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Is social dispersal stressful? A study in male crested macaques (*Macaca nigra*)

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Summary

In gregarious species, dispersal events represent one of the most dramatic changes in social life and environment an animal will experience during life due to increased predation risk, aggression from unfamiliar conspecifics and the lack of social support. However, little is known about how individuals respond physiologically to dispersal and whether this process is stressful for the individuals involved. We therefore studied the physiological stress response during dispersal in the crested macaque, a primate species in which males often change groups. Over a period of 14 months and 14 dispersal events in 4 groups, we determined faecal glucocorticoid metabolite (FGCM) levels during the process of immigration into a new group and examined a variety of factors (e.g. male age, rank achieved, number of males in the group) potentially affecting FGCM levels during this process. We found that FGCM levels were significantly elevated in the first few days upon immigration, after which levels returned quickly to baseline. FGCM response levels upon immigration were significantly and positively influenced by the number of males in the group. The rank a male achieved upon immigration, aggression received, as well as the proximity to other males did not significantly influence FGCM levels. Our data confirm previous findings on other species demonstrating that in crested macaques immigration into a new social group is associated with an acute endocrine stress response. However, given that stress hormone levels remained elevated only for a short period of time, we do not expect males to experience high physiological costs during immigration. Given our limited knowledge on the physiological responses to dispersal in animals, this study contributes to our understanding of dispersal more generally, and particularly inter-individual differences in the stress response and the potential physiological costs associated with these.

Introduction

In gregarious animals, certain individuals leave their natal group as a consequence of selective pressures from within-group competition and inbreeding avoidance (Clutton-Brock and Lukas, 2012; Crnokrak and Roff, 1999; Henzi and Lucas, 1980; Pusey, 1987). The dispersal of an individual from one social unit to another is likely to represent one of the most fundamental shifts in social life and environment in an animal's life (Smale et al., 1997). Dispersal is often accompanied by substantial benefits such as access to unrelated females but may also come with a number of costs to the individual involved. For instance, in comparison to philopatric individuals, conspecifics transferring from one group or territory to another often face increased predation risk and restricted access to known food resources (e.g. Alberts and Altmann, 1995; Pärt, 1995; Ridley et al., 2008). Furthermore, upon arrival in a new social group, dispersing individuals may face an elevation in aggression (Teichroeb et al., 2011; Ydenberg et al., 1988), the need to establish a permanent residency and dominance rank, and unfamiliar competitors – which require them to adapt their behaviour accordingly (Smale et al., 1997). Dispersal from a social group and immigration into a new group represent therefore potentially highly stressful and costly life-phases for the dispersing individual. To date, however, information on the physiological response and potential costs of such dispersal events and, in particular, how stressful the different phases (i.e. transition, immigration, membership in new group) associated with dispersal events are, is scarce. Such information, however, would be important to better understand the implications of this event on the health, survival and consequently the fitness of individuals.

Generally, as an adaptation to cope with the new environment, increased predation risk, and social challenges during the transition period and upon immigration, dispersing individuals can be expected to show a physiological stress response, i.e. an activation of the hypothalmo-pituitary-adrenal (HPA) axis and the secretion of increased levels of stress hormone (i.e. glucocorticoid; GC) (e.g. Sapolsky, 2002). This stress response enables the mobilization of energy for immediate use (e.g. 'flight or fight')(Sapolsky, 2002). Enhanced cognition, analgesia, and sensory function, as well as decreased pain perception are further adaptive consequences of an acute physiological stress response increasing the chances to overcome stressful and life-threatening situations (Nelson, 2005; Sapolsky, 1992). A direct link between a physiological stress response and such situations (e.g. increased predation risk, food scarcity) has been shown in diverse taxa (e.g. amphibians: Narayan et al., 2013; mammals: Sheriff et al., 2009). Studies investigating the direct link between time spent outside a social group and its effect on GC levels are, however, scarce. To our knowledge, the only study to examine this relationship, showed that subordinate male meerkats (*Suricata suricatta*) show increased stress hormone levels when conducting extra-territorial forays (Young and Monfort, 2009). Elevated GC levels upon immigration into a new social group, the second important phase individuals face during dispersal, has been reported for a number of primate species (long-tailed macaques (*Macaca fasicularis*): van Schaik et al., 1991; yellow baboons (*Papio cynocephalus*): Alberts et al., 1992; chacma baboons (*Papio ursinus*): Bergman et al., 2005; gray-cheeked mangabeys (*Lophocebus albigena*): Arlet et al., 2009; but see black howler monkeys (*Alouatta pigra*): van Belle et al., 2009) and thus seems to be a more common pattern, at least in this taxon.

However, the duration of these elevations is mostly unknown despite its potential implication on an individual's health and fitness. Whereas a short term (i.e. acute) increase in GCs is adaptive and allows an individual to better cope with a stressful situation, long-term or frequent activation of the HPA axis may lead to chronically elevated GC levels that can lead to physiological costs and thus compromise fitness due to its diverse negative effects on health (e.g. immunosuppression, decreased growth, impaired reproduction), potentially even leading to death (Sapolsky, 1992; Balm, 1999; Nelson, 2005). As such, individuals that are better able to downregulate their physiological stress response experience diminished health and fitness impairments. If a stress response is purely adaptive, a rise in GC levels is expected to be followed by a quick return to baseline levels.

Although current data on the stress response following dispersal is extremely limited due to the difficulty to predict the timing of dispersal and immigration, we expect that both intrinsic factors (e.g. fighting ability/rank achieved) and extrinsic factors (e.g. number of males in the new group, aggression received) will predict inter-individual stress responses for the dispersing males. In primates, males are the dispersing sex in the majority of species, and newly-arriving individuals of high fighting ability often challenge resident males to achieve a high rank in the new group, often at the expense of high risks such as severe injuries (Marty et al., 2016; Drews 1995). Immigrants with comparably lower fighting ability often only achieve a lower rank in the new group and develop strategies with which to circumvent contest, in this way reducing the potential costs of immigration (Clarke et al., 2008: Marty et al., 2016). Given these differences in immigration costs, males of different fighting ability can be expected to also differ in their stress hormone responses upon immigration.

Once arrived in a new group, a successful competition for mates does not only depend on intrinsic factors such as fighting ability but also on the number of competitors. We assume that the number of competitors and therefore the degree of male-male competition may have an influence on the males FGCM levels. FGCM levels are expected to be positively associated with the number of competitors in the group. Alternatively, males may circumvent competition by avoiding proximity to other males and becoming peripheral (Harcourt 1987).

To investigate individual stress levels during dispersal and immigration, we studied crested macaques (*Macaca nigra*). Crested macaques are an excellent model species as males are known to disperse from one group to another several times in their life (Marty et al., 2016). This species has a high reproductive skew leading to high male-male competition and a steep linear hierarchy (Neumann et al., 2011; Reed et al., 1997)(Engelhardt et al. under review). Reproduction in crested macaques is moderately seasonal whereby females can give birth year round. Crested macaques are endemic to the island of Sulawesi (Indonesia) and show a social system typical for cercopithecines with groups consisting of several males and females. As in most primates, females are philopatric whereas males disperse after reaching their physical prime (Marty et al., in press). Predation risk during the transition between groups seems to be low (Marty et al., 2016). Immigrations into a new group are non-random and many males are observed to immigrate into a group around the same time as other males. Males who achieve a high rank upon immigration (i.e. high fighting ability) are mainly young adult males dispersing from their natal group for the first time, immigrate independent of other males, are more likely to get injured, and can expect high future reproductive success. Males who achieve a low rank upon immigration (low fighting ability) align their immigration to a recent change in the alpha male position (exclusively conducted from new immigrants) (Marty et al., in press).

The overall aim of this study was to investigate the pattern of stress hormone output shown by dispersing males of a gregarious primate, and, by doing so, to examine whether dispersal events are associated with long-term physiological costs to these animals. We also investigated the effect of a variety of factors potentially influencing stress hormone output during immigration. Generally, we posed the following predictions: for solitary living males in the transition between two social groups (transient males), we predict that (1) they will show higher FGCM levels than individuals living in a group (resident males). Directly upon immigration, we predict (2) elevated FGCM levels in the immigrants. However, due to differences in a males' physical condition and the target group constellation, inter-individual differences in the magnitude of the physiological stress response are expected. Specifically, we therefore predict (3) high rank achievers who usually challenge the top rank position upon immigration (Marty et al, 2016.) to initially show higher FGCM levels than males who achieve a low rank. Regarding male-male competition, we expect (4) males immigrating into groups with comparably more competitors, and males that spend more time in the vicinity of such males upon immigration (5) to experience higher FGCM levels.

Methods

Study subjects and study site

Four groups of wild crested macaques were studied from November 2011 until January 2013 (15 months) in the Tangkoko Reserve in North Sulawesi, Indonesia (1°33'N, 125°10'E) as part of an ongoing long term project (Macaca Nigra Project, www.macaca-nigra.org). The reserve ranges from sea level to 1350m and comprises 8867 ha of lowland rainforest (Collins et al.1991; Rosenbaum et al.1998). The groups live in a mixture of regenerating former gardens, secondary forest, and undisturbed primary forest. Two of the observed groups (R1 and R2) have been periodically studied during the last ten years (O'Brien and Kinnaird, 1997; Reed et al., 1997) and continuously since 2006 (e.g. Neumann et al., 2010), whereas the two other groups (PB and R3) were habituated in 2008 and 2010, respectively (for more details see Marty et al. 2016). All individuals were fully habituated to the presence of human observers and individually known by the observers. All dispersing males were fully adult and none of the males included in this study were considered to be of old age (i.e. worn down canines, skinny body and slower movement).

Behaviour data collection

Upon the arrival of a new male in a group, 12 new immigrating males were followed all day, from one sleeping tree to the next sleeping tree. Two of these males immigrated twice into different groups (Table 1). Whenever possible, these males were followed on a daily basis for the first 14 days. Focal data were collected using all occurrence event sampling of all agonistic and affiliative behaviours with group members, as well as scan sampling every 5 minutes recording position, nearest neighbours, and activity (Altmann, 1974). The number of adult males in proximity was measured using the data on males within 10m which was collected every 5 min during a focal observation. A daily average of adult males within 10m was used for further analyses. All interactions between the focal animal and other group members were entered into handheld computers (Psion Workabout Pro G2) using spread-sheet software (PTab Spreadsheet v.3.0; Z4Soft). In addition, data on four solitarily roaming males was collected while following them for a total of 111 focal hours. Overall, behavioural data was collected during more than 2300 focal hours.

Table 1: Study male ID, number of observed immigrations, ID of the group the males immigrated into, rank achieved upon immigration, and number of samples within the first 14 days following immigration.

ID	Group	Rank	no of samples
BN	R1	0.07	10
OL	PB	1.00	15
QL	PB	1.00	4
UL	PB	0.78	19
AN	R1	0.00	5
JL	PB	0.00	16

JM	PB	1.00	21
LL	R1	0.63	18
PL	R3	0.33	29
CN	R3	0.00	26
NL *	PB	1.00	2
NL**	R3	1.00	17
FL *	PB	0.11	21
FL**	R2	0.00	14

* = first observed dispersal, ** = second observed dispersal

Rank: Standardized rank between zero and one, 0.00 represents the highest rank, 1.00 the lowest

Determination of the dominance hierarchy

All displacements (approach/leave interactions) and agonistic dyadic aggressive interactions between males with a clear winner/loser outcome were considered in order to quantify dominance hierarchies. Depending on the available data, interactions within the first three to six months (depending on the group tenure of the males) after the immigration were included into analysis in order to obtain an accurate rank for the newly immigrated male. Dominance rank was assessed using corrected normalized David's score (de Vries et al., 2006), using the package "Steepness" (Leiva and de Vries, 2011) in R (R Development Core Team, 2009) based on a matrix of proportions of wins calculated for each dyad. All ranks were standardized between 0 and 1 with the lower number representing a higher rank (see also Marty et al. 2016).

Sample collection

Urine-uncontaminated faecal samples were collected from transient males, newly immigrated males, and resident males. Samples from newly immigrated males were collected continuously for the first 14 days upon immigration into the new group. If possible, one sample was collected each in the morning, noon, and late afternoon. The freshly defecated faeces were homogenized before an aliquot of 2-3g was placed in a polypropylene tube (Hodges and Heistermann, 2011). Samples were directly stored in a cool box filled with ice until they were placed in a freezer (-18°C) after return to camp. Overall, 217 faecal samples from immigrating males within the first 14 days upon immigration were collected. An additional 187 faecal samples were collected from the immigrating males after the initial 14 days. Furthermore, 130 samples from eight non-natal resident males were collected during the study period for comparison. Eight samples from four transient males were also collected. The first fecal samples for these transient males were collected two days after these males were detected to account for the 1-2 day lag time in fecal glucocorticoid metabolite excretion, thus ensuring the hormone levels represent days the male was not in a group.

Hormone analyses

For analysis, all faecal samples were freeze-dried and pulverized and an aliquot of 0.05 – 0.08g of the faecal powder was extracted with 3ml of 80% methanol in water (Palme et al., 2013) as described in detail by Heistermann et al. (1995). Faecal extracts were analysed for immunoreactive 11β-hydroxyetiocholanolone, a major metabolite of cortisol in the feces of primates (e.g. Heistermann et al., 2006; Marty et al., 2015) by using enzyme immunoassay (Ganswindt et al., 2003). The assay, carried out as described in Heistermann et al. (2004), has been validated for monitoring adrenocortical activity in numerous primate species of all major taxa, including several species of macaques (Fichtel et al., 2007; Heistermann et al., 2006; Ostner et al., 2008; Shutt et al., 2012; Weingrill et al., 2011). The assay was recently also validated for use in crested macaques by confirming the presence of high amounts of 11β-hydroxyetiocholanolone in the feces of the species, and demonstrating a significant increase in levels in response to external stressors, such as injury, caught in a poachers trap or severe harassment by conspecifics (Gholib, 2011). Prior to each assay, extracts were diluted 100 to 3000 times (depending on concentration) with assay buffer to bring hormone concentrations into the working range of the assay. Sensitivity of the assays at 90% binding was 1.0 pg. Inter- and intra-assay coefficients of variation, determined by replicate measurements of high- and low-value quality controls, were 10.9% and 5.2% (high) and 14.7% and 8.1% (low), respectively. We ran each sample in duplicate and calculated mass steroid metabolite per mass fecal dried weight in ng/g.

Statistical analyses

Samples collected at least one month after the immigration were used to calculate baseline FGCM levels. In order to compare the mean FGCM levels between the four transient and the eight resident males, a Mann-Whitney-U Test was conducted in R (2.15.2)(R Development Core Team 2009) using mean FGCM levels per individual. Baseline values of an additional six males who immigrated during the study period were added as resident males to increase the sample size. To investigate factors determining FGCM levels upon immigration, a generalized linear mixed model (GLMM) with Gaussian error distribution was carried out. For this, FGCM levels were all ln-transformed to meet assumptions of a normal distribution. Hormone values in the immigrating males were adjusted (i.e. shifted) for two days to account for the time lag in FGCM excretion (Gholib, 2011). Each sample entered the analysis as the response variable (N=217). The predictor variables were: 1. number of days after immigration, 2. number of males in proximity (10m), 3. rank achieved upon immigration, 4. number of males in the group, 5.

daily aggression rate received from other males. Rank upon immigration entered the model as a control variable. Two males changed group twice within the study period and were accordingly given a number for the immigration event (1 for the first immigration, 2 for the second immigration). This immigration number, as well as the group identity entered the model as random factors. ID was treated as a nested random effect within day to control for multiple samples per day and individual. The model was conducted using the package "lmer" in R.

We checked various diagnostics of model validity and stability (Cook's distance, DFBetas, DFFits, and Variance Inflation Factors; distribution of residuals, residuals plotted against fitted values), and none of these indicated obvious influential cases, nor obvious deviations from the assumptions of normality and homogeneity of residuals (Field, 2005; Quinn and Keough, 2002). To obtain reliable p-values, a likelihood ratio test was used to compare the full model with respective reduced models using the function 'drop1'in the package 'car' (Chambers, 1992).

To compare FGCM levels of immigrating males with their respective baseline levels after the first four weeks following immigration, FGCM levels of each individual were averaged for the first 14 days following immigration and compared to the mean FGCM value recorded during the period 2-6 months after immigration using the Wilcoxon paired rank test was used. All significance levels were set at two-tailed p-values < 0.05.

Results

Although the transition period after departure from a group was not associated with elevated FGCMs for dispersing males, entry into a new group was. During the transition period, FGCM levels of the four dispersing males sampled did not significantly differ from those recorded for resident males (N=14) (Mann-Whitney-Test, U=33, p=0.32, n=18). Upon immigration, the daily mean FGCM levels of immigrating males showed a high variation within the first seven days, but did not differ from baseline values (assessed several weeks later, see above) in the second week after immigration (Wilcoxon rank sum test, W=51, P=0.24, N=24) (Figure 1).

In the GLMM investigating the factors that influence FGCM levels in response to immigration, the null model was significantly different from the full model ($chi^2=51.19$, df=12, $P<0.001$, effect size R2=0.76). The best predictors of FGCMs in immigrant males were time since arrival and number of other males in the group. Day after immigration was a significant predictor for FGCM levels in immigrating males (Table 2) with highest FGCM levels being recorded in the two days directly following immigration (Figure 1) and markedly declining levels thereafter. Mean FGCM levels were clearly less variable and consistently low following day 7. In addition, the number of males in the target group had a significant influence on the FGCM levels of the immigrating males. The higher the number of males in the target group, the higher were the immigrating males´ FGCM levels (Table 2). The rank achieved upon immigration, as well as the proximity to other males did not significantly influence FGCM levels (Table 2).

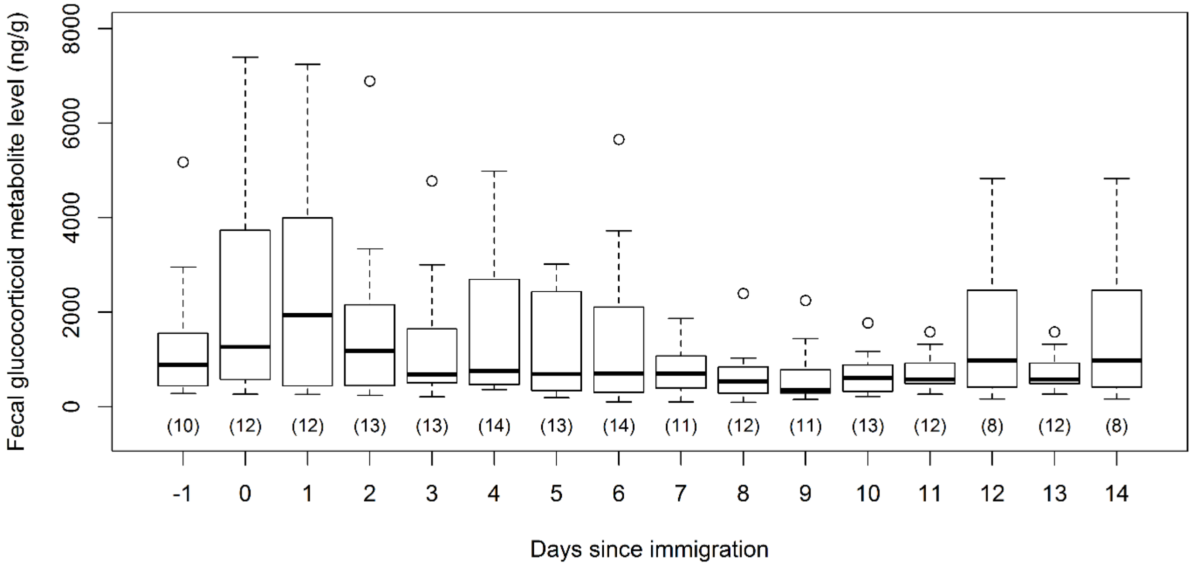

Figure 1: Boxplot of FGCM levels of immigrating males within the first 14 days upon immigration (the lines represent the minimum/maximum whereas the rectangle represents data from the lower quartile to the upper quartile, outliers above 6000 ng/g were are not shown). Numbers in brackets represent the number of males' sampled on the given day. All hormone data have already been adjusted for the FGCM excretion time lag of two days.

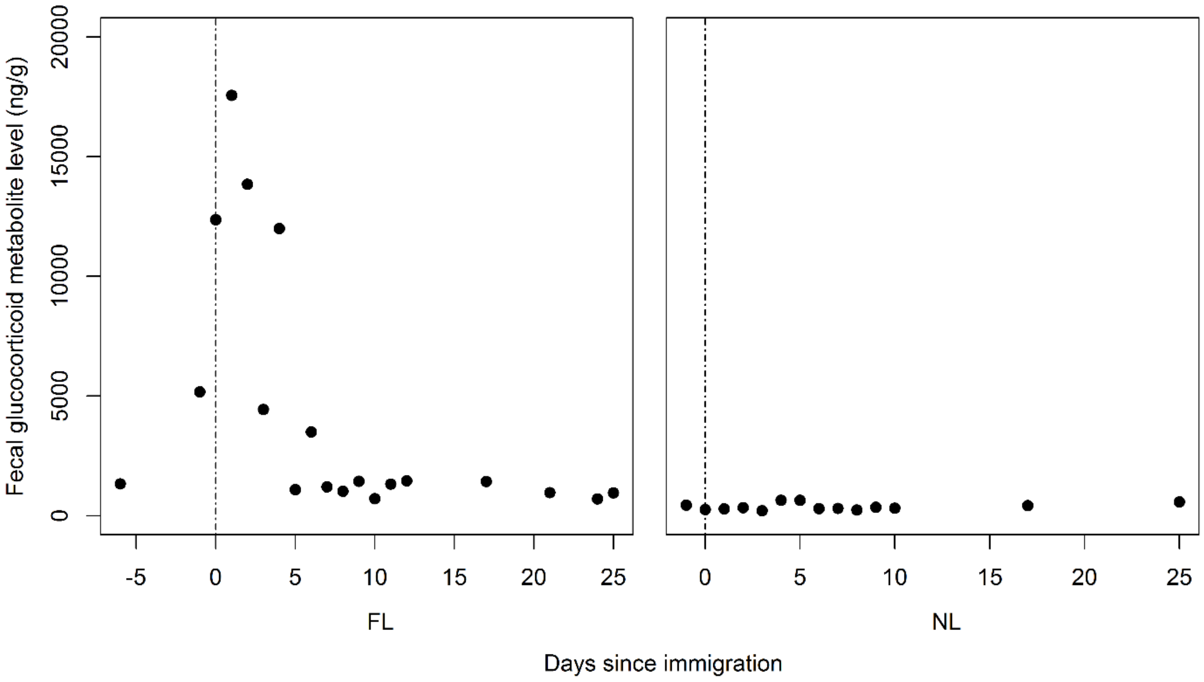

Figure 2: Visualization of the development of two males' (FL, NL) FGCM levels during and after the immigration process. Each point represents the mean daily FGCM. The line represents the time of immigration.

Table 2: Results of the GLMM testing the influence of the day after immigration, the agression received from males, the rank achived, and proximity to other males on the FGCM levels of immigrating males.

Model	Estimate	SE	t	P
Intercept	5.39	0.51	10.61	< 0.001
Day after immigration	-0.09	0.22	-4.00	**0.002**
Proximity to other males	-0.05	0.10	-0.52	0.604
Rank achieved	0.40	0.20	2.00	0.07
Number of males	0.20	0.04	5.62	**< 0.001**
Aggression received	-0.01	1.53	0.00	0.996

Discussion

In the present study, we investigated in wild crested macaques the extent to which the process of male dispersal and immigration into a new group elicits a physiological stress response (i.e. increased HPA axis activity as measured by faecal glucocorticoid (FGCM) levels) in the dispersing individuals. We also, more specifically, examined the relative importance of several factors on a male's glucocorticoid excretion upon immigration. Opposite to what we expected, there was no indication that males show elevated FGCM levels during the transition between groups. Males demonstrated, however, elevated FGCM levels during the first few days after immigration into the new group, indicating that this critical phase of the dispersal process elicits a physiological stress response. However, the period of elevated FGCM levels was relatively short even in newly immigrating males, with FGCM levels dropping to baseline already in the second week after immigration. Inter-individual variation of FGCM levels upon immigration was mainly modulated by the number of potential competitors in the new group. A high number of males in the target group was associated with comparably higher FGCM levels, suggesting a more marked stress response in the immigrating individuals under such conditions.

The combination of the absence of social partners, an increased risk of being attacked by conspecifics and predators, and life in an unknown environment is usually considered to represent a stressful situation for a dispersing individual (Smale et al., 1997; Young and Monfort, 2009). Interestingly, in our study species, dispersing individuals during the transition phase of their dispersal did not show increased FGCM levels compared to resident males. The lack of elevated FGCM levels in our roaming males may be ascribed to the small samples size that limits statistical power; thus the results need to be treated with caution. Alternatively, however, the relatively low FGCM levels in our roaming males may be related to the low predation pressure that macaques face on Sulawesi. The risk of predation in crested macaques may in principle be low for both group living animals as well as for those roaming alone. If so, our limited results suggest that the primary stressor for male crested macaques in this population may be social in nature rather than ecological (i.e., predation). In addition, aggression from conspecifics towards strangers is common also in dispersing male crested macaques and severe injuries inflicted by attacks on solitary males have been observed (personal observation). The four roaming males followed in this study, however, did not face such potentially stressful and harmful attacks during the observation time, which may add to the reasons for why FGCM levels were not elevated in these individuals. Whether the lack of encounters with other potentially threatening males was due to active avoidance strategies of the dispersing males, or just a matter of chance, remains unclear. In the few cases where we observed attacks towards solitary males, these occurred only when transient males approached a group for immigration. These attacks may therefore be part of the immigration process rather than of the transition period. However, as already mentioned, our sample size here is inherently small and results thus need to be confirmed, ideally by studying dispersing individuals prior to and during the dispersal process.

This will be an extremely challenging task though as the timing of dispersal is not predictable and following roaming males for extended periods of time is inherently difficult to do.

As expected, and corroborating findings for other primate species (Alberts et al., 1992; Arlet et al., 2009; Bergman et al., 2005), FGCM levels of male crested macaques were significantly elevated upon immigration indicating that this critical step of dispersal represents a stressful event also for our study species. Excreting stress hormones during a challenging period is clearly adaptive as it enables the mobilization of energy for immediate use (e.g., the 'flight or fight' response) (Sapolsky, 2002), presumably to cope with a new uncertain environment with potential challengers/competitors. Competition is often associated with elevated levels of physiological stress, whereby individuals do not only show elevated stress levels during, but already prior to the competitive event (e.g. *Macaca fascicularis*: Girard-Buttez et al. 2009; humans: Aubets and Segura, 1995). However, our results also show that the elevation in stress hormone output associated with immigration is not maladaptive as the increase only lasted a few days at most, helping the individual to cope with the energetic challenges associated with this process but not leading it into allostatic overload (McEwen 1998). Thus, detrimental and long-lasting effects on an individual´s health as seen under chronic conditions of stress (e.g. Sapolsky, 2002) are unlikely to occur in male crested macaques as a consequence of dispersal, at least not if immigrations are successful as they all were in our study. In recent years, it has been debated whether or not animals suffer from chronic stress in a wild environment at all or if our knowledge is too much influenced by biomedical research on captive animals (e.g. Boonstra, 2013; Wingfield and Ramenofsky, 2011). Research on fitness consequences of prolonged or chronic physiological stress in a natural setting is very limited in comparison to the overwhelming literature in biomedical research (Boonstra 2013). It has, however, been shown that the risk of extended periods of elevated stress hormone levels might affect an individual's behavioural strategy leading it to withdraw from a challenge (Girard-Buttoz et al. 2014). Our finding that dispersing between groups does not pose a long-term stressor to male crested macaques does not only show the absence of a maladaptive stress response but might also facilitate male dispersals in this species and thus explain the many immigration events we observed even for males entering at the lower end of the hierarchy.

On an individual level, our results show that differences in the magnitude of the stress response are linked to the number of potential challengers/competitors in the target group. Males who immigrated into groups with a higher number of adult males showed comparably higher FGCM levels than males who immigrated into groups with fewer potential competitors. Interestingly, not males who are actually at the highest risk of getting injured (high rank achievers) but males with the highest potential for being attacked (number of competitors) show higher FGCM levels. Even though immigration into a group with a comparable high number of males elicits a stronger physiological stress response, male crested macaques do not preferentially immigrate into groups with a lower number of competitors (Marty et al., 2016). This supports our assumption that the stress response upon immigration is purely adaptive with no negative consequences on the males' fitness. The stress response probably simply prepares the individual for upcoming potential or real challenges. The initial strong stress response may come along with rank uncertainties upon immigration and may return to homeostasis as soon as the potential of challenges declines with an increase in rank certainty. The higher the number of competitors in a group is, the longer it may take to consolidate the rank a male achieved.

Even though dispersal is often accompanied by cost in terms of injuries, predation, or starvation (e.g. Alberts and Altmann, 1995; Cheney and Seyfarth, 1983; Pärt, 1995) and represents one of the most stressful periods in a male's life (Smale et al., 1997), physiological costs might be overestimated. Both on an individual and a population level, the stress responses we observed to immigration followed the pattern expected for an adaptive response to an acute stressor. Our results do not provide any evidence of chronic stress or a prolonged stress response. Individuals in the wild may have adapted to stressful situations such as dispersal and immigration over time by using strategies to reduce costs (Marty et al., 2016). Our results suggest that inter-individual differences in the stress response are likely to be adaptive due to varying external conditions during and upon immigration.

Overall, our study demonstrates that the physiological stress response to migration shown by male crested macaques is adaptive and does most likely not carry any physiological costs. Interestingly, we did not find any statistically significant individual differences in the males' stress response suggesting that immigration is similarly stressful in this species whether males try to achieve a high or a low rank. This might explain why dispersal in crested macaques is highly dynamic with males migrating even frequently when the prospect for reproductive benefits achieved in the new group is bad. Similar studies on other species are now needed to better understand in how far duration of and inter-individual differences in the physiological stress response influence a species' migration dynamic.

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References

Adamo, S.A., Kovalko, I., Mosher, B., 2013. The behavioural effects of predator-induced stress responses in the cricket (*Gryllus texensis*): the upside of the stress response. J. Exp. Biol. 216, 4608–14.

Alberts, S.C., Altmann, J., 1995. Balancing costs and opportunities: dispersal in male baboons. Am. Nat. 145, 279–306.

Alberts, S.C., Sapolsky, R.M., Altmann, J., 1992. Behavioral, endocrine, and immunological correlates of immigration by an aggressive male into a natural primate group. Horm. Behav. 26, 167–78.

Altmann, J., 1974. Observational study of behavior: sampling methods. Behaviour 49, 227–267.

Arlet, M., Grote, M., Molleman, F., 2009. Reproductive tactics influence cortisol levels in individual male gray-cheeked mangabeys (*Lophocebus albigena*). Horm. Behav.

Aubets, J., Segura, J., 1995. Salivary cortisol as a marker of competition related stress. Sci. Sports 10, 149–154.

Balm, P.H.., 1999. Stress Physiology in Animals. Sheffield Academic Press, Sheffield.

Bergman, T., Beehner, J., Cheney, D., Seyfarth, R., Whitten, P., 2005. Correlates of stress in free-ranging male chacma baboons, *Papio hamadryas ursinus*. Anim. Behav. 70, 703–713.

Boonstra, R., 2013. Reality as the leading cause of stress: rethinking the impact of chronic stress in nature. Funct. Ecol. 27, 11–23.

Chambers, J.M., 1992. Linear models, in: Chambers, J.M., Hastie, T.J. (Eds.), Statistical Models in S. Wadsworth & Brooks.

Cheney, D.L., Seyfarth, R.M., 1983. Nonrandom Dispersal in Free-Ranging Vervet Monkeys: Social and Genetic Consequences. Am. Nat. 122, 392.

Clarke, P.M.R., Henzi, S.P., Barrett, L., Rendall, D., 2008. On the road again: competitive effects and condition-dependent dispersal in male baboons. Anim. Behav. 76, 55–63.

Clutton-Brock, T., Lukas, D., 2012. The evolution of social philopatry and dispersal in female mammals. Mol. Ecol. 472–492.

Crnokrak, P., Roff, D.A., 1999. Inbreeding depression in the wild. Heredity (Edinb). 83, 260–270.

de Vries, H., Stevens, J.M.G., Vervaecke, H., 2006. Measuring and testing the steepness of dominance hierarchies. Anim. Behav. 71, 585–592.

Drews, C., 1995. Context and patterns of injuries in free-ranging male baboons (*Papio cynocephalus*). Behaviour 133, 443–474.

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*). Horm. Behav. 51, 640–8.

Field, A., 2005. Discovering statistics using SPSS. Sage Publications, London.

Ganswindt, A., Palme, R., Heistermann, M., Borragan, S., Hodges, J., 2003. Non-invasive assessment of adrenocortical function in the male African elephant (*Loxodonta africana*) and its relation to musth. Gen. Comp. Endocrinol. 134, 156–166.

Gholib, 2011. Non-invasive hormone monitoring: Faecal androgen and glucocorticoid in male crested macaques (*Macaca nigra*) in relation to seasonal and social factors. Master thesis, Bogor Agricultural University.

Girard-Buttoz C, Heistermann M, Krummel S, Engelhardt A. 2009. Seasonal and social influences on fecal androgen and glucocorticoid excretion in wild male long-tailed macaques (*Macaca fascicularis*) Physiology & Behavior, 98 :168-175

Girard-Buttoz C, Heistermann M, Rahmi E, Agil M, Ahmad Fauzan P, Engelhardt A. 2014. Costs of mate-guarding in wild male long-tailed macaques (*Macaca fascicularis*): Physiological stress and aggression. Hormones and Behavior, 66 :637-648

Heistermann, M., Ademmer, C., Kaumanns, W., 2004. Ovarian cycle and effect of social changes on adrenal and ovarian function in Pygatherix nemaeus. Int. J. Primatol. 25, 689–708.

Heistermann, M., Finke, M., Hodges, J.K., 1995. Assessment of female reproductive status in captive-housed Hanuman langurs (*Presbytis entellus*) by measurement of urinary and fecal steroid excretion patterns. Am. J. Primatol. 37, 275–284.

Heistermann, M., Palme, R., Ganswindt, A., 2006. Comparison of different enzymeimmunoassays for assessment of adrenocortical activity in primates based on fecal analysis. Am. J. Primatol. 68, 257–273.

Henzi, S.P., Lucas, J.W., 1980. Observations on the Inter-Troop Movement of Adult Vervet Monkeys (Cercopithecus aethiops). Folia Primatol. 33, 220–235.

Hodges, J., Heistermann, M., 2011. Field endocrinology: monitoring hormonal changes in free-ranging primates, in: Field and Laboratory Methods in Primatology, A Practical Guide. Cambridge University Press, Cambridge, pp. 353–370.

Leiva, D., de Vries, H., 2011. Testing Steepness of Dominance Hierarchies.

Marty, P.R., Hodges, K., Agil, M., Engelhardt, A., 2016. Determinants of immigration strategies in male crested macaques (*Macaca nigra*). Sci. Rep. 6, 32028.

Marty, P.R., Hodges, K., Agil, M., Engelhardt, A., 2015. Alpha male replacements and delayed dispersal in crested macaques (*Macaca nigra*). Am. J. Primatol. doi: 10.1002/ajp.22448.

Marty, P.R., van Noordwijk, M.A., Heistermann, M., Willems, E.P., Dunkel, L.P., Cadilek, M., Agil, M., Weingrill, T., 2015. Endocrinological correlates of male bimaturism in wild Bornean orangutans. Am. J. Primatol. 77, 1170–1178.

McEwen BS., 1998. Stress, adaptation, and disease. Allostasis and allostatic load. Ann N Y Acad Sci. 1;840:33-44

Narayan, E.J., Cockrem, J.F., Hero, J.-M., 2013. Sight of a predator induces a corticosterone stress response and generates fear in an amphibian. PLoS One 8, e73564.

Nelson, R., 2005. An introduction to behavioiural endocrinology. Sinauer Associates, Inc., Sunderland, MA.

Neumann, C., Assahad, G., Hammerschmidt, K., Perwitasari-Farajallah, D., Engelhardt, A., 2010. Loud calls in male crested macaques, *Macaca nigra*: a signal of dominance in a tolerant species. Anim. Behav. 79, 187–193.

Neumann, C., Duboscq, J., Dubuc, C., Ginting, A., Irwan, A.M., Agil, M., Widdig, A., Engelhardt, A., 2011. Assessing dominance hierarchies: validation and advantages of progressive evaluation with Elo-rating. Anim. Behav. 82, 911–921.

O’Brien, T.G., Kinnaird, M., 1997. Behavior, diet, and movements of the Sulawesi crested black macaque (Macaca nigra). Int. J. Primatol. 18, 321–351.

Ostner, J., Kappeler, P., Heistermann, M., 2008. Androgen and glucocorticoid levels reflect seasonally occurring social challenges in male redfronted lemurs (Eulemur fulvus rufus). Behav. Ecol. Sociobiol. 62, 627–638.

Palme, R., Touma, C., Arias, N., Dominchin, M., Lepschy, M., 2013. Steroid extraction: Get the best out of faecal samples. Wiener tierärztliche Monatszeitschrift 100, 238–246.

Pärt, T., 1995. The importance of local familiarity and search costs for age- and sex-biased philopatry in the collared flycatcher. Anim. Behav. 49, 1029–1038.

Pusey, A.E., 1987. Sex-biased dispersal and inbreeding avoidance in birds and mammals. Trends Ecol.

Evol. 2, 295–9.

Quinn, G.P., Keough, M.J., 2002. Experimental Designs and Data Analysis for Biologists. Cambridge university press, Cambridge.

R Development Core Team, 2009. R: A language and environment for statistical computing.

Reed, C., O'Brien, T.G., Kinnaird, M.F., 1997. Male social behavior and dominance hierarchy in the Sulawesi crested black macaque (*Macaca nigra*). Int. J. Primatol. 18, 247–260.

Ridley, A.R., Raihani, N.J., Nelson-Flower, M.J., 2008. The cost of being alone: the fate of floaters in a population of cooperatively breeding pied babblers *Turdoides bicolor*. J. Avian Biol. 39, 389–392.

Sapolsky, R.M., 2002. Endocrinology of the stress-respone, in: Becker, J., Breedlove, S., Crews, D., McCarthy, M. (Eds.), Behavior Endocrinology. The MIT Press, Cambridge, pp. 409–450.

Sapolsky, R.M., 1992. Stress, the Aging Brain, and the Mechanisms of Neuron Death. MIT Press, Cambridge.

Sheriff, M.J., Krebs, C.J., Boonstra, R., 2009. The sensitive hare: sublethal effects of predator stress on reproduction in snowshoe hares. J. Anim. Ecol. 78, 1249–58.

Shutt, K., Setchell, J.M., Heistermann, M., 2012. Non-invasive monitoring of physiological stress in the Western lowland gorilla (Gorilla gorilla gorilla): validation of a fecal glucocorticoid assay and methods for practical application in the field. Gen. Comp. Endocrinol. 179, 167–77.

Slos, S., Stoks, R., 2008. Predation risk induces stress proteins and reduces antioxidant defense. Funct. Ecol. 22, 637–642.

Smale, L., Nunes, S., Holekamp, K., 1997. Sexually dimorphic dispersal in mammals: patterns, causes, and consequences. Adv. Study Behav. 26, 181–250.

Teichroeb, J. a., Wikberg, E.C., Sicotte, P., 2011. Dispersal in male ursine colobus monkeys (*Colobus vellerosus*): influence of age, rank and contact with other groups on dispersal decisions. Behaviour 148, 765–793.

van Belle, S., Estrada, A., Ziegler, T.E., Strier, K.B., 2009. Social and hormonal mechanisms underlying male reproductive strategies in black howler monkeys (*Alouatta pigra*). Horm. Behav. 56, 355–63.

van Schaik, C.P., Van Noordwijk, M.A., Vanbragt, T., Blankenstein, M.A., 1991. A Pilot-Study of the Social Correlates of Levels of Urinary Cortisol, Prolactin, and Testosterone in Wild Long-Tailed Macaques (*Macaca fascicularis*). Primates 32, 345–356.

Weingrill, T., Willems, E.P., Zimmermann, N., Steinmetz, H., Heistermann, M., 2011. Species-specific patterns in fecal glucocorticoid and androgen levels in zoo-living orangutans (Pongo spp.). Gen. Comp. Endocrinol. 172, 446–57.

Wingfield, J.C., Ramenofsky, M., 2011. Hormone-Behavior Interrelationships of Birds in Response to Weather, in: Brockmann, H.J., Roper, T., Naguib, M., Mitani, J.C., Simmons, L.W. (Eds.), ADVANCES IN THE STUDY OF BEHAVIOR, VOL. 43. Academic Press, Burlington, pp. 93–188.

Ydenberg, R., Giraldeau, L., Falls, J., 1988. Neighbours, strangers, and the asymmetric war of attrition. Anim. Behav. 36, 343–347.

Young, A.J., Monfort, S.L., 2009. Stress and the costs of extra-territorial movement in a social carnivore. Biol. Lett. 5, 439–441.