regulation. *Proceedings of the National Academy of Sciences* (pp. 8424-8430). <http://doi.org/10.1073/pnas.1920593117>.
- Erwin, J., & Sackett, G. (1990). Effects of management methods, social organization, and physical space on primate behavior and health. *American Journal of Primatology*, 20(1), 23-30. <http://doi.org/10.1002/ajp.1350200104>.
- Fichtel, C., Kraus, C., Ganswindt, A., & Heistermann, M. (2007). Influence of reproductive season and rank on fecal glucocorticoid levels in free-ranging male Verreaux's sifakas (*Propithecus verreauxi*). *Hormones and Behavior*, 51(5), 640-648. <https://doi.org/10.1016/j.yhbeh.2007.03.005>.
- Markowitz, H., & Shirley, L. F. (1987). Artificial prey as behavioral enrichment devices for felines. *Applied Animal Behaviour Science*, 18(1), 38-63. [https://doi.org/10.1016/0168-1591\(87\)90252-8](https://doi.org/10.1016/0168-1591(87)90252-8).
- Miller, D. B., & Jame, P. O. (2002). Neuroendocrinology of the stress-response. *Behavioral Endocrinology*, 51(6), 287-324. <http://doi.org/10.1053/meta.2002.33184>.
- Newton-Fisher, N. E. (2006). Female coalitions against male aggression in wild chimpanzees of the Budongo Forest. *International Journal of Primatology*, 27(6), 1589-1599. <https://doi.org/10.1007/s10764-006-9087-3>.
- Panisa, T. (2011). Promoting behavior and developing integrated quality of life for animals. *Integrated conservation of the survival of all living things. 5th Zoo Wildlife Seminar*.
- Poole, T. B. (1991). *Primate responses to environmental change*. Springer Book Archive. https://doi.org/10.1007/978-94-011-3110-0_19.
- Romero, L. M., & Remage-Healey, L. (2000). Daily and Seasonal Variation in Response to Stress in Captive Starlings (*Sturnus Vulgaris*): Glucose. *General and Comparative Endocrinology*, 119(1), 60-68. <https://doi.org/10.1006/gcen.2000.7492>.

- Rothschild, M. D., Serfass, T. L., Seddon, W. L., Hegde, L., & Fritz R. S. (2008). Using Fecal Glucocorticoids to Assess Stress Levels in Captive River Otters. *Journal of Wildlife Management*, 72(1), 138-142. <https://doi.org/10.2193/2005-700>.
- Sapolsky, R. M. (1992). Cortisol concentrations and the social significance of rank instability among wild baboons. *Psychoneuroendocrinology*, 17(6), 701-709. [https://doi.org/10.1016/0306-4530\(92\)90029-7](https://doi.org/10.1016/0306-4530(92)90029-7).
- Setchell, J. M., Charpentier, M., & Wickings, E. J. (2005). Mate guarding and paternity in mandrills: factors influencing alpha male monopoly. *Animal Behaviour*, 70(5), 1105-1120. <https://doi.org/10.1016/j.anbehav.2005.02.021>.
- Setchell, J. M., Smith, T., Wickings, E. J., & Knapp, L. A. (2010). Stress, social behaviour, and secondary sexual traits in a male primate. *Hormones and Behavior*, 58(5), 720-728. <https://doi.org/10.1016/j.yhbeh.2010.07.004>.
- Soma, K. K. (2006). Testosterone and Aggression: Berthold, birds and beyond. *Journal of Neuroendocrinology*, 18(7), 543-551. <http://doi:10.1111/j.1365-2826.2006.01440>.
- Whitten, P. L., Stavisky, R., Aureli, F., & Russell, E. (1998). Response of fecal cortisol to stress in captive chimpanzees (*Pan troglodytes*). *American Journal of Primatology*, 44(1), 57-69. [https://doi.org/10.1002/\(SICI\)1098-2345\(1998\)44:1<57:AID-AJP5>3.0.CO;2-W](https://doi.org/10.1002/(SICI)1098-2345(1998)44:1<57:AID-AJP5>3.0.CO;2-W).
- Whiten, A., Goodall, J., McGrew, W. C., Nishida, T., Reynolds, V. S. Y., Tutin, C. E. G., Wrangham, R. W., & Boesch, C. (2001). Charting cultural variation in chimpanzees. *Behaviour*, 138(11), 1481-1516. <http://doi.org/10.1163/156853901317367717>.
- Wurbel, H., Stauffacher, M., von-Holst, D. (1996). Stereotypies in laboratory mice-Quantitative and qualitative description of the ontogeny of wire gnawing and jumping in ICR and ICR nu-mice. *Ethology International Journal of Behavioral Biology*, 102(5), 371-385. <https://doi.org/10.1111/j.1439-0310.1996.tb01133.x>.
- Yoshinai, K., & Michael, E. B. (2021). *Handbook of hormones comparative endocrinology for basic and clinical research*. Academic Press.
- Young, R. J. (2003). *Environmental enrichment for captive animals*. Blackwell Science.