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SOCIAL STATUS AND AVAILABILITY OF FEMALES DETERMINE PATTERNS OF SPERM ALLOCATION IN THE FOWL

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Abstract.—Where sperm competition occurs, the number and quality of sperm males inseminate relative to rival males influences fertilization success. The number of sperm males produce, however, is limited, and theoretically males should allocate sperm according to the probability of gaining future reproductive opportunities and the reproductive benefits associated with copulations. However, the reproductive opportunities and value of copulations males obtain can change over their lifetime, but whether individuals respond to such changes by adjusting the way they allocate sperm is unclear. Here we show that, in the fowl, *Gallus gallus*, dominant males, which have preferential access to females, modulate the number of sperm they ejaculate according to the availability of females. When presented with two females, dominant males allocated more sperm to higher quality females, whereas when females were on their own, only copulation order had an affect on their sperm numbers. In contrast, subordinate males, whose mating activity is restricted by dominant males, allocated high numbers of sperm to initial copulations, irrespective of female availability. We further show, by manipulating male social status, that sperm allocation is both phenotypically plastic, with males adjusting their patterns of sperm allocation according to their dominance rank, and intrinsic, with males being consistently different in the way they allocate sperm, once the effects of social status are taken into account. This study suggests that males have evolved sophisticated patterns of sperm allocation to respond to frequent fluctuations in the value and frequency of reproductive opportunities.

Key words.—Dominance, reproductive opportunities, sexual selection, sperm allocation, sperm competition.

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In numerous species, females are promiscuous, leading to the ejaculates of males competing for fertilizations (Parker 1970; Smith 1984; Birkhead and Møller 1998; Simmons 2001). Where sperm competition arises, the number of sperm males inseminate relative to rivals plays a key role in determining fertilization success (Martin et al. 1974; Petersen et al. 2001; Neff et al. 2003; Garcia-Gonzalez and Simmons 2005), and this has been shown to be instrumental in shaping the evolution of a wide range of reproductive traits (Smith 1984; Andersson 1994; Birkhead and Møller 1998; Simmons 2001). For example, both within and across species, more intense sperm competition has been found to result in the evolution of larger testes, which increases sperm production and promotes success in sperm competition (Stockley et al. 1997; Hosken et al. 2001; Pitcher et al. 2005; Ramm et al. 2005). However, sperm production is costly (Dewsbury 1982; Olsson et al. 1997; Van Voorhies 1992) and the number of sperm males ejaculate can be limited (Squires et al. 1979: Nakatsuru and Kramer 1982; Birkhead et al. 1995), constraining male reproductive success (Preston et al. 2001). Under such circumstances, sexual selection is expected to lead to the evolution of strategic sperm allocation with males adjusting the number of sperm they inseminate according to the reproductive value of copulations and the probability and cost of acquiring future reproductive opportunities (Parker 1982, 1990a,b, 1998; Reinhold et al. 2002; Wedell et al. 2002).

The reproductive opportunities available to males often depend on the social environment and can be influenced by factors such as male social status and the number of available females (Parker 1983; Shapiro et al. 1994; Warner et al. 1995; Johnstone et al. 1996; Jennions and Petrie 1997; Widemo and Saether 1999). Social status mediates access to females, placing dominant and subordinate males in favored and disfavored roles, respectively (LeBoeuf 1974; Cheng and Burns 1988; Rudolfsen et al. 2006). Theoretically, favored and disfavored mating roles are expected to lead to the evolution of alternative ejaculate strategies and it is predicted that males in disfavored roles will allocate more sperm to initial copulations due to a lower probability of obtaining extra copulations and a higher risk of facing sperm competition (Parker 1990a, 1998; Ball and Parker 2000; Reinhold et al. 2002). In contrast, males in favored roles are expected to evolve more prudent sperm allocation strategies, adjusting ejaculate size according to the reproductive value of copulations because the probability of gaining future copulations is high (Parker 1990a, 1998; Ball and Parker 2000; Reinhold et al. 2002). Examining the interaction between targets of pre- and postcopulatory sexual selection, such as social status and ejaculate size, is therefore important in understanding the evolution of alternative reproductive strategies and how genetic diversity is maintained within the sexes.

The value of copulations can differ in a number of ways, and empirical research has shown that males allocate sperm according to female quality (Shapiro et al. 1994; Yusa 1994; Yasui 1996; MacDiarmid and Bulter 1999), the risk and intensity of sperm competition (Linley and Hinds 1975; Marconato and Shapiro 1996; Jivoff 1997; Hunter et al. 2000; Nicholls et al. 2001; Pound and Gage 2004), and the number of sperm males have previously inseminated in females (Pizzari et al. 2003). Nevertheless, the reproductive opportunities and the value of copulations males acquire can vary, for example with changes in social status (Setchell and Dixson 2002; Setchell and Lee 2003; Rudolfsen et al. 2006) and the quality of available females (Shapiro et al. 1994), and sexual selection is predicted to favor males that adjust their patterns

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of sperm allocation accordingly (Parker 1990a, 1998). However, the way variation in reproductive opportunities has shaped the evolution of male ejaculate strategies and whether patterns of sperm allocation are phenotypically plastic or fixed within males requires further research.

The aim of this study was therefore to experimentally test the hypothesis that individual males adjust the number of sperm they allocate to copulations of different reproductive value according to probability of gaining future reproductive opportunities, using the model system of the fowl. The fowl, *Gallus gallus*, is a highly promiscuous species living in socially structured groups in which sperm competition is intense (Collias and Collias 1996; Pizzari et al. 2002). Dominant males acquire more copulations than subordinate males (Guhl et al. 1945; Cheng and Burns 1988), which is due in part to dominant males restricting the copulation activity of subordinates (Pizzari et al. 2002), but also due to females preferring to copulate with dominant males (Pizzari and Birkhead 2000). Females further enhance the probability of dominant males fertilizing their eggs by ejecting the sperm of subordinate males (Pizzari and Birkhead 2000). In addition, females vary in their reproductive quality, and males are attracted to females with large ornaments (a fleshy appendage on the head called the comb) which are in better condition and produce eggs of greater mass and yolk content (Pizzari et al. 2003; Cornwallis 2004). The reproductive opportunities available to males are therefore dependent upon their social status, and the reproductive value of copulations is linked to female comb size.

We have previously shown that dominant and subordinate males differ in the way they allocate sperm under different levels of sperm competition and to females with different comb sizes (Pizzari et al. 2003). However, male social status and the number and reproductive quality of available females frequently changes (Collias and Collias 1996) and it is unknown whether individual males respond to such changes by adjusting their patterns of sperm allocation. Here we manipulate the number of reproductive opportunities in two ways: through (1) female availability, and (2) male social status, to test the prediction that individual males allocate more sperm to higher quality females when more reproductive opportunities are available.

MATERIALS AND METHODS

Study Population

We studied a free-ranging population of fowl that are behaviorally and morphologically very similar to red jungle fowl (Harrison 1987; Schütz and Jensen 2001) at Tovetorp Zoological Field Station, University of Stöckholm during April to July 2002 and May to August 2003. Males were randomly assigned to pairs and kept in aviaries (6 × 6 m) separated using wire netting from groups of four females in adjoining aviaries (6 × 8 m). Male dominance was assessed through pairwise interactions (Clutton-Brock et al. 1979). The birds were fully habituated to human presence and natural ejaculates were obtained by presenting males with live females fitted with plastic harnesses and held in a soliciting position (Pizzari et al. 2003). Following copulation, ejaculates were collected and measured using a Gilson (Middleton, WI) pipette and stored in 5% formalin solution for later counting, which was done using a standard protocol (Bakst and Cecil 1997; Pizzari et al. 2003). The comb size of females was measured every two weeks by taking a digital image against a standardized background with a scale under standardized light conditions. Each image was calibrated against the scale and the area (mm^2) of the comb was calculated using Adobe Photoshop (San Jose, CA). Body mass was also measured every two weeks to the nearest 10 g, and body size was measured at the start and the end of the breeding season using principal component one (PC1) of a principal components analysis of tarsus, wing, and head measurements. PC1 explained 79% of the variation in these morphological traits and an average of the two PC1 values that were obtained from measurements at the start and the end of the breeding season was used in all analyses. For body mass and comb size, the measurements used in analyses were those taken nearest to the time to when females were presented to males.

Availability of Females

The influence of the availability of females on male sperm allocation was assessed in two ways: first, on the number of sperm allocated to initial copulations, irrespective of female quality, and second, on the number of sperm allocated to attractive and unattractive females, as indicated by their comb size (Pizzari et al. 2003).

Part 1: Sperm allocated to initial copulations.—Males were exposed in a randomized order to two treatments. In both treatments males were chosen at random with respect to their social status. In the first treatment (single) males were presented with a single female and allowed to copulate once. In the second treatment (paired) males were presented with two females who were two meters apart from each other and equidistant from the male, who was at least one meter away. The male was prevented from copulating with one female, chosen at random, by placing a small wire cage over her. The male was allowed to copulate with the uncaged female once. This experiment was carried out using 20 males who were exposed to the two treatments and were replicated once in the single treatment and up to three times in the paired treatment. Variation in the number of sperm males allocated to females presented singly and in pairs was analyzed using a generalized linear mixed model (GLMM) with male social status and female presentation (1 = single, 2 = pair) as fixed effects and male identity nested within group, defined as the subject of repeated measure, and year as random effects. Sperm numbers followed a Poisson distribution, which is typical for count data (Quinn and Keough 2002), and therefore the model was defined with a Poisson error structure.

Part 2: Sperm allocated to attractive and unattractive females.—Sperm allocated to attractive and unattractive females was assessed when females were presented in: (a) isolation and (b) pairs. (a) Presentation of females in isolation: Males were presented with a single female, chosen at random, and allowed to copulate once. Following the first copulation, males were presented with a second female and again allowed to copulate once. Males were then sexually rested for 48 h, which is the time required for males to replenish their sperm reserves (Etches 1996), and, consistent with this idea, the number of sperm that dominant and subordinate males ejaculated during first trials was not significantly different from that ejaculated 48 h later (GLMM: total number of sperm as response variable, trial and status as a fixed factor, and male nested within group as a random factor: trial, $F_{1,115} = 1.59$, $P = 0.21$; status, $F_{1,115} = 0.36$, $P = 0.55$; trial × status, $F_{1,114} = 0.29$, $P = 0.59$). After 48 h of sexual rest, males were presented with the same two females, but the order in which they copulated with females was reversed. This was to control for the effects of copulation order on sperm numbers. This experiment was conducted using 20 males, each replicated once. Data from first copulations were the same as that used in the single treatment in part (1). (b) Presentation of females in pairs: Males were presented with two females simultaneously, but prevented from copulating with one female, chosen at random, by placing a wire cage over her. Males were allowed to copulate once with the uncaged female. Females were then presented again, but the cage was placed over the female that previously received a copulation, allowing the male to copulate with the second female. The male therefore copulated with each female once. After 48 h of sexual rest the protocol was repeated but the order in which males copulated with the females was reversed. This experiment was carried out using the same 20 males as in (a). Males were replicated up to three times each with different pair of females, and data from first copulations were the same as that used in the paired treatment in part (1). In all experiments, males were isolated from their pair male 30 min prior to the presentation of females. In (a) and (b), the two females were ranked according to their relative comb sizes: large or small. We analyzed variation in the relative number of sperm that males allocated to large-combed females (number of sperm ejaculated with large-combed female − number of sperm ejaculated with small-combed female/total number of sperm ejaculated) using a general linear mixed model with male social status, female presentation, copulation order, female body size, and body mass as fixed effects and male identity nested within male group (defined as the subject of repeated measure) and year as random effects. The effect of female comb size on the number of sperm that males ejaculated was analyzed by testing the relative number of sperm that dominant and subordinate males allocated to large-combed females (mean ± SE) against zero using least-square means from the GLMM.

Manipulation of Social Status

At the beginning of each breeding season pairs of males were established (2002: $n_{\text{pairs}} = 8$; 2003: $n_{\text{pairs}} = 8$). Halfway through each breeding season social status was experimentally manipulated by switching males between pairs. Male pairs were randomly chosen two at a time, and the two dominant males from these pairs were placed together, resulting in one male decreasing in status and becoming subordinate. The same was done with the two subordinates from these pairs, leading to one subordinate becoming dominant. Overall, this resulted in 16 males changing status. After the manipulation, groups were left for one week to acclimatize. In 2003, two males became sick and could not be used. One male, who was only replicated once when subordinate and deviated from other males, was excluded from the analysis as a significant outlier (Cook's distance = 1.23). Three males used in both 2002 and 2003 changed status between years, giving a total of 16 males that were used in the analysis. In 2002, male sperm allocation patterns were measured twice before and twice after the status manipulation and in 2003 twice before and once after using the experimental protocol described in Part 2(a) (Availability of females). We analyzed variation in the relative number of sperm allocated to large-combed females when males were dominant and subordinate using a GLMM with male social status, direction of status change (1 = dominant to subordinate, 2 = subordinate to dominant), and copulation order as fixed effects and male identity (defined as the subject of repeated measure) and year as random effects. We also analyzed variation in the relative number of sperm allocated to first copulations (number of sperm ejaculated during first copulation − number of sperm ejaculated during second copulation/total number of sperm ejaculated) when males were dominant and subordinate using a GLMM with male social status, direction of status change, and female comb rank (1 = large, 2 = small) as fixed effects and male identity (defined as the subject of repeated measure) and year as random effects.

All analyses were conducted using SAS version 9.1 (SAS Institute, Cary, NC), and all results are given in the figure legends.

RESULTS

Availability of Females

Consistent with the idea that males are more prudent with their sperm when more reproductive opportunities are available, dominant males allocated less sperm to females when females were presented in a pair than when females were on their own (Fig. 1). In contrast, subordinate males did not vary the number of sperm they ejaculated according to whether females were presented singly or as a pair (Fig. 1).

The differences in the number of sperm that dominant and subordinate males allocated to initial copulations influenced the number of sperm they ejaculated during subsequent copulations. When females were presented in pairs, dominant males allocated significantly more sperm than subordinate males to females with large combs during second copulations (Fig. 2A). This resulted in dominant males, over two copulations, allocating more sperm to large-combed females than to females with small combs (see Fig. 2A legend). However, the relative number of sperm that subordinate males transferred to females with large and small combs was not significantly different (see Fig. 2A legend). There was a significant interaction between male social status, the number of females presented, and copulation order, indicating that the difference in the sperm allocated by dominant and subordinate males to females when they were presented in pairs disappeared when they were presented singly (Fig. 2). When females were presented singly, the number of sperm that dominant males allocated to first and second copulations with large-combed females did not differ from that of subordinate males (see Fig. 2B). Furthermore, this meant that neither dominant nor subordinate males allocated more sperm to females with large combs when females were presented on their

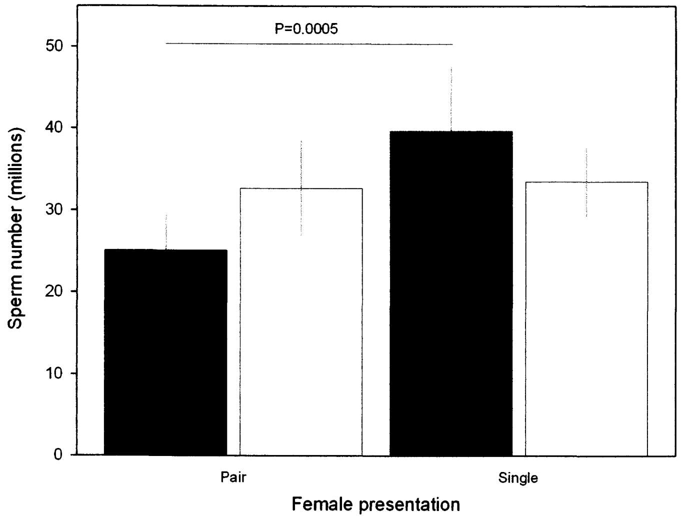

FIG. 1. Number of sperm allocated (mean ± SE) by dominant (filled bars) and subordinate males (empty bars) in a single copulation to females when they were presented in a pair (pair) and when presented on their own (single). Dominant males allocated more sperm to females when they were presented in isolation (least-square means: $P = 0.0005$), whereas the number of sperm that subordinate males ejaculated was unaffected by whether females were presented as pairs or singly (status, $F_{1,105} = 0.34$, $P = 0.559$; female presentation, $F_{1,105} = 5.89$, $P = 0.016$; status × female presentation, $F_{1,105} = 5.52$, $P = 0.021$).

own (Fig. 2B legend). This suggests that, irrespective of the availability of females, subordinate males consistently allocate higher numbers of sperm to initial copulations, whereas dominant males allocate more sperm to the highest quality female when more females are available.

Manipulation of Social Status

Manipulating social status had a marked effect on the way males allocated sperm to females with large combs and initial copulations. When males were dominant they allocated significantly more sperm to females with large combs (Fig. 3A) and significantly less sperm to first copulations than when they were subordinate (Fig. 3B). The direction in which males changed status (dominant to subordinate or subordinate to dominant) had no effect on the relative number of sperm males transferred to females with large combs or initial copulations (see Fig. 3 legend). The relative number of sperm males allocated to females with large combs and initial copulations when they were dominant was also positively related to the number of sperm they allocated when they were subordinate (Fig. 3): males that transferred more sperm during initial copulations (averaged over dominance ranks = sperm ejaculated when dominant + sperm ejaculated when subordinate/2) transferred significantly less sperm to females with large combs (Fig. 4). This indicates that, in addition to males adjusting their sperm allocation patterns according to their social status, there were also inherent differences between males, and allocating more sperm to current copulations was traded off against allocating sperm to females of the highest quality.

DISCUSSION

The results of this study demonstrate that males adjust their patterns of sperm allocation in response to changes in the availability of reproductive opportunities, which in the fowl is determined by male social status and the number of accessible females. These results have important implications for our understanding of the evolution of alternative reproductive strategies and the maintenance of genetic variation within populations.

The availability of reproductive opportunities, as determined by the number of soliciting females, elicited social status–dependent patterns of sperm allocation. Subordinate males consistently allocated more sperm to initial copulations regardless of female quality or whether females were presented in pairs or on their own. This was not due to the inability of individual males to adjust their sperm numbers, since when they became dominant they preferentially allocated sperm to large-combed females. Therefore, sexual selection may favor males when in subordinate positions to increase the number of sperm they allocate to initial copulations, irrespective of the availability of females, because the probability of acquiring subsequent copulations is low due to dominant males preventing copulation (Parker 1983; Reinhold et al. 2002; Wedell et al. 2002). Models of sperm allocation have demonstrated that males should allocate more sperm to initial copulations, largely due to the uncertainty of gaining extra copulations (Freyer et al. 1998; Reinhold et al. 2002), and males in a wide variety of species ejaculate the majority of their sperm reserves during first copulations (Squires et al. 1979; Pitnick and Markow 1994; Birkhead et al. 1995). Selection for allocating more sperm to initial copulations may be further intensified by the fact that in some species female propensity to remate, and hence sperm competition, is reduced by receiving larger ejaculates (Cook and Wedell 1999).

Theoretically, as the probability of obtaining subsequent copulations increases, and given there is variance in female quality (Parker 1983; Reinhold et al. 2002), males are expected to adjust their allocation patterns, transferring more sperm to the highest quality females (Reinhold et al. 2002). The probability of dominant males gaining copulations with additional females is higher than that of subordinate males (Guhl et al. 1945; Cheng and Burns 1988; Pizzari and Birkhead 2000), and thus it is expected that sexual selection will favor dominant males that strategically allocate sperm according to the reproductive value of copulations (Parker 1998; Reinhold et al. 2002; Wedell et al. 2002). In accordance with theory, when dominant males were presented with pairs of females, males allocated more sperm to females of higher quality, despite there still being strong effects of copulation order. Nonetheless, under natural conditions the composition of social groups frequently changes (Collias and Collias 1996), and even the probability of dominant males gaining future copulations will be low when the number of females in groups declines. Consistent with this idea, dominant males ceased to allocate more sperm to higher quality females when females were presented on their own and transferred greater numbers of sperm to initial copulations. This suggests that, mechanistically, males may use the number of females vi-

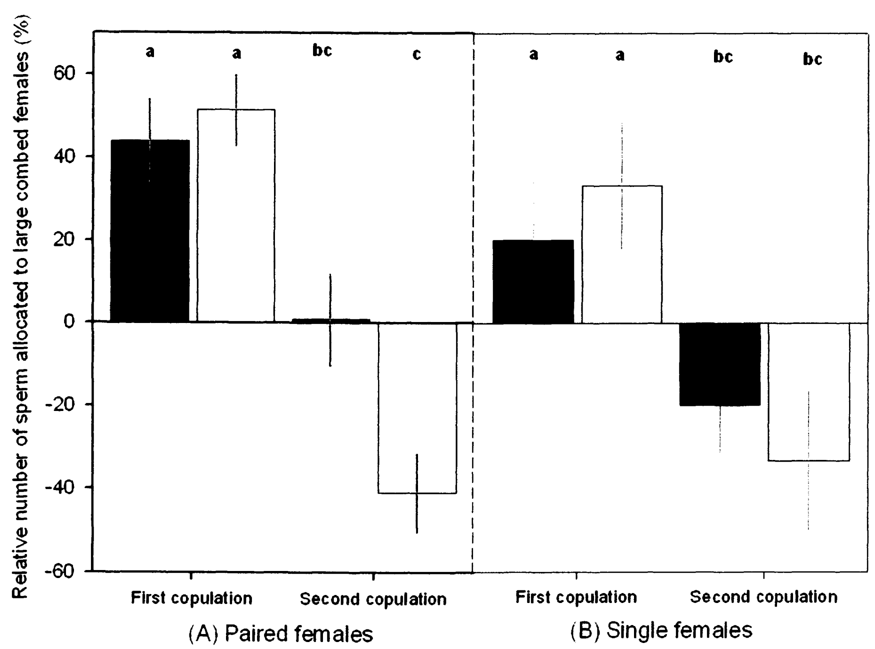

FIG. 2. Relative number of sperm (mean ± SE) allocated to females with relatively large combs by dominant (filled bars) and subordinate males (empty bars) when females were presented in (A) pairs and (B) singly. Lowercase letters represent significant differences between bars (least-square means $P < 0.05$). The dashed line separates bars that represent when females were presented as pairs and as singles. (A) When females were presented in pairs, dominant males allocated significantly more sperm to large-combed females during second copulations than did subordinate males (least-square means $P < 0.05$). This resulted in dominant, but not subordinate, males allocating more sperm over two copulations to large-combed females (relative sperm number (mean ± SE) tested against zero using least-square means from generalized linear mixed model (GLMM): dominant males, 15.20 ± 0.69, $t = 2.20$, $n = 10$, $P = 0.031$; subordinate males, 0.51 ± 0.07, $t = 0.70$, $n = 10$, $P = 0.49$). (B) When females were presented singly, dominant and subordinate males did not differ in the number of sperm they allocated to large-combed females during first or second copulations (least-square means $P > 0.05$). Consequently, when females were presented singly, neither dominant nor subordinate males allocated more sperm to females with large combs (relative sperm number mean ± SE tested against zero using least-square means from GLMM: dominant males, 0.035 ± 1.15, $t = 0.03$, $n = 10$, $P = 0.98$; subordinate males, 0.0092 ± 1.15, $t = 0.01$, $n = 10$, $P = 0.99$) leading to a significant interaction between status, female presentation, and copulation order (GLMM: status, $F_{1,110} = 0.69$, $P = 0.41$; female presentation, $F_{1,110} = 1.56$, $P = 0.21$; copulation order, $F_{1,110} = 63.40$, $P < 0.0001$; status × female presentation × copulation order, $F_{4,110} = 2.72$, $P = 0.033$).

sually present to assess the probability of obtaining future reproductive opportunities and adjust their patterns of sperm allocation accordingly. Similar findings have been documented in the bluehead wrasse, *Thalassoma bifasciatum*, in which males with higher mating success release fewer sperm per spawning event than less successful males, and fertilization rates are dependent upon female body size, a correlate of fecundity, and whether spawning occurs in groups or in pairs (Shapiro et al. 1994; Warner et al. 1995). The reproductive success of males in favored mating roles may therefore be dependent on the availability of females, whereas that of males in disfavored roles will be more influenced by the presence of other males, resulting in the evolution of status-specific sperm allocation patterns that are expressed according to current social conditions.

Status-specific patterns of sperm allocation may theoretically evolve to be phenotypically plastic, with males adjusting the way they allocate sperm according to their mating role (conditional strategy), or fixed, with males remaining constant over their lifetime (alternative strategy) (Gross 1996). Whether sexual selection favors the evolution of conditional or alternative strategies of sperm allocation is likely to depend upon the frequency with which males occupy different social positions and the net benefits and costs associated with adjusting patterns of sperm allocation. In the fowl, male social status frequently changes (Collias and Collias 1996; Cornwallis 2004), generating variation in copulation success (Guhl et al. 1945; Cheng and Burns 1988), and the number of sperm males have available for ejaculation influences their fornication success, but can be limited (Martin et al. 1974; Pizzari et al. 2003). These conditions are likely to have promoted the evolution of phenotypically plastic patterns of sperm allocation that were evident in this study, with males changing the relative number of sperm they transferred to copulations according to their social status. This is consistent with recent research on Artic charr, *Salvelinus alpinus*, which revealed that males respond to changes in social status by adjusting their sperm quality (Rudolfsen et al. 2006).

Despite males being phenotypically plastic in their patterns of sperm allocation, after the effects of social status had been taken into account, there were consistent differences in the way males allocated sperm to copulations. Males appeared to face a trade-off between allocating sperm to initial copulations and to females of higher quality. This relationship may have been accentuated by the fact that male behavioral choice for females was removed in this study, resulting in males copulating with the females they least preferred first 50% of the time. It is therefore possible that, under natural mating conditions, males may circumvent this trade-off between allocating sperm to first copulations and to females

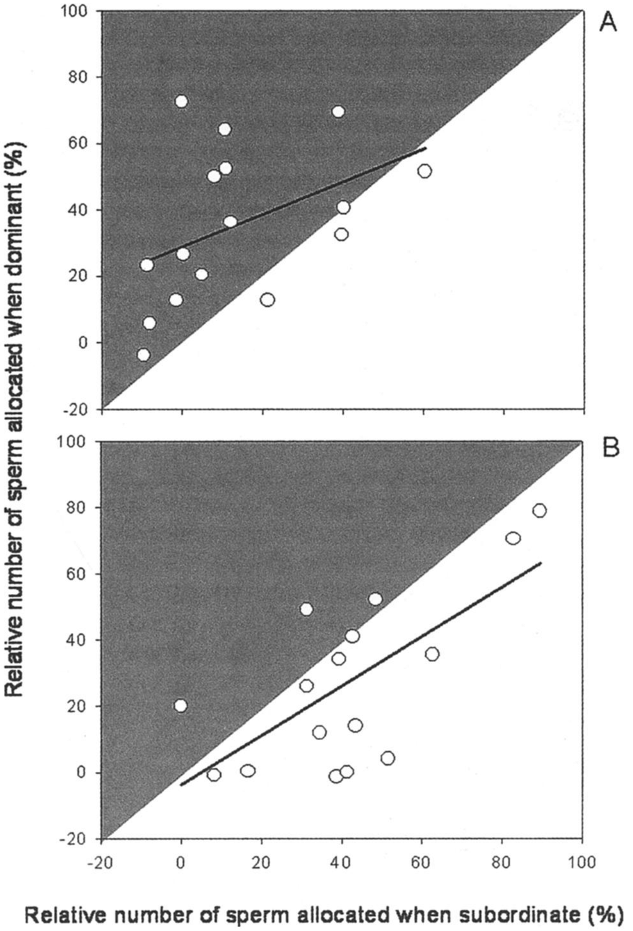

FIG. 3. Relative number of sperm males allocated to (A) females with large combs, and (B) first copulations when dominant (y-axis) and when subordinate (x-axis). Points in the gray areas indicate that males allocated greater numbers of sperm when they were dominant than the number they allocated when subordinate. Lines represent best linear fits. (A) After controlling for the effects of copulation order, males allocated significantly more sperm to females with relatively large combs when they were dominant versus when they subordinate (status, $F_{1,113} = 6.67$, $P = 0.011$; copulation order, $F_{1,113} = 44.47$, $P < 0.0001$; direction of status change, $F_{1,114} = 0.61$, $P = 0.4353$). There was also a tendency for the number of sperm that males allocated to large-combed females when dominant to be correlated to the number of sperm they allocated when subordinate (Pearson's correlation: $r = 0.44$, $n = 16$, $P = 0.087$). (B) After controlling for the effects of female comb size, males allocated significantly less sperm to first copulations when they were dominant versus when they were subordinate (status, $F_{1,113} = 3.90$, $P = 0.051$; comb rank, $F_{1,113} = 21.71$, $P < 0.0001$; direction of status change, $F_{1,114} = 0.93$, $P = 0.3370$). Furthermore, the amount of sperm that males allocated to first copulations when they were dominant was significantly correlated to when they were subordinate (Pearson's correlation: $r = 0.68$, $n = 16$, $P = 0.004$).

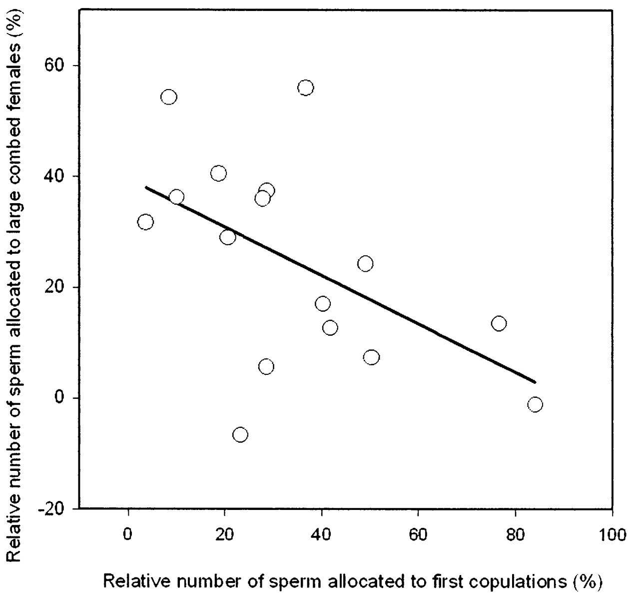

FIG. 4. The relationship between relative number of sperm allocated to large-combed females and first copulations. Datapoints represent sperm allocation by individual males averaged across when they were dominant and when they were subordinate. The relative number of sperm that males allocated to large-combed females was significantly negatively correlated to the number of sperm they allocated to first copulations (Pearson's correlation: $r = -0.53$, $n = 16$, $P = 0.035$).

with larger combs by preferentially copulating with large-combed females. Nevertheless, under natural conditions the copulation success of males is constrained by the activity of females and other males (Pizzari et al. 2002) and it is unlikely that males consistently acquire copulations with preferred females. The trade-off between allocating sperm to initial copulations and to future opportunities of higher reproductive value may consequently reflect the interaction between pre- and postcopulatory sexual selection. Genetic variance in precopulatory sexually selected traits, such as social status, will determine the probability of males obtaining subsequent copulations and also govern the risk and intensity of sperm competition that males face, which play key roles in postcopulatory sexual selection and in the evolution of sperm allocation (Parker 1982, 1990a,b, 1998; Reinhold et al. 2002; Wedell et al. 2002). Genetic covariance between pre- and postcopulatory sexually selected traits may therefore arise, and where trade-offs exist between such traits, the genetic covariance will be negative, facilitating the evolution of alternative reproductive strategies.

In the fowl, despite social status changing, there is a heritable component to dominance (Craig et al. 1965; Etches 1996). Therefore, genes coding for social status may to some extent determine the reproductive opportunities and sperm competition that males experience and, in turn, facilitate the evolution of heritable patterns of sperm allocation. However, the heritability of sperm allocation and genetic covariance with precopulatory sexually selected traits, including social status, does remain to be investigated, and it is possible that differences between males in this study were the result of maternal effects (Sheldon 2000) or past social experience. Regardless of the underlying causes it appears that patterns of sperm allocation are composed of an intrinsic element, on top of which, phenotypic plasticity enables males to respond to variation in reproductive opportunities.

In summary, where variance in reproductive success is linked to the number of sperm that males transfer to females, sperm resources are limited and reproductive opportunities frequently change, postcopulatory sexual selection is expected to lead to the evolution of phenotypically plastic patterns of sperm allocation, for which this study provides empirical evidence. These results also have important repercussions for the evolution of female reproductive strategies, because the number of sperm females obtain, and, in turn, the paternity of their eggs, is dependent on the presence of other females and the relative social competitive ability of males. Changes in social dynamics, combined with trade-offs in the number of sperm that males can allocate to copulations, may therefore lead to the evolution of alternative reproductive strategies that are phenotypically plastic and that maintain genetic variance within populations.

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LITERATURE CITED

Andersson, M. 1994. Sexual selection. Princeton Univ. Press, Princeton, NJ.

Bakst, M. R., and H. C. Cecil. 1997. Techniques for semen evaluation, semen storage, and fertility determination. 4. Sperm motility and metabolism VII. In vitro sperm-egg interaction assay utilising inner perivitelline layer from laid chicken eggs. Poultry Science Association, Savoy, IL.

Ball, M. A., and G. A. Parker. 2000. Sperm competition games: a comparison of loaded raffle models and their biological implications. J. Theor. Biol. 206:487–506.

Birkhead, T. R., and A. P. Møller. 1998. Sperm competition and sexual selection. Academic Press, London.

Birkhead, T. R., F. Fletcher, E. J. Pellatt, and A. Staples. 1995. Ejaculate quality and the success of extra-pair copulations in the zebra finch. Nature 377:422–423.

Cheng, K. M., and J. T. Burns. 1988. Dominance relationship and mating behavior of domestic cocks: a model to study mate-guarding and sperm competition in birds. Condor 90:697–704.

Clutton-Brock, T. H., S. D. Albon, R. M. Gibson, and F. E. Guiness. 1979. The logical stag: adaptive aspects of fighting in red deer (*Cervus elaphus* L.). Anim. Behav. 27:211–225.

Collias, N. E., and E. C. Collias. 1996. Social organization of a red junglefowl, *Gallus gallus*, population related to evolutionary theory. Anim. Behav. 51:1337–1354.

Cook, P. A., and N. Wedell. 1999. Non-fertile sperm delay female remating. Nature 397:486.

Cornwallis, C. K. 2004. The reproductive tactics of male and female fowl, *Gallus gallus*. Ph.D. diss. University of Sheffield, Sheffield, U.K.

Craig, J. V., L. L. Ortman, and A. M. Guhl. 1965. Genetic selection for social dominance ability in chicks. Anim. Behav. 13: 114–131.

Dewsbury, D. A. 1982. Ejaculate cost and male choice. Am. Nat. 119:601–610.

Etches, R. J. 1996. Reproduction in poultry. CAB International, Oxford, U.K.

Freyer, T., C. Cannings, and G. T. Vickers. 1998. Sperm competition. I. basic model, ESS and dynamics. J. Theor. Biol. 196: 81–100.

Garcia-Gonzalez, F., and L. W. Simmons. 2005. Sperm viability matters in insect sperm competition. Curr. Biol. 15:271–275.

Gross, M. R. 1996. Alternative reproductive strategies and tactics: diversity within sexes. Trends. Ecol. Evol. 11:92–98.

Guhl, A. M., N. E. Collias, and W. Allee. 1945. Mating behavior and the social hierarchy in small flocks of white leghorns. Physiol. Zool. 18:365–390.

Harrison, B. 1987. Den Svenska dvärghönan II. Sven. Rasf. Tidsr. 1:12–14.

Hosken, D. J., T. W. J. Garner, and P. I. Ward. 2001. Sexual conflict selects for male and female reproductive characters. Curr. Biol. 11:1–20.

Hunter, F. M., R. Harcourt, M. Wright, and L. Davis. 2000. Strategic allocation of ejaculates by male Adélie penguins. Proc. R. Soc. Lond. B 267:1541–1545.

Jennions, M. D., and M. Petrie. 1997. Variation in mate choice and mating preferences: a review of causes and consequences. Biol. Rev. Camb. Philos. Soc. 72:283–327.

Jivoff, P. 1997. The relative roles of predation and sperm competition on the duration of the post-copulatory association between the sexes in the blue crab, *Callinectes sapidus*. Behav. Ecol. Sociobiol. 40:175–185.

Johnstone, R. A., J. D. Reynolds, and J. C. Deutsch. 1996. Mutual mate choice and sex differences in choosiness. Evolution 50: 1382–1391.

LeBoeuf, B. J. 1974. Male-male competition and reproductive success in elephant seals. Am. Zool. 14:163–176.

Linley, J. R., and M. J. Hinds. 1975. Quantity of the male ejaculate influences by female unreceptivity in the fly, *Culicoides melleus*. J. Insect Physiol. 21:281–285.

MacDiarmid, A. B., and M. A. Bulter. 1999. Sperm economy and limitation in spiny lobsters. Behav. Ecol. Sociobiol. 46:14–24.

Marconato, A., and D. Y. Shapiro. 1996. Sperm allocation, sperm production and fertilisation rates in the bucktooth parrotfish. Anim. Behav. 52:971–980.

Martin, P. A., T. J. Reimers, J. R. Lodge, and P. J. Dzuik. 1974. The effect of ratios and numbers of spermatozoa mixed from two males on the proportion of offspring. J. Reprod. Fertil. 39: 251–258.

Nakatsuru, K., and D. L. Kramer. 1982. Is sperm cheap? Limited male fertility and female choice in the lemon tetra (Pisces, Characidae). Science 216:753–755.

Neff, B. D., P. Fu, and M. R. Gross. 2003. Sperm investment and alternative mating tactics in bluegill sunfish (*Lepomis macrochirus*). Behav. Ecol. 14:634–641.

Nicholls, E. H., T. Burke, and T. R. Birkhead. 2001. Ejaculate allocation by male sand martins *Riparia riparia*. Proc. R. Soc. Lond. B 268:1265–1270.

Olsson, M., T. Madsen, and R. Shine. 1997. Is sperm really so cheap? Costs of reproduction in male adders, *Vipera berus*. Proc. R. Soc. Lond. B 264:455–459.

Parker, G. A. 1970. Sperm competition and its evolutionary consequences in the insects. Biol. Rev. Camb. Philos. Soc. 45: 525–567.

———. 1982. Why are there so many tiny sperm? Sperm competition and the maintenance of two sexes. J. Theor. Biol. 96: 281–294.

———. 1983. Mate quality and mating decisions. Pp. 141–166 *in* P. Bateson, ed. Mate choice. Cambridge Univ. Press, NY.

———. 1990a. Sperm competition games: raffles and roles. Proc. R. Soc. Lond. B 242:120–126.

———. 1990b. Sperm competition games: sneaks and extra-pair copulations. Proc. R. Soc. Lond. B 242:127–133.

———. 1998. Sperm competition and the evolution of ejaculates: towards a theory base. Pp. 3–54 *in* T. R. Birkhead and A. P. Møller, ed. Sperm competition and sexual selection. Academic Press, London.

Petersen, C. W., R. R. Warner, D. Y. Shapiro, and A. Marconato. 2001. Components of fertilization success in the bluehead wrasse, *Thalassoma bifasciatum*. Behav. Ecol. 12:237–245.

Pitcher, T. E., P. O. Dunn, and L. A. Whittingham. 2005. Sperm competition and the evolution of testes size in birds. J. Evol. Biol. 18:557–567.

Pitnick, S., and T. A. Markow. 1994. Male gametic strategies: sperm size, testes size and the allocation of ejaculate among successive mates by the sperm-limited fly *Drosophila pachea* and its relatives. Am. Nat. 143:785–819.

Pizzari, T., and T. R. Birkhead. 2000. Female fowl eject sperm of subdominant males. Nature 405:787–789.

Pizzari, T., D. P. Froman, and T. R. Birkhead. 2002. Pre- and post-insemination episodes of sexual selection in the fowl. Heredity 89:112–116.

Pizzari, T., C. K. Cornwallis, H. Løvlie, S. Jakobsson, and T. R. Birkhead. 2003. Sophisticated sperm allocation in male fowl. Nature 426:70–74.

Pound, N., and M. J. G. Gage. 2004. Prudent sperm allocation in Norway rats, *Rattus norvegicus*: a mammalian model of adaptive ejaculate adjustment. Anim. Behav. 68:819–823.

Preston, B. T., I. R. Stevenson, P. J. M. and K. Wilson. 2001. Dominant rams lose out by sperm depletion: a waning success in siring counters a ram's high score in competition for ewes. Nature 409:681–682.

Quinn, G. P., and M. J. Keough. 2002. Experimental design and data analysis for biologists. Cambridge Univ. Press, Cambridge, U.K.

Ramm, S. A., G. A. Parker, and P. Stockley. 2005. Sperm competition and the evolution of male reproductive anatomy in rodents. Proc. R. Soc. Lond. B 272:949–955.

Reinhold, K., J. Kurtz, and L. Engquist. 2002. Cryptic male choice: sperm allocation strategies when female quality varies. J. Evol. Biol. 15:201–209.

Rudolfsen, G., L. Figenschou, I. Folstad, H. Tveiten, and M. Figenschou. 2006. Rapid adjustments of sperm characteristics in relation to social status. Proc. R. Soc. Lond. B 273:325–332.

Schütz, K. E., and P. Jensen. 2001. Effects of resource allocation on behavioural strategies: a comparison of red junglefowl (*Gallus gallus*) and two domesticated breeds of poultry. Ethology 107:753–765.

Setchell, J. M., and A. F. Dixson. 2002. Developmental variables and dominance rank in adolescent male mandrills (*Mandrillus sphinx*). Am. J. Primatol. 56:9–25.

Setchell, J. M., M. Charpentier, and E. J. Wickings. 2005. Sexual selection reproductive careers in mandrills (*Mandrillus sphinx*). Behav. Ecol. Sociobiol. 58:474–485.

Shapiro, D. Y., A. Marconato, and T. Yoshikawa. 1994. Sperm economy in a coral reef fish *Thalassoma bifasciatum*. Ecology 75:1334–1344.

Sheldon, B. C. 2000. Differential allocation: tests, mechanisms and implications. Trends. Ecol. Evol. 15:397–402.

Simmons, L. W. 2001. Sperm competition and its evolutionary consequences in the insects. Princeton Univ. Press, Princeton, NJ.

Smith, R. L. 1984. Sperm competition and the evolution of animal mating systems. Academic Press, Orlando, FL.

Squires, E. L., B. W. Pickett, and R. P. Amann. 1979. Effect of successive ejaculation on stallion seminal characteristics. J. Reprod. Fertil. 27:7–12.

Stockley, P., M. J. G. Gage, G. A. Parker, and A. P. Møller. 1997. Sperm competition in fish: the evolution of testis size and ejaculate characteristics. Am. Nat. 149:933–954.

Van Voorhies, W. A. 1992. Production of sperm reduces nematode lifespan. Nature 360:456–458.

Warner, R. R., D. Y. Shapiro, A. Marconato, and C. W. Peterson. 1995. Sexual conflict: males with highest mating success convey the lowest fertilization benefits to females. Proc. R. Soc. Lond. B 262:135–139.

Wedell, N., M. J. G. Gage, and G. A. Parker. 2002. Sperm competition, male prudence and sperm-limited females. Trends. Ecol. Evol. 17:313–320.

Widemo, F., and S. A. Saether. 1999. Beauty is in the eye of the beholder: causes and consequences of variation in mating preferences. Trends. Ecol. Evol. 14:26–31.

Yasui, Y. 1996. Males of a mite, *Macrocheles muscadomesticae*, estimate of a female's value on the basis of her age and reproductive status. J. Insect. Behav. 9:517–524.

Yusa, Y. 1994. Factors regulating sperm transfer in ahermaphroditic sea hare, *Aplysia parvula* Morch, 1863 (Gastropoda: Opisthobranchia). J. Exp. Mar. Biol. Ecol. 181:213–221.

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