Citation for published version:
Kosztolányi, A, Küpper, C, Chastel, O, Parenteau, C, Yilmaz, KT, Miklósi, A, Szekely, T & Lendvai, AZ 2012, 'Prolactin stress response does not predict brood desertion in a polyandrous shorebird', Hormones and Behavior, vol. 61, no. 5, pp. 734-740. https://doi.org/10.1016/j.yhbeh.2012.03.011

DOI:
10.1016/j.yhbeh.2012.03.011

Publication date:
2012

Document Version
Peer reviewed version

Link to publication

Brood desertion in a polyandrous shorebird: A role of prolactin and corticosterone?

András Kosztolányi[a], Clemens Küpper[b], Olivier Chastel[c], Charline Parenteau[c],
K. Tuluhan Yılmaz[d], Ádám Miklósi[a], Tamás Székely[e], Ádám Z. Lendvai[f]

a Department of Ethology, Eötvös Loránd University,Budapest 1117, Pázmány Péter sétány 1/c, Hungary

b Museum of Comparative Zoology, Department of Organismic and Evolutionary Biology, Harvard University, Cambridge MA 02138, USA

c Centre d'Études Biologiques de Chizé, Centre National de la Recherche Scientifique, Villiers-en-Bois 79360, France

d Department of Landscape Architecture, Çukurova University, Mithat Özhan Blvd, 01330 Adana, Turkey

e Department of Biology and Biochemistry, University of Bath, Bath BA2 7AY, UK

f Institute of Biology, College of Nyíregyháza, Sóstói út 31/b, Nyíregyháza 4400, Hungary

Correspondence: András Kosztolányi
tel.: +36 1 381 2179;
fax: +36 1 381 2180;
email: andras.kosztolanyi@gmail.com

ABSTRACT

One of the fundamental principles in life-history theory is that parents have to balance their resources carefully between current and future offspring. Deserting the dependent young is a dramatic life-history decision that saves resources for future reproduction, however, may cause failure of the current brood. Despite its importance for sexual conflict theory and breeding system evolution, the underlying mechanisms of brood desertion are largely unknown. We investigated two candidate hormones that may influence brood desertion: prolactin (‘parental hormone’) and corticosterone (‘stress hormone’) in the Kentish plover *Charadrius alexandrinus*. In this small polyandrous shorebird brood desertion occurs naturally; after hatching of the precocial chicks either sex (more often the female) may desert the brood and mate with a new partner shortly after desertion. We measured hormone levels of parents at hatching using the standard capture and restraint protocol, and subsequently followed the broods to determine whether a parent deserted the chicks. We investigated whether hormone levels were different between adult males and females, and between deserting and caring parents. We found no evidence that either baseline or stress-induced prolactin levels predicted brood desertion. Stress-induced corticosterone levels were generally higher in females, however, the individual hormone levels did not explain the probability of brood desertion. We propose that in species where frequent brood desertion is part of the natural breeding system, desertion is a rapid dynamic process that is mediated not only by hormone levels, but also by swiftly changing social environment, i.e. by number and quality of available mates for remating.

Keywords: social behavior, neuroendocrine system, parental care, brood desertion, corticosterone, prolactin, capture and restraint, shorebird, Kentish plover

INTRODUCTION

Brood desertion, i.e. when a parent stops caring and abandons its dependent young, is a dramatic resolution of the life-history trade-off between current and future reproduction. Desertion may entail both costs (e.g. reduced offspring survival) and benefits (e.g. increased survival or remating and successive reproduction of the parent, Houston *et al.*, 2005; Székely *et al.*, 1996). Understanding brood desertion is important because it has implications on population parameters (such as the operational sex ratio, OSR) and life-history evolution, and because it is a key idea in sexual conflict resolution in a parental care context (Clutton-Brock, 1991; Lessels, 1999; Székely *et al.*, 2000; Székely *et al.*, 2007). However, the physiological mechanisms that trigger brood desertion remain poorly understood.

Circulating hormones are thought to be the primary physiological mediators of life-history trade-offs (Flatt and Heyland, 2011; Ketterson and Nolan Jr, 1999; Sinervo and Svensson, 1998). For example, when individual survival prospects are compromised due to threats by predators, pathogens, food shortage, inclement weather etc. (commonly referred to as stressors), vertebrates respond by the activation of the hypothalamus-pituitary-adrenal cortex (HPA) axis, which elevates the circulating levels of glucocorticoids (reviewed by Wingfield and Sapolsky, 2003). The glucocorticoids in turn promote behaviours that help the immediate survival and concurrently repress those that do not, most notably reproduction. Because of these antagonistic effects, it has been hypothesised that corticosterone, the avian glucocorticoid mediates the survival-reproduction trade-off in birds (Ricklefs and Wikelski, 2002). Recent studies have supported the predictions of this hypothesis by showing that corticosterone levels are modulated according to the relative importance of the current reproduction compared with survival (Bókony *et al.*, 2009; Heidinger *et al.*, 2006; Lendvai and Chastel, 2008; Lendvai *et al.*, 2007).

However, corticosterone may not be the only hormonal modifier of survival-reproduction trade-offs. The vertebrate stress response is complex and involves several physiological agents. The circulating levels of prolactin also change during the stress response; for example, standard handling stress induces a significant decrease in its plasma concentrations in several species (Angelier *et al.*, 2007; Angelier *et al.*, 2009a; Heidinger *et al.*, 2010; Riou *et al.*, 2010). Since prolactin is actively involved in the regulation of parental behaviour in birds and facilitates incubation and brooding behaviours (Adkins-Regan, 2005; Adkins-Regan *et al.*, 2010), it has been recently suggested to play a key role as the physiological mediator of the trade-off between current parental care vs. future reproduction (Angelier and Chastel, 2009; Chastel *et al.*, 2005). Specifically, it has been proposed that the magnitude of prolactin decrease in response to a standardised stress protocol may reflect the willingness and/or ability to maintain parental care. Therefore the prolactin stress response may be interpreted as a proximate signal of parental investment (Angelier and Chastel, 2009).

Our aim in this study was to investigate the underlying proximate mechanisms of natural brood desertion. We investigated a small shorebird, the Kentish plover *Charadrius alexandrinus* with well-known brood care behaviour including brood desertion (Amat *et al.*, 1999; Kosztolányi *et al.*, 2006; Lessells, 1984). After hatching of the precocial chicks, either the male or the female parent may desert the brood, although the frequency of brood desertion differs between sexes and populations (Araceli Arguelles Tico *et al.* unpubl data). This high flexibility of parental behaviour and the amenability of this species for experimental manipulations make the Kentish plover an ideal species to investigate the hormonal background of brood desertion.

We investigated four hypotheses regarding the role of prolactin and corticosterone in the regulation of brood desertion behaviour. In our study population brood desertion by females is 11 times more frequent than desertion by males (Kosztolányi *et al.*, 2006).

Therefore we asked first, whether this sex difference in desertion may be a consequence of lower prolactin levels in females than in males. In precocial species, concentrations of circulating levels of prolactin either drops sharply after hatching of the chicks (Dittami, 1981; Goldsmith, 1982; Goldsmith and Williams, 1980; Hall and Goldsmith, 1983; Wentworth *et al.*, 1983), or remains elevated after hatching and may decrease slowly until the chicks become thermally independent (Boos *et al.*, 2007; Gratto-Trevor *et al.*, 1990; Oring *et al.*, 1986; Oring *et al.*, 1988). Since Kentish plovers spend a significant amount of time on brooding their downy chicks (Székely and Cuthill, 1999), we expected that in this species the prolactin levels remain high after hatching and decline gradually as chicks grow and need less brooding. If females have already lower baseline prolactin levels at hatching of their chicks than males do, gradual decrease in their circulating prolactin after hatching may reach a threshold where the motivation for tending the chicks becomes insufficient and desertion occurs. Therefore, we predicted that baseline prolactin levels are lower in females than in males. Second, we tested whether individual desertion decisions can be predicted by the prolactin stress response. If the magnitude of decrease in prolactin in response to a standardised stressor can be regarded as a surrogate measure of parental investment (Angelier and Chastel, 2009), we predicted that females deserting their brood would have a stronger prolactin stress response (i.e. lower stress-induced prolactin levels) than females that do not desert. Third, since the hormone corticosterone has been proposed as a mediator of life-history trade-off between reproduction and survival, we tested whether the corticosterone levels differ between sexes. Finally, we asked whether stress-induced corticosterone levels predict desertion decision in females. We predicted that females have higher baseline corticosterone levels than males, and deserting females have higher stress-induced corticosterone levels than non-deserting females.

METHODS

Study area and field methods

Fieldwork was carried out at Lake Tuzla (36°43’ N, 35°03’ E), southern Turkey in two years (2009: 24 April –25 June, 2010: 29 April – 24 June; see details about the study site and field methodology in Kosztolányi *et al.*, 2006; Lendvai *et al.*, 2004). Both parents were captured by funnel traps either at the nest at hatching or with the chicks after hatching, whereas chicks were captured either in the nest scrape immediately after hatching, or at the first encounter (capture date of adults relative to hatching date of their clutch; 0.5 ± 0.21 days (mean ± SE), range: -2 – 6 days, $n = 82$ adults). Adults were ringed with a metal ring and an individual combination of three colour rings. Chicks were ringed by two rings, one metal ring and a colour ring (the same colour within a family). We measured the body mass, the left and right tarsus length of each captured bird. From adults blood samples were taken for hormone assays using the standard capture and restraint protocol (Wingfield, 1994): 1.4 ± 0.06 minutes (mean ± SE, range: 0.8 – 3.4, $n = 82$ adults) after capture about 150 µl (in 2009) or 210 µl (in 2010) blood was collected from the brachial vein and the bird was then placed in a cloth bag. Neither corticosterone nor prolactin levels at the first blood sampling were related to the time elapsed from installation of the trap to blood sampling (Pearson correlations, corticosterone: $r = -0.07$, $n = 39$, $p = 0.676$; prolactin: $r = 0.03$, $n = 82$, $p = 0.785$) or from capture to blood sampling (corticosterone: $r = 0.15$, $n = 39$, $p = 0.367$; prolactin: $r = 0.04$, $n = 82$, $p = 0.739$), therefore these samples are referred to as baseline. A second blood sample was collected 30.7 ± 0.10 minutes (range: 29.7 – 35.8) after the first blood sample. Blood samples were centrifuged and the plasma was stored at -20 °C until analysis. One female was sampled in both years. For this female only the brood from 2010 was retained in the dataset. After hatching, the broods were visited regularly and the sex and number of attending parents and the number of chicks were recorded. We attempted to follow broods until the chicks perished or were 25 days old when they were considered as fledged. If a parent was not present at the brood at two consecutive visits we considered the parent as deserted. As desertion by males was rare (see results), we analysed desertion decision only in females. Ten broods fledged at least one chick, whereas all chicks died in four broods at 0.8 ± 0.48 (mean ± SE) days of age. Twenty-six broods of which five had been deserted by the female were followed only until 12.4 ± 1.25 days either because the fieldwork ended or because we were not able to locate the family.

Hormone assays

Hormone concentrations were determined in duplicate aliquots from 50 µl plasma sample by radioimmunoassay at Centre d'Études Biologiques de Chizé (CEBC), France. Prolactin measurements were available for $n = 41$ pairs (in 2009: 21 pairs, in 2010: 20 pairs). Plasma concentrations of prolactin were determined by a heterologous radioimmunoassay at the CEBC as detailed in Cherel *et al.* (1994). Pooled plasma samples of Kentish plovers produced a dose–response curve that paralleled chicken prolactin standard curves ('AFP 4444B', source: Dr Parlow, NHPP Harbor-UCLA Medical Center, Torrance, CA, USA). Therefore, the cross-reactivity of the chicken prolactin antibody with prolactin was equivalent in both species and this heterologous assay could be used to assess relative concentrations of Kentish plover prolactin. The detection limit of the assay was 19.26 ng/ml and the lowest measurement was 194.142 ng/ml. The samples were run in two assays (intra-assay coefficient: 13%, inter-assay coefficient: 24%).

Blood for corticosterone assay was collected from $n = 20$ pairs in 2010, however, the plasma was not enough to run the assay in case of one male. Total corticosterone concentrations were measured in one assay as described in Lendvai *et al.* (2011). The intra-assay coefficient of variation was 7.07% for 10 duplicates. The minimum detectable level of corticosterone was 0.28 ng/ml (lowest measurement: 7.78 ng/ml).

Statistical analyses

All data processing and statistical analyses were performed in the R computing environment (version: 2.1.4.0, R Development Core Team, 2011). First we checked the possible effect of potential confounding variables: season (measured as days since 1 March), brood age, parental sex and size and time of the day. Second, we used mixed-effects models including stress (first/second bleeding) and sex as factors, the confounding variables having a significant effect in the preliminary analyses and all second order interactions as fixed effects. The repeated measurements of an individual were controlled for by including ring ID as random structure in the models (Pinheiro and Bates, 2000). We carried out model selection using AIC values (Venables and Ripley, 2002). As several broods were not followed until fledging (see above), the effect of hormone levels on desertion was analysed using Cox regression. In these models desertion was the terminal event and non-deserted broods were censored cases. Neither baseline, nor stress-induced prolactin levels differed between years (t-tests, baseline: $t_{80} = 0.997$, $p = 0.322$, stress-induced: $t_{80} = 0.527$, $p = 0.600$), therefore we pooled and analysed prolactin data from both years. One male showed an unusual response on capture stress in corticosterone levels (see white symbol on Fig. 3), therefore we repeated all analyses excluding the hormone measurements of this male from the dataset. However, removal did not change our conclusions and we only present the results of the analysis of the full data set.

RESULTS

Parental care and prolactin

We observed 12 desertions in 41 broods. In 11 cases, the female deserted the family, and in one case the male. Median time of female desertion was day 9 post hatching (range: 2 – 24 days). Large females deserted the brood more often than small ones (Cox regressions, $n = 40$, tarsus length: b = 1.079, $p = 0.009$), and the probability of desertion tended to decrease over the breeding season (hatching date: b = -0.063, $p = 0.071$).

Baseline prolactin levels did not differ between the sexes (t-test, $t_{80} = 0.029$, $p = 0.977$), and was not influenced by size of parents (Pearson correlations, sexes combined, $n = 82$, tarsus length: $r = -0.12$, $p = 0.289$; body mass: $r = -0.01$, $p = 0.954$). Baseline prolactin levels decreased with the age of the chicks, however, neither hatching date nor time of the day influenced baseline prolactin levels (Pearson correlations, sexes combined, $n = 82$, brood age: $r = -0.28$, $p = 0.012$; hatching date: $r = 0.17$, $p = 0.122$; time of the day: $r = 0.16$, $p = 0.159$).

As expected, circulating prolactin levels decreased in response to the capture restraint stress in both sexes, however, the sexes did not differ in their stress-induced prolactin level (Fig. 1, t-tests, stress: $t_{162} = 14.605$, $p < 0.001$; sex : $t_{80} = 0.616$, $p = 0.540$). The mixed-effects model analysis gave consistent results with t-tests (Table 1).

The stress-induced prolactin levels of caring females were higher than those of deserting females (Fig. 1, $t_{38} = 2.064$, $p = 0.046$), however, the caring females group also included females of broods where the chicks died or where the brood was not followed until fledging of the chicks. Survival analyses showed that stress-induced prolactin levels did not influence the probability of desertion in females (Cox regression controlling for female size, $n = 40$, tarsus: $b = 1.117$, $p = 0.009$; stress-induced prolactin: $b = 0.001$, $p = 0.712$). The plot of observed caring history of broods against female stress-induced prolactin levels revealed that although we did not observe desertion among females with the highest stress-induced prolactin levels, several long-caring females had relatively low stress-induced prolactin levels (Fig. 2).

Corticosterone

Baseline corticosterone levels did not differ between adult males and females (t-test, t_{37} = 1.147, $p = 0.259$). Corticosterone levels did not change with parental size, age of the brood, hatching date or capture time (Pearson correlations, sexes combined, $n = 39$, all $p \geq 0.173$). Capture stress induced a significant increase in corticosterone levels (Fig. 3, Table 1, t-test, t_{76} = 11.758, $p < 0.001$), and females had higher stress-induced corticosterone levels than males ($t_{37} = 2.239$, $p = 0.031$).

Stress-induced corticosterone levels did not predict desertion behaviour (Cox regression controlling for female size, $n = 19$, tarsus: $b = 1.320$, $p = 0.036$; stress-induced corticosterone: $b = -0.003$, $p = 0.892$).

DISCUSSION

Prolactin levels are associated with parental care in many species including birds, mammals and fish (reviewed by Adkins-Regan *et al.*, 2010; Angelier and Chastel, 2009; Rall *et al.*, 2004; Ziegler *et al.*, 2009), although in our study, prolactin levels at (or near) hatching of the eggs did not predict brood desertion in Kentish plovers. Prolactin may be unrelated to the deserting decision, and this behaviour could be regulated by alternative pathways. Conversely, it is possible that prolactin does influence the probability of desertion behaviour, although we failed to find this relationship because the decrease in prolactin levels does not take place at hatching but happens only shortly before desertion which occurs at varying chick ages.

Brood desertion is an adaptive strategy if the benefits associated with desertion outweigh the costs (Clutton-Brock, 1991; McGraw *et al.*, 2010; Székely *et al.*, 1996). Such benefits may be either the increased survival or the possibility to rapidly remate and increase reproductive success. Although prolactin has been documented to play a role in clutch abandonment or brood desertion (Chastel and Lormée, 2002; Groscolas *et al.*, 2008; Spee *et al.*, 2010), and in temporary egg neglect (Angelier *et al.*, 2007), desertion in these species occurs exclusively at severe energy deficit, so probably operates as part of an emergency reaction and serves increased survival. In one study, clutch desertion was related to reduced prolactin levels in response to partial clutch loss, however, here desertion decision was also triggered by cues of nest predation (Hall, 1987). In contrast, brood desertion in the Kentish plover (and in its close relative, the snowy plover *Charadrius nivosus*) is part of the natural breeding system, and deserting parents often remate and lay new clutches (Székely and Williams, 1995; Warriner *et al.*, 1986). Therefore, it is possible that these two types of desertion are controlled by different hormonal pathways, and that the decision of desertion is made independently of prolactin levels in plover.

Elevated prolactin levels are known to induce parental behaviours, especially for persistent incubation and brooding (Adkins-Regan *et al.*, 2010; Angelier and Chastel, 2009; Buntin, 1996); therefore it would require specific adaptations if birds were to leave their brood and initiate a new clutch with elevated prolactin levels, especially, as prolactin have also antagonist effects on luteinizing hormone (LH) and gonadal steroids, which are necessary for rebreeding (Sharp *et al.*, 1998).

If, on the other hand, prolactin plays a role in the regulation of brood desertion, it must be temporarily decoupled from the hormone levels we measured close to hatching. Consistently with the idea that prolactin reflects the amount of active parental care, we found that baseline prolactin decreased after the hatching and as the chicks became more independent, similarly to other polygamous shorebirds (e.g. Wilson’s phalaropes *Phalaropus tricolor,* Oring *et al.*, 1988; red-necked phalaropes *Phalaropus lobatus,* Gratto-Trevor *et al.*, 1990). Nevertheless, female Kentish plovers did not have lower baseline prolactin levels than males, therefore the different desertion patterns between the sexes cannot be the consequence of females having already lower prolactin levels at hatching. Furthermore, the slope of the decrease in prolactin after hatching was similar in males and in females (results not shown), which suggests that if plummeting prolactin levels are responsible for the sex-biased desertion pattern, than the fall of prolactin levels should happen later, potentially only shortly before desertion.

For example, in Eurasian penduline tits *Remiz pendulinus,* another bird species where desertion by either sex is part of the natural breeding system, desertion is a rapid process, and the behaviour of the parents shortly before they leave does not predict their decision (van Dijk *et al.*, 2007). It is plausible that in species such as the penduline tit or the Kentish plover, where there is an intensive sexual conflict over parental care and both sexes may potentially desert (Székely *et al.*, 2006), selection should favour physiological mechanisms that allow for rapid shifts in behaviour. On the contrary, in species, where desertion occurs as a response to an energetic crisis, clutch or brood abandonment seems to be slow. For instance, an association of low prolactin levels, progressive decrease in nest attendance and transitory neglecting of the eggs preceding definitive desertion or clutch failure have been observed in king penguins *Aptenodytes patagonicus* (Groscolas *et al.*, 2008), red-footed boobies *Sula sula* (Chastel and Lormée, 2002), snow petrels (Angelier *et al.*, 2007) and black-legged kittiwakes (Angelier *et al.*, 2009b).

The rapid transition between care and desertion may explain why the results did not support our second prediction, namely that deserting females will respond more strongly to a standardised stress than females that continue to provide care. By measuring the prolactin stress response, we may have measured the parental investment of the parents at the day of sampling (i.e. the end of the incubation or the beginning of chick rearing) which may not have been different for females that later decided to desert or to care.

Corticosterone is considered as an important mediator of the life-history between reproduction and survival, and therefore the levels of this hormone may affect fitness (Blas *et al.*, 2007; Bonier *et al.*, 2009; Ricklefs and Wikelski, 2002; Wingfield *et al.*, 1995). The biological effect of baseline and stress-induced levels are different, with baseline levels having essentially metabolic effects, whereas at the higher stress-induced levels corticosterone may trigger significant shifts in behaviour in concert with other hormones (Angelier *et al.*, 2009b; Hau *et al.*, 2010; Wingfield and Sapolsky, 2003). Here, we found that baseline corticosterone levels did not differ between the sexes but stress-induced corticosterone levels were higher in females than males. The fact that baseline levels were similar in the two sexes corroborates that female desertion in Kentish plovers is not driven by an energy deficit but reflects a reproductive strategy. On the other hand, higher stress-induced corticosterone levels in females compared with males indicate that the value of a given reproduction is lower for the potentially multibrooded polyandrous females than for the males. However, individual corticosterone levels in females were not related to desertion probability. Recent studies have shown that natural variation in corticosterone levels are negatively related to post-stress parental care (Lendvai and Chastel, 2010; Miller *et al.*, 2009). Here we found an eightfold variation in baseline and a threefold variation in stress-induced corticosterone levels, but this extensive variation had very little power explaining the probability of desertion in individual females.

Taken together, we found that sex differences in stress-induced corticosterone, but not prolactin levels, were consistent with the higher frequency of brood desertion in females. However, individual variation in either the ‘stress hormone’ corticosterone, or the ‘parental hormone' prolactin measured near hatching of the young was not related to brood desertion probability. These results show that brood desertion by female Kentish plovers is not the consequence of their low overall parental commitment. We propose that desertion is a rapid process in this species which may be triggered by sudden increase in mating opportunities rather than a general reduction in commitment to care. Desertion may then be orchestrated by corresponding changes in hormone levels. The decisive test for this hypothesis would require manipulating prolactin secretion or its binding to receptors, or inducing desertion behaviour by manipulating mating opportunities in natural habitats of plovers.

ACKNOWLEDGMENTS

The study was supported by the Hungarian Scientific Research Fund (OTKA, K81953, PD76862). Fieldwork was supported by a grant from the Hungarian – Turkish Intergovernmental S&T Cooperation Programme (OMFB-00581/2009, TUBITAK-108Y329). CK was supported by a DAAD fellowship. At the CEBC, we thank Colette Trouvé for her technical assistance in prolactin and corticosterone assays. We are grateful to Peter Sharp for discussions and useful advices on the methods.

REFERENCES

Adkins-Regan, E., 2005. Hormones and Animal Social Behavior. Princeton University Press, Princeton.

Adkins-Regan, E., DeVoogd, T.J., Moore, J.M., 2010. Social behaviour and bird song from a neural and endocrine perspective. In: T. Székely, A.J. Moore, J. Komdeur (Eds.), Social Behaviour: Genes, Ecology and Evolution, Cambridge University Press, Cambridge, pp. 59–84.

Amat, J.A., Fraga, R.M., Arroyo, G.M., 1999. Brood desertion and polygamous breeding in the Kentish plover *Charadrius alexandrinus*. Ibis 141, 596–607.

Angelier, F., Chastel, O., 2009. Stress, prolactin and parental investment in birds: A review. Gen. Comp. Endocrinol. 163, 142–148.

Angelier, F., Moe, B., Blanc, S., Chastel, O., 2009a. What factors drive prolactin and corticosterone responses to stress in a long-lived bird species (snow petrel *Pagodroma nivea*)? Physiol. Biochem. Zool. 82, 590–602.

Angelier, F., Clément-Chastel, C., Weckler, J., Gabrielsen, G.W., Chastel, O., 2009b. How does corticosterone affect parental behaviour and reproductive success? A study of prolactin in black-legged kittiwakes. Funct. Ecol. 23, 784–793.

Angelier, F., Moe, B., Weimerskirch, H., Chastel, O., 2007. Age-specific reproductive success in a long-lived bird: do older parents resist stress better? J. Anim. Ecol. 76, 1181–1191.

Blas, J., Bortolotti, G.R., Tella, J.L., Baos, R., Marchant, T.A., 2007. Stress response during development predicts fitness in a wild, long lived vertebrate. PNAS 104, 8880–8884.

Bonier, F., Martin, P.R., Moore, I.T., Wingfield, J.C., 2009. Do baseline glucocorticoids predict fitness? TREE 24, 634–642.

Boos, M., Zimmer, C., Carriere, A., Robin, J., Petit, O., 2007. Post-hatching parental care behaviour and hormonal status in a precocial bird. Behav. Processes 76, 206–214.

Buntin, J.D., 1996. Neural and hormonal control of parental behaviour in birds. In: J.S. Rosenblatt, C.T. Snowdon (Eds.), Advances in the Study of Behavior, vol. 25., Academic Press, New York, pp. 161–213.

Bókony, V., Lendvai, Á.Z., Liker, A., Angelier, F., Wingfield, J.C., Chastel, O., 2009. Stress response and the value of reproduction: Are birds prudent parents? Am. Nat. 173, 589–598.

Chastel, O., Lacroix, A., Weimerskirch, H., Gabrielsen, G., 2005. Modulation of prolactin but not corticosterone responses to stress in relation to parental effort in a long-lived bird. Horm. Behav. 47, 459–466.

Chastel, O., Lormee, H., 2002. Patterns of prolactin secretion in relation to incubation failure in a tropical seabird, the red-footed booby. Condor 104, 873–876.

Cherel, Y., Mauget, R., Lacroix, A., Gilles, J., 1994. Seasonal and fasting-related changes in circulating gonadal steroids and prolactin in king penguins, *Aptenodytes patagonicus*. Physiol. Zool. 67, 1154–1173.

Clutton-Brock, T.H., 1991. The Evolution of Parental Care. Princeton University Press, Princeton, New Jersey.

Dittami, J.P., 1981. Seasonal changes in the behavior and plasma titers of various hormones in barheaded geese, *Anser indicus*. Z. Tierpsychol. – J. Comp. Ethol. 55, 289–324.

Flatt, T., Heyland, A., 2011. Mechanisms of Life History Evolution: The Genetics and Physiology of Life History Traits and Trade-Offs. Oxford University Press, New York.

Goldsmith, A.R., 1982. The Australian black swan (*Cygnus atratus*): prolactin and gonadotropin secretion during breeding including incubation. Gen. Comp. Endocrinol. 46, 458–462.

Goldsmith, A.R., Williams, D.M., 1980. Incubation in mallards (*Anas platyrhynchos*): changes in plasma-levels of prolactin and luteinizing hormone. J. Endocrinol. 86, 371–379.

Gratto-Trevor, C.L., Oring, L.W., Fivizzani, A.J., Elhalawani, M.E., Cooke, F., 1990. The role of prolactin in parental care in a monogamous and a polyandrous shorebird. Auk 107, 718–729.

Groscolas, R., Lacroix, A., Robin, J., 2008. Spontaneous egg or chick abandonment in energy-depleted king penguins: A role for corticosterone and prolactin? Horm. Behav. 53, 51–60.

Hall, M., 1987. Nesting success in mallards after partial clutch loss by predators. J. Wildl. Manage. 51, 530–533.

Hall, M.R., Goldsmith, A.R., 1983. Factors affecting prolactin secretion during breeding and incubation in the domestic duck (*Anas platyrhynchos*). Gen. Comp. Endocrinol. 49, 270–276.

Hau, M., Ricklefs, R.E., Wikelski, M., Lee, K.A., Brawn, J.D., 2010. Corticosterone, testosterone and life-history strategies of birds. Proc. R. Soc. Lond. B. 277, 3203–3212.

Heidinger, B., Chastel, O., Nisbet, I., Ketterson, E., 2010. Mellowing with age: older parents are less responsive to a stressor in a long-lived seabird. Funct. Ecol. 24, 1037–1044.

Heidinger, B.J., Nisbet, I.C.T., Ketterson, E.D., 2006. Older parents are less responsive to a stressor in a long-lived seabird: a mechanism for increased reproductive performance with age? Proc. R. Soc. Lond. B. 273, 2227–2231.

Houston, A.I., Székely, T., McNamara, J.M., 2005. Conflict between parents over care. TREE 20, 33–38.

Ketterson, E.D., Nolan Jr, V., 1999. Adaptation, exaptation, and constraint: a hormonal perspective. Am Nat 154, S4–S25.

Kosztolányi, A., Székely, T., Cuthill, I.C., Yılmaz, K.T., Berberoğlu, S., 2006. Ecological constraints on breeding system evolution: the influence of habitat on brood desertion in Kentish plover. J. Anim. Ecol. 75, 257–265.

Lendvai, Á.Z., Bókony, V., Chastel, O., 2011. Coping with novelty and stress in free-living house sparrows. J. Exp. Biol. 214, 821–828.

Lendvai, Á.Z., Chastel, O., 2008. Experimental mate-removal increases the stress response of female house sparrows: The effects of offspring value? Horm. Behav. 53, 395–401.

Lendvai, Á.Z., Chastel, O., 2010. Natural variation in stress response is related to post-stress parental effort in male house sparrows. Horm. Behav. 58, 936–942.

Lendvai, Á.Z., Giraudeau, M., Chastel, O., 2007. Reproduction and modulation of the stress response: an experimental test in the house sparrow. Proc. R. Soc. Lond .B. 274, 391–397.

Lendvai, Á.Z., Kis, J., Székely, T., Cuthill, I.C., 2004. An investigation of mate choice based on manipulation of multiple ornaments in Kentish plovers. Anim. Behav. 67, 703–709.

Lessells, C.M., 1984. The mating system of Kentish plovers *Charadrius alexandrinus*. Ibis 126, 474–483.

Lessells, C.M., 1999. Sexual conflict in animals. In: Keller L. (Ed.), Levels of Selection in Evolution. Princeton University Press, Princeton, pp. 75–99.

McGraw, L., Székely, T., Young, L.J., 2010. Pair bonds and parental behaviour. In: T. Székely, A.J. Moore, J. Komdeur (Eds.), Social Behaviour: Genes, Ecology and Evolution, Cambridge University Press, Cambridge, pp. 271–301.

Miller, D.A., Vleck, C.M., Otis, D.L., 2009. Individual variation in baseline and stress-induced corticosterone and prolactin levels predicts parental effort by nesting mourning doves. Horm. Behav. 56, 457–464.

Oring, L.W., Fivizzani, A.J., Colwell, M.A., Elhalawani, M.E., 1988. Hormonal changes associated with natural and manipulated incubation in the sex-role reversed Wilson's phalarope. Gen. Comp. Endocrinol. 72, 247–256.

Oring, L.W., Fivizzani, A.J., Elhalawani, M.E., 1986. Changes in plasma prolactin associated with laying and hatch in the spotted sandpiper. Auk 103, 820–822.

Pinheiro, J.C., Bates, D.M., 2000. Mixed-Effects Models in S and S-PLUS. Springer-Verlag, New York, NY.

R Development Core Team, 2011. R: A language and environment for statistical computing. R Foundation for Statistical Computing, ISBN 3-900051-07-0, URL http://www.R-project.org, Vienna, Austria.

Rall, M.K., Liljander, M., Borg, B., 2004. Prolactin diminishes courtship behaviour and stimulates fanning in nesting male three-spined sticklebacks, *Gasterosteus aculeatus*. Behaviour 141, 1511–1519.

Ricklefs, R.E., Wikelski, M., 2002. The physiology/life-history nexus. TREE 17, 462–468.

Riou, S., Chastel, O., Lacroix, A., Hamer, K.C., 2010. Stress and parental care: Prolactin responses to acute stress throughout the breeding cycle in a long-lived bird. Gen. Comp. Endocrinol. 168, 8-13.

Sharp, P.J., Dawson, A., Lea, R.W., 1998. Control of luteinizing hormone and prolactin secretion in birds. Comp. Biochem. Physiol. C. 119, 275–282.

Sinervo, B., Svensson, E., 1998. Mechanistic and selective causes of life history trade-offs and plasticity. Oikos 83, 432–442.

Spee, M., Beaulieu, M., Dervaux, A., Chastel, O., Le Maho, Y., Raclot, T., 2010. Should I stay or should I go? Hormonal control of nest abandonment in a long-lived bird, the adelie penguin. Horm. Behav. 58, 762–768.

Székely, T., Cuthill, I.C., 1999. Brood desertion in Kentish plover: the value of parental care. Behav Ecol 10, 191–197.

Székely, T., Kosztolányi, A., Küpper, C., Thomas, G.H., 2007. Sexual conflict over parental care: a case study of shorebirds. J. Ornithol. 148, S211–S217.

Székely, T., Thomas, G.H., Cuthill, I.C., 2006. Sexual conflict, ecology, and breeding systems in shorebirds. Bioscience 56, 801–808.

Székely, T., Webb, J.N., Cuthill, I.C., 2000. Mating patterns, sexual selection and parental care: an integrative approach. In: M. Apollonio, M. Festa-Bianchet, D. Mainardi (Eds.), Vertebrate Mating Systems. World Science Press, Singapore, pp. 194–223.

Székely, T., Webb, J.N., Houston, A.I., McNamara, J.M., 1996. An evolutionary approach to offspring desertion in birds. In: V. Nolan Jr, E.D. Ketterson (Eds.), Current Ornithology. Plenum Publisher, New York, pp. 271–330.

Székely, T., Williams, T.D., 1995. Costs and benefits of brood desertion in female Kentish plovers, *Charadrius alexandrinus*. Behav. Ecol. Sociobiol. 37, 155–161.

van Dijk, R.E., Szentirmai, I., Komdeur, J., Székely, T., 2007. Sexual conflict over parental care in Penduline Tits *Remiz pendulinus*: the process of clutch desertion. Ibis 149, 530–534.

Venables, W.N., Ripley, B.D., 2002. Modern Applied Statistics with S. Springer, New York, NY.

Warriner, J.S., Warriner, J.C., Page, G.W., Stenzel, L.E., 1986. Mating system and reproductive success of a small population of polygamous snowy plovers. Wilson Bull. 98, 15–37.

Wentworth, B.C., Proudman, J.A., Opel, H., Wineland, M.J., Zimmermann, N.G., Lapp, A., 1983. Endocrine changes in the incubating and brooding turkey hen. Biol. Reprod. 29, 87–92.

Wingfield, J.C., 1994. Modulation of the adrenocortical response to stress in birds. In: K. Davey, R. Peter, S. Tobe (Eds.), Perspectives in Comparative Endocrinology. National Research Council of Canada, Ottawa, Canada, pp. 520–528.

Wingfield, J.C., O'Reilly, K.M., Astheimer, L.B., 1995. Modulation of the adrenocortical responses to acute stress in arctic birds: a possible ecological basis. Am. Zool. 35, 285–294.

Wingfield, J.C., Sapolsky, R.M., 2003. Reproduction and resistance to stress: when and how. J. Neuroendocrinol. 15, 711–724.

Ziegler, T.E., Prudom, S.L., Zahed, S.R., Parlow, A.F., Wegner, F., 2009. Prolactin's mediative role in male parenting in parentally experienced marmosets (*Callithrix jacchus*). Horm. Behav. 56, 436–443.

FIGURE LEGENDS

Fig. 1. Individual baseline and stress-induced prolactin levels and boxplots of hormone levels in the Kentish plover. For each box, the central line represents the median, and the bottom and the top of the box are the lower and upper quartiles, respectively. The whiskers extend to the lowest and highest observations, respectively. The open circles denote a male with unusual change in corticosterone levels (see Methods).

Fig. 2. Stress-induced prolactin levels in females of Kentish plover broods and the observed caring history of these broods (40 biparental or female deserted broods). Each brood is represented by a horizontal line (or dot if observed only on one day); solid line indicates biparental, whereas broken line indicates male-only care.

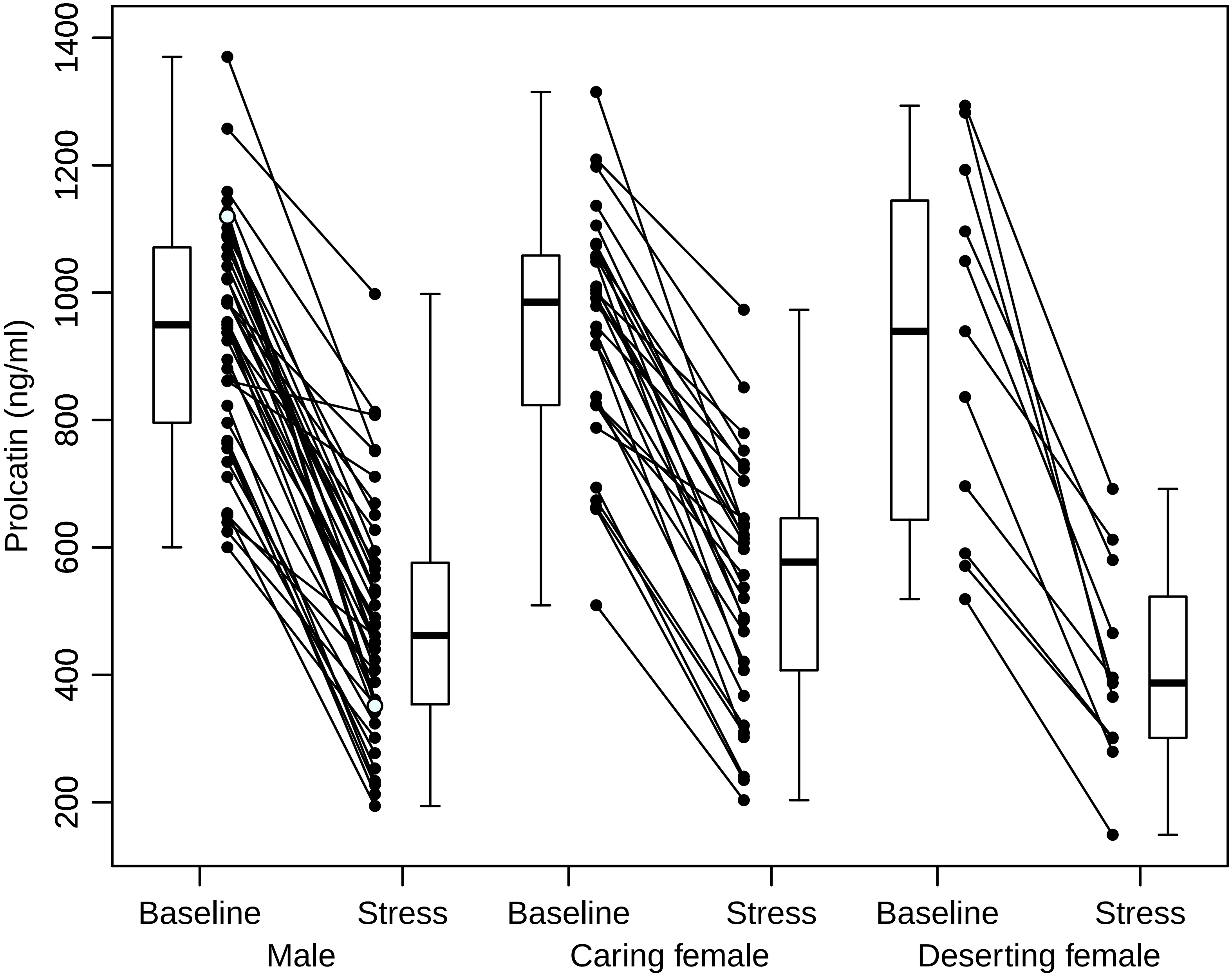
Fig. 3. Individual baseline and stress-induced corticosterone levels and boxplots of hormone levels in the Kentish plover. Boxplots as in Fig. 1. The open circles denote a male with unusual corticosterone levels (see Methods).

Table 1. Parameter estimates from the minimal mixed-effects models for prolactin (ng/ml) and corticosterone (ng/ml) plasma levels in the Kentish plover after AIC-based model selection.

	Prolactin					**Corticosterone**				
	value	**se**	**df**	***t***	***p***	**value**	**se**	**df**	***t***	***p***
Brood age	-32.53	9.81	80	3.32	0.001					
Sex						-8.96	4.05	37	2.21	0.033
Stress	-438.84	17.80	81	24.65	<0.001	44.72	3.29	38	13.59	<0.001

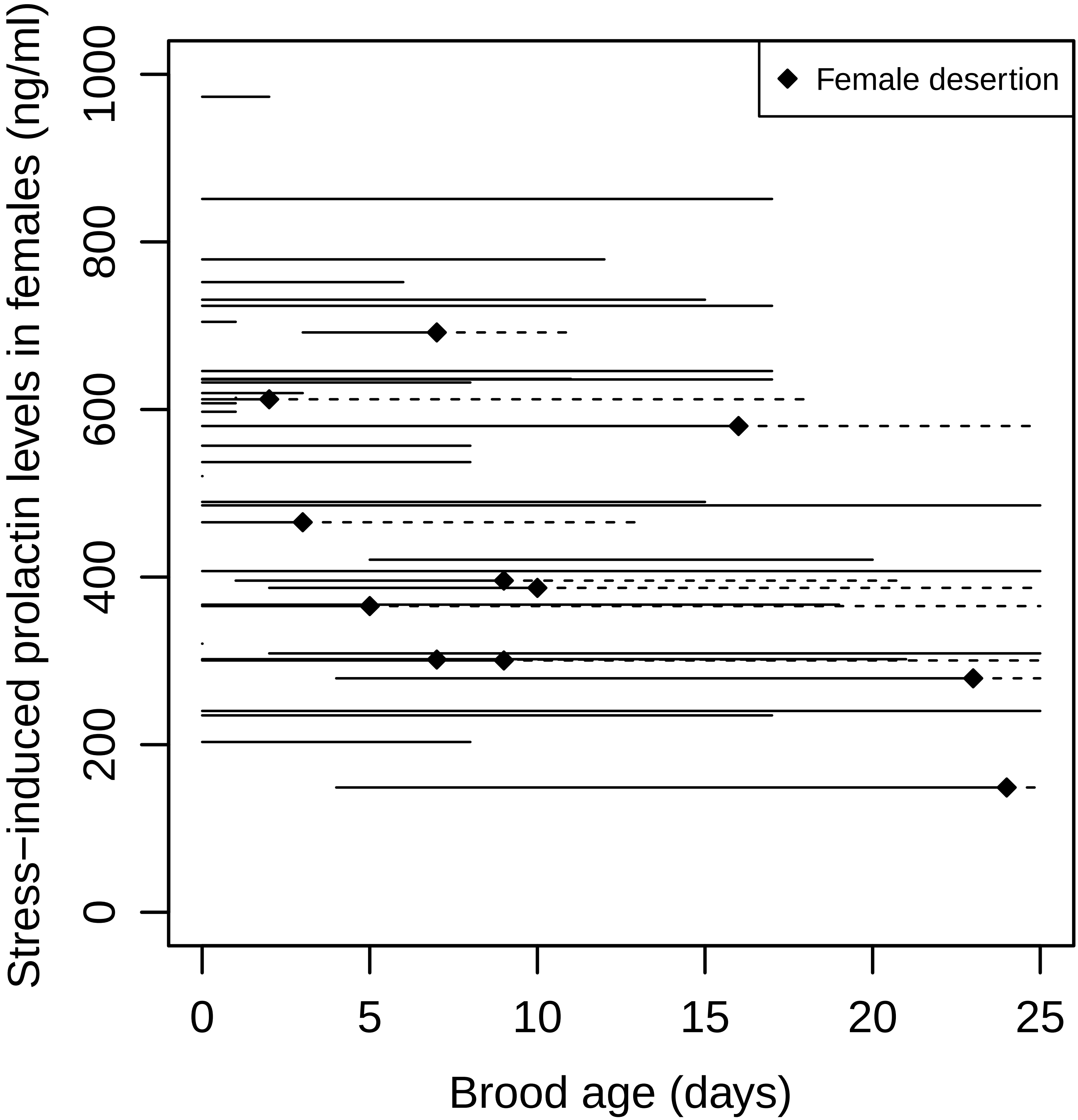
The initial model for prolactin contained brood age, sex and stress and all second order interactions, and for corticosterone sex, stress and sex × stress interaction.

Fig. 1



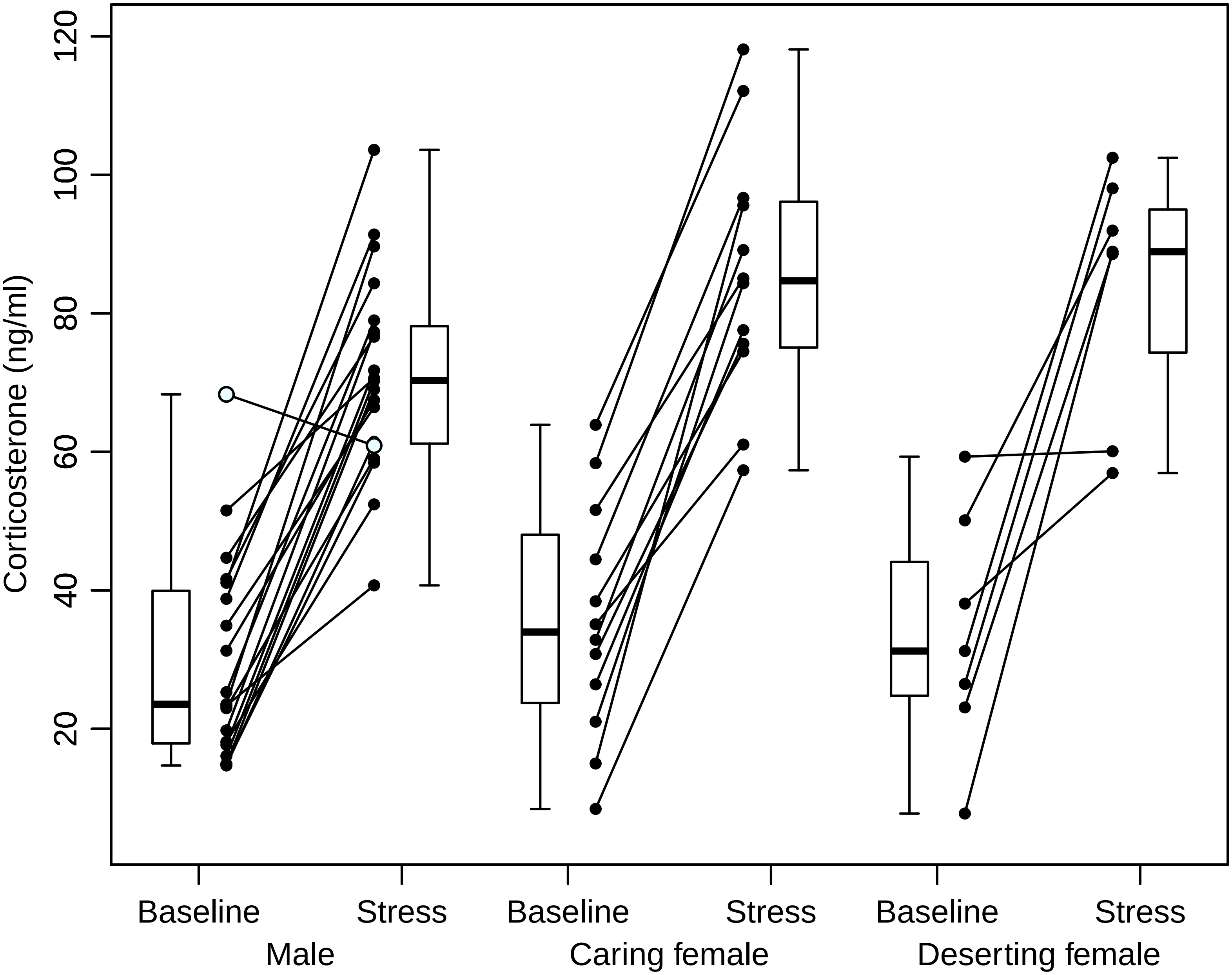
1400
1200
1000
800
600
400
200
Prolcatin (ng/ml)
Baseline
Stress
Baseline
Stress
Baseline
Stress
Male
Caring female
Deserting female

Fig. 2



Female desertion
Stress−induced prolactin levels in females (ng/ml)
0
200
400
600
800
1000
Brood age (days)
0
5
10
15
20
25

Fig. 3


Corticosterone (ng/ml)
120
100
80
60
40
20
Baseline
Stress
Baseline
Stress
Baseline
Stress
Male
Caring female
Deserting female