Supporting Information

Cant et al. 10.1073/pnas.1312626111

SI Text

A Game Theoretical Analysis of Policing by Threat. Consider the interaction between a dominant breeder and an unrelated subordinate. The subordinate must decide whether or not she should also attempt to breed, assuming that if she does so, the dominant may then choose to commit infanticide. The dominant may not be able to discriminate with perfect accuracy between its own and the subordinate's young, so we assume that if she commits infanticide, she eliminates the subordinate's set of offspring with probability d and her own offspring with probability 1 - d, where d reflects its ability to discriminate. If the offspring of only one parent survive, that parent obtains a fitness payoff of 1, whereas if both sets of offspring survive, each parent obtains a fitness payoff of 1 - k, where k (< 1) reflects the cost of competition. Breeding by the subordinate entails an additive fitness cost c_b , and infanticide by the dominant entails an additive fitness $\cot c_i$ (which we assume is <1, so that infanticide is favored when discrimination is perfect).

What is the subgame perfect solution of this game (that is, the stable equilibrium outcome in a sequential game, assuming that incredible threats and promises are ignored)? In the second step, the dominant does best to commit infanticide provided that $d - c_i > c_i$ 1 - k. If this condition is not met, then the subordinate should certainly breed in the first step, because there is no plausible threat of infanticide; if the condition is met, and the threat of infanticide is real, the subordinate should nevertheless attempt to breed, provided that $1 - d - c_b > 0$. The solution of the game thus features infanticide when discrimination by the dominant, d, lies in the intermediate range $1 + c_i - k < d < 1 - c_b$. If d falls below the lower bound of this range, then the subordinate will breed but the dominant will nevertheless refrain from killing any young because her ability to identify her own offspring is too poor. If d falls above the upper bound of this range, then the subordinate will refrain from breeding because the threat of infanticide is too great. This is the zone where the threat of infanticide is sufficient to suppress subordinate reproduction. Note that as offspring become more costly (i.e., as c_b increases), as the cost of competition diminishes (i.e., as k decreases) and as the cost of infanticide grows (i.e., as c_i increases), the zone of infanticide shrinks. Ultimately, if $c_i + c_b > k$ (i.e., if the total cost of subordinate breeding and dominant infanticide exceeds the cost of competition, then infanticide will never occur. Fig. S1 shows the subgame perfect outcomes of the model for illustrative parameter values ($k = 0.2; c_i = 0.025$).

How does the model relate to our empirical findings? Our finding that banded mongoose females did not suffer immediate fitness costs when subordinates added to the communal litter indicates that the cost of cobreeding k is relatively low in this species. According to our model, therefore, subordinate banded mongoose females can escape the threat of infanticide by introducing a relatively small degree of maternity uncertainty (specifically, by reducing maternity discrimination d below $1 - k + c_i$, where c_i is the cost of committing infanticide). Escape from suppression is more difficult for social mammals in which there are substantial costs k of cobreeding (e.g. refs. 1 and 2). Variation within and across species in the costs of cobreeding and the costs of producing offspring determine the extent to which threats can be effective at suppressing reproduction among subordinates, and hence both the degree of reproductive skew and the frequency of infanticide. Because the cost of producing offspring is generally much higher in vertebrates compared with

insects, there is greater potential for policing by threats rather than acts of infanticide in the former than the latter.

Hormone Sample Collection, Extraction, and Assay Methods. Fecal samples were collected from 13 adult female banded mongooses (five treated with Depo-Provera and eight untreated) in two social groups 10 wk after pack oestrus. All samples were immediately placed on ice and transferred to a -20 °C freezer within 3 h. Samples were then transferred to the United Kingdom on ice and stored in a -20 °C freezer before hormone extraction. Hormones were extracted from fecal samples following thawing and manual homogenization using a wet-weight shaking extraction adapted from Walker et al. (3).

Fecal progesterone and estradiol metabolites were analyzed by modified previously described enzyme immunoassays (EIA) [Young et al. (4) adapted from Munro and Stabenfeldt (5)]. Each EIA used an antibody (monoclonal antiserum progesterone metabolite CL425; and polyclonal antiserum estradiol R0008, supplied by C. J. Munro, University of California, Davis, CA), horseradish peroxidase-conjugated label [progesterone and estradiol; prepared according to Munro and Stabenfeldt (5)] and standards (progesterone and estradiol; Sigma-Aldrich).

For the progesterone EIA, antiserum was diluted at 1:10,000 for progesterone in coating buffer (0.05M NaHCO3, pH 9.6) loaded 50 μ L per well on a 96-well Nunc-Immuno Maxisorp (Thermo-Fisher Scientific) microtiter plate, covered with a plate sealer, and left overnight at 4 °C. Plates were washed five times (0.15M NaCl, 0.05% Tween 20), standards (progesterone, 0.78–200 pg per well) or samples were loaded (50 μ L per well) onto each plate and the horseradish peroxidase conjugate was diluted in EIA buffer to 1:35,000 and added 50 μ L per well.

For the estradiol EIA, nonspecific goat anti-rabbit γ -globulin (IgG; Sigma, R2004) was diluted in coating buffer and loaded 1.0 µg in 250 µL per well on Nunc-Immuno Maxisorp (Thermo-Fisher Scientific) microtiter plates and left overnight at room temperature. The nonspecific IgG was then discarded and 300 µL per well of Tris blocking buffer (0.02 M Trizma, 0.300 M NaCl, 1.0% BSA, pH 7.5) was added and incubated for a minimum of 2 h at room temperature. Plates were washed five times (0.15 M NaCl, 0.05% Tween 20) and EIA buffer (0.1 M NaPO₄, 0.149M NaCl, 0.1% BSA, pH 7.0) was loaded at 50 µL per well. Standards (estradiol, 7.8–2,000 pg per well) or samples (diluted in EIA buffer) were loaded 20 µL per well, the horseradish peroxidase conjugate diluted in EIA buffer to 1:25,000 was added at 50 µL per well.

Following incubation in full light for 2 h at room temperature, plates were washed five times and incubated with 100 μ L per well of room temperature substrate [0.4 mM 2,2'-azino-di-(3-ethylbenzthiazoline sulfonic acid) diammonium salt, 1.6 mM H₂O₂, 0.05 M citrate, pH 4.0) and left to develop at room temperature full light.

The progesterone antiserum CL425 cross-reacted with several progesterone metabolites. This cross-reactivity was determined using binding inhibition curves, and calculated as the standard concentration (wt/wt) at 50% binding divided by the concentration of the competitive antigen at 50% binding, expressed as a percentage. Cross-reactivity results were as follows: 4-pregnen-3, 20-dione (progesterone) 100%; 4-pregnen-3 α -ol-20-one 188%; 4-pregnen-3 β -ol-20-one 172%; 4-pregnen-11 α -ol-3,20-dione 147%; 5 α -Pregnan-3 β -ol-20-one 94%; 5 α -Pregnan-3 α -ol-20-one 64%; 5 α -Pregnan-3, 20-dione 55%; 5 β -Pregnan-3 β -ol-20-one 12.5% and

 ${\leq}10\%$ for all other metabolites tested (6, 7). The estradiol-2 R0008 antiserum cross-reacted with Estradiol 17 β 100%, estrone 0.73% and estrone sulfate <0.01% progesterone <0.01% testosterone <0.01% cortisol <0.01% corticosterone <0.01% androstenedione <0.01% .

These progesterone and estradiol assays were validated for measuring progesterone and estradiol metabolites in female banded mongoose feces by parallelism and accuracy check. Serial dilutions of banded mongoose fecal extract yielded a displacement curve parallel to the progesterone standard curve [sample percent binding = 7.16 + 0.82 (standard percent binding), $R^2 = 0.92$, $F_{1,7} = 86.06$, P < 0.001]. There was no evidence of

- Clutton-Brock TH, et al. (2001) Cooperation, control, and concession in meerkat groups. Science 291(5503):478-481.
- Creel SR, Waser PM (1991) Failures of reproductive suppression in dwarf mongooses (Helogale parvula): Accident or adaption. Behav Ecol 2(1):7–15.
- Walker SL, Waddell WT, Goodrowe KL (2002) Reproductive endocrine patterns in captive female and male red wolves (*Canis rufus*) assessed by fecal and serum hormone analysis. *Zoo Biol* 21(4):321–335.
- Young KM, et al. (2004) Noninvasive monitoring of adrenocortical activity in carnivores by fecal glucocorticoid analyses. Gen Comp Endocrinol 137(2):148–165.

matrix interference, as addition of diluted fecal extract to progesterone standards did not alter the amount expected [observed = 0.92 + 1.57 (expected), $R^2 = 0.99$, $F_{1,7} = 631.87$, P < 0.001]. Serial dilutions of banded mongoose fecal extract yielded a displacement curve parallel to the estradiol standard curve [sample percent binding = 15.88 + 0.92 (standard percent binding), $R^2 = 0.94$, $F_{1,7} = 55.47$, P < 0.001]. There was no evidence of matrix interference, as addition of diluted fecal extract to estradiol standards did not alter the amount expected [observed = 1.13 + 3.87 (expected), $R^2 = 0.99$, $F_{1,7} = 9323.86$, P < 0.001].

- Munro C, Stabenfeldt G (1984) Development of a microtitre plate enzyme immunoassay for the determination of progesterone. J Endocrinol 101(1):41–49.
- Walker SL, Smith RF, Jones DN, Routly JE, Dobson H (2008) Chronic stress, hormone profiles and estrus intensity in dairy cattle. *Horm Behav* 53(3):493–501.
- Watson R, et al. (2013) Development of a versatile enzyme immunoassay for noninvasive assessment of glucocorticoid metabolites in a diversity of taxonomic species. *Gen Comp Endocrinol* 186(7):16–24.



Fig. S1. Stable outcomes in the two-step policing model as a function of the dominant's discrimination ability d and the cost of producing young for subordinates c_b . Infanticide is predicted to occur only in the shaded zone. (A) Threat not credible: in this zone the dominant will not commit infanticide so the subordinate can breed without risk. (B) Threat ineffective: in this zone the dominant kills some offspring but this does not deter the subordinate from breeding. (C) Threat effective: here the threat of infanticide is sufficient to suppress subordinate reproduction. (D) Subordinate abstains: in this zone breeding does not pay for the subordinate because the cost of producing young is too high.