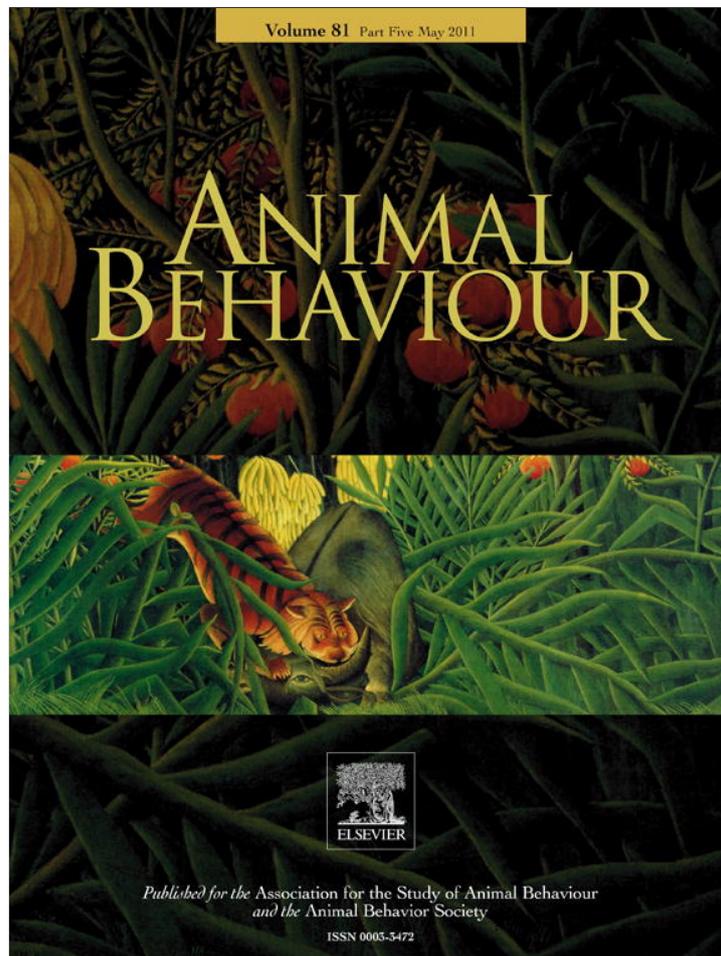


Provided for non-commercial research and education use.
Not for reproduction, distribution or commercial use.



This article appeared in a journal published by Elsevier. The attached copy is furnished to the author for internal non-commercial research and education use, including for instruction at the authors institution and sharing with colleagues.

Other uses, including reproduction and distribution, or selling or licensing copies, or posting to personal, institutional or third party websites are prohibited.

In most cases authors are permitted to post their version of the article (e.g. in Word or Tex form) to their personal website or institutional repository. Authors requiring further information regarding Elsevier's archiving and manuscript policies are encouraged to visit:

<http://www.elsevier.com/copyright>



Contents lists available at ScienceDirect

Animal Behaviour

journal homepage: www.elsevier.com/locate/anbehav

The energetics of male–male endurance rivalry in free-ranging rhesus macaques, *Macaca mulatta*

James P. Higham^{a,b,*}, Michael Heistermann^{b,1}, Dario Maestriperi^a^a Institute for Mind and Biology, The University of Chicago^b Reproductive Biology Unit, German Primate Centre, Göttingen

ARTICLE INFO

Article history:

Received 17 November 2010

Initial acceptance 16 December 2010

Final acceptance 31 January 2011

Available online 5 March 2011

MS. number: A10-00806R

Keywords:

endurance rivalry

energy balance

feeding competition

male–male competition

reproductive strategies

In many animal species, males engage in endurance rivalry, competing for mating opportunities over extended durations. In the rhesus macaque, a seasonally breeding primate species, energetically costly mating behaviours such as consortship lead to greater reproductive success, and the ability of males to improve their body condition before the mating season may be crucial in determining their ability to use such strategies. Here, we explore relationships between male dominance rank, behaviour, body mass index (BMI) and energetics (assessed through urinary C-peptide of insulin levels, UCPs) in free-ranging rhesus macaques during a 6-month birth season and subsequent 6-month mating season. We analysed measures of general activity, mating behaviours and restlessness, here defined as the rate of change between behaviours. During the birth season, high-ranking males spent more time feeding on high-energy food, were less restless and spent less time travelling than low-ranking males. Restlessness and travelling time were both negatively correlated with male BMI and UCPs. Males in good condition during the birth season were in good condition at the onset of the mating season. During the mating season, high-ranking males participated in more consortships, which were positively correlated with copulatory activity. Copulation was negatively correlated with UCP levels, demonstrating the cost of such mating activities. As a consequence, by the end of the mating season, high-ranking males were in the worst condition. Our results suggest that male rhesus macaques engage in competitive endurance rivalry, and that male condition built during the previous birth season is an integral part of mating success.

© 2011 The Association for the Study of Animal Behaviour. Published by Elsevier Ltd. All rights reserved.

In many animal species, males must compete with one another for mating opportunities over long periods ('endurance rivalry': Andersson 1994). A key component of a male's success in endurance rivalry is his physical condition; males with good body condition may be able to sustain costly mating activities over longer periods than males in poorer condition. For example, male grey seals, *Halichoerus grypus*, with greater body fat and energy are able to sustain mating for longer (Lidgard et al. 2005); male bullfrogs, *Rana catesbeiana*, in better condition can sustain longer chorus tenures (Judge & Brooks 2001); and larger male green anoles, *Anolis carolinensis*, with better body condition are able to court females for longer periods than smaller males (Jenssen & Nunez 1998). To increase success in endurance rivalry, males can improve their own body condition, adopt strategies that reduce the condition of their rivals, or both.

Many species of nonhuman primates live in multimale, multi-female social groups in which mating is highly promiscuous, competition between males for reproductive opportunities is intense, and in which this competition takes place over an extended mating season. The consequences of endurance rivalry for male condition can be dramatic. For example, in provisioned rhesus macaques, males lose an average of 10–12% of body mass (Bernstein et al. 1989) and 50% of skinfold fat (Bercovitch & Nürnberg 1996) during the mating season. The energetic stress of mating competition undoubtedly contributes to the marked increase in mortality for adult males (but not for adult females) during the mating season when compared to the birth season (Hoffman et al. 2008).

Rhesus macaque males that undertake energetically costly reproductive strategies such as long consortships (involving repeated mating and extended mate guarding) have greater reproductive success (Berard et al. 1994). In turn, only males that build sufficient body condition during the birth season may be capable of maintaining costly consortships during the subsequent mating season (Bercovitch 1997). During the birth season, adult males are therefore expected to maximize their energy intake and resting time, while minimizing energetically costly activities such as travel,

* Correspondence: J. P. Higham, Institute for Mind and Biology, The University of Chicago, 940 East 57th Street, Chicago, IL 60637, U.S.A.

E-mail address: jhigham@uchicago.edu (J. P. Higham).

¹ M. Heistermann is at the Reproductive Biology Unit, German Primate Centre, Kellnerweg 4, Göttingen 37077, Germany.

in order to build body condition. If individual differences in male body condition during the birth season are maintained into the subsequent mating season, then acquiring good condition during the birth season may be a crucial aspect of successful male mating strategies (Bercovitch 1997).

Activities other than travelling can result in significant energy expenditure among primates. Studies of humans have shown that overfed individuals burn two-thirds of their extra energy intake through fidgeting (nonfunctional movement) and other non-exercise activities (Levine et al. 1999). Fidgeting, specifically, contributes significantly to energetic expenditure (Levine et al. 2000), and has been linked to interindividual differences in weight and in the tendency to gain and lose mass (Levine et al. 1999; Marra et al. 2007). In humans, fidgeting is often accompanied by restlessness, which includes the tendency to initiate new activities frequently (i.e. be 'constantly on the go'; Dunn & Kronenberger 2003) and an inability to rest. Both fidgeting and restlessness have been linked to anxiety (e.g. Mehrabian & Friedman 1986; Heerey & Kring 2007; Kavan et al. 2009).

In primates, social anxiety may be related to dominance rank (Maestriperi et al. 1992), such that we might expect individuals of lower rank to be more restless than individuals of higher rank. Furthermore, rank may also determine access to food, especially highly desirable food types. Such rank-related differences in behaviour may affect the ability of males to build body condition before the mating season, and in turn, affect their ability to compete for access to fertile females through endurance rivalry. Unfortunately, few studies of free-ranging primates have examined the energetic components of male mating strategies, due to the past unavailability of reliable noninvasive measures of energetic condition. In particular, there is little direct evidence that the relationship between rank and mating success in promiscuous male primates may be mediated by body condition and energetic variables.

We conducted a study of dominance rank, body condition, energetics and behaviour among adult male rhesus macaques on Cayo Santiago over a 1-year period, which included a 6-month birth season and the following 6-month mating season. To assess energetic status over this period, we measured the concentrations of urinary C-peptide (UCPs), a small polypeptide cleaved in an equimolar ratio from proinsulin as it is converted to insulin (Sherry & Ellison 2007; Emery Thompson & Knott 2008; Deschner et al. 2008). UCP levels are positively correlated with energetic intake estimates (Emery Thompson & Knott 2008) and body mass (Deschner et al. 2008), such that higher UCP levels indicate better body condition. Recent studies using this marker have shown that variation in UCP levels tracks food availability, with levels decreasing as food abundance declines (fruit, chimpanzees, *Pan troglodytes*: Emery Thompson et al. 2009; leaves, colobus monkeys, *Colobus guereza*: Harris et al. 2010). However, to our knowledge, no study has yet used this technique to investigate the metabolic costs and benefits of reproductive strategies in free-ranging primates. During the 6-month birth season, we combined UCP measures with data on behavioural activity (feeding, travelling, resting), and a novel measure of behavioural restlessness, which we operationally defined as the rate at which individuals change their behavioural state, for example, from resting to travelling to resting to travelling again (see *Methods*). We trapped males at the end of the birth season and weighed and measured them to determine their relative body condition at this time. We then studied males during the 6-month mating season, recording the same variables as in the birth season, in addition to mating behaviours.

We predicted that, during the birth season, high-ranking males would spend more time feeding, feed more on high-energy monkey chow, rest more, travel less, and be less restless than low-

ranking individuals. We also predicted that, as a consequence of their higher energy intake and lower energy expenditure, high-ranking males would have higher levels of UCPs than low-ranking males during the birth season, and higher body mass indices at the end of the birth season. In addition, we predicted that male body mass indices and UCP levels would be related to behaviour, with higher levels of feeding, feeding on chow and resting being associated with higher levels of UCPs and higher body mass indices. Conversely, higher levels of travelling and higher levels of restlessness should be associated with lower levels of UCPs and lower body mass indices. We predicted that male condition during the birth season would predict male condition during the following mating season. During the mating season, we predicted that mating would be negatively correlated with feeding behaviour, demonstrating that mating constrains feeding. We predicted that the above relationships between dominance rank, behaviour and energetic condition would also be found during the early parts of the mating season. However, we predicted that we would additionally find strong negative relationships between measures of mating effort and UCP levels, providing direct evidence of the costs of mating and reproductive strategies. Consistent with this, we expected that the relationship between rank and UCP levels would be reversed by the end of the mating season, with high-ranking males having lower UCP levels due to increased mating activity. The overall goal of our study was to provide evidence that male rhesus macaques engage in endurance rivalry, with male mating success determined by rank and mediated by energetic condition.

METHODS

Study Site and Population

This study took place on Cayo Santiago, a 15.2 ha island located 1 km off the coast of Puerto Rico. A rhesus macaque colony, currently managed by the Caribbean Primate Research Center (CPRC) of the University of Puerto Rico, was established on this island in 1938 from free-ranging individuals captured in India (Rawlins & Kessler 1986), with no new individuals introduced to the population since then except through births. The animals are provisioned daily with commercial monkey chow, made available in several feeding corrals on the island. In this population, birth seasonality has changed over the last 50 years, but there is currently a 6-month mating season from March to August, followed by a 6-month birth season from September to February (Hoffman et al. 2008). All births occur during this birth season, with peak births currently occurring in October and November. Data presented in this study were collected during the October 2008–February 2009 birth season and the March 2009–August 2009 mating season. During this time the population included approximately 1000 individuals living in six social groups. All data collection took place in Group R, which consisted of an average of 268 individuals (range 243–307) during the study period (group size changed as a result of births, deaths and male emigration/immigration). We collected dominance rank data (see below for rank calculations) on all males in Group R, which numbered up to 44, but with only around 35 males usually seen with sufficient regularity to consider them real group members, and to observe them interact with other males with sufficient regularity to include them in dominance rankings. We collected other behavioural data and urine samples on a subset of 15 focal males during the birth season (mean age = 13.9 years, range 8.8–21.8 years) and 20 males during the mating season (mean age = 12.9 years, range 5.5–22.4 years). Ranks of focal males ranged from 1 to 34 (birth season) and 1–35 (mating season), so encapsulating the full range of ranks in

the group. Data on male ages were available for all males from long-term records.

Behavioural Data Collection and Processing

We undertook 30 min continuous focal observations (Altmann 1974) of adult male behaviour using the same ethogram and methods for both seasons. We recorded all activity, such as feeding, resting, travelling (all locomotion) and self-directed behaviours, and all social interactions, including grooming and agonistic interactions such as chasing and lunging. During the mating season, we also recorded copulation and ejaculatory copulation (considered separately, since rhesus macaques are serial-mount ejaculators). We also documented the presence of consortships between males and females, which we defined as occurring when males and females maintained close (<2 m) association, with exclusive mating (see Berard et al. 1994 for a similar definition). For social and sexual behaviours, we recorded the identity of the individual(s) interacting with the focal, and whether the focal was the actor or recipient in the interaction. For other behaviours (where there was no interactant) we recorded, on a continuous basis, the identity of the focal subjects' nearest neighbour within 2 m. For feeding behaviour, we recorded both total feeding time and feeding time on high-energy, highly desirable monkey chow (the most commonly eaten food type) separately. We analysed the following behaviours in relation to rank, body condition and UCP levels: feeding, feeding on monkey chow only, resting, travelling, copulation, ejaculatory copulation and restlessness (defined below). As we wanted to assess our novel measure of restlessness and whether it might be related to anxiety, we determined whether self-scratching, a validated index of anxiety (Maestriperi et al. 1992; Troisi 2002), was correlated with restlessness and dominance rank. We undertook a minimum of 1 h of focal behavioural observations per week (although usually more) on all males. Observations for each male were balanced between two time blocks, from 0700 to 1000 hours, and from 1000 to 1430 hours, such that each male was followed at least once within each time block each week. Total observation time was 605 ± 22 min per male (birth season) and 1676 ± 167 min per male (mating season). Data cannot usually be collected before 0700 hours or after 1430 hours on Cayo Santiago because of restrictions on access to the island.

Behavioural data were collected using Behaviour software loaded onto a Psion hand-held Workabout. Data were parsed into Access (Microsoft Corp., Redmond, WA, U.S.A.), and Access queries were then used to generate behavioural rates and durations. Behavioural data were analysed as the percentage of known time (feeding, feeding on chow, resting, travelling, copulation, ejaculatory copulation) or rates per hour of observation (restlessness = rate of behavioural change, see below; and rates of self-scratching, copulation, ejaculatory copulation). We recorded both rates and durations of copulations and ejaculatory copulations, as copulatory duration is highly variable and this variation may be important.

We operationally defined restlessness as the rate at which individuals changed their behavioural state, for example, from resting to travelling to resting to travelling again. Such changes in behavioural state could be spontaneous, caused by others indirectly (e.g. by dominant individuals creating anxiety and agitation in subordinates), or caused by others directly (e.g. by dominant individuals forcing subordinates to move). To obtain measures of rate of behavioural change that were as independent of social effects as possible, we treated the data in three ways. First, we created a score using the raw behavioural data, which included all behavioural changes. Second, we removed all dominance interactions as if they did not take place, so that the rate of behavioural change could not be affected by them. For example, a behavioural string of 'rest –

being chased by another male – rest' would not contain any behavioural changes as the animal was considered resting throughout this whole period in this index. Third, we removed all social interactions and all self-directed behaviours as if they never took place, and focused solely on transitions between the three major behavioural state types of rest, travel and feed. In this analysis, for example, a behavioural string 'travel – groom – self-groom – chase – travel' would not count as a behavioural change, but the string 'travel – chase – self-groom – groom – rest' would include one behavioural change (travel to rest). We undertook these separate analyses so that, in addition to utilizing the raw behavioural data, we also analysed indices that were as independent of direct dominance interactions, self-directed behaviours or social integration (relative frequency of social interaction) effects as possible. The results of all analyses involving the rate of behavioural change did not differ qualitatively regardless of which of these three indices we used. Therefore, we present only the analyses of rate of change between the three major behavioural state types (rest, travel and feed), hereafter 'restlessness'.

Calculation of Dominance Rank

We created a separate dominance rank for the birth season and the mating season, based on the following 'winner–loser' dominance interactions: fear grins (winner is the individual grinned at); avoidances (movement away from an oncoming individual; winner is the individual avoided); displacements (individual physically displaced by another, usually at a food or water source; winner is the displacer); and threats, chases and lunges (winner is the aggressor in all cases). During the birth season, we recorded 643 such interactions involving all adult (≥ 5.5 years) males. During the following mating season, ranks changed significantly from 1 June due to the occurrence of a period of revolutionary male–male coalitions (Higham & Maestriperi 2010). However, as mating mainly took place during March–May, we used 812 interactions between all adult males recorded during these 3 months to construct a main mating season hierarchy. Finally, we also calculated an end of mating season ranking to assess male condition by rank at the end of the mating season. To do this we utilized interactions from 1 July until the end of data collection (ignoring interactions from June, the month of main rank instability, which produced inconsistent hierarchies). We compiled interactions from each period into winner–loser matrices, and used MatMan 1.1 (Noldus Information Technology, Wageningen, The Netherlands) to create our separate hierarchies. There was a large percentage of unknown relationships in both seasons (birth season, 69%; mating season, 71%), due to a large number of males, including many peripheral males, that interacted only rarely with most males in the group. Despite this, following 10 000 iterations, significant linear hierarchies were produced (linearity test using Landau's linearity index corrected for unknown relationships, birth season, $P = 0.011$; mating season, $P = 0.003$, end of mating season, $P = 0.049$), which were highly directionally consistent (birth and main mating seasons, DCI = 0.91; end of mating season, DCI = 0.93). The ranks formed by this process were used in further analyses.

Collection of Urine Samples and Analysis of Urinary C-peptide of Insulin

We collected 68 urine samples during the second half of the birth season (3 November 2008–14 January 2009), between 0720 and 1340 hours, with 78% of samples collected between 0720 and 1020 hours. We collected 289 samples during the mating season between 0713 and 1615 hours, with 61% collected between 0720 and 1020 hours (see below for sample numbers by male). Urine

samples were pipetted off the ground or other substrate (e.g. leaves, rocks) with 1 ml disposable transfer tubes (WVR), only after a male was directly observed to urinate. Urine was placed into 2 ml eppendorf safe-lock microcentrifuge tubes (VWR, West Chester, PA, U.S.A.) and placed on ice. The sample was checked for cleanliness, and if there was any particulate matter in the sample, this was allowed to settle to the bottom. The supernatant urine was then pipetted off and this was placed in a fresh microcentrifuge tube. This process was repeated until the sample was clean. Each day, at the end of data collection (usually either at 1130 or 1430 hours), samples were returned to the CPRC laboratory in Punta Santiago, Puerto Rico. Samples were frozen at -80°C until transportation on ice packs to the German Primate Centre (DPZ) in Göttingen, Germany, where the c-peptide analyses were conducted. All samples arrived at DPZ frozen.

We analysed UCPs using a commercial C-Peptide ELISA Kit (Art. No. RE 53011, IBL International GmbH, Hamburg, Germany), which we validated for use in macaques. We first demonstrated analytical validation, and then undertook biological validations, showing significant correlations between levels of urinary and serum c-peptide, measures of body mass and condition, and responsiveness to feeding changes (Girard-Buttoz et al., *in press*). Prior to assay, urine samples were diluted between 1:2 and 1:10 (depending on the UCP level and amount of urine available) with IBL sample diluent (Art. No. RE 53017) to bring the samples into the working range of the assay, and 100 μl of the diluted urine was then assayed using the provided protocol of the manufacturer. Assay sensitivity was 0.064 ng/ml. Interassay coefficients of variation (CV) calculated from the measurement of low-, middle- and high-value quality controls run in each assay were 8.1%, 13.0% and 13.2%, respectively, while intra-assay CV values reported were 6.5%, 6.7% and 5.1%, respectively.

From the 357 samples analysed, 145 were below assay sensitivity. Those males in good body condition (high BMI) typically gave samples that were easily detectable. In contrast, males in poor condition (low BMI) gave samples that hovered around the detectability mark, sometimes being above assay sensitivity, and sometimes dipping below it. For these values, we assumed the maximum value possible, which was the value of assay sensitivity (0.064 ng/ml). Note that this is a very conservative approach to our data as it means that in samples of low (undetectable) concentration we slightly overestimated their concentrations, therefore reducing variation in our data set (slightly overestimating the levels of males in poor condition). UCP values were adjusted for variation in urine concentration by indexing them to the level of urinary creatinine (Erb et al. 1970) measured according to the method described by Bahr et al. (2000). One sample with a creatinine value of less than 0.05 mg/ml was discarded (as in, for example, Muller & Wrangham 2004). Following correction for differences in creatinine concentration, values are presented as c-peptide (ng)/creatinine (mg). After samples from the same male but from different times on the same day were averaged, we were left with a data set of 64 (birth season) and 267 (mating season) different 'male-days' (unique male-day combinations). We had a mean \pm SE per male of 4.3 ± 0.4 (range 2–12) male-days for the birth season and 13.3 ± 1.6 (range 3–28) male-days for the mating season. These were then averaged to provide mean values for each male for each season. Finally, we also used a subsample of 118 of the mating season values that came from 1 June until the end of the study to create one final set of male means. This was used solely to assess male condition by rank at the end of the mating season.

Collection of Morphometric Data

Between 22 January and 24 February 2009 (i.e. at the end of the birth season), 13 males (all of which represented birth season

focals, and 12 of which were mating season focals) were captured during the annual colony trapping procedures. Trained staff members captured these males between 0830 and 1100 hours in a 100 m² feeding corral provisioned with monkey chow, netting or capturing the monkeys by hand, transferring them to a holding cage (0.62 \times 0.42 \times 0.62 m), and then to a field laboratory where they remained overnight. The following morning, veterinary technicians anaesthetized the males with ketamine (approximately 10 mg/kg via IM injection), and we weighed the anaesthetized males in a standard hanging scale graded in tenths of a pound. We used a 1 m ruler with 1 mm gradations to measure crown–rump length of each male in a standardized position with his back fully straight. We calculated body mass index (BMI) for each male by dividing mass (kg) by crown–rump length squared (m²) (Campbell & Gerald 2004).

Statistical Analysis

We analysed mean values of each variable per male over the course of the birth season, and separately over the course of the mating season. Behavioural variables were normally distributed, and therefore correlations between variables were assessed with Pearson's coefficient. However, as Pearson's correlations are unsuitable for correlating variables when one is a dominance rank (i.e. an ordinal rather than continuous scale), analyses involving male rank were assessed using Spearman's rank correlations. For analyses linking behaviour to BMI and UCP levels, we also undertook partial correlations that controlled for rank.

To test for possible confounding effects of age, we tested both linear and quadratic relationships between rank and male age (as previous studies of macaques have found both types of relationship; e.g. linear: Paul 1989; quadratic: Takahashi 2002). Rank was unrelated to age among both the 15 birth season focal subjects and the 20 mating season subjects either linearly (two-tailed Spearman rank correlation: birth season: $r_s = 0.211$, $N = 15$, $P = 0.451$; mating season: $r_s = 0.075$, $N = 20$, $P = 0.759$), or in the form of a quadratic relationship (two-tailed quadratic regression: birth season: $R^2 = 0.256$, $P = 0.170$; mating season: $R^2 = 0.193$, $P = 0.179$). As such, age effects were not considered further in our data analyses and we present correlations without controlling for age. Since we had clear and specific predictions in all our analyses, all probabilities are one tailed, unless otherwise noted. Probability values below 0.05 were considered significant. Mean birth season UCP values were affected by two outliers (both >2 SDs above the mean of other males); all mean UCP data were therefore log transformed for analyses, following which data were normally distributed. All other data were normally distributed (Kolmogorov–Smirnov tests: all $P > 0.05$). We undertook all statistical tests in SPSS 18.0 (SPSS Inc., Chicago, IL, U.S.A.).

RESULTS

Birth Season

During the birth season, there was a negative but nonsignificant relationship between rank and feeding time ($r_s = -0.404$, $N = 15$, $P = 0.068$), and a significant negative correlation between rank and feeding time on chow only ($r_s = -0.443$, $N = 15$, $P = 0.049$), with higher-ranking individuals feeding more on chow than lower-ranking individuals did. Contrary to our prediction, there was no significant relationship between rank and time spent resting ($r_s = 0.207$, $N = 15$, $P = 0.230$). However, lower-ranked males spent more time travelling than higher-ranked males did ($r_s = 0.682$, $N = 15$, $P = 0.003$). Restlessness was also highly significantly correlated with male dominance rank, with higher-ranked males

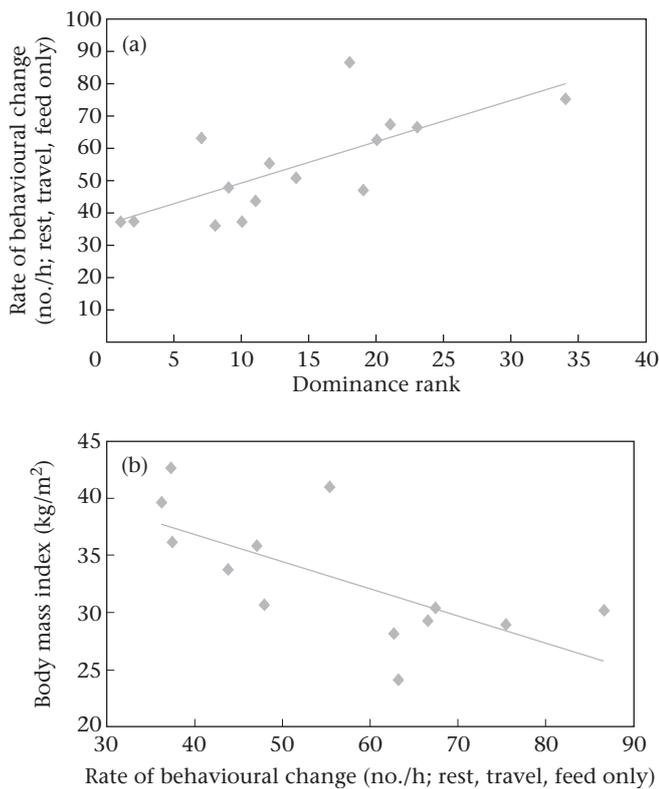


Figure 1. Relationships between restlessness (presented as mean rate of behavioural change between major state categories) and (a) male dominance rank and (b) body mass index (BMI) in free-ranging rhesus macaques.

changing behaviours less often than lower-ranked males ($r_s = 0.721$, $N = 15$, $P = 0.001$; Fig. 1a). To determine whether this effect was due to low-ranked males being more frequently disturbed by the presence of other individuals around them, we calculated the mean percentage of time spent alone by each male. This was highly correlated with male rank, such that lower-ranked males spent more time alone ($r_s = 0.864$, $N = 15$, $P < 0.001$). Therefore, it appears unlikely that the higher restlessness of lower-ranked males was mainly the result of disturbance by other individuals. Furthermore, rate of self-scratching was also positively correlated with restlessness ($r_{13} = 0.580$, $P = 0.012$), as well as with rank ($r_s = 0.693$, $N = 15$, $P = 0.002$), such that lower-ranked individuals self-scratched at higher rates.

As predicted, higher-ranked males had higher body mass indices than low-ranked males ($r_s = -0.527$, $N = 13$, $P = 0.032$). There was no significant relationship between BMI and percentage time spent feeding ($r_{11} = -0.020$, $P = 0.474$), feeding on chow only ($r_{11} = 0.315$, $P = 0.148$), or resting ($r_{11} = 0.165$, $P = 0.296$). However, there was a significant negative correlation between percentage time spent travelling and BMI ($r_{11} = -0.550$, $P = 0.023$), such that males that travelled more during the birth season had lower body mass indices than those that travelled less. Restlessness was the strongest predictor of BMI ($r_{11} = -0.678$, $P = 0.006$), such that higher restlessness was related to lower BMI (Fig. 1b). In partial correlations controlling for rank, restlessness was still significantly correlated with BMI ($r_{10} = -0.536$, $P = 0.036$), but travelling time was not ($r_{10} = -0.297$, $P = 0.174$).

As predicted, there was a significant relationship between male rank and mean UCP levels, with higher-ranked individuals having higher levels ($r_s = -0.461$, $N = 15$, $P = 0.042$). Mean UCP levels were not significantly correlated with time spent feeding, feeding on chow only, or resting (all $P > 0.1$). There were significant relationships between mean UCP levels and both time spent travelling

($r_{13} = -0.525$, $P = 0.023$) and restlessness ($r_{13} = -0.535$, $P = 0.020$). However, neither relationship was significant after controlling for rank effects using partial correlations (both $P > 0.1$).

Mating Season

Mating occurred over a 6-month period from March to August, but peaked between March and May. Mating activity was negatively correlated with feeding time (copulation rate: $r_{18} = -0.384$, $P = 0.047$; copulation duration: $r_{18} = -0.435$, $P = 0.028$; ejaculatory copulation rate: $r_{18} = -0.553$, $P = 0.006$; ejaculatory copulation duration: $r_{18} = -0.343$, $P = 0.070$). Mean mating season and birth season UCP levels were significantly positively correlated ($r_{11} = 0.608$, $P = 0.027$), suggesting that individual differences in male condition during the birth season are maintained to the onset of the following mating season. High-ranking males spent far more time consorting females than low-ranking males ($r_s = -0.754$, $N = 20$, $P < 0.001$; Fig. 2), but rank did not predict measures of copulation (all $P > 0.1$). However, time spent in consortship was itself positively correlated with measures of copulation (copulation rate: $r_{18} = 0.637$, $P = 0.001$; copulation duration: $r_{18} = 0.589$, $P = 0.003$; ejaculatory copulation rate: $r_{18} = 0.416$, $P = 0.034$; ejaculatory copulation duration: $r_{18} = 0.550$, $P = 0.006$). In contrast to the birth season, high-ranking males spent less time feeding than low-ranking males ($r_s = 0.514$, $N = 20$, $P = 0.012$), but similar to the birth season, high-ranking males were less restless than low-ranking males ($r_s = 0.528$, $N = 20$, $P = 0.020$). High-ranking males did not spend more time resting than low-ranking males ($r_s = -0.351$, $N = 20$, $P = 0.070$), and neither travelling time nor time spent feeding on chow only were significantly related to rank (both $P > 0.1$).

Consistent with our prediction, we found significant negative relationships between UCP levels and measures of copulation (copulation rate: $r_{17} = -0.408$, $P = 0.028$; copulation duration: $r_{17} = -0.369$, $P = 0.055$; ejaculatory copulation rate: $r_{17} = -0.395$, $P = 0.042$; ejaculatory copulation duration: $r_{17} = -0.570$, $P = 0.004$). However, after controlling for rank using partial correlations, only ejaculatory copulation duration was significantly negatively correlated with UCP levels over and above the effects of rank ($r_{16} = -0.507$, $P = 0.016$; Fig. 3). No other behaviours predicted mating season UCP levels (all $P > 0.1$). By the end of the mating season, high-ranking males had lower levels of UCPs than low-ranking males ($r_s = 0.465$, $N = 18$, $P = 0.026$).

DISCUSSION

Our results provide evidence that free-ranging rhesus macaque males engage in endurance rivalry, with dominance rank,

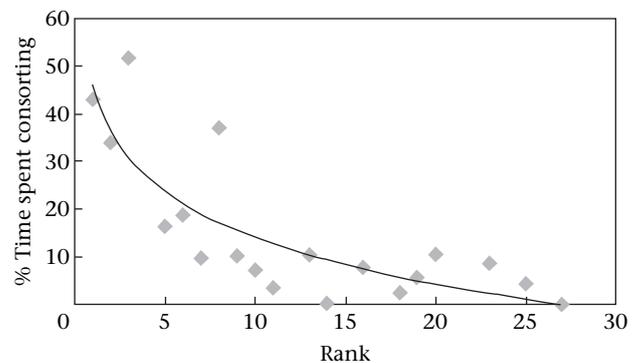


Figure 2. Relationship between time spent in consortship and dominance rank in free-ranging rhesus macaques.

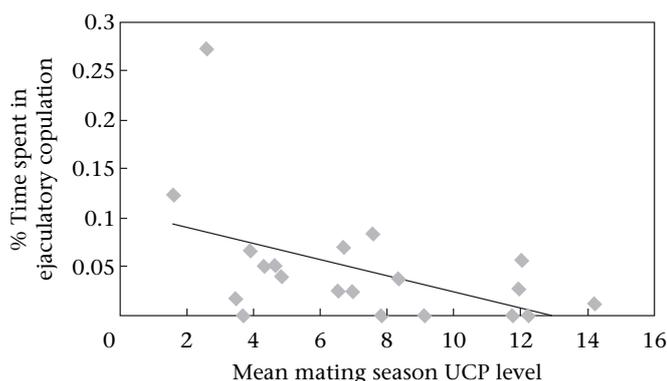


Figure 3. Relationship between time spent in ejaculatory copulation and mean mating season urinary C-peptide (UCP) of insulin levels in free-ranging rhesus macaques.

behaviour, body condition and energetic status all playing important roles in determining mating success. During the birth season, high-ranking males were less restless, travelled less, fed more on high-energy monkey chow, had higher levels of UCPs and were in better body condition than low-ranking males. Given that weight gain and loss in an individual are a direct consequence of relationships between energetic intake and expenditure, it seems highly likely that the observed differences in body condition between high- and low-ranking males may have resulted from differences in feeding behaviour and behavioural energy expenditure. Since restlessness was the only behavioural measure found to impact body condition above and beyond the effects of rank, our birth season data suggest that the rate at which an individual cycles through his/her behavioural repertoire might provide important information about the relationship between behaviour and energy use in free-ranging primates.

Previous studies of captive rhesus macaques have shown that higher-ranked males are fatter than lower-ranked males (Bercovitch & Nürnberg 1996), and that males lose substantial body mass and fat during the mating season (Bernstein et al. 1989; Bercovitch 1992; Bercovitch & Nürnberg 1996). However, the data in these studies of captive populations precluded thorough explorations of relationships between behaviour, dominance rank and body condition (Bercovitch 1997). During the mating season, in free-ranging populations, there is variation in male reproductive strategies both within and between individuals, with costly behaviours such as mate guarding increasing male reproductive success (Berard et al. 1994). In our study, male energetic condition during the birth season was correlated with condition during the mating season. The negative correlation between mating activity and feeding during the mating season suggests that mating acts as a significant constraint on the ability of males to spend time foraging. Therefore, it is primarily during the birth season that males have the opportunity to build condition. High-ranking males were in the best condition in the birth season, and only these males adopted consortship as a mating strategy. Consortship, in turn, was positively correlated with measures of copulatory activity, which was significantly negatively correlated with UCP levels. By the end of the mating season, high-ranking males had the lowest UCP levels. Our results are consistent with the hypothesis that male rhesus macaques engage in endurance rivalry, with rank-related differences in body condition and energetic status during the birth season directly determining differences in the ability of different males to undertake costly mating activities during the 6-month mating season. Similar differences in condition have been shown to determine differences in male ability to sustain mating in grey seals (Lidgard et al. 2005), mating chorus tenures in bullfrogs (Judge & Brooks 2001) and courtship periods in male green anoles

(Jenssen & Nunez 1998). As such, this type of phenomena could be widespread in the animal kingdom, although it is often not well documented. Endurance rivalry may also be seen as a type of 'carry-over effect', in which processes in one season directly determine success in another (Harrison et al. 2011).

During the mating season, only measures of mating predicted measures of energetic expenditure, suggesting that variation in mating overrides the importance of variation in any other behaviours in determining energetic condition during this period. In contrast, a number of behaviours were related to male rank during the birth season, and measures of energetic expenditure (travelling, restlessness) were also related to measures of body condition (BMI, UCP levels). However, only restlessness was related to any measure of body condition after controlling for rank effects. Moving from one state to another should use more energy than remaining in one state. When an animal is travelling it has momentum and it is easier to keep travelling and then rest than it is to stop, start, stop again and start again, even if the overall time spent resting and travelling are the same. This is because energy has to be repeatedly put into stopping the animal's forward movement against its momentum, and then restarting it, against its resting state. In addition, fidgeting and restlessness generally occur together (e.g. Dunn & Kronenberger 2003) and both are associated with increased energy consumption (e.g. Levine et al. 1999, 2000). As such, it could also be that our restlessness measure is acting more generally as a measure of fidgeting and nonexercise related activity.

There are two closely related mechanisms that may account for restlessness in lower-ranked individuals: greater disturbance by others, or greater levels of agitation and anxiety. By focusing solely on changes in behaviour between major state types (so ignoring social and self-directed behaviours), by showing that lower-ranked individuals spend more time alone (in fact, low-ranked individuals spent a majority of their time alone, and so were far less disturbed by others) than higher-ranked individuals, and by showing that rate of behavioural change is correlated with self-scratching (an index of anxiety in primates), we have provided evidence that differences in restlessness may reflect, at least in part, a higher level of anxiety. Clearly, however, the two mechanisms are not necessarily independent: greater disturbance by others is likely to lead to greater anxiety. Whatever the exact mechanism, lower-ranked males are more restless than higher-ranked males, and there appears to be direct energetic consequences of this.

In rhesus macaques and numerous other primate species, high-ranking individuals attack low-ranking individuals at high rates, often seemingly at random (Miller 1997; Silk 2002; Maestripieri 2007). It has been suggested that such random attacks are beneficial to attackers because they cause chronic activation of the stress response in victims, leading to health and fertility problems for lower-ranked individuals (Silk 2002). Here, we suggest a further consequence of such attacks; the anxiety that they cause may also lead to increased restlessness and energy expenditure. Anxiety-related energy expenditure, in turn, may reduce the ability of low-ranking males to gain weight and improve body condition, and hence to compete sustainably for females over time during the mating season (endurance rivalry). Thus, the results of our study add a dimension of complexity to the social competitive strategies of male primates and could lead to a better understanding of variability in mating and reproductive success.

Acknowledgments

We thank Doreen Hess, Jenna Goldfein, Maria Rakhovskaya, Alison Vitale and the staff of the Caribbean Primate Research Center for logistical support in the field and assistance with animal capturing and handling. We are deeply indebted to John Addicott for

writing the data parser and for helping to create our Access database. We are also extremely grateful to Andrea Heistermann for analysing the urine samples, and to Tara Mandalaywala and the Caribbean Primate Research Center for assisting with their transportation to Germany. Lauren Brent, Antje Engelhardt, Stuart Semple and anonymous referees provided helpful comments on a previous version of the manuscript. This study was conducted in accordance with the NIH Guide for the Care and Use of Laboratory Animals. The protocol for this study was approved by the Institutional Animal Care and Use Committee, Medical Sciences Department, University of Puerto Rico. This research was supported by NIH grant R21-AGO29862 to D.M., and by an IPS research grant to J.H. This publication was made possible by grant number CM-5-P40RR003640 from the National Institutes of Health (NIH) National Center for Research Resources (NCRR) to the Caribbean Primate Research Center of the University of Puerto Rico. Its contents are solely the responsibility of the authors and do not necessarily represent the official views of NCRR or NIH.

References

- Altmann, J. 1974. Observational study of behaviour: sampling methods. *Behaviour*, **49**, 227–267.
- Andersson, M. 1994. *Sexual Selection*. Princeton, New Jersey: Princeton University Press.
- Bahr, N. I., Palme, R., Möhle, U., Hodges, J. K. & Heistermann, M. 2000. Comparative aspects of the metabolism and excretion of cortisol in three individual nonhuman primates. *General and Comparative Endocrinology*, **117**, 427–438.
- Berard, J. D., Nürnberg, P., Epplen, J. T. & Schmidtke, J. 1994. Alternative reproductive tactics and reproductive success in male rhesus macaques. *Behaviour*, **129**, 177–201.
- Bercovitch, F. B. 1992. Estradiol concentrations, fat deposits, and reproductive strategies in male rhesus macaques. *Hormones and Behavior*, **26**, 272–282.
- Bercovitch, F. B. 1997. Reproductive strategies of rhesus macaques. *Primates*, **38**, 247–263.
- Bercovitch, F. B. & Nürnberg, P. 1996. Socioendocrine and morphological correlates of paternity in rhesus macaques (*Macaca mulatta*). *Journal of Reproduction and Fertility*, **107**, 59–68.
- Bernstein, I. S., Weed, J. L., Judge, P. G. & Ruehlmann, T. E. 1989. Seasonal weight changes in male rhesus macaques (*Macaca mulatta*). *American Journal of Primatology*, **18**, 251–257.
- Campbell, B. C. & Gerald, M. S. 2004. Body composition, age and fertility among free-ranging female rhesus macaques (*Macaca mulatta*). *Journal of Medical Primatology*, **33**, 70–77.
- Deschner, T., Kratzsch, J. & Hohmann, G. 2008. Urinary C-peptide as a method for monitoring body mass changes in captive bonobos (*Pan paniscus*). *Hormones and Behavior*, **54**, 620–626.
- Dunn, D. W. & Kronenberger, W. G. 2003. Attention-deficit/hyperactivity disorder in children and adolescents. *Neurologic Clinics*, **21**, 933–940.
- Emery Thompson, M. & Knott, C. D. 2008. Urinary C-peptide of insulin as a non-invasive marker of energy balance in wild orangutans. *Hormones and Behavior*, **53**, 526–535.
- Emery Thompson, M., Muller, M. N., Wrangham, R. W., Lwanga, J. S. & Potts, K. B. 2009. Urinary C-peptide tracks seasonal and individual variation in energy balance in wild chimpanzees. *Hormones and Behavior*, **55**, 299–305.
- Erb, R. E., Tillson, S. A., Hodgen, G. D. & Plotka, E. D. 1970. Urinary creatinine as an index compound for estimating rate of excretion of steroids in the domestic sow. *Journal of Animal Science*, **30**, 79–85.
- Girard-Buttoz, C., Higham, J. P., Heistermann, M., Wedegärtner, S., Maestriperi, D. & Engelhardt, A. In press. Urinary c-peptide of insulin as an energetic marker in macaques. *PLoS ONE*.
- Harris, T. R., Chapman, C. A. & Monfort, S. L. 2010. Small folivorous primate groups exhibit behavioral and physiological effects of food scarcity. *Behavioral Ecology*, **21**, 45–56.
- Harrison, X. A., Blount, J. D., Inger, R., Norris, D. R. & Bearhop, S. 2011. Carry-over effects as drivers of fitness differences in animals. *Journal of Animal Ecology*, **80**, 4–18.
- Heerey, E. A. & Kring, A. M. 2007. Interpersonal consequences of social anxiety. *Journal of Abnormal Psychology*, **116**, 125–134.
- Higham, J. P. & Maestriperi, D. 2010. Revolutionary coalitions in male rhesus macaques. *Behaviour*, **147**, 1889–1908.
- Hoffman, C. L., Ruiz-Lambides, A. V., Davila, E., Maldonado, E., Gerald, M. S. & Maestriperi, D. 2008. Sex differences in survival costs of reproduction in a promiscuous primate. *Behavioral Ecology and Sociobiology*, **62**, 1711–1718.
- Jenssen, T. A. & Nunez, S. C. 1998. Spatial and breeding relationships of the lizard, *Anolis carolinensis*: evidence of intrasexual selection. *Behaviour*, **135**, 981–1003.
- Judge, K. A. & Brooks, R. J. 2001. Chorus participation by male bullfrogs, *Rana catesbeiana*: a test of the energetic constraint hypothesis. *Animal Behaviour*, **62**, 849–861.
- Kavan, M. G., Elsasser, G. N. & Barone, E. J. 2009. Generalized anxiety disorder: practical assessment and management. *American Family Physician*, **79**, 785–791.
- Levine, J. A., Eberhardt, N. L. & Jensen, M. D. 1999. Role of nonexercise activity thermogenesis in resistance to fat gain in humans. *Science*, **283**, 212–214.
- Levine, J. A., Schleusner, S. J. & Jensen, M. D. 2000. Energy expenditure of non-exercise activity. *American Journal of Clinical Nutrition*, **72**, 1451–1454.
- Lidgard, D. C., Boness, D. J., Bowen, W. D. & McMillan, J. I. 2005. State-dependent male mating tactics in the grey seal: the importance of body size. *Behavioral Ecology*, **16**, 541–549.
- Maestriperi, D. 2007. *Macchiavellian Intelligence: How Rhesus Macaques and Humans Have Conquered the World*. Chicago: The University of Chicago Press.
- Maestriperi, D., Schino, G., Aureli, F. & Troisi, A. 1992. A modest proposal: displacement activities as an indicator of emotions in primates. *Animal Behaviour*, **44**, 967–979.
- Marra, M., Pasanisi, F., Montagnese, C., De Filippo, E., De Caprio, C., de Magistris, L. & Contaldo, F. 2007. BMR variability in women of different weight. *Clinical Nutrition*, **26**, 567–572.
- Mehrabian, A. & Friendman, S. L. 1986. An analysis of fidgeting and association individual differences. *Journal of Personality*, **54**, 406–429.
- Miller, G. F. 1997. Protean primates: the evolution of adaptive unpredictability in competition and courtship. In: *Machiavellian Intelligence II: Extensions and Evaluations* (Ed. by A. Whiten & R. W. Byrne), pp. 312–340. Cambridge: Cambridge University Press.
- Muller, M. N. & Wrangham, R. W. 2004. Dominance, aggression and testosterone in wild chimpanzees: a test of the 'challenge hypothesis'. *Animal Behaviour*, **67**, 113–123.
- Paul, A. 1989. Determinants of male mating success in a large group of Barbary macaques (*Macaca sylvanus*) at Affenberg Salem. *Primates*, **30**, 461–476.
- Rawlins, R. G. & Kessler, M. J. (Eds). 1986. *The Cayo Santiago Macaques: History, Behavior & Biology*. Albany, New York: SUNY Press.
- Sherry, D. S. & Ellison, P. T. 2007. Potential applications of urinary C-peptide of insulin for comparative energetic research. *American Journal of Physical Anthropology*, **133**, 771–778.
- Silk, J. B. 2002. Practice random acts of aggression and senseless acts of intimidation: the logic of status contests in social groups. *Evolutionary Anthropology*, **11**, 221–225.
- Takahashi, H. 2002. Changes in dominance rank, age, and tenure of wild Japanese macaque males in the Kinkazan A troop during seven years. *Primates*, **43**, 133–138.
- Troisi, A. 2002. Displacement activities as a behavioral measure of stress in nonhuman primates and human subjects. *Stress*, **5**, 47–54.