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House sparrows (*Passer domesticus*) adjust their social status position to their physiological costs

Karin M. Lindström[a,*], Dennis Hasselquist[b], Martin Wikelski[a]

[a]*Department of Ecology and Evolutionary Biology, Princeton University, 106 Guyot Hall, Princeton, NJ 08544, USA*
[b]*Department of Animal Ecology, Lund University, Ecology Building, 223 63 Lund, Sweden*

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Abstract

For group-living animals, the maintenance of a position in the social hierarchy may be associated with physiological costs such as increased stress and energy expenditure or suppressed immune functions. In this study, we experimentally manipulated the social status of house sparrows so that each bird experienced two social environments in random sequence: being dominant and subordinate. For 14 males, we investigated how corticosterone concentrations, energy expenditure and immune functions were affected by these changes in social status position. We found that the cost of maintaining a social status position differed between individuals and were related to individual body size. Birds with small body size had increased costs in terms of increased stress responses and reduced cell-mediated immune responses while being experimentally kept as dominants, while birds with large body size had increased costs while they were subordinates. We also found that birds with increased energetic and immunological costs as *dominants* obtained a low status position in the large group, while birds with increased costs as *subordinates* obtained a high status position in the large group. In summary, we found that the costs associated with the maintenance of social status position differed between individuals and was related to the individuals' body size. Furthermore, in a large group, individuals maintained a social status position that minimized energetic and immunological costs.

Keywords: Social status position; Physiological cost; Glucocorticoid; Stress; Immunocompetence; *Passer domesticus*

Introduction

Social hierarchies exist among most group-living organisms. The maintenance of a social status position within the group is an important trait for the individual because it determines the priority of access to resources. Because the maintenance of a social status position can be associated with costs (Goymann and Wingfield, 2004), an individual has to balance the potential benefits of maintaining a social status position with the costs. Social status maintenance can be costly because the competitive interactions with other members of the group can induce social 'stress', commonly quantified as an elevation of glucocorticoid hormones released by the hypothalamic pituitary–adrenocortical (HPA) axis (Abbott et al., 2003; Goymann and Wingfield, 2004; Sapolsky et al., 2000; Wingfield et al., 1997). Secondly, an individual's social status can also influence the exposure to stressors, such as starvation or predation. A recent review showed that dominant individuals generally have higher glucocorticoid concentrations compared to those of subordinates in species with cooperate breeding systems (Sands and Creel, 2004, see also Creel, 2001; Creel et al., 1996). A reason for this could be that it is resource demanding to monopolize breeding efforts. In other study systems, like for example winter flocks of birds, glucocorticoid concentrations can instead be higher in subordinate individuals (Creel, 2001; Hegner and Wingfield, 1987). Such patterns could occur because subordinates are more likely to be excluded from food sources or hiding places or because they are harassed by dominants (Barnard et al.,

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* Corresponding author. Present address: Department of Population Biology, Evolutionary Biology Centre, Uppsala University, Norbyvägen 18 D, 752 36 Uppsala, Sweden.

E-mail address: karin.lindstrom@ebc.uu.se (K.M. Lindström).

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1998; Creel et al., 1996; Hegner and Wingfield, 1987; Sloman et al., 2001). Thus, glucocorticoid concentrations can be elevated both in dominants and subordinates under different circumstances.

The finding that, for example, an individual of low status has elevated glucocorticoid concentrations in the field, can have several potential explanations. First, it could be because the maintenance of a low status position is associated with high levels of social stress. Second, the pattern could occur because low status individuals are in poor body condition as a result of their low status. Because an individual's social status position can be related to its physical condition, it is difficult to disentangle cause and effect when investigating correlations between an individual's social status position and glucocorticosteroids concentrations.

Individuals may also differ in their ability to cope with social conflicts (Bartolomuccia et al., 2003; Sapolsky, 1995). Research on primates and birds has emphasized that individuals can have different styles of coping with social stressors, and their individual coping style can influence their response (Carere et al., 2001; Verbeek et al., 1999; Virgin and Sapolsky, 1997). For individuals of a wide variety of species, body size is an important predictor of social dominance because a large body size can be a competitive advantage in physical conflicts (Andersson, 1994). In addition, a large body size can influence an individual's motivation to obtain a dominant social status because individuals with large body sizes have higher absolute energetic requirements (Bryant and Tatner, 1991). Thus, if the availability of resources becomes limited, an individual of large size would reach a negative energetic balance faster than a small animal.

The exposure to stress can be physiologically costly via several pathways. If an elevation of glucocorticoid concentrations is pronounced and persistent, it can impair reproduction (Moore and Jessop, 2003; Wingfield and Sapolsky, 2003) or lead to an increased mortality rate (Romero and Wikelski, 2001). Thus, from an evolutionary perspective, an animal is expected to have evolved behavioral responses to avoid prolonged stress exposure. Chronic stress can be the result of an imbalance in an organism's energy budget (Goymann and Wingfield, 2004) and lead to a down-regulation of the immune system (Apanius, 1998; Hillgarth and Wingfield, 1997) and increased susceptibility to diseases (Barnard et al., 1998; Nelson et al., 2002). Furthermore, relationships between an individual's social status and both energy expenditure (Bryant and Newton, 1994; Hogstad, 1987; Reinertsen and Hogstad, 1994; Senar, 1990) and immune responses or disease resistance (Apanius, 1998; Lindström, 2004; Zuk and Johnsen, 2000) have been found. Because elevation of glucocorticoid hormone concentrations, increased metabolic expenditure and suppressed immunity all represent physiological costs that can be inter-related and because previous studies show inconsistency in regards to which social status position is most costly to maintain, we designed an experiment to disentangle these factors.

Our approach was to perform an experiment on house sparrows (*Passer domesticus*) under standardized conditions in aviaries and measure the costs associated with the maintenance of a dominant and subordinate social status position. The birds in the study were kept isolated before the experiment so that any differences in body condition that may be a result of social status differences would be minimized. We then experimentally manipulated social status and measured corticosterone concentrations, energy expenditure and two types of immune responses both when a bird was in a high and in a low status position. With this approach, we could test several hypotheses.

First, we tested if the maintenance of a certain social status position would be associated with increased physiological costs. If so, we expected to find increased costs (elevated corticosterone levels, increased energy expenditure and suppressed immune responses) in one particular social status position. Because previous studies have shown that both a dominant and a subordinate social status position can be associated with higher costs, we made no a priori prediction of the direction of this relationship.

Second, we tested the hypothesis that birds with a large body size would be better adapted to cope with the social challenge of maintaining a dominant position. Here, we predicted that the physiological cost of maintaining a dominant status position would be related to an individual's body size. Because a large body size can give a competitive advantage, we expected birds of large body size to have a lower cost of maintaining a dominant position compared to small birds.

Finally, we examined if individuals were behaviorally adapted to minimize their physiological cost when the choice of social status position was less restricted. Here, our expectation was that individuals that had experienced increased costs when placed in a dominant social status position in the experiment would reduce these costs by maintaining a low status position in a larger group.

Material and methods

In September and October 2001, 28 experimental male house sparrows were caught in the Princeton area (40,35°N: 74,66°W) using mistnets. After capture, all birds were banded and aged as yearlings or older according to Svensson (1992). We used 18 previously wild caught house sparrows that had been held in captivity for a maximum of 4 months as social partners (see below). After capture, all birds were kept in randomly mixed groups of 5–10 in outdoor aviaries (2 × 2 × 1.5 m). Two weeks before the experiment started, all birds were removed from the aviaries and placed in individual indoor cages (0.5 × 0.5 × 0.4 m) each containing perches and sand. Birds were fed ad libitum, and water was exchanged each day throughout the experiment. Birds were fed a mixture of seeds (millet, thistle, safflower), and as food supplements, birds were given fresh fruit, meal worms, egg and spinach leaves. In the aviary, all cages were visually isolated from each other, but birds could still be in auditory contact. Throughout the experiment, birds were kept on a constant day length cycle (11 h L:13 h D) similar to the prevailing outdoor light conditions when the birds were transferred from the outdoor aviaries and a constant ambient temperature (20°C). The study was done in accordance with the guidelines specified in the United States by the National Institutes of Health *Guide for the Care and Use of Laboratory Animals*.

We measured the tarsus length of the right leg (to the nearest 0.1 mm) of each bird with digital callipers. All birds were marked with color bands, their body mass was measured (to the nearest 0.1 g) with a Pesola spring balance before the experiment started and twice during the experiment, as they were removed from the metabolic chambers. We used a set of measurements for body mass taken before the experiment started to calculate body size (see below) because this measurement represented the body mass a bird had obtained without any influence of the social status treatment. The motivation for using only male house sparrows for this study was that we also aimed to investigate the signaling function of the male bib badge. Several studies have shown that the social status of male house sparrows can be predicted by the size of the black bib badge (e.g. Hein et al., 2003; Liker and Barta, 2001; Møller, 1987), and we measured the size of this badge by tracing the contours of the black feather area twice onto a transparent paper that was cut out and weighed (to the nearest 0.01 g).

Experimental design

The experiment was performed between Nov 2001 and Jan 2002. We randomly assigned each of the 28 birds to either control or social status treatment. For each bird receiving the social status treatment (n = 14), we placed an additional male, the social partner bird, in their cage. Experimental birds received reverse treatments and thus served as their own controls during the experiment. Nevertheless, we kept an additional group of control birds (n = 14) that continued to be kept on their own in their cages. The reason for including single birds as a second control group was to be able to detect changes in the measured variables over time, e.g. making it possible to detect any changes in corticosterone levels that could be due to habituation to the aviary environment. In the beginning of the experiment, we measured the social status (as described below) of all birds in the social status treatment group and classified each bird as being either dominant or subordinate to its social partner each day for three consecutive days, starting 3 days after their new social partner had been introduced. After 2 weeks of treatment, we began taking measurements of body mass, metabolic expenditure, corticosterone levels and immunocompetence (as described below) of experimental birds and controls.

After this first set of measurements, birds in the social status treatment group were assigned a new social partner in order to change their social status. Because an individual's social status is a relative concept, an individual's social status can be changed by changing the social environment. Thus, birds that had previously been dominants were now assigned a social partner to which they were subordinate, and previously subordinate birds were assigned a partner to which they were dominant. We used information on body size, age and the outcome of previous encounters to predict the hierarchy in the novel pairs. The change in social status was accomplished by exchanging the social partner birds between cages, while all experimental birds remained in their respective cages throughout the experiment. In some cases, we also used previously unassigned birds as social partners to obtain a desired social status situation for a treatment bird. Because all social partner birds had been kept separately, these encounters resulted in new social contacts. We checked if the social status treatment birds had actually switched social status by measuring their social status (as described below) in the new dyads daily during three consecutive days, starting 3 days after their new social partner had been introduced. At this time, 10 out of 14 of the social status changes had been successful, whereas 4 partner birds had to be replaced by new birds to accomplish the status switch. When all birds had obtained a new status, 2 weeks passed before all measurements were repeated using the same protocol as before. After the experiment, all the 28 experimental birds (birds in the social status treatment group and controls) were divided into four groups and placed in larger cages (100 × 50 × 70 cm) each housing seven birds. These four groups had a similar age composition, but otherwise birds were distributed randomly between groups. Each bird's social status within these groups (large group social status) was measured (as described below) after 1 week had passed, during three consecutive days. Since some of the experimental birds had been trapped at the same sites and birds could have encountered each other in the outdoor aviaries, these social contacts may not have been novel. Still, the birds had not been in contact with each other for at least 3 months.

Immune challenges

To measure humoral immunocompetence, we immunized all birds with keyhole limpet hemocyanin (KLH) (Sigma-Aldrich, St. Louis, Missouri, USA). This protein has immunostimulatory properties and activates the cellular and the humoral immune response of birds and other animals (Harris and Markl, 1999; Hasselquist et al., 1999). To perform the immunizations, we emulsified 1 mg KLH/ml diluted in sterile H_2O together with 1 ml of Freund's incomplete adjuvant (Sigma-Aldrich), and a total volume of 70 μl was injected subcutaneously in fat tissue in the thorax.

Birds were immunized with KLH a total of three times, once before the experiment started and then twice during the experiment so that the second and third injections that were used to compare antibody responses were secondary and tertiary booster injections. Injections and blood samples were taken the forth week after each treatment started. Since the social status treatment was randomized in respect to order, a subset of the birds in the social status treatment group ($n = 10$) received their secondary challenge while they were in a subordinate position and their tertiary injection in the dominant position, and for the remaining subset ($n = 4$), the order of treatments was the opposite. We determined antibody titer 5 days after the secondary and tertiary challenge. This day corresponded to the mean peak of secondary antibody production. We also analyzed blood antibody content 13 days after the primary challenge and 5, 7 and 9 days after the second and third KLH injection (data not shown). The samples were taken by extracting 50–100 μl blood from the jugular vein with a 27 G syringe. Samples were kept cold and centrifuged for 5 min at 5000 rpm within 2 h of sampling. The plasma section was stored frozen at −20°C until analyzed. Antibody titers were analyzed in an ELISA assay following a protocol described previously (Hasselquist et al., 1999, 2001).

Delayed-type hypersensitivity responses were used for measurements of cell-mediated immune responses (CMI), and we used phytohemagglutinin (PHA) as antigen. In this test, PHA was diluted to the concentration 1 mg/ml in phosphate-buffered saline solution (PBS), and 100 μl of this solution containing 0.1 mg PHA (Sigma-Aldrich) was injected subcutaneously into the right wing web with a 27 G syringe. The injections were given the forth week after each treatment started. The response was measured as the difference in wing-web thickness before and 24 h after the PHA injection, measured with a thickness gauge (Precision Graphic Instruments, Inc. Spokane, Washington State, USA) to the nearest micrometer. Both of these techniques have been used in other studies of wild passerines (e.g. Hasselquist et al., 1999, 2001; Martin et al., 2005) and in combination, they provide reliable measurements of the magnitude of two types of immune responses.

Social status

To determine the social status of the birds, we observed their behaviors from a hidden observation spot behind a screen in the aviary placed 2 m from the cages. The screen was placed so that the observer could enter and exit the room without being seen by the birds. Before each observation period, the water and seeds bowls were removed for 2 h, and the observations started after reintroducing the bowls. Before the observations, the aviary door was opened once to simulate a human exit from the aviary. This procedure was performed to give an impression that the observer had left the aviary. As a result, birds immediately become more active and displayed a larger repertoire of behaviors (i.e. feeding, singing, social interactions). The observer took notes on behaviors displayed during social interactions between birds in the social status treatment group and their social partners. These social interactions were minor conflicts taking place at perches or around the food and water bowls. In these social interactions, winners and losers were assigned. Winners were birds that displayed dominant behaviors such as a threatening posture or a physical attack, while the other bird, the loser, responded with a subordinate behavior such as a flying or hopping withdrawal (Senar, 1990). Alternatively, if the bird that had initiated the threat was forced to withdraw, he was considered a loser, and the other bird was assigned as a winner. When one of the birds in a pair had won a significantly higher number of social interactions as determined by a binomial test, it was classified as the dominant bird of the cage that day. Social status measured in this way was highly repeatable in the same pairs between subsequent test days (93%, $n = 28$).

We also assigned each experimental bird a large group social status (ranging from 1–7) based on his social status position in a group that consisted of seven experimental birds. Observations of the large groups were made in similar way as described above. As before, we removed the food and water bowl of the cages and replaced them after 2 h. After reintroducing the food and water bowl, pair-wise social interactions that took place in the cages were observed. Each bird was identified with a unique color band, and winners and losers of social dyads were determined until we could assign a dominant or subordinate status individual in dyad comparisons. From these data, we constructed a matrix of pair-wise dominance relationships within each group. Because the number of possible pair-wise combinations was large, we used a simplified technique for the construction of the dominance hierarchy that included the assumption that dominance hierarchies were linear within the four groups. Thus, for example, if it had been determined that A was dominant over B, and B was dominant over C, we made the assumption that A would also be dominant over C. Although this technique did not take into account the full potential complexity of the dominance hierarchy, the method should produce a good index of the social status of each bird in the groups because linear dominance hierarchies are the most commonly observed social structure of groups of less than 10 individuals (Jameson et al., 1999). All observations that we made supported the assumption that hierarchies were linear also in our groups. Using the information from the matrixes, each experimental male was assigned a large group social status ranging from 1 (lowest in the group) to 7 (highest in the group). A large group social status was assigned each day in three consecutive observation days. The large group social status measured in this way was highly consistent (97%, $n = 28$) between days.

Corticosterone concentrations

We measured the magnitude of stress responses at two occasions for each bird, 2 weeks after a new social partner was introduced for experimental birds. Stress responses were measured by taking a series of 50 μl blood samples from the wing vein 3, 30 and 60 min after entering the experimental room. In four consecutive days, 7 randomly selected experimental birds per day were sampled for corticosterone and then placed individually in metabolic chambers for measurement of metabolic rate (see below). To standardize the procedure, birds were left undisturbed at least 3 h before each measurement session, always taking place at the same time of the day (15:00–16:00 h). Blood samples were taken at the same time of day, and we used a constant photo period to minimize seasonal and diurnal variation in corticosterone concentrations that has been reported from house sparrows (Rich and Romero, 2001). Blood samples were kept cold, centrifuged at 6000 × *g* for 5 min, the plasma section separated and stored at −20°C until assayed. The concentration of corticosterone in each sample was determined in duplicates in a direct radio immunoassay (RIA), performed on 5–20 μl of plasma, according to the methods described previously (Tarlow et al., 2001; Wingfield et al., 1992). We used corticosterone antibodies (B3-163 Endocrine Sciences, Calabasas, CA). The variation between assays was 15%, and the average variation within assays was 8%. Values were adjusted individually for recoveries, and the average recovery was 78%. Corticosterone concentrations are reported separately in Tables 1, 2. As a measure of the integrated corticosterone exposure, we calculated the magnitude of the stress response (ng corticosterone per hour) for each individual by adding the mean concentrations between 3 and 30 min measurement to the mean concentration between 30 and 60 min. The stress response showed strong positive correlations with corticosterone concentrations at each time point (3 min: $r_s = 0.58$, 30 min: $r = 0.92$, 60 min: $r = 0.78$, all $P < 0.001$, $n = 52$).

Table 1
Paired comparisons of the control group in the first and the second measurement

Response variable (unit)	Control						
	First	SD	Second	SD	Stats	*P*	*n*
BM (g)	25.8	1.5	25.4	1.3	$t = 2.23$	0.04	14
CMI (μm)	17.7	7.8	18.5	9.0	$t = -0.31$	0.76	14
KLH (o.d.)	26.6	17.3	42.6	11.1	$t = -3.6$	0.003	14
RMR (ml O_2/3 min)	46.8	6.2	65.7	32.8	$t = 2.05$	0.07	10
Stress response (ng cort/h)	30.1	12.4	31.3	13.9	$t = -0.23$	0.81	12
3 min (ng cort)	4.7	4.3	3.9	3.1	$Z = 0.41$	0.67	12
30 min (ng cort)	19.7	8.7	19.1	7.5	$t = 0.16$	0.87	12
60 min (ng cort)	15.9	8.6	20.3	11.3	$t = -1.19$	0.25	12

Group-wise means and standard deviations are given for body mass (BM), cell-mediated immune responses (CMI), standardized humoral immune responses (KLH), resting metabolic rate (RMR), stress response (mean cort/h), corticosterone concentration at 3, 30 and 60 minutes. Test statistics and *P* values are from paired *t* test (*t*) and Wilcoxon's matched pair tests (*Z*).

Table 2
Paired comparisons of experimental birds in the subordinate (Sub) and dominant (Dom) treatment

Response variable (unit)	Treatment						
	Sub	SD	Dom	SD	Stats	*P*	*n*
BM (standardized)	−0.24	0.98	0.08	1.25	$t = -0.85$	0.41	14
CMI (μm)	17.5	8.9	20.5	12.1	$t = -0.91$	0.35	14
KLH (standardized)	−0.01	1.1	0.17	1.3	$t = -0.55$	0.59	14
RMR (ml O_2/3 min)	51.2	21.1	55.2	12.4	$t = 0.49$	0.63	11
Stress response (ng cort/h)	28.5	11.1	27.8	10.6	$t = 0.27$	0.79	12
3 min (ng cort)	5.7	4.3	5.2	5.5	$Z = 0.23$	0.82	12
30 min (ng cort)	18.8	8.4	17.1	6.8	$t = 0.73$	0.48	12
60 min (ng cort)	13.8	9.1	16.1	8.8	$t = -0.77$	0.46	12

Group-wise means and standard deviations are given for standardized body mass (BM), cell-mediated immune responses (CMI), standardized humoral immune responses (KLH), resting metabolic rate (RMR), stress response (mean cort/h), corticosterone concentration at 3, 30 and 60 minutes. Test statistics and *P* values are from paired *t* test (*t*) and Wilcoxon's matched pair tests (*Z*).

Resting metabolic rate

We measured the resting metabolic rates 2 weeks after the start of each new social status treatment started by placing seven birds per measurement day in transparent plastic chambers (4 l), each containing two perches, seed and water trays. One chamber was always kept empty to obtain basal gas levels, and the system was calibrated every 70 min. Outside air was pumped through the chambers at a constant flow rate (600 ml/min), controlled by a mass-controlled flow meter. The CO_2 and O_2 content in the outflowing air were analyzed by sucking in a continuous subsample of air through a CO_2 and an O_2 analyzer (Sable Systems Inc., Henderson, Nevada, USA). We measured CO_2 and O_2 concentration every second for 10 min for each chamber. Air collection was alternated between chambers throughout the night, so that each chamber was measured at 80-min intervals. Resting metabolic rate for each bird was determined as the lowest 3-min O_2 consumption measured during the night, usually occurring between 3 and 4 am. During one of the nights, the system was malfunctioning, and we did not obtain reliable data from the 7 birds in this group, thus reducing the sample size. As could be expected, individuals resting metabolic rate showed positive covariance with body mass ($r = 0.45$, $P = 0.005$, $n = 36$).

Statistical analyses

All data were tested for normality with a Kolmogorov–Smironov test. When the data fulfilled the assumption of normality, we used parametric tests, and for other data, we used non-parametric tests. We tested for effects of treatment on the response variables using within-subject paired tests where data obtained from experimental birds in the dominant position were compared to those obtained in the subordinate position. We tested for effects of time in the control group with paired tests. In cases where time had a significant effect, we used data that were standardized when examining the effects of treatment to remove any influence of time.

For each bird in the experimental group, we calculated the "cost of dominance" by comparing values measured in the dominant position to those measured in the subordinate position. We considered increases in stress responses and energy expenditure and decreases in the magnitude of the cellular or humoral immune response to represent costs. In this calculation, the costs of dominance could also be negative (see Figs. 1, 2). A negative costs of dominance mean that an individual had increased costs in the subordinate status position.

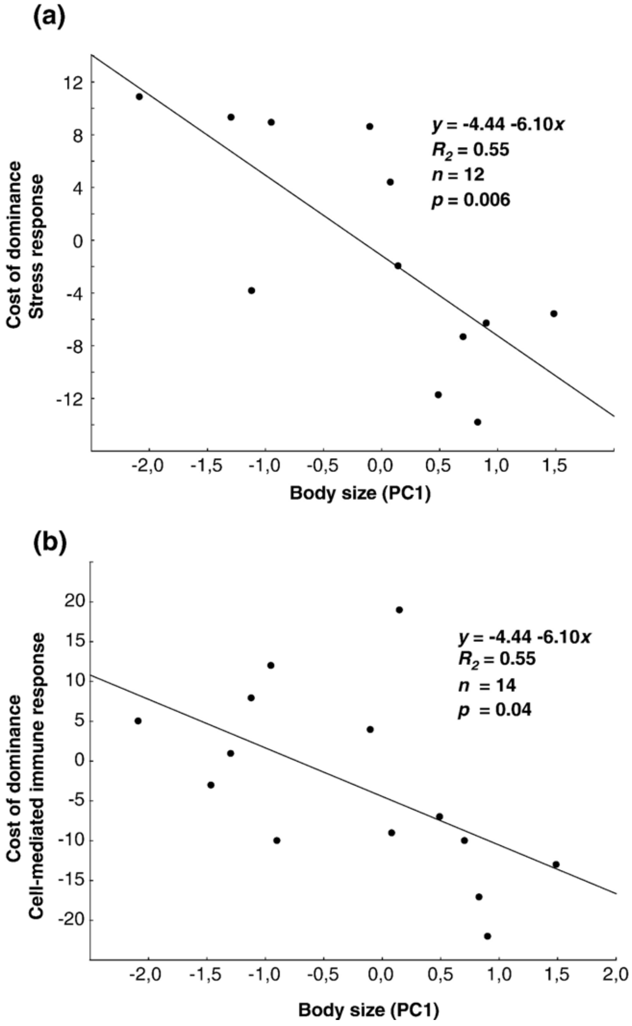

Fig. 1. The costs of dominance as a function of body size. Panel (a) shows the cost of dominance in terms of an increased stress response (ng corticosterone during 1 h), and panel (b) shows the costs of dominance in terms of a decreased cell-mediated immune responses (μm). Positive values are increased costs in the dominant position, and negative values are increased costs in the subordinate position. Body size is represented by PC1, which was the first principal component of variation for body mass and tarsus length.

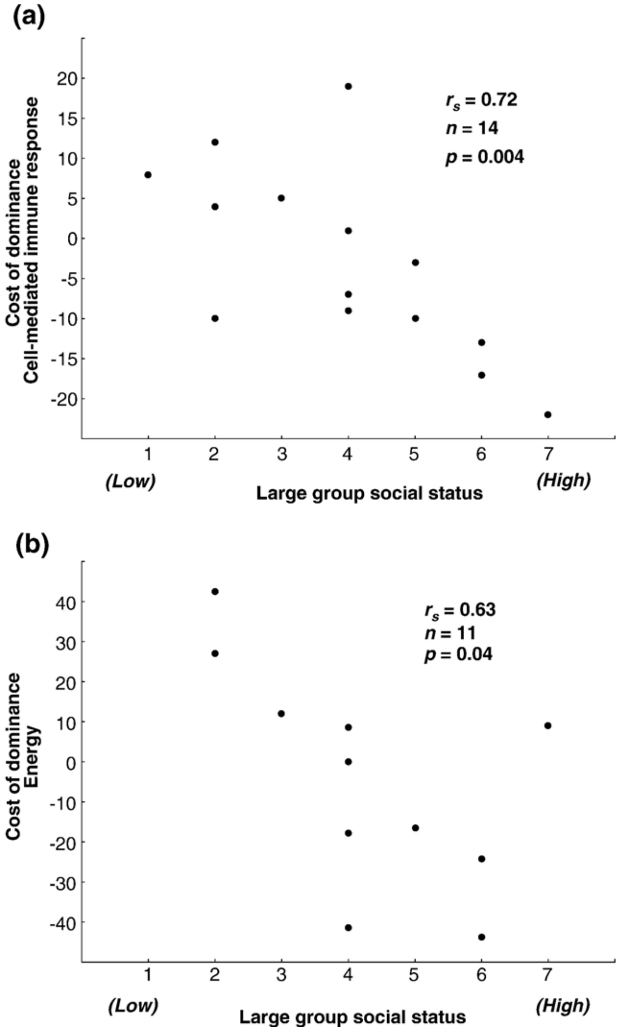

Fig. 2. The relationship between the costs of dominance and social status obtained in a group. Panel (a) shows the costs of dominance in terms of a decreased cell-mediated immune response (μm), and panel (b) shows the costs of dominance in terms of increased resting metabolic rate (ml O_2 consumed in 3 min). Positive values are increased costs in the dominant position, and negative values are increased costs in the subordinate position.

To estimate body size, we performed a principal component analyses that included body mass at the start of the experiment and tarsus length. Body mass and tarsus length were significantly correlated ($r = 0.51$, $P = 0.005$, $n = 28$). The first principal component, PC1, was positively related (0.87) to tarsus and body mass and explained 76% of the variance, thus we used this as an index of body size.

We used regression models to test whether body size could predict within-individual changes in energy expenditure, stress responses, humoral immune responses and CMI responses (dependent variables), and the presented R^2 values are coefficients of determination. We used non-parametric correlations to test for an association between costs of dominance and large group social status. r_s are

Spearman rank correlation coefficients. All statistical analyses were done with Statistica for Windows.

Results

Correlates of social status

We tested for correlations between large group social status (1–7) and badge size, age and body mass. Our measurements of badge size showed high repeatability ($r = 0.93$, $n = 27$), and there was large variation in badge size between males (range: 4.1–7.2 cm^2). Still, social status (large group status 1–7) was not significantly related to the size of the badge ($r_s = 0.10$, $P = 0.60$, $n = 28$). Instead, social status showed non-significant tendencies to correlate with age ($r_s = 0.36$, $P = 0.06$, $n = 28$) and body mass ($r_s = 0.36$, $P = 0.06$, $n = 28$).

Changes within controls

During the course of the experiment, there was a significant reduction of body mass among controls from the first to the second measurement (Table 1: BM). Antibody titers were significantly increased in the tertiary immune response compared to the secondary response (Table 1: KLH), while there was no significant change in the strength of the CMI (Table 1: CMI). There was a non-significant tendency for an increase in resting metabolic rate from the first to the second measurement (Table 1: RMR). We found no significant increase of corticosterone concentrations at 3, 30 or 60 min between the first and the second measurement (Table 1: 3 min, 30 min, 60 min). Stress responses did not change between measurements (Table 1).

General responses to the social status treatment

Because we had detected significant changes in body mass and humoral immune responses during the course of the experiment, we used data that were standardized within measurements for these variables when testing for an effect of treatment. In these comparisons, we found no significant effect of treatment on body mass, standardized humoral immune responses, resting metabolic rate, CMI, corticosterone concentrations at 3, 30 or 60 min or stress responses (Table 2).

Individual responses and body size

Although we found no general effect of the social status treatment, we also wanted to examine the possibility that birds in the experimental group responded to the social status treatment, but in different directions. We examined this hypothesis by testing for relationships between each individuals cost of dominance and their body size. For stress responses, we found that the cost of dominance was significantly related to an individual's body size, and as predicted, birds with a small body size had higher cost of dominance ($R^2 = 0.55$, $P = 0.006$, $df = 1,10$; Fig. 1a). We also found that costs of dominance in terms of suppressed CMI responses were significantly predicted by body size ($R^2 = 0.31$, $P = 0.04$, $df = 1,12$; Fig. 1b), as birds of small body size had suppressed CMI responses in the dominant position. For resting metabolic rate ($R^2 = 0.02$, $P = 0.69$, $df = 1,9$) and humoral immune responses ($R^2 = 0.10$, $P = 0.27$, $df = 1,12$), there was no significant relationship between the cost of dominance and body size.

Costs of dominance and large group social status

We also tested if the individual's costs of dominance were associated with the social status a bird obtained in a larger group consisting of 7 individuals. In terms of CMI responses, we found a significant negative relationship between the costs of dominance and an individual's large group social status ($r_s = -0.72$, $P = 0.004$, $n = 14$; Fig. 2a), and a high immunological costs of dominance was associated with a low social status in the large group. There was also a significant relationship between the costs of dominance in terms of resting metabolic rate and large group social status ($r_s = -0.63$, $P = 0.04$, $n = 11$; Fig. 2b), and birds with high energetic costs of dominance in the experiment maintained a low social status in the large group. For stress responses ($r_s = -0.44$, $P = 0.15$, $n = 12$) and humoral immune responses ($r_s = 0.11$, $P = 0.69$, $n = 14$), there was no significant relationship between the costs of dominance and large group social status.

Discussion

In this experiment, we found no support for the prediction that physiological costs of social status maintenance were generally increased in a dominant or a subdominant social status position (Table 2). In post-hoc power analyses, we found that we had high power (0.95–0.99) to detect medium sized changes in physiological costs (i.e. stress responses, body mass, energy expenditure, immune responses) but low power (0.27–0.30) to detect small changes with the repeated-measures design used in this experiment (Buchner et al., 1997). Thus, we conclude that any general physiological costs of maintaining high or low social status that acted uniformly on all individuals were either non-existent or small. Although no studies have examined this question experimentally, several other studies have reached a similar conclusion. For example, in a study on dark eyed juncos *Junco hyemalis*, no relationship between corticosterone concentrations and social status was found (Holberton et al., 1989). In primates, a recent meta-analyses showed that there was no consistent relationship between social status and stress responses (Abbott et al., 2003). A study on greylag geese (Kotrschal et al., 1998) showed that corticosterone concentrations were higher in non-breeding subordinates in the winter, while glucocorticoid concentrations instead were elevated for dominants in the breeding season.

We found that costs associated with social status maintenance differed between individuals. We found support for the prediction that house sparrows of large body size had lower costs of dominance. Moreover, changes in stress responses could be predicted by an individual's body size, and birds of large body size had reduced stress responses while they were maintaining a dominant status position, whereas their stress responses increased as subordinates. Changes in corticosterone concentrations can be understood by considering the allostatic load imposed on an individual (Goymann and Wingfield, 2004; McEwen and Wingfield, 2003). When energetic demands are increased or the supply of energy is reduced, the allostatic load can increase to a point where the levels of stress hormones become elevated. In the present study, food was supplemented ad libitum. Still, because the access to food can be restricted by dominants, birds could have perceived the food supply as more uncertain in a subordinate position (Ekman and Hake, 1990). Thus, the corticosterone concentration increase in birds of large body size in a subordinate status position could have been due to an expectation of an energy deficit, caused by their higher absolute metabolic demands. In addition, we found that large birds had suppressed CMI responses when placed in the subordinate position. Because glucocorticosteroids can down-regulate immune responses, this result could be a direct consequence of the increased magnitude of the stress responses (Berger et al., 2005; Martin et al., 2005).

An alternative explanation for the increased stress responses and decreased immune responses observed in large birds as subordinates could be that the birds of large body size were not accustomed to maintaining a subordinate social status position. Our data suggest that body size could be related to social status in this group of house sparrows, and large birds could therefore have been accustomed to maintaining a high social status in the wild. From other studies of animals in social hierarchies, it is known that social reorganizations in general (Cristol, 1995) and social defeat in particular can be strong stressors (Abbott et al., 2003; Bartolomuccia et al., 2003; Dugatkin, 1997).

In contrast to large birds, small birds experienced increased cost, like increased corticosterone concentrations, while maintaining a dominant social status position. For small birds, it could still be beneficial to maintain a dominant position if the increased costs are counterbalanced by some benefits (Ricklefs and Wikelski, 2002). In the wild, there could be several potential benefits associated with being dominant, including priority of access to food, high quality territories or females. Although these potential benefits may have served as a motive, it is unlikely that birds in this study actually enjoyed any of these benefits in the aviary environment.

We also found support for the prediction that physiological costs influenced the social status position that an individual obtained in a larger group where social status position was less restricted. We found that birds that had experienced suppressed CMI and increased basal metabolic rates when put in a *dominant* position were those that maintained a low social status in the large group, while birds that had reduced CMI and increased metabolic rates in a *subordinate* position were those that kept a high status position in the large group. These results suggest that individual house sparrows chose to maintain a social status position in the large group that minimized their immunological and energetic cost.

We found no evidence in this study that black bib badges predict an individual's social status. Instead, social status showed positive trends of relationships with age and body mass. These results contrasts with previous field studies on social dominance in house sparrows where male bib badge has been identified as the only predictor of male social status (Hein et al., 2003; Møller, 1987). The reason for the lack of correlation between badge size and social status in this study could be that our study was performed in the non-breeding season when the breast bib is not fully exposed. The absence of females in this study could also have affected this result. Two recent studies on house sparrows suggest that the signaling function of the male bib badge is most important in social interactions between males and females (Hein et al., 2003; Liker and Barta, 2001). Social signals like the black breast bib are primarily used to settle minor conflicts and to give a first visual impression of an individual's competitive ability (Maynard Smith and Harper, 1988). Because birds in this study had been exposed to each other during more long-term interactions, sparrows could have used other more direct cues to estimate competitive ability.

Although this study focused on estimating costs associated with social interactions among males, male–female and female–female interactions are also frequent in winter flocks of house sparrows (Hein et al., 2003; Jawor, 2000; Liker and Barta, 2001). In previous studies on mixed-sex flock of house sparrows, it has been found that both males and females are as frequently involved in aggressive social interactions, and conflicting results regarding the sex difference in social status have been reported (Jawor, 2000; Liker and Barta, 2001). Thus, social interactions among females or between males and females could entail the same physiological costs as those observed in this study for males.

For animals that live in groups, any change in stress responsiveness that is associated with social status maintenance is likely to be important because an individual's social status has to be maintained on a daily basis and over long periods of time. In this study, we found that social status maintenance in house sparrows affected stress responsiveness, and individuals of different body size responded differently to the same social situation. Small individuals had increased physiological costs of maintaining a dominant social status position, while large individuals had increased costs as subordinates. Finally, when the choice of social status position was less restricted, we found that individuals chose to maintain a social status position that minimized their physiological costs. Thus, the house sparrows behavioral decisions appeared to be influenced by their physiological state.

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