

Supplementary Materials:

The influence of stress hormones and aggression on cooperative behaviour in subordinate meerkats

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Supplementary Materials, Methods & Results

(a) Validating methods to manipulate subordinate cortisol levels

We provisioned subordinate meerkats with either cortisol (10 mg/kg, hydrocortisone, Sigma H4126), mifepristone (40 mg/kg, Sigma M8046), or oil vehicle (100% coconut oil). We chose these dosages based upon previous studies in meerkats and other species and our own pilot studies reported herein (Figs. S1-S3). We based our dosage of cortisol on a previous study in meerkats (62) that showed that an injection of ~20 mg/kg caused plasma cortisol levels to be ~11x higher than individuals injected with saline (they were sampled ~2 hours after injection). We cut the dosage by 50% (10 mg/kg) in order to produce a less steep increase in circulating glucocorticoid concentrations. Our pilot experiments confirmed that this dosage caused a significant, but less severe, increase in plasma glucocorticoid (cortisol) concentrations (Fig. S2) in addition to a long-term elevation in faecal glucocorticoid metabolite concentrations (Fig. S3). We based our dosage of mifepristone on previous studies in mammals (30 mg/kg: 70; 50 mg/kg: 69) and birds (10 or 50 mg/kg in birds: 71) that used similar dosages of mifepristone. These studies show that these mifepristone dosages could affect behaviour (71), antagonize the glucocorticoid receptor (GR) but not cause toxicity (72), but are sufficient to bind to GRs as evident by their effects on raising circulating glucocorticoid levels (by exerting negative feedback on the hypothalamic-pituitary-adrenal axis: 69,73).

To prepare the dosages, cortisol (70 mg each time) or mifepristone (28 mg) was weighed on an analytical balance, and 100% coconut oil was added to achieve a standardized concentration for cortisol (0.07 mg/1 μ l of coconut oil) and mifepristone (0.14 mg/ μ l), the solutions were vortexed to suspend the cortisol or mifepristone, and then stored at 4° C prior to

use. New dosages were made at least every 7 d. Prior to provisioning meerkats with the dosages, we thoroughly vortexed and then injected 100 μ l of the emulsions into the thorax of a dead scorpion (*Opisthophthalmus* spp.), and fed the meerkats the scorpions. Meerkats were fed in the morning soon after foraging started. Control treatments were just 100 μ L of 100% coconut oil.

Effects of treatments on plasma glucocorticoid levels

To assess how our treatments affected plasma glucocorticoid levels, we obtained blood samples from subordinates prior to starting and during the treatments and measured plasma glucocorticoid (cortisol) levels using established protocols (7, 12, 15). Meerkats sampled during the treatment period were blood sampled 1-5 hours after they consumed the treatments (mean = 3.45 hrs). We confirmed that there were no statistically significant differences in plasma glucocorticoid (cortisol) levels prior to treatment (Fig. S2) and that subordinate meerkats fed cortisol had significantly higher plasma glucocorticoid levels than those fed mifepristone or the controls sampled on the same day that they were provisioned with their treatment (Fig. S2).

Effects of treatments on faecal glucocorticoid metabolite levels

We measured the effects of the treatments on faecal glucocorticoid metabolite (fGCM) levels, which represent an average level of circulating glucocorticoid levels experienced over a ~24-48 hr period (6, 74). Faecal samples (n=26) for fGCM monitoring were collected from 15 subordinate meerkats over the course of the experimental treatment period (range = 1-10 days after the treatments started, median = 4.5 d). Samples were taken immediately after the animal left the defecation spot, placed on wet ice, and stored frozen within 2-6 h after collection at -20° C until further processing. For fGCM extraction, carried out at the Endocrine Research Laboratory, Faculty of Veterinary Science, University of Pretoria, South Africa, frozen faeces were lyophilized, subsequently pulverized and fGCMs extracted from 0.10–0.11 g of faecal

powder using 3 ml 80% methanol in water (full details provided in 75). One ml of the extract was oven dried over night at 55 to 58° C (76) and shipped to the Endocrinology Laboratory of the German Primate Centre, Göttingen, Germany, for hormone analysis.

Faecal extracts were measured for immunoreactive 11 β -hydroxyetiocholanolone, a major metabolite of cortisol in meerkats (74). Its measurement has been validated and proven reliable in reflecting adrenocortical activity in meerkats (74,76). Prior to assay, dried faecal extracts were reconstituted in 0.5 ml 80% methanol in water by sonication in a water bath for 5 minutes, followed by 30 seconds of vortexing (76-77). Reconstituted extracts, diluted 1:10 to 1:300 (depending on concentration) in assay buffer, were then taken to assay according to the procedure described elsewhere (78). Sensitivity of the assay was 1 pg/well. Intra-assay coefficients of variation (CV) determined by repeated measurements of high and low value quality controls were 5.3% and 7.0%, respectively, and inter-assay CV values were 11.0% and 16.4% (n=2 assays). All fGCM concentrations reported are expressed as ln ng/g dry faecal matter.

During the treatment period, fGCM levels of subordinates fed cortisol were significantly higher than those fed mifepristone or the controls (Fig. S3). Taken together, these results indicate that the dosages of cortisol we fed to subordinate meerkats entered their bloodstream and elevated plasma glucocorticoid levels (Fig. S2) and that meerkats fed cortisol experienced significantly higher overall exposure to glucocorticoids than those fed mifepristone or the controls (Fig. S3). This is similar to previous studies in other mammal species where provisioning individuals with exogenous glucocorticoids each day for 6-28 continuous days caused sustained elevations in plasma glucocorticoids (79-81) or fGCM levels (80) during the treatment period. Finally, we confirmed that these experimental increases in cortisol levels were

not causing abnormal changes in meerkat behaviour, as indicated by the lack of treatment effects on meerkat activity levels (Fig. S5).

Validation of mifepristone dosage

We performed a separate set of experiments to confirm that mifepristone affected the expression of behaviours stimulated by exogenous cortisol, likely by antagonizing the GR and therefore lowering GR activity. To do this, we fed subordinate males cortisol (10 mg/kg, n=8), cortisol (10 mg/kg) plus mifepristone (40 mg/kg, n=4), or oil vehicle (n=6) for one day and then assessed the effects of these treatments on their foraging success during that day of treatment. Subordinates fed cortisol by itself had significantly higher foraging success compared to the controls on day 1 of treatment (Fig. S1) but meerkats fed cortisol along with mifepristone exhibited foraging success that was similar to the controls (Fig. S1) and lower but not significantly lower than those fed cortisol by itself (Fig. S1). Administration of mifepristone by itself for 10 continuous days during either babysitting or pupfeeding also lowered foraging success compared to those fed cortisol (Fig. S4). These results are consistent with previous studies that show that experimental increases in glucocorticoid concentrations promote food-seeking behaviour or food-intake (82-86) and that administration of mifepristone by itself can reduce food-seeking behaviour or food-intake (69,71,85). Overall, this indicates that our dosages of mifepristone reduced the activity of the glucocorticoid receptor and caused behavioural changes.

Mifepristone is a potent glucocorticoid receptor antagonist, but it is also a progesterone receptor antagonist (72,87). Although we cannot rule out the possibility that the effects of mifepristone on babysitting or pupfeeding behaviour were caused by its anti-progesterone receptor effects rather than its anti-glucocorticoid receptor effects, we think this is unlikely for

the following reasons. First, experimental administration with cortisol increased food-intake but co-administration with mifepristone prevented this change in behaviour (Fig. S1) and administration of mifepristone by itself reduced food-intake compared to those fed cortisol (Fig. S4). This indicates that mifepristone altered food-seeking behaviour that is known to be influenced by the activity of glucocorticoid receptors (see 69, 71-72 and 82-86). Second, we had *a priori* evidence in subordinate males that there should be a negative association between increased glucocorticoid receptor activity and babysitting (15) but a positive association between glucocorticoid receptor activity and pupfeeding (12). We found support for these associations when we reduced glucocorticoid receptor activity with mifepristone. Finally, if mifepristone caused changes in behaviour due to its anti-progesterone effects, we would expect that its effects would be most obvious in pregnant females. We only conducted our experiments in subordinate females that were not pregnant and in subordinate males. Consequently, the behavioural consequences of mifepristone on babysitting and pupfeeding behaviour were most likely caused by a reduction in glucocorticoid receptor activity rather than a reduction in progesterone receptor activity.

The dosages of mifepristone we used (40 mg/kg) were within the range that has been used in other species (see above) and also not known to be toxic (72). Nonetheless, we confirmed that our mifepristone dosages were not causing abnormal changes in behaviour by documenting subordinate behaviour during focal behavioural observations. In Fig. S5, we show that treating subordinates with mifepristone did not cause abnormal changes in their activity levels. Our own observations during the experiments (B. Dantzer, pers. obs.) also indicated that these dosages did not cause observable sickness behaviour or vomiting.

(b) Additional Details of Statistical Analyses

Effects of experimental treatments on babysitting and pupfeeding

We first used an information-theoretic approach (50) to examine the degree of support among different models to explain variation in 1) babysitting contributions (generalized linear mixed-effects model [GLMM] with binomial errors), 2) the probability that pupfeeding occurred during our focal observations (GLMM with binomial errors), and 3) the proportion of biomass that was found by subordinates and subsequently fed to pups foraging with the group (i.e., ‘generosity’: GLMM with binomial errors). We included sex and treatment and, in some models, an interaction term between sex of the subordinate and treatment to identify if there were sex-specific effects of the treatments on babysitting or pupfeeding. Although we aimed to provision all subordinates their treatments prior to starting the focal observations (focals), in some cases this was not possible and focals were conducted ≥ 24 hrs after the last treatment in 64 of 230 total focals. Consequently, in the two pupfeeding models (probability of pupfeeding during focals and the proportion of biomass found that was fed to pups), we also included a binary covariate for whether the subordinate was fed or not prior to the focals occurring and, in some models, an interaction term between treatment and whether the subordinate was fed prior to the focals. We did not include this latter covariate in the babysitting models because we used the average relative babysitting contribution over the entire treatment period as the response variable.

Group size, litter size, and age of the subordinate may affect subordinate contributions to babysitting (34,35) and pupfeeding (34,41). Foraging success and body mass may also influence babysitting and pupfeeding (34, 35). In some of our initial babysitting models (Table S1), we included a covariate for the average age-corrected body mass of treated subordinates during the treatment period. Average age-corrected body mass during the treatment period were the

residuals from a general linear model containing average morning body mass during the treatment period as the response variable and average age during the treatment period as the independent variable. In the initial models for the probability of observing pupfeeding during the behavioural focals (Table S2), we included a covariate for the total biomass of food found during the focal session to control for the effects of variation in foraging success on the probability of observing pupfeeding. We included some of these variables in our initial model selection procedures shown in Tables S1-S3. Relatedness is not associated with variation in pupfeeding (88) or babysitting (35) and including it in these models would have restricted our sample sizes (we did not have full relatedness information for all individuals) so we did not include it in our models shown in Tables S1-S3. Our model selection procedures revealed that the best model (with lowest AIC_c shown in Tables S1-S3) for babysitting and pupfeeding was the null model that did not contain any biological predictor variables. Because we were interested in the effects of the experimental treatments on babysitting and pupfeeding, we focus on and only report results from models (Tables S1-S3) containing biological predictor variables with the lowest AIC_c (model in bold face font in Tables S1-S3) in the main text and use these results for our interpretations. Because a second model containing biological predictor variables (i.e., not the null model) was within $\Delta AIC_c < 2$ in Tables S1-S3, we also include results from model averaging (52) from these top models in Tables S10-S12 (using the zero method: 52).

Causes of variation aggression directed at subordinates

We used AIC_c model selection procedures to identify the top model of those considered to explain variation in aggression by dominant females directed at subordinate meerkats (Table S7). In these GLMMs (Poisson error structure), we included characteristics of the individual (sex, age at sampling [days of age], age-corrected body mass), social group (group size, group size²,

proportion of adult group members (>12 months of age) that were females [group sex ratio]), and a 4-level categorical variable reflecting group stage, which indicated whether there were pups at the burrow being babysat, pups foraging with the group, no pups in the group and the dominant female was pregnant, or no pups in the group and the dominant female was not pregnant. We also included a fixed effect for relatedness between the subordinates and dominant female defined as whether the dominant female was either the mother or full sister of the subordinate or not (two-level categorical variable). Group size was the total number of individuals in the group and group sex ratio was the proportion of females in the group during the *ad libitum* behavioural observation session. To identify if there were sex-specific effects of the dominant female pregnancy status or the presence of pups at the burrow or foraging with the group on aggression directed at subordinates, we included an interaction term between sex and group stage (no pups and dominant female not pregnant, no pups but dominant female pregnant, pups at burrow being babysat, pups foraging with the group). In some models, we also included interactions between sex and age and sex and age-corrected body mass to investigate whether aggression directed at subordinates was elevated in older or heavier females but not males. We included a covariate for season to indicate whether the amount of aggression directed at subordinates differed between the summer (September-April) or winter (May-August) months and, in some models, an interaction between sex and season. All models included an offset for the total amount of time of the behavioural observation session (time was ln transformed) as the total amount of time we observed each subordinate or dominant was variable.

Associations between babysitting and pupfeeding and aggression received

We also used AIC_c model selection procedures to identify the top model of those considered to examine associations between the amount of aggression subordinates received

from dominant females and their 1) babysitting contributions (Table S8) and 2) the probability that pupfeeding occurred during our *ad libitum* behavioural observations (Table S9). In these GLMMs (binomial error structure), we included characteristics of the individual (sex, age, age-corrected body mass), social group (group size, total number of pups foraging with the group and being pupfed), and the amount of aggression per hour that the subordinate received from the dominant female over the previous month ($\ln+1$ transformed for the babysitting models). In some models, we also included interactions between sex and age, age-corrected body mass, group size, or the amount of aggression received. The response variable in these models was either the relative proportion of babysitting contributions provided by that individual or a binary response variable to indicate whether pupfeeding did or did not occur in the behavioural observation sessions. The model for the effects of dominant female aggression on subordinate pupfeeding behaviour included an offset for the total amount of time observing the group as there was variation in how much time we observed each subordinate.

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Supplementary Figures S1-S5 and Tables S1-S12

Figure S1. We confirmed that mifepristone was antagonizing glucocorticoid receptors by conducting a separate set of experiments where we fed subordinate males cortisol, cortisol plus mifepristone, or oil vehicle for one day and assessed their effects on food-intake (foraging success during the day of treatment). Subordinates that were fed cortisol ($n=8$) had significantly higher foraging success (grams of prey biomass consumed per hour) than controls fed oil vehicle ($n=6$, $t=-2.92$, $P=0.01$) but subordinates fed cortisol along with mifepristone did not have higher foraging success than the controls ($n=4$, $t=-0.99$, $P=0.34$). Subordinates fed cortisol by itself had slightly higher foraging success than those fed cortisol plus mifepristone ($t=-1.53$, $P=0.15$). Together, these data suggest that our dosages of mifepristone lowered the activity of the glucocorticoid receptor because they prevented exogenous glucocorticoids from increasing food-seeking behaviour (as was observed in those fed cortisol). Data are from an experiment separate from those shown in Fig. S4 where subordinate males were treated with one of the three treatments (treated for 1 d) and their foraging success on the day of treatment was quantified by weighing them in the morning prior to the treatments starting and when foraging commenced and in the evening when foraging ended. We quantified foraging success as the evening body mass minus the morning body mass and estimated the number of grams gained of food consumed per hour as we had the time of each body mass measure. Results are from a general linear model with foraging success as the response variable (grams of prey biomass consumed per hour) and treatment as the independent variable.

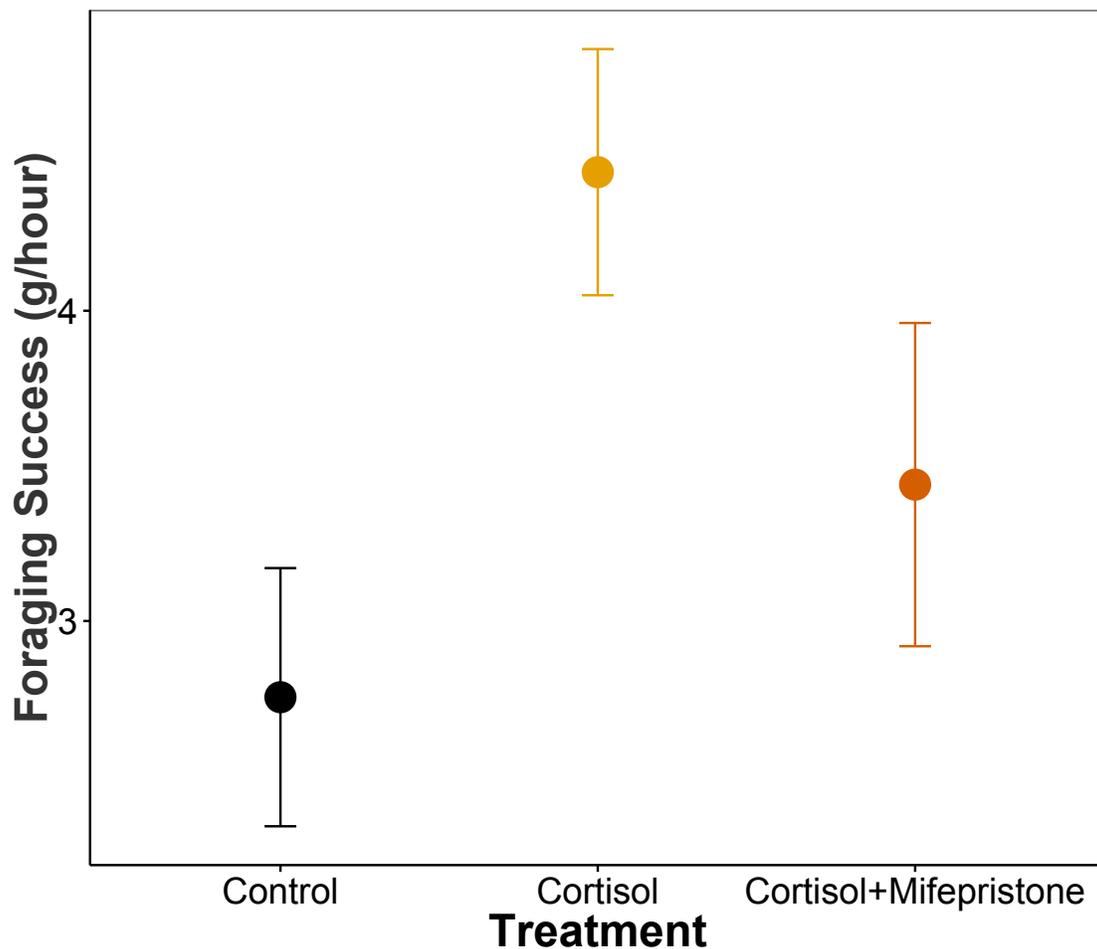


Figure S2. Prior to the treatments starting (“Before”), plasma glucocorticoid (cortisol) levels from subordinate meerkats that were to be treated with cortisol (n=12) were similar to those from subordinates that were to be treated with mifepristone (n=9, $t=0.85$, $P=0.39$) or oil vehicle (controls, n=7, $t=-0.47$, $P=0.38$). Plasma cortisol levels in subordinates that were to be treated with mifepristone did not differ compared to the controls ($t=1.21$, $P=0.23$). During the treatment period (“During”), subordinates fed cortisol (n=8) had significantly higher plasma cortisol levels compared to those fed mifepristone (n=5, $t=-3.05$, $P=0.004$) and the controls (n=7, $t=-3.62$, $P=0.001$). Plasma cortisol levels in subordinates being fed mifepristone did not differ compared to those from the controls ($t=0.41$, $P=0.68$). Meerkats sampled during the treatment period were blood sampled 1-5 hours after they consumed the treatments (mean = 3.45 hrs). Results are from a general linear mixed-effects model that contained treatment (mifepristone, control, cortisol), treatment period (before or during treatment), sex, sample acquisition time (ranging from 1-7 min, median=4 min), and a two-way interaction between treatment and treatment period. We also included random intercept terms for individual identity and group because we had repeated samples on the same individuals from the same groups (48 samples from 36 individuals in 11 different groups). A separate model that included an interaction term between sex, treatment, and treatment period confirmed that there were no sex-specific effects of the treatments on plasma cortisol levels prior to or during the treatments (interaction between sex x treatment x treatment period, $F_{1,33} = 0.03$, $P=0.86$). Boxplots show median (solid horizontal line), mean (grey diamonds), and first (25%) and third (75%) quartiles.

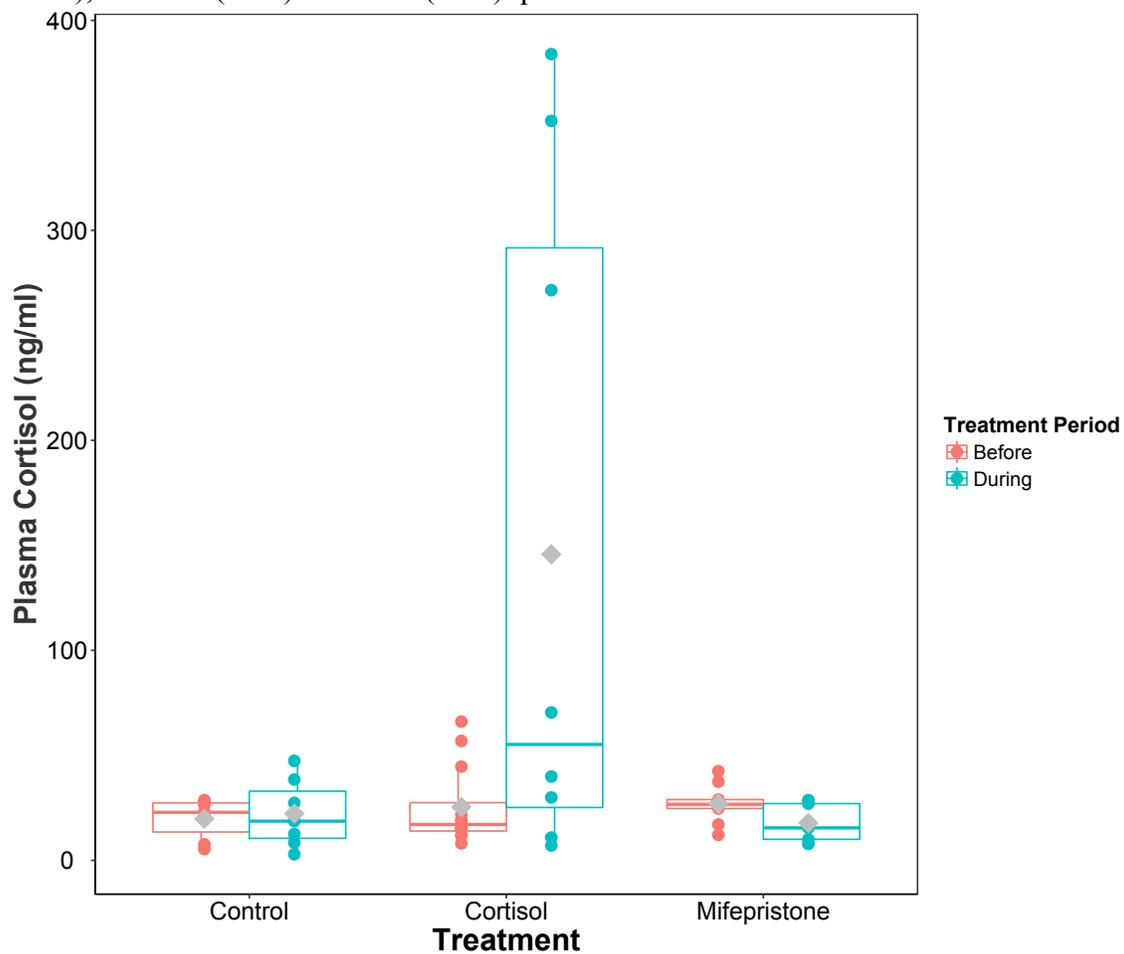


Figure S3. Subordinates that were being treated with cortisol (n=9) had significantly higher faecal glucocorticoid metabolite levels than those fed mifepristone (n=9, $t=-5.74$, $P<0.0001$) and the controls (n=8, $t=-4.34$, $P=0.0003$). There were no differences between subordinates that were being treated with mifepristone and the controls ($t=-1.44$, $P=0.17$). The increase in faecal glucocorticoid metabolites for those treated with cortisol was within the biologically-relevant range in meerkats (89). Results are from a general linear mixed-effects model that included treatment, sex, and a 3-level categorical variable for the time of day the sample was collected (see 74) and random intercept terms for individual identity and group (26 samples from 15 individuals in 8 groups). A separate model that included an interaction term between treatment and sex confirmed that there were no sex-specific effects of the treatments on faecal glucocorticoid metabolite levels ($F_{2,19} = 0.52$, $P=0.6$). Boxplots show median (solid horizontal line), mean (grey diamonds), and first (25%) and third (75%) quartiles.

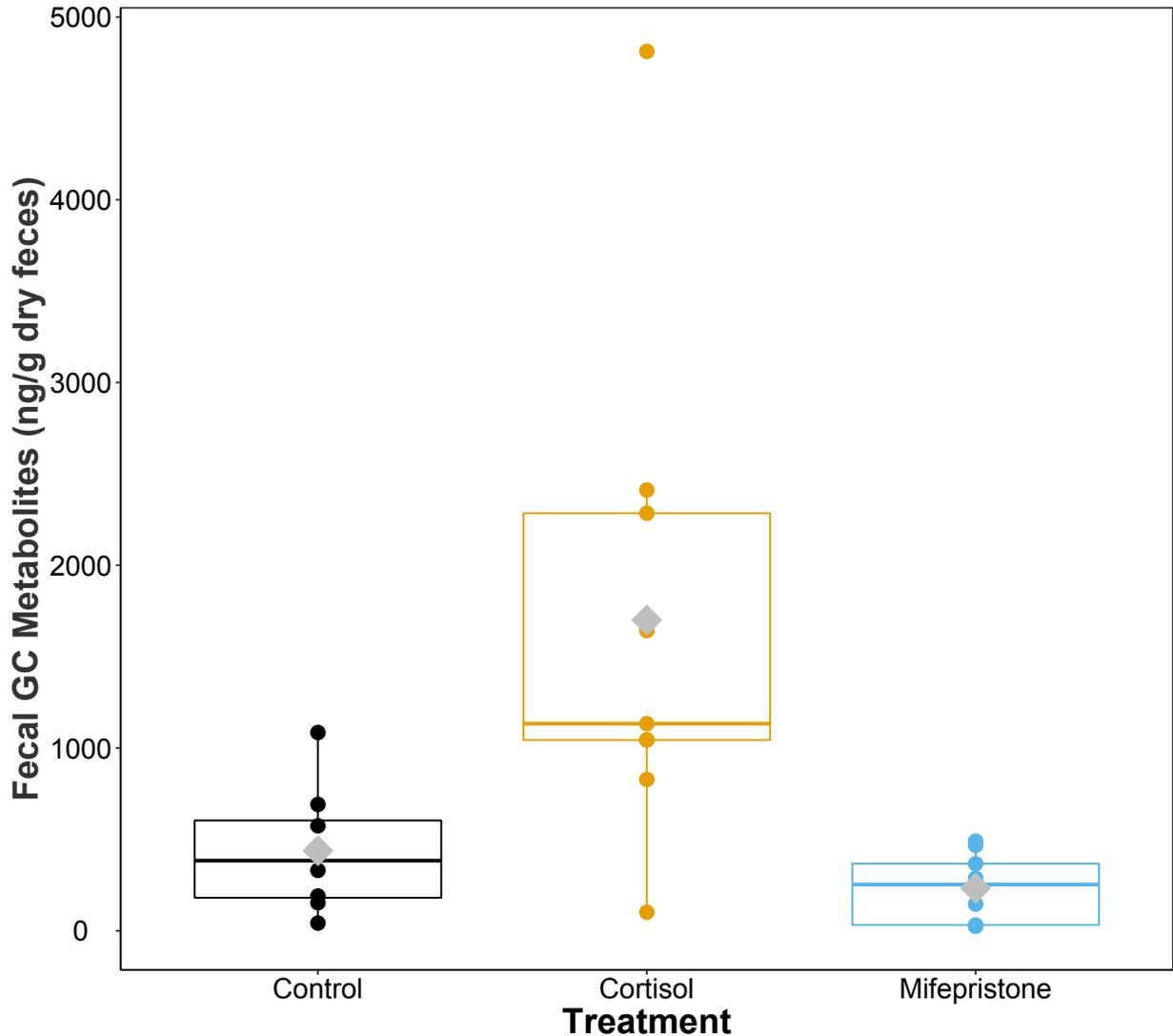


Figure S4. Subordinate meerkats fed mifepristone ($n=20$) during babysitting or pupfeeding tended to have lower foraging success than those fed cortisol ($n=21$, $t=1.92$, $P=0.06$) but not controls fed oil vehicle ($n=16$, $t=1.32$, $P=0.19$). Foraging success did not differ between subordinates fed cortisol and the controls ($t=-0.47$, $P=0.64$), suggesting that although short-term increases in plasma cortisol levels increased foraging success (Fig. S1), treatment with cortisol for 10 days did not. Values on y-axis represent average foraging success (change in body mass from morning to mid-afternoon divided by the number of hours) averaged over the entire treatment period. Results are from a general linear mixed-effects model with foraging success (grams of prey biomass consumed per hour) as the response variable, sex and treatment as fixed effects, and individual identity as a random intercept term because of repeated observation on the same individuals.

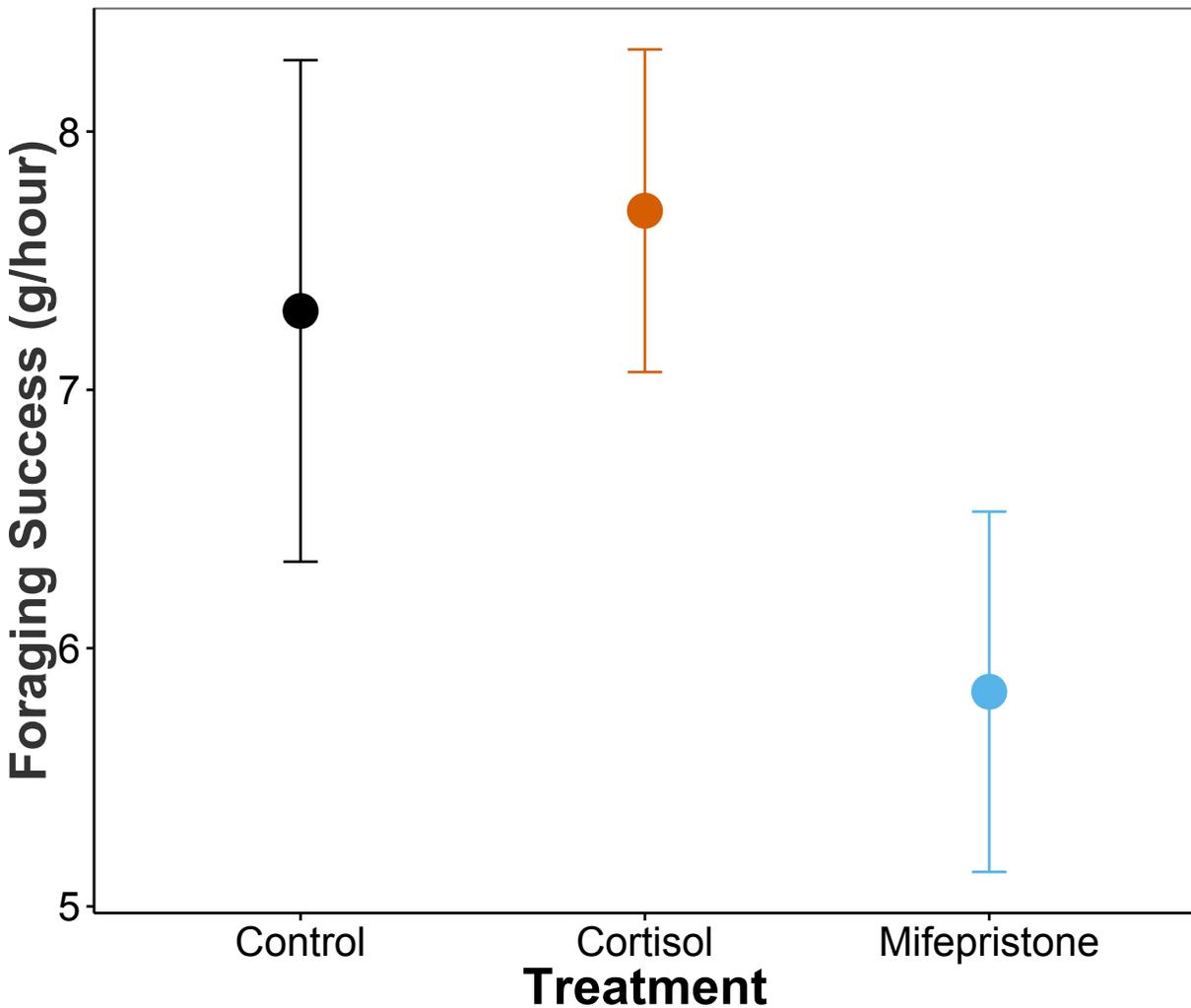


Figure S5. Our experimental treatments did not affect the average amount of time subordinate meerkats spent (A) moving ($F_{2,232} = 2.3$, $P=0.11$) or (B) scrabbling for food ($F_{2,203} = 0.87$, $P=0.42$) during 20 min continuous focal behavioural observations collected during the babysitting and pupfeeding treatment periods. Results are from a general linear mixed-effects model with time spent moving or scrabbling as the response variable (ln transformed), sex, treatment, treatment period (babysitting, pupfeeding), and interaction terms between sex and treatment or treatment period as fixed effects, and individual identity as a random intercept term because of repeated observation on the same individuals. There was no evidence of sex-specific effects of the treatments on the behaviours or that the effects of the treatments on the behaviours depended upon whether the meerkats were treated during the babysitting or pupfeeding treatment periods as indicated by the lack of significant interactions between treatment and period (babysitting or pupfeeding) in any of these comparisons (P-value at least >0.21 for all interaction terms). Sample sizes were the following: mifepristone (3140 min focal behavioural data on 27 individuals), cortisol (2840 min on 22 individuals), controls (2480 min on 23 individuals).

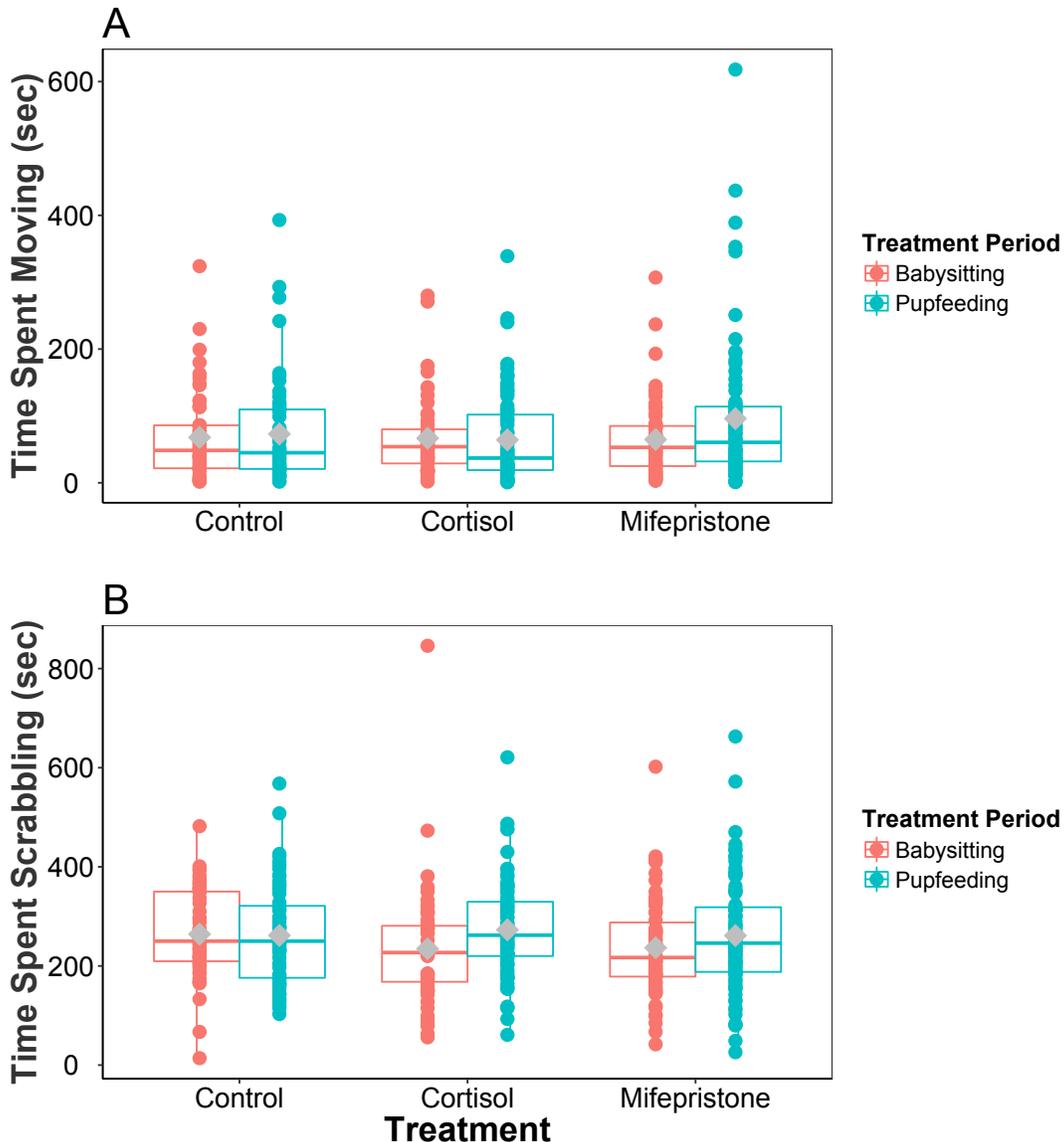


Table S1. AIC_c model selection for identifying the effects of the treatments on relative babysitting contributions exhibited by subordinate meerkats fed cortisol (n=9), mifepristone (n=9), or oil vehicle (controls, n=8). All models were generalized linear mixed-effects models (binomial errors) that were fit by maximum likelihood and included a random intercept terms for litter (L) and an observation level random effect (obs). Results from the model presented in the main text shown in bold. Model averaging results for the two top models (italicized) containing biological variables with <2 ΔAIC_c are shown in Table S10.

Fixed Effects	Random Effects	K	AIC_c	ΔAIC_c
Null	L, obs	3	130.1	0
<i>S+T</i>	<i>L, obs</i>	6	<i>133.2</i>	<i>3.07</i>
<i>GS+S+T</i>	<i>L, obs</i>	7	<i>134.8</i>	<i>4.74</i>
S+T+S*T	L, obs	8	136.8	6.65
A+BM+S+T	L, obs	8	139.5	9.42
GS+S+T+S*T	L, obs	9	140.7	10.59
GS+A+BM+S+T	L, obs	9	141.6	11.52
GS+S+T+S*GS+S*T	L, obs	10	144.3	14.21
A+BM+S+T+S*T	L, obs	10	145.3	15.24
A+BM+S+T+S*A+S*BM+S*T	L, obs	12	156.7	26.62
GS+A+BM+S+T+GS*S+S*A+S*BM+S*T	L, obs	14	165.5	35.42

“S” is sex of the subordinate, “A” is age of the subordinate, “T” is treatment, “GS” means group size during the babysitting period, “BM” is the age-corrected body mass.

Table S2. AIC_c model selection for identifying the effects of the treatments on the probability that pupfeeding occurred during our behavioural focal observations. All models were generalized linear mixed-effects models (binomial errors) that were fit by maximum likelihood and included random intercept terms for identity of the subordinate meerkat (ID), litter identity (L), and observer that collected the behavioural data (O). Results from the model presented in the main text shown in bold. Model averaging results for the two top models (italicized) containing biological variables are shown in Table S11.

Fixed Effects	Random Effects	<i>K</i>	AIC_c	ΔAIC_c
Null	ID, L, O	4	306.3	0
<i>F+S+T+S*T</i>	<i>ID, L, O</i>	<i>10</i>	<i>311.6</i>	<i>5.35</i>
<i>F+S+T</i>	<i>ID, L, O</i>	8	313.1	6.84
F+S+LS+S+T+S*T	ID, L, O	12	314.1	7.85
F+A+FS+S+T	ID, L, O	10	314.8	8.51
F+S+T+F*T+S*T	ID, L, O	12	315	8.69
F+S+T+F*T	ID, L, O	10	316.5	10.21
F+GS+LS+S+T+F*T	ID, L, O	12	317.1	10.78
F+GS+LS+A+FS+S+T+F*T	ID, L, O	14	317.2	10.89
F+A+FS+S+T+F*T+S*T	ID, L, O	14	317.5	11.24
F+GS+LS+S+T+F*T+S*T	ID, L, O	14	317.7	11.39
F+A+FS+S+T+F*T	ID, L, O	12	318.4	12.17
F+GS+LS+A+FS+S+T+F*T+S*T	ID, L, O	16	319.2	12.96

“F” indicates whether the subordinate was fed or not on the same day of the behavioural observations, “S” is sex of the subordinate, “A” is age of the subordinate, “T” is treatment, “GS” means group size during the pupfeeding period, “LS” is the litter size of pups that were foraging with the group, “FS” is the foraging success of the subordinate during the focal observation session (total grams of prey biomass found). Sample sizes for each treatment were as follows: controls (n=7 females, n=7 males), cortisol (n=10 females, n=4 males), and mifepristone (n=8 females, n=7 males) whose behaviour was recorded for 400-1200 min (see main text).

Table S3. AIC_c model selection for identifying the effects of the treatments on the proportion of biomass that was found by subordinates and subsequently fed to pups foraging with the group (“generosity”) during our behavioural focal observations. All models were generalized linear mixed-effects models (binomial errors) that were fit by maximum likelihood and included a random intercept term for identity of the subordinate meerkat (ID), litter identity (L), and observer (O). Results from the model presented in the main text shown in bold. Model averaging results for the two top models (italicized) containing biological variables are shown in Table S12.

Fixed Effects	Random Effects	<i>K</i>	AIC_c	ΔAIC_c
Null	ID, L, O	4	257.4	0
<i>F+S+T+S*T</i>	<i>ID, L, O</i>	<i>10</i>	<i>263.2</i>	<i>5.8</i>
<i>F+S+T</i>	<i>ID, L, O</i>	8	264.6	7.2
F+A+S+T	ID, L, O	9	265.3	7.9
F+S+T+F*T+S*T	ID, L, O	12	265.4	8.0
F+GS+LS+S+T+S*T	ID, L, O	12	265.6	8.2
F+S+T+F*T	ID, L, O	10	266.4	9.0
F+GS +LS+S+T+F*T+S*T	ID, L, O	13	266.9	9.5
F+A+S+T+F*T	ID, L, O	11	267.2	9.8
F+GS+LS+S+T+F*T+S*T	ID, L, O	14	267.9	10.5
F+GS+LS+A+S+T+F*T+S*T	ID, L, O	15	269.5	12.1
F+GS+LS+S+T+F*T	ID, L, O	12	269.7	12.3
F+GS+LS+A+S+T+F*T	ID, L, O	13	270	12.6

Variable abbreviations are the same as described for Tables S1 and S2. Sample sizes for each treatment were as follows: controls (n=7 females, n=7 males), cortisol (n=10 females, n=4 males), and mifepristone (n=8 females, n=7 males) whose behaviour was recorded for 400-1200 min (see main text).

Table S4. AIC_c model selection for identifying the causes of variation in the amount of aggression subordinate meerkats received from the dominant female. All models were generalized linear mixed-effects models (Poisson error structure) that were fit by maximum likelihood and included random intercept terms for identity of the subordinate meerkat (I), social group (G), dominant female (DF), year (Y), and the *ad libitum* behavioural observation session (SS). Models included an offset for observation time. Results from top model (in bold) presented in Table S7.

Fixed Effects	Random Effects	K	AIC_c	ΔAIC_c
S+A+BM+GST+R+SR+GS+GS²+SN+S*A+S*B M+S*GST	I, G, DF, Y, SS	22	18690.2	0
S+A+BM+GST+R+SR+GS+GS ² +SN+S*A+S*BM	I, G, DF, Y, SS	19	18693.3	3.0
S+A+BM+GST+R+SR+GS+GS ² +SN+S*A+S*BM +S*GST+S*R+S*SR+S*GS+S*GS ²	I, G, DF, Y, SS	26	18694.9	4.7
S+A+BM+GST+R+SR+GS+GS ² +SN+S*GST+S*R +S*SR+S*GS+S*GS ²	I, G, DF, Y, SS	24	18762.4	72.1
S+A+BM+GST+R+SR+GS+GS ² +SN	I, G, DF, Y, SS	17	18771.5	81.2
S+A+BM	I, G, DF, Y, SS	10	18806.4	116.2
S+A+BM+GST+R+SR+GS+GS ² +SN+S*GST	I, G, DF, Y, SS	20	18847.1	156.9
S+GST+R+SR+GS+GS ² +SN	I, G, DF, Y, SS	15	19250.9	560.7
Null	I, G, DF, Y, SS	6	19343.1	652.9

“S” is sex of the subordinate, “A” is age of the subordinate, “BM” is the age-corrected body mass, “GST” or group stage is a 4-level-level categorical variable to indicate whether there were pups at the burrow being babysat, pups foraging with the group that were being pupped by subordinates, no pups in the group and the dominant female was pregnant, or no pups in the group and the dominant female was not pregnant, “R” is relatedness of the subordinate to the dominant female, “SR” is the proportion of females in the group (group sex ratio), “GS” means group size during the pupfeeding period and “GS²” is a quadratic term for group size, and “SN” is season. Data used in these analyses were collected from >110,483 hours of behavioural data collected over 19 years that provided us with rates of aggression by 98 dominant females towards 1520 subordinates (713 females, 807 males) in 40 different groups.

Table S5. AIC_c model selection for identifying how the amount of aggression subordinate meerkats received from the dominant female affected their relative babysitting contributions. All models were generalized linear mixed-effects models (binomial error structure) that were fit by maximum likelihood and included random intercept terms for identity of the subordinate meerkat (I), litter (L), dominant female (DF), and year (Y). Results from top model presented in Table S8.

Fixed Effects	Random Effects	K	AIC_c	ΔAIC_c
S+A+BM+GS+Agg+S*A+S*BM+S*GS+S*Agg	I, L, DF, Y	15	39188.3	0
S+A+BM+GS+Agg+S*Agg	I, L, DF, Y	12	39338.2	149.9
S+A+BM+GS+Agg	I, L, DF, Y	11	39343.3	155
S+A+BM+Agg+S*A+S*BM	I, L, DF, Y	12	39618.1	429.8
S+A+BM+Agg+S*Agg	I, L, DF, Y	11	39759.1	570.8
S+A+BM+Agg	I, L, DF, Y	10	39762.7	574.4
S+GS+Agg+S*GS+S*Agg	I, L, DF, Y	11	40404.5	1216.2
S+GS+Agg+S*Agg	I, L, DF, Y	10	40439.9	1251.6
S+GS+Agg	I, L, DF, Y	9	40449.6	1261.3
S+Agg+S*Agg	I, L, DF, Y	9	40724.5	1536.2
S+Agg	I, L, DF, Y	8	40731.5	1543.2
Null	I, L, DF, Y	5	40789.4	1601.12

Variable abbreviations are the same as described for Table S4 except here “Agg” is the amount of aggression the subordinate received from the dominant female (number of assertions received over observation time). Data used in these analyses are from 9410 estimates of babysitting contributions from 1487 subordinate meerkats during 639 different litters produced by 96 different dominant females over the 19 years.

Table S6. AIC_c model selection for identifying how the amount of aggression subordinate meerkats received from the dominant female affected the probability of pupfeeding during *ad libitum* behavioural observations. All models were generalized linear mixed-effects models (binomial error structure) that were fit by maximum likelihood and included random intercept terms for identity of the subordinate meerkat (I), litter (L), dominant female (DF), and year (Y). Models included an offset for observation time. Results from top model presented in Table S9.

Fixed Effects	Random Effects	K	AIC_c	ΔAIC_c
LS+S+A+BM+GS+Agg+S*A+S*BM+S*GS+S*Agg	I, L, DF, Y	15	6460.8	0
LS+S+A+BM+GS+Agg+S*Agg	I, L, DF, Y	12	6466.1	5.27
LS+S+A+BM+GS+Agg	I, L, DF, Y	11	6467.1	6.28
LS+S+A+BM+Agg+S*A+S*BM	I, L, DF, Y	12	6500.2	39.42
LS+S+A+BM+Agg+S*Agg	I, L, DF, Y	11	6507.6	46.78
LS+S+A+BM+Agg	I, L, DF, Y	10	6508.9	48.13
LS+S+GS+Agg+S*Agg	I, L, DF, Y	10	6813.2	352.41
LS+S+GS+Agg	I, L, DF, Y	9	6815.0	354.21
LS+S+GS+Agg+S*GS+S*Agg	I, L, DF, Y	11	6815.1	354.3
LS+S+Agg+S*Agg	I, L, DF, Y	9	6840.7	379.86
LS+S+Agg	I, L, DF, Y	8	6842.8	381.98
Null	I, L, DF, Y	5	7101.0	640.16

Variable abbreviations are the same as described for Table S5 but here “LS” refers to the litter size or number of pups that were foraging with the group. Data used in these analyses were from 7619 estimates of the probability of exhibiting pupfeeding by 1626 subordinates towards pups from 544 different litters produced by 92 dominant females over 20 years.

Table S7. Results from a generalized linear mixed-effects model to examine the causes of variation in the amount of aggression subordinate meerkats received from the dominant female. This model (Poisson error structure) included random intercept terms for identity of the subordinate meerkat, litter, dominant female, year, and the *ad libitum* behavioural observation session. An offset for observation time was included. Reference for the categorical variables described below and in parentheses where applicable. Significant variables in bold.

Fixed Effect	b ± SE	z	P-value
Intercept	-9.6 ± 0.12	-77.8	<0.0001
Sex (Male)	-0.8 ± 0.09	-8.5	<0.0001
Age	0.77 ± 0.04	19.2	<0.0001
Body mass	0.34 ± 0.03	9.53	<0.0001
Group stage			
Babysitting groups	-0.59 ± 0.19	-3.06	0.0022
Pupfeeding groups	-0.23 ± 0.11	-2.01	0.044
Dom. Fem. Not Pregnant, No Pups	-0.54 ± 0.15	-3.7	0.0002
Dom. Fem. Mom or Sister? (No)	0.08 ± 0.08	1.02	0.31
Sex ratio	-0.05 ± 0.05	-0.88	0.38
Group size	0.08 ± 0.21	0.39	0.69
Group size²	-0.45 ± 0.22	-2.06	0.039
Season (Winter)	0.08 ± 0.09	0.8	0.42
Sex (Male) x Age	-0.49 ± 0.05	-8.9	<0.0001
Sex (Male) x Body mass	-0.11 ± 0.05	-1.92	0.055
Sex (Male) x Group stage			
Sex (Male) x Babysitting groups	0.55 ± 0.2	2.73	0.006
Sex (Male) x Pupfeeding groups	0.16 ± 0.11	1.53	0.13
Sex (Male) x Dom. Fem. Not Preg., No Pups	0.27 ± 0.16	1.69	0.09

“Dom. Fem. Not Pregnant, No Pups”, “Babysitting groups”, and “Pupfeeding groups” described the group stage (GST in Table S4) where the reference level was that the dominant female was pregnant and there were no pups in the group. “Dom. Fem. Mom or Sister?” was a two-level categorical variable to indicate whether the dominant female in the group where the subordinate was observed was either the mom or sister of the subordinate (reference level was that the dominant female was not the mother or full sister). “Season” was a two-level categorical variable to indicate whether the behavioural data were collected in the summer or winter months (reference level was summer). Reference level for “Sex” was female. The results from this model are based upon 58568 estimates of the amount of aggression 1520 subordinates received from 98 dominant females in 40 different social groups over 19 years during 5120 *ad libitum* behavioural observation sessions.

Table S8. Results from a generalized linear mixed-effects model to examine how the amount of aggression subordinate meerkats received from the dominant female affected their babysitting contributions. This generalized linear mixed-effects model (binomial error structure) included random intercept terms for identity of the subordinate meerkat, litter, dominant female, and year. Reference for the categorical variables described below and in parentheses where applicable. Significant variables in bold.

Fixed Effect	b ± SE	z	P-value
Intercept	-2.49 ± 0.04	-60.8	<0.0001
Sex (Male)	-0.31 ± 0.03	-11.2	<0.0001
Age	0.36 ± 0.02	20.2	<0.0001
Body mass	0.26 ± 0.02	14.6	<0.0001
Group size	-0.39 ± 0.02	-21.2	<0.0001
Aggression received	-0.003 ± 0.007	-0.48	0.63
Sex (Male) x Age	-0.21 ± 0.02	-10.4	<0.0001
Sex (Male) x Body mass	-0.06 ± 0.02	-2.9	0.004
Sex (Male) x Group size	-0.03 ± 0.02	-1.96	0.049
Sex (Male) x Aggression received	-0.013 ± 0.01	-0.94	0.34

Reference level for sex was female. “Aggression received” is the amount of aggression the subordinate received from the dominant female (number of assertions received over observation time). The results from this model are based upon 9410 estimates of babysitting contributions from 1487 subordinate meerkats during 639 different litters produced by 96 different dominant females over the 19 years.

Table S9. Results from a generalized linear mixed-effects model to examine how the amount of aggression subordinate meerkats received from the dominant female affected the probability of observing pupfeeding during *ad libitum* behavioural observations. This generalized linear mixed-effects model (binomial error structure) included random intercept terms for identity of the subordinate meerkat, litter, dominant female, and year. An offset for observation time was included. Reference for the categorical variables described below and in parentheses where applicable. Significant variables in bold.

Fixed Effect	b ± SE	z	P-value
Intercept	1.8 ± 0.36	4.98	<0.0001
Litter size	0.32 ± 0.1	3.28	0.001
Sex (Male)	-0.62 ± 0.1	-5.9	<0.0001
Age	-0.02 ± 0.09	-0.26	0.79
Body mass	1.02 ± 0.09	11.91	<0.0001
Group size	-0.68 ± 0.12	-5.57	<0.0001
Aggression received	-0.01 ± 0.04	-0.26	0.8
Sex (Male) x Age	0.26 ± 0.1	2.57	0.01
Sex (Male) x Body mass	-0.23 ± 0.09	-2.44	0.014
Sex (Male) x Group size	-0.03 ± 0.09	-0.33	0.74
Sex (Male) x Aggression received	0.14 ± 0.09	1.59	0.11

Reference level for sex was female. “Aggression received” is the amount of aggression the subordinate received from the dominant female (number of assertions received over observation time). The results from this model are based upon 7619 estimates of the probability of exhibiting pupfeeding by 1626 subordinates towards pups from 544 different litters produced by 92 dominant females over 20 years.

Table S10. Results from model averaging from the two models containing biological predictor variables with the lowest AIC_c from Table S1 regarding the treatment effects on babysitting behaviour. These generalized linear mixed-effects models included random intercept terms for identity of the litter and an individual-level for observation. Reference for the categorical variables described below. Significant terms in bold and based upon whether 95% CI overlaps 0.

Fixed Effect	b ± SE	95% CI
Intercept	-1.69 ± 0.14	-1.97 - -1.4
Sex (Male)	0.16 ± 0.18	-0.23 - 0.54
Treatment		
Control	-0.46 ± 0.23	-0.93 - 0.02
Cortisol	-0.47 ± 0.18	-0.9 - -0.04
Group size	-0.04 ± 0.07	-0.29 - 0.05

“Treatment” was a three-level categorical variable to indicate whether the behavioural data were collected from control individuals or individuals treated with cortisol or mifepristone (reference level). Reference level for “Sex” was female.

Table S11. Results from model averaging from the two models containing biological predictor variables with lowest AIC_c from Table S2 regarding the treatment effects on the probability of pupfeeding behaviour. These generalized linear mixed-effects models included random intercept terms for identity of the subordinate meerkat, litter, and observer. Reference for the categorical variables described below. Significant terms in bold and based upon whether 95% CI overlaps 0.

Fixed Effect	b ± SE	95% CI
Intercept	0.37 ± 0.59	-0.8 – 1.54
Treated prior to focal?	0.04 ± 0.36	-0.66 – 0.75
Sex (Male)	-1.35 ± 0.83	-2.99 – 0.29
Treatment		
Control	-0.79 ± 0.69	-2.16 – 0.56
Cortisol	-0.84 ± 0.75	-2.33 - 0.64
Sex (Male) x Treatment		
Sex (Male) x Control	-0.49 ± 1.0	-0.49 – 3.22
Sex (Male) x Cortisol	1.59 ± 1.35	0.45 – 4.25

“Treatment” was a three-level categorical variable to indicate whether the behavioural data were collected from control individuals or individuals treated with cortisol or mifepristone (reference level). Reference level for “Sex” was female.

Table S12. Results from model averaging from the two models containing biological predictor variables with lowest AIC_c from Table S3 regarding the treatment effects on the proportion of biomass that was found by subordinates and subsequently fed to pups foraging with the group (“generosity”) during our behavioural focal observations. These generalized linear mixed-effects models included random intercept terms for identity of the subordinate meerkat, litter, and observer. Reference for the categorical variables described below. Significant terms in bold and based upon whether 95% CI overlaps 0.

Fixed Effect	b ± SE	95% CI
Intercept	-2.66 ± 0.58	-3.81 – -1.51
Treated prior to focal?	0.2 ± 0.32	-0.42 – 0.83
Sex (Female)	0.41 ± 0.68	-0.93 – 1.76
Treatment		
Control	0.37 ± 0.49	-0.58 – 1.33
Cortisol	1 ± 0.67	-0.32 – 2.33
Sex (Female) x Treatment		
Sex (Female) x Control	-0.36 ± 0.64	-1.96 – 0.89
Sex (Female) x Cortisol	-1.2 ± 1.05	-3.31 – -0.29

Variable descriptions as in Table S11. Reference level for “Sex” was male.