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RESEARCH ARTICLE

Bigger Groups Have Fewer Parasites and Similar Cortisol Levels: A Multi-Group Analysis in Red Colobus Monkeys

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If stress and disease impose fitness costs, and if those costs vary as a function of group size, then stress and disease should exert selection pressures on group size. We assessed the relationships between group size, stress, and parasite infections across nine groups of red colobus monkeys (*Procolobus rufomitratus*) in Kibale National Park, Uganda. We used fecal cortisol as a measure of physiological stress and examined fecal samples to assess the prevalence and intensity of gastrointestinal helminth infections. We also examined the effect of behaviors that could potentially reduce parasite transmission (e.g., increasing group spread and reducing social interactions). We found that cortisol was not significantly related to group size, but parasite prevalence was negatively related to group size and group spread. The observed increase in group spread could have reduced the rate of parasite transmission in larger groups; however, it is not clear whether this was a density-dependent behavioral counter-strategy to infection or a response to food competition that also reduced parasite transmission. The results do not support the suggestion that gastrointestinal parasitism or stress directly imposed group-size-related fitness costs, and we cannot conclude that they are among the mechanisms limiting group size in red colobus monkeys. Am. J. Primatol. 70:1072–1080, 2008.

Key words: colobus; group size; parasite; cortisol; disease transmission

INTRODUCTION

Among social animals, group size results from a complex set of interacting factors including predation pressure, food competition, and social considerations, many of which are well studied, particularly among primates. Other factors, such as endocrine responses and infectious diseases, may also be important but comparatively little empirical work has been done on their relationship to group size in primates [Alexander, 1974; Anderson & May, 1979; Nunn & Altizer, 2006; Pride, 2005b; but see Freeland, 1976, 1979; Pride 2005b]. As a result, our understanding of the determinants and effects of group size among primates is incomplete.

Stress: Fitness Effects and Relationship to Group Size

A stressor is anything that disrupts an individual's allostatic balance, such as injury, illness, or the threat of predation [Sapolsky, 1994]. To restore balance, the body initiates a stress response that involves the central nervous and endocrine systems [Sapolsky, 1994; Selye, 1979]. This response mobilizes energy for immediate use and is a highly effective means of coping with acute stressors; however, under chronic stress, this natural response can lead to fitness costs, because as energy is diverted elsewhere, essential functions such as growth, reproduction, and immunity are compromised [Sapolsky, 1994].

Cortisol, a steroid produced in the adrenal cortex, is a key hormone involved in the stress response [Sapolsky, 1994; Selye, 1979; Wingfield & Romero, 2001]. Blood serum and fecal cortisol levels have often been used as a measure of stress, and it has been well demonstrated that prolonged stress, as indicated by cortisol levels, has negative effects on fitness and is associated with reduced survival, fecundity, and immunity [Bercovitch & Ziegler,

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2002; Boonstra & Singleton, 1993; Creel et al., 2002; Ferin, 1999; Moberg, 1985; Muehlenbein, 2006; Pride, 2005a; Romero & Wikelski, 2001]. Because cortisol is part of the body's general stress response [Sapolsky, 1994; Selye, 1979], it reflects the combined effects of all causes of stress, including social, nutritional, disease-related, and reproductive stress [Pollard, 1995; Sapolsky, 1994].

Because many stressors are known to vary with group size, cortisol may provide a general index of overall stress levels in groups of different sizes, and thus of the fitness costs associated with variation in group size [Pride, 2005b]. Indeed, cortisol has been shown to be related to group size, food availability, and feeding effort in mammals [Boonstra & Singleton, 1993; Cavigelli, 1999; Chapman et al., 2007; Foley et al., 2001], birds [Raouf et al., 2006; Wasser et al., 1997], and reptiles [Romero & Wikelski, 2001]. For example, Pride [2005b] found that ringtailed lemurs (*Lemur catta*) experienced the least stress in medium-sized groups, compared with larger or smaller groups. Pride concluded that there was an optimal group size, but that the optimum varied with habitat type and food availability, which suggests that food competition (which may lead to social and nutritional stress) is among the mechanisms by which group size imposes a stress cost. Goymann and Wingfield [2004] found that cortisol levels were related to the allostatic load (measured as energy costs) associated with social status in many taxa. Extending this logic, cortisol levels should increase in larger groups if increasing group size leads to increasing energetic costs (e.g., travel costs associated with food competition).

Parasite Infections: Fitness Effects and Relationship to Group Size

There is a large body of empirical evidence demonstrating the negative fitness consequences of parasitic infections [reviewed in Nunn & Altizer, 2006], which include sickness, compromised nutritional status, suppressed immunity, decreased fecundity, and death. Although mild infections may have little effect on the host, negative effects increase with the intensity of infection or with parasite species richness [Nunn & Altizer, 2006]. Here, we focus on gastrointestinal helminths because they can be noninvasively studied in fecal samples. The most commonly observed helminths in wild primates are nematodes, which include species of *Enterobius* (pinworms, superfamily Oxyuroidea), *Trichuris* (whipworms, superfamily Trichuroidea), *Strongyloides* (threadworms, superfamily Rhabditoidea), and strongyles (order Strongylida) [Nunn & Altizer, 2006]. Primates become infected by ingesting feces or contaminated substrates (soil, vegetation) that contain third-stage larvae (strongyles) or first-stage larvae in eggs (*Enterobius*, *Trichuris*), or through skin contact with infective larvae (*Strongyloides*). Group living increases the probability of transmission by increasing the probability of coming into contact with contaminated substrates [Anderson, 2000; Nunn & Altizer, 2006].

Freeland [1979] considered parasite population dynamics relative to host group size in terms of island biogeography theory [MacArthur & Wilson, 1967; Simberloff, 1974]. He suggested that host social groups are analogous to biological islands, and that parasite population size and diversity should be affected by host group size and by the rate of migration of parasites between groups (through intergroup contact and host dispersal). In short, larger more connected groups of hosts should support larger and more diverse parasite populations than smaller more isolated groups.

Understanding the relationship between parasite infections and group size is complicated by a number of confounding factors. First, parasite infections and stress levels are interdependent. Parasite burdens, species richness, and pathogenic effects may be amplified when infections co-occur with nutritional, social, or reproductive stress because energy deficiencies and chronic stress can depress immune function and weaken the host's ability to fight infection [Appleton & Henzi, 1993; Bush et al., 2001; Hausfater & Watson, 1976; Koski et al., 1999; Nunn & Altizer, 2006; Padgett & Glaser, 2003]. In turn, the nutrient demands of the parasite and the energetic cost of mounting an immune response to infection can further compromise nutritional status and can cause or increase stress [Anderson & May, 1979; Bush et al., 2001; Koski & Scott, 2001; Sheldon & Verhulst, 1996].

Second, depending on their life cycle and transmission mode, parasites may create selection pressure for either larger or smaller groups [Freeland, 1976; Nunn & Altizer, 2006]. For example, ectoparasites and parasites transmitted by mobile hosts (e.g., malaria) may decrease in prevalence or intensity with increasing group size owing to a dilution effect and/or grooming behaviors [Bordes et al., 2007; Freeland, 1976; Mooring & Hart, 1992; Nunn & Altizer, 2006]. Conversely, both intrinsic disease risk and infection rates for many parasites that are directly transmitted or transmitted via an intermediate host or an infected substrate (e.g., the intestinal helminths considered here, viruses, and protozoa) should increase with group size owing to increasing proximity and contact rates among individuals and the increased probability of contact with contaminated substrates [Altizer et al., 2003; Anderson & May, 1979; Arneberg, 2002; Arneberg et al., 1998; Brown et al., 2001; Freeland, 1976, 1979]. Indeed, empirical data largely support this relationship in within-species (but not necessarily between-species) comparisons: for birds and mammals, the prevalence, diversity, and severity of

helminth, viral, and protozoan infections have been shown to increase with population density or group size, particularly in host species with stable groups [Altizer et al., 2003; Brown et al., 2001; Chapman et al., 2005; Cote & Poulin, 1995; Ezenwa, 2004; Freeland, 1979; Nunn et al., 2003; Shields & Crook, 1987; Stoner & Gonzalez di Pierro, 2005].

Third, the fitness costs of infection should create selective pressure for the evolution of immunological and behavioral counter-infection adaptations [Freeland, 1976; Nunn & Altizer, 2006]. Behavioral strategies, such as reducing contact rates and increasing interindividual spacing, may reduce the likelihood of infection and may obscure the expected relationships between group size and infections [Freeland, 1976; Nunn & Altizer, 2006].

Fourth, the social and ranging behaviors of the host species must be considered [Ezenwa, 2004; Nunn & Dokey, 2006]. The degree of home range overlap, the type and frequency of between-group contact, and immigration events may all influence the transmission of parasites between groups and may reduce intergroup differences [Altizer et al., 2003; Freeland 1979, 1980]. However, unless levels of between-group contact are very high, the effect of group size should not be obscured because smaller group sizes will impose limits on parasite population growth [Freeland, 1979]. Similarly, differences in the intensity of range use may affect transmission risk by altering the duration of contact with contaminated substrates and/or the likelihood of exposure to novel pathogens from other groups [Nunn & Dokey, 2006].

Finally, spatial and temporal variation in environmental factors may affect the transmissibility, intensity, and pathogenicity of parasite infections. Resource distribution and availability will affect the nutritional status of hosts and thus their immune response, and climatic conditions (temperature, humidity, rainfall) will affect egg survival and thus the probability of transmission via contaminated substrates [Freeland, 1976; Nunn & Altizer, 2006; Roepstorff et al., 2001; Stoner, 1996].

Objectives

We conducted a multi-group study of the costs of increasing group size in folivorous red colobus monkeys (*Procolobus rufomitratu*) in Kibale National Park, Uganda. We assessed the relationships between group size, stress, and parasite infections. We used fecal cortisol as a measure of physiological stress and fecal egg counts to measure the incidence and intensity of intestinal helminth infections. We expected that increasing group size would be associated with increasing stress levels and increasing parasite infections. By measuring indices of parasite infections and stress levels, we assessed the degree to which they interact. We also examined the relationship between parasite infections and social behavior because changes in group spread or social contact may affect transmission rates. We controlled for ecological variation by simultaneously collecting data on groups that occupied overlapping home ranges.

In a separate paper [Snaith & Chapman 2008], we presented behavioral measures of food competition from the same study groups and time period. We found evidence suggesting that within-group food competition led to increased foraging effort in larger groups. This may lead to compromised nutrition and may be associated with increased stress and reduced immunity to parasitism. We also found that larger groups spread out more and may have spent less time engaged in social interactions. These behavioral differences may be attributable to food competition, but may also represent counter-strategies to parasite transmission. In addition, we found that there were fewer offspring relative to the number of adult females in larger groups, which may simply be due to the energetic cost of food competition, but may also be related to additional physiological costs associated with stress and infectious disease.

METHODS

We followed nine groups of red colobus monkeys in Kibale National Park, Uganda, during May and June 2006. Group size and composition were determined based on daily counts of group members. To reduce potential confounds associated with temporal and spatial ecological variation, all groups occupied overlapping home ranges, and all groups were observed during a 2-month period. Five groups were followed simultaneously during May, and four groups were followed during June. Each group was followed for at least 22 complete consecutive days (6:30 a.m. until at least 7:00 p.m.; mean 27 days; maximum 33 days) for a total of 215 complete follow days. Group spread and the percent time engaged in social behavior are used as indices of social contact. Group spread (m^2) was calculated as the area of an ellipse defined by the distance between the most distantly separated monkeys along two perpendicular axes. To normalize for group size we also divide group spread by the number of individuals in a group (m^2/individual). This measure of group spread assumes the monkeys are distributed in a single horizontal plane and does not account for vertical distribution in the canopy. Group spread was measured each half hour by pacing the length and width of the group. The percent of time engaged in social behavior was calculated from half-hourly activity scan data. Field methods for behavioral variables are fully described in Snaith and Chapman (2008).

Fecal samples were collected to assess fecal cortisol levels and parasite infections. We aimed to collect samples from five individuals per group per

day, but daily sampling varied from zero to five per group. Individuals from which samples were collected were identified to age–sex class by observing defecation, but individual recognition was not possible. To avoid confounds associated with age, sex, and reproductive status [Bercovitch & Clarke, 1995; Cristobal-Azkarate et al., 2007; Festa-Bianchet, 1989; Hausfater & Watson, 1976; Klein & Nelson, 1999; Lloyd, 1983; Millspaugh & Washburn, 2004] and diurnal variation in hormone clearance [Millspaugh & Washburn, 2004; Sousa & Ziegler, 1998], samples were collected before 10:00 a.m. and only from adult males and females with infants.

Samples were collected immediately after defecation, placed into individual vials, and frozen within 5 hr [Millspaugh & Washburn, 2004]. Samples were thoroughly mixed [Millspaugh & Washburn, 2004] before half a gram of each sample was removed and prepared for cortisol analysis in the field using the citrate buffer and ethanol technique [Chapman et al., 2006; Gould et al., 2005]. Samples were then sent to the National Primate Research Center at the University of Wisconsin–Madison for measurement of cortisol and metabolites using the methods outlined in Ziegler et al. [1995]. Fecal cortisol levels are presented as ng cortisol and metabolites/g dry feces. Dry weights were determined for each sample in the field by oven drying half a gram of each sample to a constant weight and subtracting the dry weight from the wet weight to determine percent water content.

A portion of each sample was removed and stored in formalin for parasite analysis at McGill University. Half a gram of sample was processed using the formalin–ethyl acetate sedimentation concentration procedure [Garcia, 1999]. Parasite eggs were counted, photographed, and identified based on their size, shape, color, and content. We were able to identify eggs to the level of superfamily and sometimes genus. Infections were described in terms of prevalence (the proportion of samples infected), density (the number of eggs per sample), average density (mean density across all samples), and richness (the number of unique parasite species in a sample) [terms following Bush et al., 1997; Nunn & Altizer, 2006]. Because we could not identify individual monkeys, our measure of prevalence represents the proportion of *samples* infected, rather than the proportion of *individuals* infected as it is normally defined [Bush et al., 1997; Nunn & Altizer, 2006]. This measure of prevalence may produce either inflated (if sampling is biased by repeated sampling of infected individuals) or deflated (because infected individuals may not shed eggs in every defecation) estimates [Huffman 1997; Rothman 2008]. Although fecal analysis is the only noninvasive approach available for the study of gastrointestinal parasitism in wild primates [Gillespie, 2006], fecal egg counts may not provide a reliable measure of the actual nematode burden, because variation in egg counts may be affected by a variety of factors including parasite oviposition patterns, host fecal output and water content, and clustering of eggs in feces [Anderson & Schad, 1985; Hall, 1981; Rothman 2008]. Thus, though extreme variation in fecal egg counts may indicate different parasite burdens, small differences are not likely meaningful, and although we present egg density values by group, we do not include measures of density in our statistical analyses. Furthermore, our egg counts may be low because we froze our samples, which may destroy some eggs [Roepstorff et al., 2001]. This limitation will prevent direct comparisons with other studies, but should not bias between-group comparisons of density or prevalence, because all samples were treated in the same manner. Measures of species richness, however, may be biased if some egg species occurred only in some groups and were more likely to be destroyed by freezing than others; however, we were able to detect the diagnostic stages of parasite species found in similar studies of red colobus in Kibale where feces were not frozen [Chapman et al., 2005; Gillespie et al., 2005].

Average values were calculated to characterize the parasite infections and stress levels of each group and results are presented as group-level values. Although we have more than 400 fecal samples, replicate samples from within groups are not independent. To avoid pseudoreplication, we used group means to test for a treatment effect of group size ($n = 9$) [c.f. Hurlbert, 1984]. We used Pearson correlations to test whether group size was related to cortisol, parasite prevalence, or parasite richness, and whether parasite prevalence was related to group spread or percent time social. Because we previously demonstrated a positive relationship between group spread and group size [Snaith & Chapman 2008], we conducted a partial correlation to statistically control for the effect of group size when examining the relationship between prevalence and group spread. Because we found no difference in cortisol levels across groups, we did not statistically control for its effect when examining parasite relationships.

Because we ran multiple comparisons, we reduced α using the Benjamini and Yekutieli modified False Discovery Rate method, which has been shown to be a meaningful experiment-wise correction for multiple pairwise tests that reduces Type I error while maintaining statistical power [Narum, 2006]. Five pairwise comparisons were made involving group size and five were made against parasite prevalence, calling for $\alpha = 0.022$ for all tests [Narum, 2006].

All field and laboratory methods were approved by McGill Animal Care Committee, the Uganda Wildlife Authority, and the Uganda National Council for Science and Technology.

RESULTS

Group size varied from 25 to 127 individuals (mean = 65). We analyzed 477 samples for fecal cortisol (mean = 53 per group, range 36–92). Average group cortisol levels ranged from 93 to 208 ng/g dry feces (mean = 162 ng/g) and was not significantly related to group size ($r = 0.100$, $P = 0.798$) (Fig. 1).

We analyzed 442 samples for parasite infection (mean = 49 per group, range 38–65). We found eggs of *Trichuris sp.*, *Strongyloides sp.*, *Colobenterobius sp.*, *E. colobi*, and other strongyles (Table I). There were 206 infected samples, giving an overall infection prevalence of 47%. Across groups, infection prevalence varied from 12 to 68% (mean = 42%), and was negatively related to group size ($r = -0.934$, $P < 0.001$). This relationship appears to be primarily driven by the variation in *Trichuris* prevalence (Table I). Overall, maximum species richness was 3 (mean = 0.50, range 0–3) and was not significantly related to group size ($r = -0.274$, $P = 0.475$). Density ranged from 0 to 97 eggs per sample. Overall average density was 3.58 and average density across groups ranged 1.3–7.1 eggs per sample.

The percent time engaged in social activity varied from 5.1 to 10.2% (mean = 7.2%). There was no statistically significant relationship between parasite prevalence and percent social time ($r = 0.617$, $P = 0.076$), although a weak trend may exist.

Across groups, average group spread varied from 299 to 10,746 m^2 (Fig. 2), or from 7 to 85 m^2/individual (Fig. 2). There was a positive relationship between group spread and group size ($r = 0.817$, $P = < 0.001$) and a negative relationship between group spread and parasite infection prevalence ($r = -0.782$, $P = 0.013$). When group size was statistically controlled, group spread and parasite prevalence were not statistically related ($r = 0.136$, $P = 0.748$).

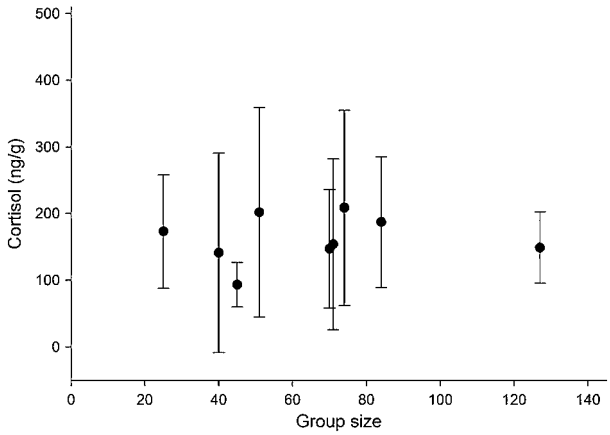

Fig. 1. Mean and standard error of cortisol levels (ng/g) across nine groups of red colobus in Kibale, Uganda.

TABLE I. Parasite Infection Prevalence and Densities Across Groups

		Trichuris		Strongyles		*Strongyloides*		*Colobenterobius*		*Enterobius*	
Group size	*N* Samples	Prevalence	Mean density	Prevalence	Mean density	Prevalence	Mean density	Prevalence	Mean density	Prevalence	Mean density
127	47	0.11	3.26 (0–97)	0.02	0.04 (0–1)	0.00	0.00 (n/a)	0.00	0.00 (n/a)	0.01	0.02 (0–1)
84	48	0.21	1.38 (0–10)	0.01	0.02 (0–1)	0.01	0.02 (0–1)	0.00	0.00 (n/a)	0.00	0.00 (n/a)
74	65	0.42	4.38 (0–37)	0.01	0.03 (0–2)	0.01	0.01 (0–1)	0.00	0.00 (n/a)	0.00	0.00 (n/a)
71	38	0.31	4.08 (0–28)	0.03	0.05 (0–1)	0.00	0.00 (n/a)	0.01	0.08 (0–3)	0.00	0.00 (n/a)
70	50	0.37	4.00 (0–32)	0.04	0.06 (0–1)	0.00	0.00 (n/a)	0.00	0.00 (n/a)	0.00	0.00 (n/a)
51	47	0.45	3.02 (0–27)	0.04	0.04 (0–1)	0.00	0.00 (n/a)	0.00	0.00 (n/a)	0.00	0.00 (n/a)
45	48	0.56	3.35 (0–41)	0.02	0.02 (0–1)	0.00	0.00 (n/a)	0.00	0.00 (n/a)	0.00	0.00 (n/a)
40	50	0.65	7.02 (0–89)	0.08	0.06 (0–1)	0.00	0.00 (n/a)	0.00	0.00 (n/a)	0.00	0.00 (n/a)
25	49	0.60	1.22 (0–24)	0.04	0.02 (0–1)	0.08	0.04 (0–1)	0.00	0.00 (n/a)	0.00	0.00 (n/a)

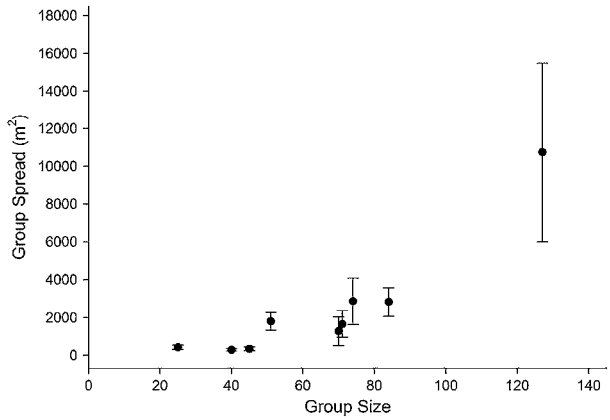

Fig. 2. Mean and standard deviation of group spread (m^2) across nine groups of red colobus in Kibale, Uganda.

DISCUSSION

Physiological stress, as indicated by fecal cortisol, was not significantly related to group size, which is puzzling because we previously demonstrated that larger groups experienced more food competition (increased day range, reduced foraging efficiency) and had fewer offspring per female than smaller groups [Snaith & Chapman, 2008], which led to the expectation that we would observe greater stress in larger groups. This result suggests that larger groups do not necessarily experience higher stress levels and we cannot conclude that physiological stress is among the mechanisms limiting group size or affecting the reproductive success of females in larger groups. ''However it is possible that stress is more important during periods of food shortage, and that females may suffer group size-related increases in cortisol (and decreases in fecundity) that were not captured during this study.''

We predicted that social and density-dependent transmission would lead to higher intestinal helminth infection rates in larger groups [Altizer et al., 2003; Freeland, 1976; Loehle, 1995; Moller et al., 1993; Nunn & Altizer, 2006]. Surprisingly, we found a negative relationship between parasite infection prevalence and group size in red colobus monkeys. Freeland [1976] reasoned that behavioral adaptations should evolve to reduce transmission rates and may obscure the expected relationship between group size and infection levels. Taking this logic further, if such counter-strategies increase with group size or infection risk, then a negative relationship between group size and infection rates may be observed. Our results provide support for this contention; group spread was negatively related to parasite infection prevalence, which may account for the unexpected negative relationship between group size and parasite infections. However, group spread covaried with group size, and when group size was statistically controlled, there was no significant relationship between prevalence and group spread. Although such covariation is expected if group spread is a density-dependent strategy, interpretation of the biological importance of group spread is difficult.

There was no relationship between parasite prevalence and the amount of time engaged in social behavior, which is perhaps not surprising given that most of the parasites in question are not transmitted between individuals through direct social contact, but require a period of time to develop [Anderson, 2000].

Overall, bigger groups spread out more and had fewer parasites, possibly owing to reduced environmental contamination and/or reduced contact with contaminated substrates. This was most evident in the largest group, which displayed very large spread, and very low parasite and cortisol values (Figs. 1 and 2). These relationships require further examination to improve our understanding of disease transmission dynamics in primate social groups.

In our behavioral analysis of food competition in this study population [Snaith & Chapman 2008], we suggested that the increase in group spread was a behavioral response to mitigate the energetic costs of food competition in larger groups [c.f. Clutton-Brock & Harvey, 1977; Dunbar & Dunbar, 1988; Janson & Goldsmith, 1995], and here we suggest that it may be a behavioral counter-strategy to parasite infection [c.f. Freeland, 1976]. We cannot determine the direction of causation; it is possible that group spread increases due to food competition and as a by-product reduces parasite transmission rates in larger groups. Or, transmission risk could directly create selective pressure for density-dependent adjustment of group spread. The negative relationship between group size and parasite infection prevalence suggests that parasite infections may not directly impose group-size-related costs that limit group size in this species. However, if group spread is adjusted to reduce parasite transmission, and if increasing group spread is associated with fitness costs, then parasite disease risk may impose an indirect cost by creating selection pressure for a costly behavior.

Parasite infections and stress levels are interdependent. Increasing stress can cause increasing susceptibility to infection due to compromised immunity, while infections can simultaneously increase stress levels by compromising nutrition and imposing costs associated with the immune response [Koski & Scott, 2001; Nunn & Altizer, 2006; Sapolsky, 1994]. Our finding that cortisol was not significantly related to group size may thus be related to the reduced parasite infection prevalence observed in larger groups. Less parasite-related stress may have counteracted the effect of other stressors in larger groups, resulting in lower cortisol levels. Alternately, if stress was lower in larger

groups for some other reason, the reduced parasite levels may have been due to better immune function resulting from lower cortisol levels. The causative direction of this relationship requires further investigation. Furthermore, the cortisol results must be considered relative to the behavioral measures, because if group spread increased as a behavioral mechanism to reduce stress associated with food competition or social pressures in large groups, this may help explain why cortisol does not increase with group size.

Careful studies are required to test these alternative explanations and to examine whether infection risk and food competition exert complementary selection pressures on social behavior and group size in primates. Due to our small sample size, we could not conduct more powerful multivariate tests to explore the interacting relationships among group size, group spread, stress, and parasite infections. Further work should concentrate on collecting comparable data from more groups so that such analyses may be conducted.

In a complementary study, we demonstrated that larger groups of red colobus experienced reduced foraging efficiency associated with scramble competition, and we found that female reproductive success, as indicated by the number of offspring relative to the number of females in a group, was lower in larger groups [Snaith & Chapman 2008]. We further suggested that this ecological mechanism might exert selection pressure to limit group size in some folivorous monkeys. Here, we evaluated whether parasite infections and physiological stress may exert similar pressures. Our results suggest that costs associated with parasite infections and stress did not increase with group size, and we cannot conclude that they are among the factors limiting group size in red colobus monkeys.

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