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Male reproductive tactics in the mallard, *Anas platyrhynchos*: social and hormonal mechanisms

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Abstract It has been proposed that pair status is a proximate cue influencing the mating tactics adopted by male waterfowl. Specifically, it is thought that, compared to unpaired males, paired males are more likely to force extra-pair copulations, but are less likely to court females. Currently, it is unclear how social cues interact with physiological mechanisms affecting male mating tactics. Because of its association with both sexual and aggressive behavior, testosterone may be involved in mating tactic decisions, either in association with or independently of social cues. In this study I investigated the influence of testosterone and pair status on courtship and forced copulation behavior in captive male mallards, *Anas platyrhynchos*, by staging introduction trials between a male and female that were not paired to each other. Here, forced copulation behavior was positively associated with testosterone, the first such demonstration for any species. Surprisingly, paired males were not more likely to attempt forced copulations than unpaired males, a result that was possibly an effect of captivity. Both pair status and testosterone, however, appeared to influence male courtship behavior. Unpaired males spent more time associated with females during these trials than did paired males. Moreover, there was a positive correlation between testosterone and the time that males spent associated with females. This relationship held for both paired and unpaired males. As both forced copulation and extra-pair courtship are polygynous tactics, these results add to the growing body of evidence that testosterone is involved in polygynous behavior.

Keywords Courtship · Forced copulation · Mallard · Polygyny · Testosterone

Introduction

There are generally two insemination tactics open to males of many waterfowl species during the breeding season. A male may court a female to establish a pair bond and ensure 'consensual' copulations, or he may force copulations on a female outside of a pair bond. McKinney et al. (1983) pointed out that during the breeding season forced extra-pair copulation (FEPC) attempts in most waterfowl species are performed primarily by paired males and that unpaired males are instead more likely to court females. The authors concluded that FEPC is a secondary insemination tactic for paired males, and this hypothesis has received some support (e.g., Afton 1985; McKinney and Evarts 1998). Yet, it is unclear how a social cue such as pair status might lead to differences in mating tactics in waterfowl. There are three likely hypotheses: (1) the pairing process causes physiological changes in the male that alter the probability that he will exhibit FEPC or courtship behavior; (2) another trait independently affects both the likelihood for a male to become paired as well as his tendency to exhibit each mating tactic (courtship or FEPC), but pairing itself does not alter the probability of exhibiting FEPC or courtship behavior; and (3) physiological changes caused by pair status somehow interact with independent physiological mechanisms that underlie mating tactics such that the effects of either mechanism alone are amplified or attenuated. In this study I investigate possible social and physiological mechanisms underlying the mating tactics of male mallards, *Anas platyrhynchos*, to better understand the patterns of FEPC and courtship behavior observed in waterfowl.

FEPC behavior

Forced extra-pair copulation is obvious and pronounced in waterfowl (reviewed by McKinney and Evarts 1998)

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and appears to be the predominant polygynous tactic open to male waterfowl. Males of few species are able to attract and retain more than one mate, and then only rarely (for examples, see Sorenson 1991; Johnsgard and Carbonell 1996). No female of any waterfowl species has ever been observed to solicit extra-pair copulations during the breeding season (McKinney and Evarts 1998). Thus, for most males of most waterfowl species, FEPC appears to be the only polygynous tactic available.

One possible physiological mechanism involved in FEPC behavior involves the hormone testosterone (T). Paired males have higher breeding season T levels than do unpaired males (Hirschenhauser et al. 2000; Davis 2002), and as mentioned above, males that exhibit FEPC behavior in the wild are often paired (reviewed by McKinney and Evarts 1998). Although it is currently unknown if T is involved in sexual aggression, several lines of evidence are consistent with this hypothesis. First, T is associated with both male–male aggression (Etienne 1964; Balthazart and Stevens 1975; Schmedemann and Haase 1984) and male sexual behavior (Etienne 1964; Balthazart 1976, 1978; Balthazart and Stevens 1975, 1976) in mallards. Second, FEPC in waterfowl is not observed until the breeding season, when T reaches an annual maximum (Balthazart and Hendrick 1976). Importantly, pair status does not appear to influence breeding season T levels in mallards (Davis 2002). Therefore, if T is associated with FEPC behavior, the association would likely be independent of any effects of pair status on T.

Courtship behavior

In contrast to FEPC behavior, courtship peaks during the winter months and is observed at lower frequency during the spring breeding season (Johnsgard 1960). Courtship, courtship displays and pair formation are well described for the mallard (e.g., Weidmann 1956; Johnsgard 1961; von de Wall 1963; Weidmann and Darley 1971; Davis 1997). Courtship display groups usually comprise multiple males and a single female. Both the female and males give a series of preliminary displays, followed by a dramatic and synchronous burst of highly ritualized displays given by one or more of the males. Male displays are directed at the female (Simmons and Weidmann 1973; Davis 1997) or agonistically at other males (Davis 1997), and females can indicate preference for a given male (Weidmann 1956; Johnsgard 1961; McKinney 1992) by a display termed “inciting” (Lorenz 1953). This female display causes the preferred male or mate to attack intruding males in some species, although typically not in mallards.

There is evidence that T may be positively associated with the performance of courtship displays (Phillips and McKinney 1962; Etienne 1964; Schmedemann and Haase 1985) in mallards. Androgens are correlated with the frequency of courtship activity prior to the breeding season (Balthazart 1976; Balthazart and Hendrick 1979;

Klint et al. 1989), but it is unknown if this relationship holds for both paired and unpaired males. As noted earlier, unpaired males are more likely than paired males to court females during the breeding season (McKinney et al. 1983; but see McKinney and Stolen 1982; Sorenson 1992), but have lower T levels during the breeding season than paired males (Hirschenhauser et al. 2000; Davis 2002). Thus, it seems paradoxical for courtship behavior to be positively associated with T if courting males during the breeding season are unpaired males with low T. Alternatively, it is possible that high T males court more, but the effect is more pronounced or only holds for unpaired males. Such a synergistic interaction would be consistent with the third hypothesis outlined above. In this study I investigate the involvement of T and pair status in the expression of both FEPC and courtship behavior.

Methods

Some of the methods used in this study are reported elsewhere (Davis 2002), and therefore are described here only briefly. Details not crucial to the results of the current study have been omitted.

Study site and population

Experiments were conducted at the Hillebrand Rare Bird Ranch in Cross Plains, Wisconsin over 2 years. In the first year of the study (beginning spring 1997), I raised ducklings from eggs collected from nests on the property or purchased as day-old ducklings from Whistling Wings Mallard Farm in Hanover, Illinois. I used only first-year birds in the study to control for age and experience effects. In the second year of the study, I used offspring of the year 1 birds. In both years, birds were housed in mixed sex groups until the pairing treatment began in the fall.

In the fall of each year, all males and females were individually marked with a colored nasal marker over each nostril, as well as two colored leg bands that were patterned left to right identically to the nasal markers. This redundant method of marking facilitated identification. All birds were pinioned and housed in covered, wire mesh outdoor pens, the sizes of which were manipulated to control for bird density. Birds had unlimited access to food and to water in small wading pools, which were heated in winter to prevent freezing. The birds were exposed to ambient light and temperatures throughout the study.

Pairing treatments

Although similar, pairing treatments differed between years 1 and 2 to investigate bidirectional effects of hormones and pair formation (Davis 2002). The basic difference between years, as described below, was the manner in which pairs formed. In general, males in both years were placed in groups into their home pens in November. Females were introduced to the pens in early December, as described below.

In year 1, I randomly assigned males ($n=34$) to one of four home pens in which males either would have access to an equal number of females (two pens, each with eight or nine males) or no access to females (two pens, also with eight or nine males). Thus, in year 1 pair status was predetermined for each male depending on pen assignment, but the identity of the pairs was not predetermined. Each pen initially was visually isolated from all other pens by means of opaque barriers; after approximately 3 weeks, the barriers were removed, allowing individuals visual access to all

other pens. All males in the 'paired' pens were paired by the beginning of the breeding season. Three unpaired males were removed from the study in year 1 owing to harassment by other males; thus the final sample sizes were 17 for paired males and 14 for unpaired males.

In November of year 2, all males ($n=36$) were placed into a single pen with a limited number of females ($n=18$) introduced in December, and thus neither pair status nor pair identities were predetermined in year 2. Unpaired birds were removed from this pen at the beginning of the breeding season and 12 unpaired males were randomly selected to remain in the study as unpaired males. These 12 unpaired birds were placed in an adjacent pen, and, similarly to year 1 were physically but not visually isolated from the paired birds for the remainder of the study. In this manner, about half of the males in each year were paired by the beginning of the breeding season in spring (year 1: paired $n=17$, unpaired $n=14$; year 2: paired $n=12$, unpaired $n=12$).

In both years, birds in the pens containing females were observed, using 3-min focal animal sampling, 3–5 times per week from the time females were introduced until the spring introduction trials commenced. The main purpose of these observations was to identify pairs, but agonistic behavior, including FEPC behavior, was also recorded. The assessment of FEPC behavior is described below.

Spring introduction trials

In the spring of each year, one or two introduction trials were conducted per day. First, one or two randomly selected males were captured, and blood was sampled from each male. Then one male was placed alone in a test pen, while the second male was temporarily returned to his home pen to reduce stress effects. The test pen measured 2.5×7 m, and contained about equal parts land and pond water, which varied from 0.3 m to 0.6 m deep. After a 30-min acclimation period, a female was introduced to the test pen, and the birds were observed and videotaped from a blind for 30 min (year 1) or 20 min (year 2). Each female was already paired to another male in the study and her breeding condition (prelaying, laying or incubating) was recorded to assess any possible effect on male behavior during the trial. No female solicited copulations from the test male. Each male was tested only once, and females were randomly selected to be used in the trials with the following caveats: (1) the female could not be the mate of the test male; (2) the female was not from the same pen as the test male in year 1, so that the familiarity between the test male and female was similar for paired and unpaired males that year; and (3) females were not used in more than two trials. The test male and female were returned to their home pens immediately following the trial. If a second trial was scheduled that day, the second male was recaptured, placed into the test pen immediately after the first trial, and the second trial was conducted after the 30-min acclimation period. Trials were conducted from 31 March to 15 May 1998 in year 1, and from 29 March to 14 April 1999 in year 2. Possible effects of this sampling protocol were investigated separately and are described below.

There were several differences between years in these introduction trials. Because all FEPC behavior in year 1 occurred during the first few minutes in all trials in which the behavior occurred, trials were shortened to 20 min in year 2. More importantly, there was a difference in the source of the females used in each year. In year 1, as mentioned above, there were two pens of paired birds and two pens of unpaired birds, and the females used in the introduction trials were never from the same home pen as the test male. Birds from each pen had visual access to birds in all other pens, and so likely had some familiarity with each other. In year 2, females used in the introduction trials came from the same pen as the paired males because there was only one pen of paired birds. However, as the unpaired males in year 2 had recently been removed from that pen, their familiarity with the females was similar to that of the paired males. Thus, there likely were differences in familiarity of birds in the introduction trials between years but not within years.

Assessment of FEPC and courtship behavior

Forced copulation attempts generally follow a predictable, hierarchical pattern, and may be terminated at any step, often owing to successful resistance by the female or her mate if present. Data were recorded on each of these steps during the introduction trials: (1) chasing of the female; (2) grabbing her feathers; (3) mounting her; (4) and/or copulating with her. To minimize the influence of the female's ability to resist FEPC attempts on the categorization of males, a male was categorized as a 'Forcer' if he exhibited any of these types of behavior; otherwise, he was categorized as a 'Nonforcer'. Because chasing is a more ambiguous measure of FEPC behavior, I analyzed the data both with and without males that only chased the female.

Mallard courtship display bouts involve direct male–male competition as well as courtship (McKinney 1992), and one of the major displays, the Down-Up, is often directed at other males in the courting party (Davis 1997). This fact may account for the observation that males generally do not perform any of the major courtship displays in the absence of other males (Bossemma and Krujit 1982; Krujit et al. 1983; Brodsky et al. 1988; personal observation). Instead a lone male indicates interest in a female by remaining as close to her as possible (Krujit et al. 1983; Brodsky et al. 1988), a behavior that is sufficient to form pair bonds in the absence of displays (McKinney 1992). Therefore, to assess male courtship behavior I measured the time during the trials that a male spent within three body lengths of the female by reviewing videotapes of each of the trials. Though not strictly courtship behavior per se, this measure, called time near female (TNF), likely represents a male's interest in forming a pair bond. Time near female is reported as a proportion of the total time of the trial because the length of the trials varied between years.

Handling effects

Acute stress often leads to increased corticosterone (Cort) levels within several minutes, which can in turn quickly depress gonadal steroid hormone levels (reviewed by Sapolsky et al. 2000). Therefore, blood samples were always taken as quickly as possible, as described below. Additionally, I investigated the effects of handling on hormone levels during the spring introduction trials by closely simulating the handling and 30-min delay in behavioral assessment that occurred in the trials. To that end, blood was sampled twice, 30 min apart, from 14 randomly selected males in year 1 between 23 and 27 March 1998. As the introduction trials would be in an unfamiliar test pen, each male was placed in a different, unfamiliar pen following the first sampling, and was sampled again 30 min later. Males were returned to their home pen after the second blood sample was drawn.

Blood sampling and hormone assays

Before taking a blood sample, an assistant and I stood outside of the home pen and first visually located the randomly determined target male. We then entered the pen, quickly captured the male and drew a 2.5-ml blood sample from the jugular vein using a heparinized syringe and 22-gauge needle. I recorded both the time of day the sample was taken and the length of time it took to obtain the sample (sample duration). All males were bled between 1 and 3 h post-sunrise throughout the entire study to minimize the effects of diurnal fluctuations in T seen in this species (Balthazart 1976). The samples were centrifuged on-site for 20 min, and the plasma portions were collected and kept on ice until they could be stored in a –20°C freezer later that day.

All samples were analyzed at the UW Regional Primate Research Center Assay Laboratory for T and Cort in the summer following their collection. I extracted the samples and separated the T and Cort fractions by celite chromatography (Abraham et al. 1972) before assaying using radioimmunoassay techniques (Robinson et al. 1975; Moore 1984) with internal recoveries. Tes-

tosterone antibody was purchased from Holly Hill Farms in Hillsboro, Oregon and the Cort antibody was purchased from Endocrine Sciences Products of Calbasas Hills, California. Serial dilutions of the plasma pool ($n=7$) gave parallelism to the standards for T and Cort with no differences in slopes ($P>0.05$). Accuracy for T and Cort was 104.0% and 99.2%, respectively. Sensitivities of the assays were 5 pg/tube and 36 pg/tube for T and Cort, respectively.

I analyzed two levels of pooled samples for each hormone for the samples from year 1. The intra- and inter-assay coefficients of variation (CV) for Cort were 5.97% and 14.84%, respectively, for the low pool, and 3.19% and 10.77% for the high pool. The intra- and inter-assay CV for T were 3.50% and 7.91%, respectively, for the low pool, and 2.02% and 7.42% for the high pool. I analyzed one pooled sample for each hormone for year 2 samples. The intra- and inter-assay CV for Cort were 4.01% and 18.48%, respectively. The intra- and inter-assay CV for T were 4.94% and 9.87%, respectively.

Data analysis

To analyze relationships between T and FEPC logistic analyses were performed using forward and backward stepwise model building. The dependent variable was FEPC, categorical predictors were year, pair status, and their interaction, and continuous predictors were Julian date, timepost–sunrise that blood samples were taken, and Cort. Models reported here were those which explained the greatest amount of variance in the data. If this criterion was similar for several models, then the simplest model was selected. Univariate tests of significance under the general linear model were developed to investigate the relationship between T and TNF. In this case, the dependent variable was TNF, categorical predictors were year, pair status, FEPC behavior, female reproductive condition and their interactions and continuous predictors were Julian date, timepost–sunrise that blood samples were taken, T and Cort. Multiple regression analyses were performed using the model developed for TNF in order to obtain the squared semi-partial correlation coefficients for T level. The squared semi-partial correlation coefficient is a measure of the percent of total variance accounted for by T. Hormone and TNF data were log-transformed to normalize data for statistical analysis.

Results

Forced copulation behavior

Overall, 21 out of 55 males were categorized as Forcers. All paired Forcers in this study also forced females other than their own mates in their home pens (no false positives during the trials), whereas 5 of 19 paired Nonforcers had forced in their home pens (five false negatives). Such comparisons could not be made for unpaired males, as there were no females in those home pens. Eleven of the 21 Forcers successfully copulated during the trials, 3 males mounted without copulation, 3 grabbed the female's feathers without mounting, and 4 only chased the female. The numbers of paired and unpaired males that attempted FEPC in each year of the study are shown in Table 1. Both year and pair status were incorporated into statistical analyses of FEPC behavior reported below.

The female used in a given trial did not appear to affect the occurrence of FEPC attempts. Females in breeding condition (laying) experienced FEPC attempts in 17 of 39 trials, and non-breeding (pre-laying or incubating)

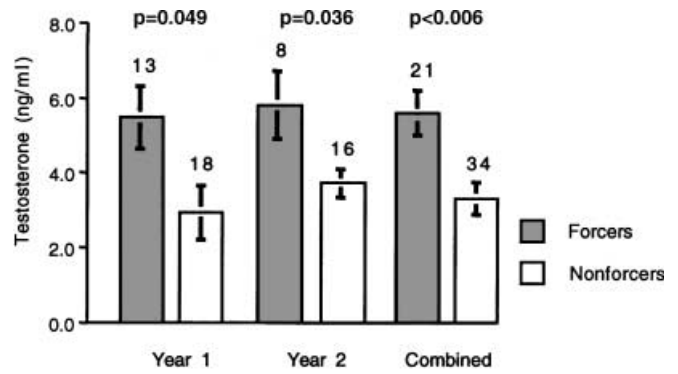


Fig. 1 Mean and SE of testosterone levels of forcing and nonforcing males. Testosterone levels of Forcers are higher than those of Nonforcers. Sample sizes are given above the bars

Table 1 Forced extra-pair copulation (FEPC) behavior by year and pair status. Y Yes, N no

	FEPC attempt? (Y/N)			
	Year 1		Year 2	
	Y	N	Y	N
Paired	6	11	4	8
Unpaired	7	7	4	8

Table 2 Logistic regression model explaining variation FEPC behavior

	Likelihood score	Wald χ^2	df (n=55)	P
Whole model	12.0		3	0.008
Univariate results				
Testosterone (T)		7.64	1	<0.006
Pair status (P)		3.49	1	0.062
T \times P		2.49	1	0.11

females experienced FEPC attempts in 4 of 16 trials; this difference was not significant ($\chi^2_1=1.66$, $P=0.20$). Males did not appear to target specific females; of the 26 females that were used in two trials each, 8 experienced no FEPC attempt, 16 experienced one FEPC attempt and only 2 experienced two FEPC attempts (Chi-square goodness of fit: $\chi^2_2=2.33$, $P>0.20$). Expected values for this last analysis were calculated from the overall probability of experiencing an FEPC attempt in a trial, where 20 FEPC attempts occurred in this subset of 52 trials. In other words, individual females were not more or less likely to experience an FEPC attempt during a trial than would be expected by chance alone.

The best model explaining FEPC behavior included T, pair status and their interaction. Of these, only T was significant (Table 2, Fig. 1). Overall, Forcers had higher T levels than Nonforcers when all data were combined and when each year was analyzed separately (year 1: Wald $\chi^2_1=3.85$, $n=31$, $P=0.049$; year 2: Wald $\chi^2_1=4.38$,

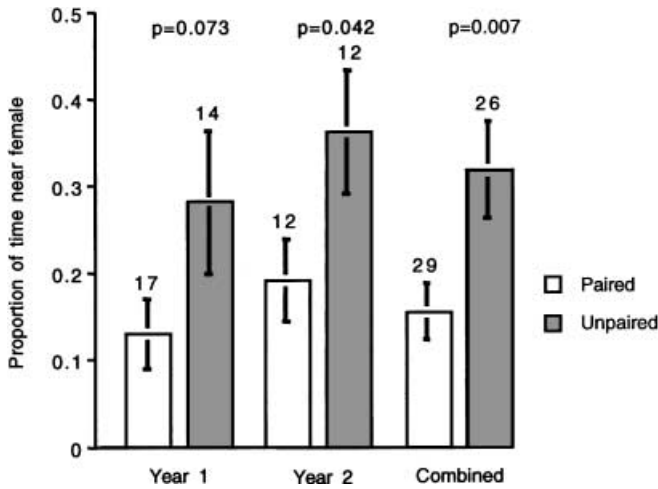


Fig. 2 Proportion of time males spent near the females during the introduction trials. Unpaired males associated with introduced females more than paired males. Sample sizes are given above the bars

$n=24$, $P=0.036$). Pair status was not significant in either year (year 1: Wald $\chi^2_1=2.79$, $n=31$, $P=0.094$; year 2: Wald $\chi^2_1=0.81$, $n=24$, $P=0.37$). The association between T and FEPC behavior was still significant when those four males that only chased the female were excluded from analysis (Wald $\chi^2_1=6.72$, $n=51$, $P=0.010$), but again, neither pair status (Wald $\chi^2_1=2.55$, $n=51$, $P=0.11$) nor the interaction term (Wald $\chi^2_1=1.68$, $n=51$, $P=0.19$) was significant.

Time near female

The mean proportion of time \pm SE during the introduction trials that an unpaired male spent within three body lengths of the female was 0.32 ± 0.011 whereas that for paired males was 0.16 ± 0.006 (Fig. 2). Forward and backward stepwise analysis yielded identical models to explain the variation in time that males associated with females (Table 3). This model was composed of three factors, each of which was significant: (1) pair status; (2) T level; and (3) Julian date. Paired males spent less time associated with females during the trials than unpaired males (Table 3, Fig. 2). This relationship was significant for year 2 ($F_{1,23}=3.30$, $P=0.042$) and not for year 1 ($F_{1,27}=2.24$, $P=0.073$). Time near female was positively correlated with T (Table 3, Fig. 3), and this relationship held for both paired ($F_{1,26}=11.2$, $P=0.002$) and unpaired males ($F_{1,23}=9.85$, $P=0.004$). Julian date was neg-

Table 4 Change in hormone levels 30 min after initial sampling. Means and confidence intervals (CI) are of back-transformed means

	Mean at t_1 (ng/ml)	95% CI	Mean at t_2 (ng/ml)	95% CI	n	P
Testosterone	1.57	(0.71, 3.45)	2.24	(1.42, 3.53)	14	0.2
Corticosterone	3.28	(2.97, 3.62)	30.7	(18.0, 52.3)	14	<0.0001

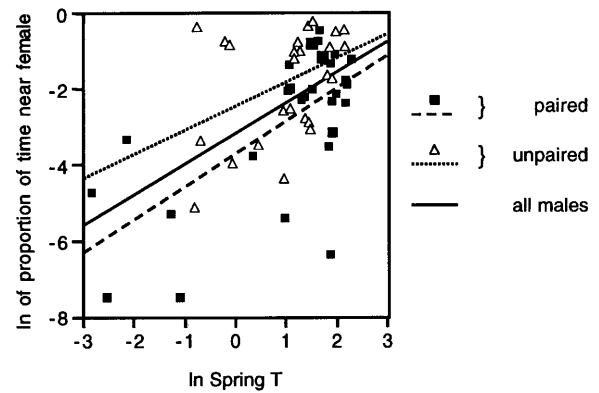


Fig. 3 Proportion of time males spent near the females during the introduction trials regressed on T-level. There is a positive correlation between T level time near female for both paired and unpaired males as well as for all males combined. The squared semi-partial correlation coefficients (analogous to r^2 values) obtained from multiple regression analyses are 0.19, 0.20 and 0.18 for paired, unpaired and all males, respectively. Data shown have undergone log transformation. See text for ANOVA details

Table 3 ANCOVA model explaining variation in time near female (courting)

	Adjusted r^2	F	df	P
Whole model	0.559	23.8	3, 51	<0.0001
Univariate results				
Pair status		8.0	1, 51	0.007
Testosterone		21.9	1, 51	<0.0001
Julian date		33.8	1, 51	<0.0001

atively correlated with the time spent associated with the female (Table 3).

Handling effects

Results reported elsewhere (Davis 2002) of nested analysis of variance with repeated measures indicated no pen effect on T in year 1 (two pens each of paired and unpaired males). Also, T levels were not significantly correlated with sampling duration in either year. Moreover, Table 4, showing the results of the March sampling, indicates that although Cort increased significantly 30 min after the initial sampling, T did not.

Discussion

Males that attempted FEPC had higher T levels than males that did not attempt FEPC, but paired males did not attempt FEPC more than unpaired males. These results are the first demonstration of a clear relationship between T and sexual coercion for any species. On the other hand, both pair status and T level were good predictors of the time that males associated with females. Unpaired males remained near females longer than paired males, and this behavior was positively correlated with T levels for both paired and unpaired males.

FEPC behavior

Testosterone was the only factor in this study that was associated with FEPC behavior. Detailed analyses of T throughout the pair formation and breeding season in these males indicated that when male pair status was determined randomly (year 1), T decreased in paired males following pair formation, but by the breeding season, the T levels of paired and unpaired males did not differ (Davis 2002). Therefore, the social milieu of pair status itself did not affect T levels during the breeding season. However, when females were able to choose from among an excess number of males (year 2), the preferred males were those who had higher breeding season T levels. Females also prefer older males (Holmberg et al. 1989), which have higher T levels than younger males (Tanabe 1992). Together, these results suggest that in the wild, paired males are more likely to have higher T levels than unpaired males. Moreover, as both FEPC behavior and pair status correlate independently with T, it is possible that the association between FEPC and pair status often observed in the wild (reviewed by McKinney and Everts 1998) is in fact a pseudocorrelation.

On the other hand, there is evidence that there may have been effects of captivity on the behavior observed in the current study. Specifically, year 2 was designed to mimic more closely conditions in the wild, where sex ratios are male biased (e.g., Humberg et al. 1978), but paired males still did not exhibit a higher FEPC frequency. This unexpected result may have been owing to aspects of the methodology used to assess FEPC behavior. For example, the staged trials may have increased the access of unpaired males to females or created motivational conflicts for the paired males who would have more control over the timing of FEPC attempts in the wild (Goodburn 1984). Therefore, although it seems clear that T was associated with FEPC, it is possible that effects of pair status on FEPC behavior were obscured in this study.

Regardless, the association between T and FEPC found here is important, in part, because of the paucity of studies on the physiological underpinnings of sexual coercion. Indeed, this question appears to have been studied previously only in humans, where complex social structure and ethical constraints on methodology

have led to inconclusive and sometimes conflicting results (e.g., Rada et al. 1976; Christiansen and Knusmann, 1987; Seim and Dwyer, 1988). For example, it may be difficult to detect a relationship between T and rape in humans because of the necessary separation between the time of the rape and the time of the study. A tight temporal association between assayed hormone level and observed FEPC behavior is one of the strengths of the current study.

Time near female

Regardless of FEPC behavior, TNF was associated with pair status, as expected based upon McKinney et al. (1983). Unpaired males spent almost twice as much time within three body lengths of the female as paired males, although this result was significant in year 2 alone and when data from both years were combined, but not in year 1 alone. As sample size was greater in year 1 than in year 2, lack of statistical power cannot reasonably account for this result in year 1. Rather, Julian date was likely a factor. Experiments were conducted over a 6 week period in year 1, but only over a 3 week period in year 2, and TNF was negatively correlated with Julian date. Therefore, as the breeding season progressed in year 1, both paired and unpaired males spent less time associated with females, which might have obscured differences between the two groups that year.

Paired males may have spent less time with females because, as with FEPC behavior, they have more conflicting interests than unpaired males. For example, paired males may be more concerned than unpaired males about returning to their home pens because of their mates. However, such conflicts between existing pair bonds and forming new ones are precisely the types of factors that may prevent paired males from courting other females in the wild.

Interestingly, TNF was also positively correlated with T level for both paired and unpaired males. Klint et al. (1989) found a positive association between the presence of Fall courtship displays and plasma dihydrotestosterone (DHT), but no significant association between display activity and T. Their study was conducted during the fall, when T is low (Balthazart and Hendrick 1976) and gonads are regressed (Johnson 1961), and this may account for the difference in results. Balthazart and Hendrick (1979) did find an association between total androgen and display activity, but it is not clear whether T, DHT or both were responsible for that result. Thus, overall, it appears that pair status affects courtship behavior of males via physiological mechanisms other than T, but that T also influences courtship activity.

T and polygyny

Extra-pair courtship (as measured here by TNF) and FEPC are types of polygynous behavior, with the latter

arguably being a more direct measure. That both were positively related to T in this study is consistent with the growing body of evidence that male T level is positively related to polygynous behavior in birds (e.g., Silverin 1980; Wingfield 1984; Ketterson et al. 1992; Chandler et al. 1994; Stoehr and Hill 2000). Moreover, in this study T was associated with potential polygynous behavior via two different behavioral strategies: extrapair courtship and forced extrapair copulations.

Conclusions

Overall, these results suggest that T is a mechanism involved in FEPC, and that the observed relationship between FEPC and pair status in the wild may be an artifact resulting from the independent association of T with both FEPC and pair status (although it is possible that other effects of pair status were not well replicated in this captive study). However, courtship decisions in mallards appear to be complex. Unpaired males associated with females more than paired males, but high T males, whether paired or unpaired, associated with females more than low T males. A link between T and polygynous behavior has been found previously in a number of avian species, but these results are the first demonstration of an association between sexual aggression and T for any species.

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