

AGGRESSION, REPRODUCTION, AND ANDROGENS IN WILD DWARF MONGOOSES: A TEST OF THE CHALLENGE HYPOTHESIS

Testosterone is classically thought of as the hormone controlling male reproduction. However, Wingfield et al. (1990; Wingfield and Moore 1987) have proposed that variation in testosterone levels may be more closely associated with temporal patterns of aggressive and paternal behavior than with changes in reproductive physiology. Specifically, the challenge hypothesis states that as the breeding season commences, testosterone levels in essentially all species rise from a nonbreeding baseline to a slightly higher breeding baseline. Breeding baseline levels of testosterone are sufficient for normal reproductive physiology and behavior but remain well below the physiological maximum, and they have little or no stimulating effect on male-male aggression and mate guarding (fig. 1 in Wingfield et al. 1990). Temporal patterns in testosterone levels within the breeding season are then predicted to differ between species, based on the amount of paternal care and the type of breeding system.

In breeding systems with high levels of paternal care (typical of monogamy), testosterone levels should increase above the breeding baseline only when aggression is triggered by challenging males or by mate guarding/mating. At other times, testosterone levels should remain at the breeding baseline so that aggression does not interfere with paternal care of offspring (Wingfield et al. 1990). In contrast, testosterone levels in breeding systems with little paternal care (typical of polygyny) should rise to near the physiological maximum and remain highly elevated throughout the breeding season, which would facilitate behavior appropriate for male-male competition. Consequently, in species with little paternal care, testosterone levels should respond weakly (if at all) to aggressive males or receptive females, because testosterone is already at or near the effective maximum. In species with considerable paternal care, testosterone should respond to behavioral challenges strongly (Wingfield et al. 1987, 1990).

Data from over 20 species of free-living birds support the above predictions clearly (Wingfield et al. 1990). Patterns of testosterone secretion in mountain spiny lizards also fit these predictions (Moore 1986), and Wingfield et al. (1990, page 830) suggest that the challenge hypothesis "may have wide application for vertebrates in general."

Mammals are conspicuous by their absence among the species for which the challenge hypothesis has been tested. Frank et al. (1985) and Wingfield et al. (1990) review several laboratory studies that suggest testosterone levels and aggression correlate in primates and rodents, but the correlation is complicated by environmental and social factors (Beach 1975; Dixon 1980; Bernstein et al. 1983;

Sachser and Prove 1984). In free-living olive baboons (*Papio anubis*) testosterone levels actually decrease during periods of aggression (and stress), with high-ranking males less affected than subordinates (Sapolsky 1984, 1985, 1987; see Wingfield et al. 1990, p. 830, for the importance of using data from the wild). Although these studies demonstrate associations between androgen levels and aggression in mammals, they do not allow a direct test of the challenge hypothesis.

In this note, we test the predictions of the challenge hypothesis using behavioral and endocrine data from a wild population of dwarf mongooses, *Helogale parvula*. Dwarf mongooses are small, social carnivores that live in cooperatively breeding packs (mean \pm SEM = 9.0 ± 0.3 individuals; Rood 1990). Within a pack, only the highest ranking female reproduces, although most adults of both sexes mate during synchronized estrous periods (Rood 1980, 1983; Creel et al. 1992; cf. Rasa 1973). Most subordinate females (88%) fail to produce litters, because of a series of rank-dependent effects on behavior and endocrine function (Creel et al. 1992). One to four litters are produced in a breeding season lasting from October through April.

In dwarf mongooses, male care of offspring is common (Rood 1978). For instance, the most frequent form of care is baby-sitting—guarding young at the den while the rest of the pack forages. Baby-sitting rates (calculated as the mean number of ≤ 5 -min bouts/h for 70 females and 79 males) did not differ between the sexes (males: 4.8 ± 0.99 ; females: 3.6 ± 0.78 ; $t = 0.93$, $P = .36$; mean \pm SEM throughout). Dwarf mongooses show considerable differentiation in parental behavior among age and sex classes (Rood 1978; Rasa 1987), and some parental behaviors are more common among females (e.g., carrying offspring during den moves), but male investment is comparable to that of monogamous species.

Given substantial paternal care, the challenge hypothesis predicts that androgen levels in dwarf mongooses should be elevated above the breeding baseline only during periods of mating or aggression. Conveniently, peaks of male-male aggression coincide with mating periods (Rasa 1973; Rood 1980): rates of aggression among males more than triple during mating periods (Creel et al. 1992).

METHODS

All data come from eight packs of individually marked dwarf mongooses (179 individuals, 82 males) living on a 25-km² study site in Serengeti National Park, Tanzania, between September 1987 and May 1989. The study area is composed of *Acacia-Commiphora* woodland, long-grass savanna, and granite outcroppings. Demography and behavior in this population have been described previously (Rood 1980, 1983, 1990; Creel and Creel 1991; Creel and Waser 1991; Creel et al. 1991). Detailed behavioral and endocrine methods are given in Rood (1980) and Creel et al. (1992). Briefly, we observed behavior daily (1,448 total h) using all-occurrences sampling to measure rates of aggressive and reproductive behavior. Quantitative dominance values (0–100, with the top-ranking individual = 100) were determined from aggressive interactions and displacements at resources, using best linear unbiased estimation (Friend et al. 1977).

We measured androgen levels by radioimmunoassay of 352 urine samples collected from 82 males. Of these, 193 were collected by trapping males and keeping them in the trap until they urinated into a pan below, usually within an hour at the trap site. Another 159 samples were collected without trapping, from a rubber pad on which the mongooses urinated in the course of normal scent marking. Trapping did not affect urinary androgen levels ($t = 0.40$, $P > .5$).

We assayed free androgens in ether extracts following arylsulfatase/beta-glucuronidase hydrolysis, using a quadruply tritiated tracer (NET-370, New England Nuclear) and antitestosterone-11-BSA (G. Niswender, Colorado State University). To control for variation in urine concentration, hormone concentrations are expressed as nanograms of androgen per milligram creatinine (Cr). Two serially diluted urine pools produced displacement curves parallel to that of serially diluted testosterone standard. Mean recovery of testosterone standards added to two urine pools was $78\% \pm 11.9\%$. Assay sensitivity was 12 pg per assay tube. The intra- and interassay coefficients of variation were 3.5% and 11.3%, respectively. Coelution trials with high-performance liquid chromatography showed that testosterone and androstanediol together accounted for 48% of all immunoreactivity found in hydrolyzed mongoose urine with this antibody. Five other compounds, including dihydrotestosterone, combined to account for the remaining immunoreactivity; each accounted for roughly equal immunoreactivity. Thus, the assay has broad specificity and measures total androgens.

Assay of androgens in urine yields concentrations that integrate changes in circulating blood androgen concentrations over a period of metabolization and pooling. Thus, slight or very short-term changes in circulating androgens might not be reflected in urinary concentrations. In *Helogale*, however, fluctuations in corticosteroids that occur over a matter of minutes to hours are readily detectable by assay of urine samples, as are fluctuations in estrogens that occur over 1–7 d. We are therefore confident that variation in androgens on the scale predicted by the challenge hypothesis would be detectable with our methods.

RESULTS AND DISCUSSION

As described above, dwarf mongooses show considerable paternal care of young, with peaks of male-male aggression during mating periods. The challenge hypothesis therefore predicts that androgens should remain at the breeding baseline except for elevations during periods of mating and aggression. Contrary to prediction, mean urinary androgen levels did not increase during periods of mating and aggression (fig. 1) for either alpha males (nonmating: 4.2 ± 0.6 ng/mg Cr; mating: 3.6 ± 0.4 ng/mg Cr, a nonsignificant decrease; $t = 1.20$, $P = .12$) or subordinates (nonmating: 3.6 ± 0.3 ng/mg Cr; mating: 3.8 ± 0.4 ng/mg Cr; $t = 0.35$, $P = .72$).

Urinary androgen levels did not correlate with mating rates (fig. 2a), rates of aggressive interaction (fig. 2b), or "winning rates" in aggressive interactions (fig. 2c). These patterns were robust (table 1), holding for all males pooled, for alpha males, and for males with nonzero rates of aggression or mating, with good sample sizes. (In contrast, we have demonstrated significant effects of behavior

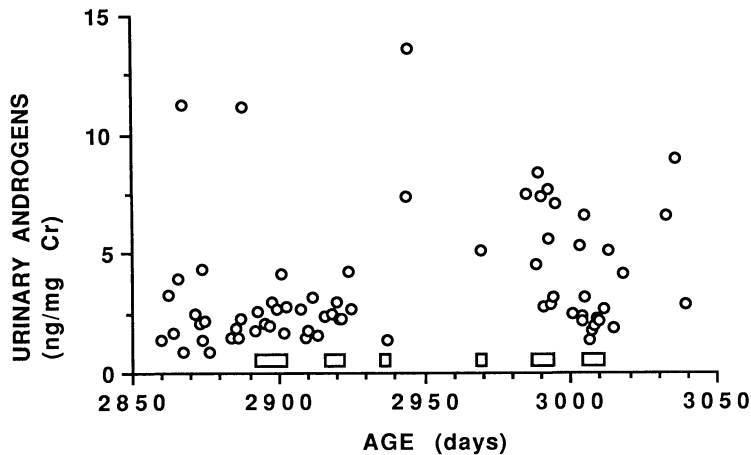


FIG. 1.—A profile of changes in urinary androgen levels for a single, dominant male dwarf mongoose over an entire breeding season (from October 4 to April 22) in Serengeti National Park, Tanzania. Mating periods are indicated by *open bars* just above the abscissa. There is no systematic increase in androgen levels during mating periods, despite heightened levels of aggressive (and sexual) behavior.

and dominance on estrogen levels in females of this population, sampling in the same way.) Multiple regression ($F = 1.46$, $df = 3$, 296 , $P = .23$) showed that androgen levels were not significantly affected by age ($t = 0.20$, $P = .83$), rank ($t = 1.51$, $P = .12$), or body mass ($t = 1.56$, $P = .12$).

Aggression was not confined to mating periods. Immigration events also led to intense male-male aggression. We used one-way ANOVA to compare the androgen levels of successful and unsuccessful immigrants, residents that were joined by immigrants, residents that repulsed immigrants, floaters, and males within 10 d of emigration. As shown in figure 3, floaters had androgen levels significantly higher than those of successful immigrants and residents that were joined by immigrants (Fisher's unprotected least significant difference). Because androgens were not elevated detectably just prior to emigration (fig. 3), elevated androgens appear to be caused by floating and not vice versa. No differences were found among other classes. Elevated androgens in floaters are consistent with the challenge hypothesis, as floaters were frequently involved in fights and chases with territorial males. Floaters' fights were also more violent than fights among pack mates, because submission did not end attacks. Unsuccessful immigrants and males that repulsed immigrants had the highest rates of aggression of all males. Their androgens were not elevated detectably above breeding baseline (fig. 3), but sample sizes were too small to allow reasonable statistical power.

An index of androgen responsiveness to aggressive or mating interactions can be made by dividing the peak androgen level (minus the nonbreeding baseline, near zero in *Helogale*) by the breeding baseline (Wingfield et al. 1990). The challenge hypothesis predicts that species with substantial paternal care should have indices of androgen responsiveness well above one, while species with little pater-

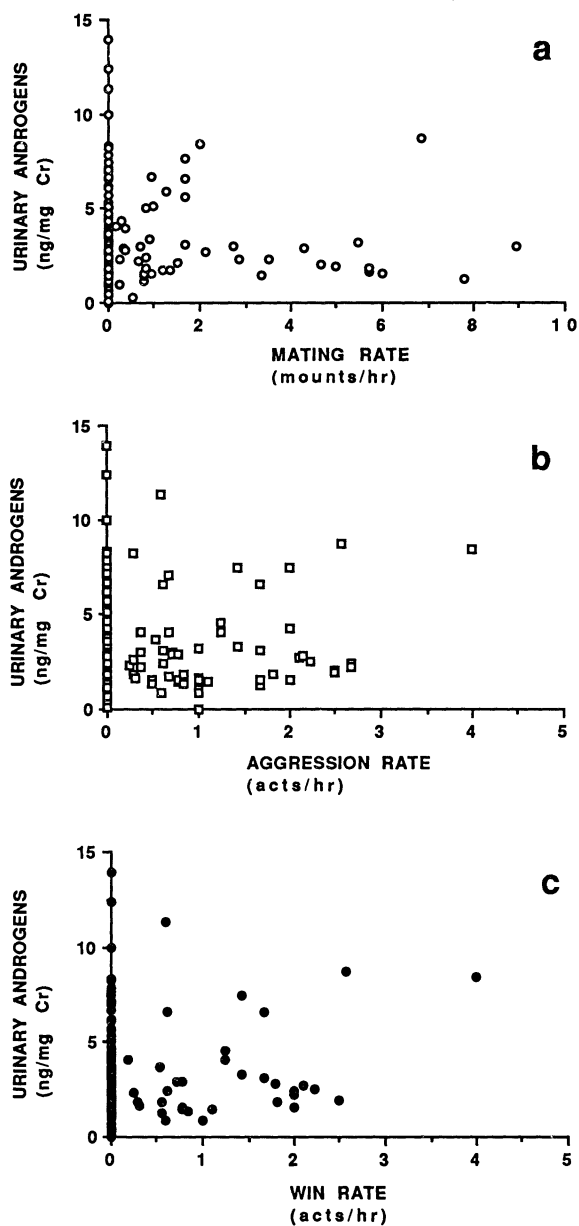


FIG. 2.—The relationships between urinary androgen concentrations of wild dwarf mongooses and hourly mating rate (a), hourly rate of aggressive encounters (b), and hourly rate of aggressive encounters won (c). Androgen levels do not correlate with the rates of these behaviors (also see table 1). Each point represents one urine sample because nested ANOVA indicated that urine samples were statistically independent units.

TABLE 1
CORRELATIONS BETWEEN URINARY ANDROGEN CONCENTRATIONS AND HOURLY RATES
OF AGGRESSION AND MATING

	CORRELATION BETWEEN ANDROGEN LEVEL AND								
	MATING RATE			AGGRESSION RATE			RATE OF AGGRESSIVE ENCOUNTERS WON		
	<i>n</i>	<i>r</i>	<i>P</i>	<i>n</i>	<i>r</i>	<i>P</i>	<i>n</i>	<i>r</i>	<i>P</i>
All males	211	-.07	.33	211	.02	.81	211	-.04	.58
Alpha males	67	-.14	.27	67	-.03	.76	67	-.03	.80
Males with rate >0	48	-.12	.38	57	.15	.28	36	-.19	.28
Alpha males with rate >0	22	-.22	.32	31	.12	.53	27	-.14	.47

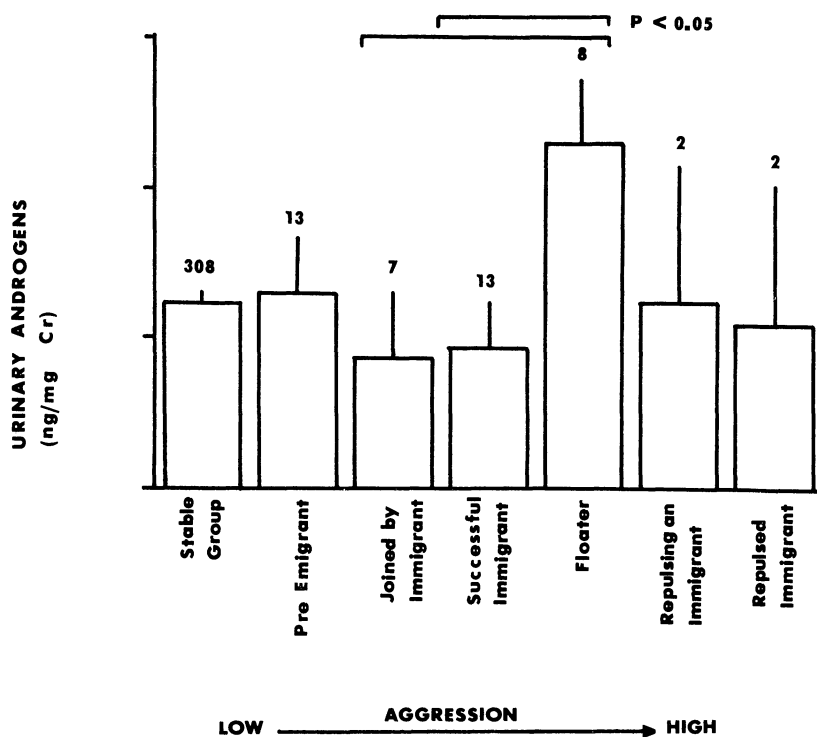


FIG. 3.—The effects of immigration, emigration, and floating on urinary androgen concentration in dwarf mongooses. Error bars show SEs. Numbers above bars are sample sizes. Horizontal lines denote significant differences, using Fisher's least significant difference. Categories are arranged from situations of low aggression to high aggression, left to right. *Stable group*, male in a pack with no immigration or emigration event in progress; *pre-emigrant*, male within 10 d of emigration; *joined by immigrant*, male in a pack that is allowing another male to join; *successful immigrant*, male in the course of successfully joining a pack; *floater*, nonterritorial male; *repulsing an immigrant*, male attacking another male that is unsuccessfully attempting to join his group; *repulsed immigrant*, male attempting to join a pack but persistently attacked and not successful in joining.

nal care should have values near one. In birds this pattern emerges as expected (Wingfield et al. 1990). We calculated this index for *Helogale*, using paired mean urinary androgen concentrations from mating and nonmating periods for each of 30 males, each sampled three or more times in each period. *Helogale*'s androgen responsiveness index was 1.10 ± 0.13 (minimum = 0.17, maximum = 3.35), not significantly greater than one ($t = 0.80$, $P = .43$). Comparing this value with data from birds, we found that *Helogale* fell into a cluster of species with little paternal care (modal index of 1) rather than the cluster of species with high levels of paternal care (modal index of 5).

Clearly, patterns of variation in androgen levels in dwarf mongooses did not correspond to those predicted by the challenge hypothesis. The reasons for this divergence from previously studied species are not clear, but some suggestions can be made based on features of dwarf mongooses' behavioral ecology that the challenge hypothesis does not explicitly address.

What features of *Helogale*'s ecology differ from previously studied species, and how might these differences affect selection on androgen levels? Perhaps the social complexity of multimale/multifemale breeding groups imposes additional selection pressures on androgen levels so that levels of aggression and parental care alone are not predictive. In packs of dwarf mongooses, the potential for sneaked copulations is high, relative to species that breed in territorial pairs or single-male polygynous groups (Creel et al. 1992). Also, matings with several males by a given female are common, and sperm competition may have an important impact on male reproductive success. Dwarf mongoose packs show rigid estrous synchrony so that all mating opportunities are concentrated into short spans of 1–7 d (Rood 1980). These three factors may select for high rates of mating and aggressive behavior. In fact, male mounting rates can exceed 80/h, and rates of aggression triple during group estrus (Creel et al. 1992). By themselves, high levels of aggression and mating should not constrain androgen levels to remain high during nonmating periods but would increase the cost of being "caught with one's androgens down."

Dwarf mongooses have as many as four sequential litters during a breeding season (5–7 mo), with an average of 2.5 litters per season. Perhaps more important, the environmental cues that affect the timing of estrous periods within a breeding season are not obvious; for instance, there is rarely any apparent synchrony in the timing of reproduction among adjacent packs. Thus, androgen levels must be elevated repeatedly, with a timing that may be difficult to predict.

Two differences between *Helogale* (and many other cooperative breeders) and previously studied species emerge: relatively strong selection for high androgens during mating periods and frequent, unpredictable mating periods. Together, these might confer a selective advantage on males with continuously elevated androgen levels, despite the need for paternal care.

Another explanation that cannot be excluded is that the physiological mechanisms of interaction between social behavior and endocrine function may differ between vertebrate classes. In birds, there are clear effects of aggression on circulating androgen levels (Wingfield et al. 1987, 1990; Wingfield and Moore

1987). Possibly, analogous effects arise in other taxa through differences not in the amount of hormone produced but in sensitivity of target tissues to endocrine stimulation. For example, olive baboon males differ in the degree to which testosterone secretion is depressed during periods of stress (Sapolsky 1984). Differences in testosterone concentration are primarily caused by differences between males in the sensitivity of interstitial cells of the testis to stimulation by luteinizing hormone (Sapolsky 1985). Similarly, mongooses might show changes through time in the sensitivity of aggressive, sexual, and paternal behavior to androgen levels in a way consistent with a broad interpretation of the challenge hypothesis.

In summary, patterns of variation in androgen levels are clearly related to social behavior and mating systems among free-living birds (Wingfield et al. 1987, 1990). As predicted by the challenge hypothesis, androgen levels are continuously high in polygynous bird species but are elevated only during periods of aggression in monogamous species. Data from free-living mammals bearing on these issues are sparse, but androgen profiles in cooperatively breeding dwarf mongooses differ from those predicted by the challenge hypothesis. It is noteworthy that androgen levels in the only cooperatively breeding bird yet tested (the white-browed sparrow weaver, *Plocepasser mahali*) also do not conform to the predictions of the challenge hypothesis (J. Wingfield, personal communication). Although the reasons for the differences are not yet clear, these results suggest that important variation may exist among taxa and within mating systems in the way that behavior and androgens interact.

ACKNOWLEDGMENTS

Our thanks to G. Perrigo, C. Snowdon, P. Waser, and K. Wynne-Edwards for helpful discussions and comments and to our friends at the Serengeti Wildlife Research Center, especially M. East and H. Hofer, for support in the field. We are grateful to D. Babu, K. Hirji, B. Maragesi, and H. Nkya for permission to work at the Serengeti Wildlife Research Center in Serengeti National Park, Tanzania. The manuscript was greatly improved through the comments of R. Allrich, R. Howard, K. Rabenold, J. Wingfield, and an anonymous reviewer. Our particular thanks to N. Marusha Creel for help in the field and lab. This research was supported by grants from the National Geographic Society, the National Science Foundation (8818040-BSR, to P. Waser), the Scholarly Studies Program of the Smithsonian Institution, Friends of the National Zoo, and a David Ross Fellowship from Purdue University.

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Submitted March 20, 1991; Revised March 19, 1992; Accepted March 24, 1992

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Associate Editor: Jeanne Altmann