

2005

Testosterone and Group Size in Cliff Swallows: Testing the “Challenge Hypothesis” in a Colonial Bird

Linda C. Smith

Richard Stockton College of New Jersey

Samrrah A. Raouf

University of Washington

Mary Bomberger Brown

University of Nebraska-Lincoln, mbrown9@unl.edu

John C. Wingfield

University of Washington

Charles R. Brown

University of Tulsa, charles-brown@utulsa.edu

Follow this and additional works at: <https://digitalcommons.unl.edu/bioscifacpub>



Part of the [Biology Commons](#), and the [Ornithology Commons](#)

Smith, Linda C.; Raouf, Samrrah A.; Brown, Mary Bomberger; Wingfield, John C.; and Brown, Charles R., "Testosterone and Group Size in Cliff Swallows: Testing the “Challenge Hypothesis” in a Colonial Bird" (2005). *Faculty Publications in the Biological Sciences*. 470.

<https://digitalcommons.unl.edu/bioscifacpub/470>

This Article is brought to you for free and open access by the Papers in the Biological Sciences at DigitalCommons@University of Nebraska - Lincoln. It has been accepted for inclusion in Faculty Publications in the Biological Sciences by an authorized administrator of DigitalCommons@University of Nebraska - Lincoln.

Published in *Hormones and Behavior* 47:1 (2005), pp. 76–82; doi:10.1016/j.yhbeh.2004.08.012
Copyright © 2004 Elsevier Inc. Used by permission.
Submitted May 17, 2004; revised July 25, 2004; accepted August 18, 2004; published online October 27, 2004.

Testosterone and Group Size in Cliff Swallows: Testing the “Challenge Hypothesis” in a Colonial Bird

Linda C. Smith,¹ Samrrah A. Raouf,^{2*} Mary Bomberger Brown,³

John C. Wingfield,² and Charles R. Brown³

1. The Richard Stockton College of New Jersey, Pomona, New Jersey, USA
2. Department of Zoology, University of Washington, Seattle, Washington, USA
3. Department of Biological Sciences, University of Tulsa, Tulsa, Oklahoma, USA

Corresponding author – C. R. Brown, email charles-brown@utulsa.edu

* Current address: Treetops Porthmadog, Gwynedd, LL49 9UG, United Kingdom

Abstract

The “challenge hypothesis” states that increases in testosterone levels of male animals during the breeding season are directly related to the extent of intrasexual competition for resources or mates that they experience. Although often tested in territorial species, the challenge hypothesis has not been evaluated for colonial animals that live in groups of different sizes and that thus experience different intensities of intrasexual competition. We measured circulating testosterone levels of male and female cliff swallows (*Petrochelidon pyrrhonota*) in southwestern Nebraska, where these birds nest in colonies of widely different sizes. Males had significantly higher testosterone levels than females, as expected. For males especially, there was a seasonal rise in testosterone levels early in the nesting cycle, corresponding to the period when birds were establishing nest ownership and egg laying, and then a fall as they switched to parental duties. Testosterone levels varied significantly with colony size; for both sexes, birds in larger colonies had higher levels of testosterone than those in smaller colonies when controlling for date. Age and body mass were not related to testosterone levels.

Higher levels of testosterone for birds of both sexes in larger colonies probably reflect greater competition for matings, often extra pair, in the more social nesting situations. The results support the predictions of the challenge hypothesis.

Keywords: coloniality, ectoparasitism, reproduction, sexual selection, social behavior

Introduction

Many aspects of reproductive physiology, morphology, and behavior in male vertebrates are influenced or controlled by the steroid hormone testosterone. Seasonal testosterone profiles in birds of various species have shown almost nondetectable levels during the nonbreeding season, with an increase to a breeding-season baseline concurrent with annual gonadal development and spermatogenesis, the onset of reproductive behavior such as territoriality, and the expression of secondary sexual characteristics (Wingfield et al., 1990, 2000). Many species, however, show increases in testosterone levels above the breeding baseline. Although this increase generally reaches a maximum physiological limit, the rise in testosterone may be short or long lasting and small or large in magnitude, and varies considerably among species.

Much of the variation in seasonal testosterone fluctuation in birds appears related to patterns of parental care and the intensity of competition among males for access to females or the resources females require. Because high levels of testosterone appear to inhibit full development of parental behavior (incubation, feeding of young), in those species in which male parental care is important, testosterone levels often drop to near or below baseline when eggs or young are present. Male-male aggression and (in some cases) mate-guarding of females, however, appear to be enhanced by high levels of testosterone, and when these activities are ongoing, many species show testosterone levels well above breeding baseline. These observations were incorporated into the "challenge hypothesis" that predicts patterns of seasonal testosterone variation in relation to mating system, pattern of parental care, and levels of intrasexual social competition (Wingfield et al., 1990). Across different bird species (over 80 so far tested; Hirschenhauser et al., 2003; Wingfield et al., 2000), the challenge hypothesis has held up well, with polygynous species in which males compete more for females and provide less parental care showing higher and longer-lasting rises in testosterone levels than in more monogamous species.

Most tests of the challenge hypothesis have been done with interspecific comparisons or among territorial species that occur in relatively low population density (Wingfield et al., 2000; cf. Ros et al., 2002). Colonial animals, on the other hand, often show extremely high local population density, and thus competition among males for females (including for extra-pair fertilizations; Birkhead and Møller, 1992; Brown and Brown, 2003; Gladstone, 1979) is potentially much greater for them than in territorial species. In addition, most colonial bird species show extensive variation in colony size, with the smallest and largest colonies within a population frequently differing in size by several orders of magnitude (Brown et al., 1990). Thus, colonially breeding birds present a natural framework for evaluating how seasonal variation in testosterone levels responds to different degrees of social competition.

In this study, we examine how levels of circulating testosterone vary among cliff swallows (*Petrochelidon pyrrhonota*) occupying colonies of different sizes. Cliff swallows show extensive variation in colony size, with birds in our study area nesting in colonies ranging from 2 to approximately 3700 nests. We used this variation to test the challenge hypothesis, asking whether males in large colonies show higher testosterone concentrations than those in smaller colonies. There is more competition for matings in the larger cliff swallow colonies, with the frequency of extra-pair copulation increasing with colony size (Brown and Brown, 1996, 2003). There may also be greater initial competition for nesting sites early in the season at larger colonies before birds have established ownership of nests or nest fragments.

Besides males, we also evaluated the variation in circulating testosterone levels in female cliff swallows. It is now known that female birds of many species routinely show detectable levels of testosterone at certain times of the nesting cycle, and some evidence indicates that testosterone in females is also associated with periods of intrasexual competition (Wingfield, 1994; Wingfield et al., 2000).

Methods

Study animal and study site

Cliff swallows are highly colonial, socially monogamous passerines that breed throughout most of western North America (Brown and Brown, 1995). They build gourd-shaped mud nests and attach them to the vertical faces of cliff walls, rock outcrops, or artificial sites such as the eaves of buildings or bridges. Their nests tend to be stacked closely together, often sharing walls, and breeding at a site is highly synchronous (Brown and Brown, 1996). Cliff swallows are migratory, wintering in southern South America, and have a relatively short breeding season in North America. They begin to arrive at our study site in late April or early May and depart by late July. Both sexes share roughly equally in building the nest, incubating, and feeding nestlings. They generally raise only one brood of (most commonly) three or four young. Cliff swallows are associated with a variety of ectoparasites, endoparasites, and viruses throughout their range (Brown and Brown, 1995; Brown et al., 2001; Monath et al., 1980; Scott et al., 1984). The ectoparasites, in particular the hematophagous swallow bug (Hemiptera: Cimicidae: *Oeciacus vicarius*), are responsible for much of the nestling mortality and nest failures that occur in our study area (Brown and Brown, 1986, 1996).

Our study site is centered at the Cedar Point Biological Station (41°13'N, 101°39'W) near Ogallala, in Keith County, along the North and South Platte Rivers, and also includes portions of Deuel, Garden, and Lincoln counties, southwestern Nebraska. We have studied cliff swallows there since 1982. There are approximately 160 cliff swallow colony sites in our 150 × 50 km study area, with about a third of these not used in a given year. Colony size varies widely; in our study area, it ranges from 2 to 3700 nests, with some birds nesting solitarily. Over a 20-year period, mean (\pm SE) colony size ($n = 1363$) was 363 (± 16) nests. Each colony site tends to be separated from the next nearest by 1–10 km but in a few cases

by ≥ 20 km. In our study area, the birds nest on both natural cliff faces and artificial structures such as bridges, buildings, and highway culverts. The study site is described in detail by Brown and Brown (1996).

Field and analysis methods

Testosterone levels were measured for cliff swallows during the 2000 and 2001 breeding seasons. As part of a long-term mark-recapture project, we mist-netted thousands of adult cliff swallows at the study colonies at intervals throughout each nesting season. Nets were placed either across one end of a culvert containing nests, with birds caught as they exited the colony, or were dropped from the top of a bridge such that the swallows would be caught as they flushed from their nests (Brown, 1998). All birds were banded with U.S. Fish and Wildlife Service bands upon initial capture, and weighed and sexed on each capture. Males were determined by the presence of a cloacal protuberance and/or absence of a brood patch; females always lacked protuberances and generally had brood patches. Blood samples were taken from a subset of the adults caught; we tried to select this subset as randomly as possible, with the principal criterion for inclusion being knowing exactly when a given bird first hit the net. Most of the birds for whom testosterone levels were measured were bled within 3 min of initial contact with a net as part of a study on corticosterone (C. Brown et al., unpublished data). However, some individuals were sampled for testosterone up to 11 min after initial contact with a net. These birds were included in this study, as the interval between capture and blood sampling had no significant effect on testosterone levels in cliff swallows (see Results). Blood samples were taken with heparinized capillary tubes after brachial vein puncture with a 26-gauge needle or lancet. Samples were initially stored on ice in a cooler, then transported back to a laboratory at the Cedar Point Biological Station where plasma was separated by microhematocrit centrifugation and harvested with a Hamilton syringe. Plasma samples were stored at -20°C until analysis. Testosterone concentration was measured by radioimmunoassay as described by Wingfield et al. (1992). Blood samples were not collected from all of our study colonies; for this work, we chose a subset of the accessible colony sites that maximized the range in colony sizes studied. All animal procedures were approved by the Institutional Animal Care and Use Committees of the University of Tulsa and the University of Nebraska–Lincoln under protocols TU-00-05 and 99-05-029, respectively.

At the time each colony was sampled, we designated the colony's stage as either early, mid, or late. The early stage was when over half of the birds in a colony were nest building, mid was when over half of the colony was incubating, and late was when over half of the colony was feeding nestlings. These periods were designated separately for each colony, and different colonies active at the same time may have been at different stages depending on when they started. These three stages, however, corresponded broadly to the early, middle, and latter parts of the breeding season. The exact nesting stage was not known for any of the adult birds sampled, as none were caught at their nests. However, the high degree of synchrony within cliff swallow colonies (Brown and Brown, 1996) ensured that most individuals could be classified accurately based on the status of the colony as a whole.

We designated age of adult birds in two ways. For those first banded as nestlings or juveniles in a past year, we knew their exact ages. The oldest bird of exact age in our sample

was 7 years old. For birds first banded as unknown-aged adults, we designated the minimum age as the number of years elapsed since banding. Thus, the minimum age of a bird first banded in the year of hormone sampling was 1, a bird first banded the previous year was 2, etc. This gave us a relative measure of a bird's age, with some individuals having a minimum age of up to 10 years. We believe that minimum age was a relatively accurate indicator of exact age, as many of the unbanded adults in our study area in a given year are in all likelihood yearlings, given the large number of adult birds (up to 10,000) banded each season.

One potentially confounding factor in analyzing testosterone levels is the presence of transient birds at some colonies. Large numbers of transients pass through some colonies (Brown and Brown, 2004a), many of them apparent nonbreeders. Transients, for a variety of reasons, might have higher or lower testosterone levels than resident birds at a site. To investigate whether this might be the case, we first identified the colonies in 2000 and 2001 where transients were suspected to occur. This was done by fitting a series of within-season survival and recapture models to the daily capture data for all birds at each site each year (Brown and Brown, 2004b). Some models contained an effect of transients while others did not (see Pradel et al., 1997). Those sites where the models with an effect of transients were the best fit (Brown and Brown, 2004b) were the ones where transients might have confounded measurements of testosterone levels. For those five colonies, we examined the seasonal capture histories of all birds whose testosterone was measured. Those caught only once at a colony during the season were considered transients and those caught at least twice were residents. While this test was conservative because some of the individuals classified as transients may have in fact been residents who just never happened to get caught again, it did allow us to determine if there were any obvious differences among known colony residents and birds that might not have lived at the site.

We repeated the ANCOVA used for the entire data set (see Results), but with residence status added, for the 239 birds designated as residents or transients from the five colonies combined. There was no significant effect of residence status on testosterone levels independent of the other variables ($P = 0.88$). Thus, because capture history at a site was not related to testosterone level, we combined data from all individuals caught at a colony and did not try to separately designate residents and transients in subsequent analyses.

In 2000 and 2001, three and four colonies, respectively, were fumigated to remove ectoparasites as part of other work (e.g., Brown and Brown, 2004a). Nests within these colonies were sprayed with a dilute solution of an insecticide, Dibrom, that was highly effective in killing swallow bugs but that had no detectable effects on the birds occupying the nests (Brown and Brown, 1996, 2004a). Nests were fumigated weekly to remove any bugs brought into the colony by transient birds. Each colony was sprayed in its entirety.

Cliff swallow colony size was defined as the maximum number of nests at a site to have contained one or more eggs. Active nests were counted at some sites by periodically checking the nest contents with a dental mirror and flashlight, whereas the colony size at other sites was estimated by counting the number of intact nests in active sections of the colony. Full details on these methods of determining colony sizes are given in Brown and Brown (1996).

We investigated the effect of colony size on testosterone levels by using a paired-colony analysis that also controlled for date-related variation in testosterone levels (see Results). Ideally, one would sample birds in all colony sizes at the same time (as we did, for example, for testis size in a mortality event; Brown and Brown, 2003). However, since this was not logistically possible when capturing and blood sampling birds, we designated for each single day or two consecutive-day interval for which two colonies were sampled during the same season, the smaller and larger colony of the pair. Comparing testosterone levels between the members of each pair allowed us to determine if colony size in a broad sense affected testosterone levels independent of date. The colonies in each pair were ones that differed in size by an average (\pm SE) 749.8 (\pm 195.8) nests, with the difference within a pair ranging from 35 to 3310 nests. If, in rare cases, we had three colonies sampled on a single day or 2-day interval, we paired the two that differed the most in size.

The set of testosterone values obtained in this study was not normally distributed, and no transformations successfully normalized it. We thus used nonparametric tests for most of our statistical analyses. In order to assess the separate effects of several independent variables on testosterone levels, we ranked the testosterone values and used the ranked values (Montgomery, 2001) in a parametric ANCOVA using Proc GLM in SAS (SAS Institute, 1990). Mean testosterone levels (\pm 1 SE) are reported.

Results

Using all birds of both sexes from all colonies ($N = 917$ individuals), we first identified with an ANCOVA the independent variables that potentially affected testosterone levels in cliff swallows. Fumigation status, nesting stage, and the interval between capture and sampling had no significant effect on testosterone levels ($P \geq 0.16$ for all). We therefore do not consider these variables further, and we combined fumigated and nonfumigated colonies for subsequent analyses. Year ($F_{1, 916} = 109.7$, $P < 0.0001$), sex ($F_{1, 916} = 113.9$, $P < 0.0001$), colony size ($F_{1, 916} = 6.72$, $P = 0.0097$), and date of sampling ($F_{1, 916} = 69.8$, $P < 0.0001$) each had a significant effect on testosterone levels independent of the others. These variables remained significant at the $P \leq 0.05$ level after applying a sequential Bonferroni correction (Rice, 1989) for multiple comparisons.

For the 2 years 2000–2001, testosterone levels of (especially) males and (to a lesser extent) females across the season showed a seasonal rise and fall (Fig. 1) consistent with the pattern seen in other songbirds. The significant effect of year on testosterone levels was brought about partly by greater testosterone concentrations among birds sampled in 2001: average testosterone levels for males were 0.84 (\pm 0.07) ng/ml and 1.43 (\pm 0.14) ng/ml in 2000 ($N = 365$) and 2001 ($N = 93$), respectively; for females, 0.27 (\pm 0.03) and 0.66 ng/ml in 2000 ($N = 380$) and 2001 ($N = 80$), respectively. Testosterone levels ranged from 0.00 to 12.17 ng/ml for males and 0.00 to 2.79 ng/ml for females in 2000; and from 0.16 to 6.23 ng/ml for males and 0.13 to 6.93 ng/ml for females in 2001. The 2 years also differed somewhat in how testosterone varied with date (Fig. 1). In 2000, testosterone in males showed a narrow spike in late May and early June, and then a fall beginning in mid-June and throughout the rest of the season. In 2001, the pattern for males was more variable with some evidence of a wider (and later) period of peak levels (Fig. 1).

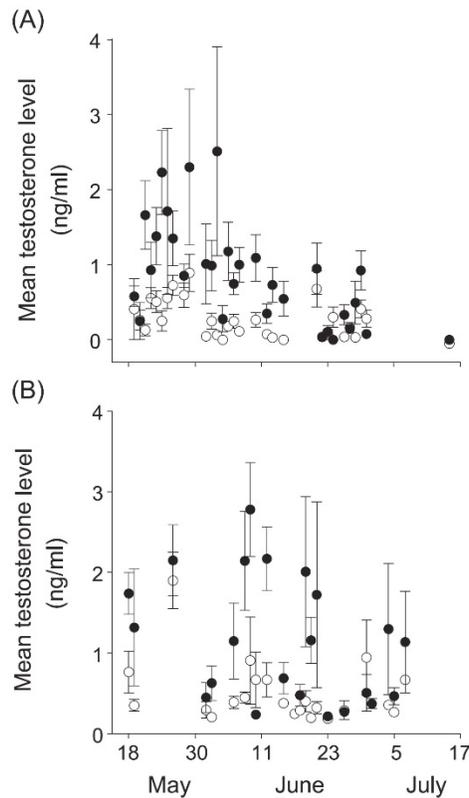


Figure 1. Mean (± 1 SE) circulating testosterone level (ng/ml) of male (●) and female (○) cliff swallows in relation to date during the (A) 2000 and (B) 2001 nesting seasons. Sample size (number of birds sampled) varied from 1 to 28 for each date. Testosterone levels varied significantly with date (see text).

For each sex, in 15 of 21 colony pairs sampled on the same day or within 2 days of each other, testosterone levels were higher in the larger colony of the pair (Fig. 2). Both sexes showed the same pattern, although the direction of the difference was not the same for each pair for males and females. The preponderance of cases with birds of the larger colony showing higher testosterone levels was significant (Fig. 2). The greatest differences between the smaller and larger colony of a pair tended to occur in early June (Fig. 2) for both sexes, the same time when testosterone levels in general peaked (Fig. 1). This analysis did not attempt to account for the magnitude of the difference in colony size between members of each pair, due to the small number of pairs relative to the wide seasonal variation in testosterone levels. Data for birds at some colonies included in the ANCOVA analysis (above) were not used in the colony-pair analysis (Fig. 2) because in some cases only one site was sampled on a single day or during a given 2-day period and thus could not be paired with a smaller or larger colony. However, the significant overall effect of colony size using all data in the ANCOVA statistically controlled for effects of date.

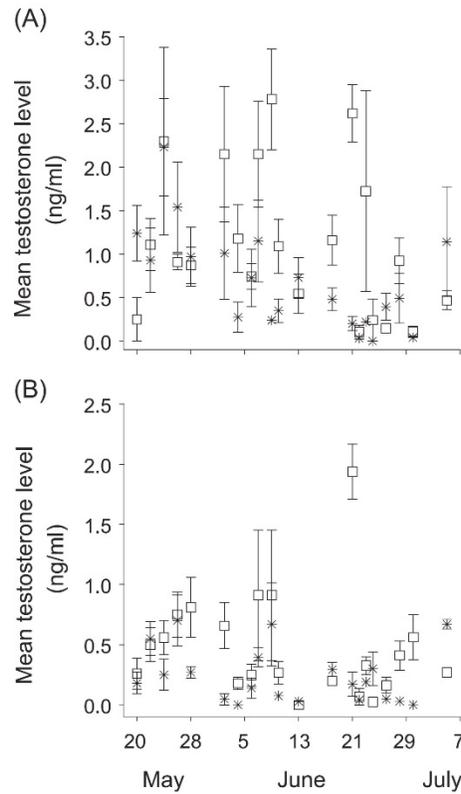


Figure 2. Mean (± 1 SE) circulating testosterone level (ng/ml) of (A) male and (B) female cliff swallows at pairs of colonies sampled on the same date (or on 2 consecutive days), in which one was larger (\square) than the other (*). Pairs sampled over 2 days are graphed both on the same date. For both males and females, in 15 of 21 colony pairs, testosterone levels were higher at the larger colony (binomial test, $P = 0.039$ for each sex).

For the subset of birds for which exact age was known, we repeated the ANCOVA and included the variable exact age. There was no significant effect of exact age on testosterone level independent of the other variables ($P > 0.09$). In similar analyses, we found no significant effect of minimum age, or of body mass during the early or late stages ($P \geq 0.71$ for all), on testosterone levels. Body mass during the mid-stage had no significant effect on testosterone levels at the $P \leq 0.05$ level when the Bonferroni correction for multiple comparisons was applied.

Discussion

Our results indicate that variation in testosterone levels of male and female cliff swallows is consistent with predictions of the challenge hypothesis, given the patterns of parental care in the species, and the study provides the first intraspecific test of the hypothesis in a colonially breeding species using groups of different sizes. Testosterone levels in each sex rise in the early part of the nesting season and then decline as both males and females

switch to parental duties (incubation, feeding of young). This is the typical pattern seen in monogamous species in which both sexes contribute extensive parental care (Wingfield et al., 1990, 2000). Interestingly, in cliff swallows, testosterone concentrations seemed determined more by broad seasonality (i.e., date) than by the specific nesting stage at a given colony site, although this might have also reflected in part our not knowing the exact nesting stage for each individual. The novel result of our study is the increased testosterone levels of both sexes in larger colonies independent of the date-related variation.

Why might testosterone levels increase with colony size in cliff swallows? The challenge hypothesis posits that increased levels of testosterone are an adaptive hormonal response to increased rates of intrasexual competition for resources and/or potential mates (Wingfield et al., 1990, 2000). While cliff swallows are not territorial (aside from defense of their own nest), there is competition among birds for nest sites within a colony, especially for intact existing nests or nest fragments that are relatively free of ectoparasites (Brown and Brown, 1996). This sort of competition occurs early in the nesting season as birds arrive at a colony site, and there is some evidence that males take a greater role than females in establishing initial ownership of nest sites (Meek and Barclay, 1996; C. and M. Brown, unpublished data). The increased levels of testosterone in both sexes among residents of the larger colonies suggest that competition for nest sites is more intense in larger cliff swallow colonies.

There is better evidence for increased competition for access to mates in the larger colonies. The incidence of extra-pair copulation attempts per capita increases with cliff swallow colony size (Brown and Brown, 1996), with females apparently often actively soliciting extra-pair matings from other resident males and from males not resident in a colony. The increased levels of testosterone, especially in males, may have been selected for by the greater mating opportunities in larger colonies, analogous to situations in normally monogamous species in which high testosterone levels are associated with shifts to polygyny (Silverin and Wingfield, 1982; Wingfield, 1984).

Male cliff swallows that perennially nest in larger colonies have larger testes (Brown and Brown, 2003). This is thought to reflect directional selection for the greater sperm-production capabilities of bigger testes, in response to the greater levels of sperm competition in large colonies. Because males must frequently copulate with their own mate as a defense against extra-pair fertilizations (male cliff swallows do not mate-guard, instead cooperating with their mate to guard the nest), there is a premium on being able to produce large volumes of sperm in this species (Brown and Brown, 2003). Thus, the increased levels of circulating testosterone in males in larger colonies could reflect only their larger testes, not any direct response to social environment or mating opportunities. Determining whether this is the case would require knowing the relationship between testis size and circulating testosterone levels in breeding individuals, which has not been studied in any songbird to our knowledge. However, that females also show higher levels of testosterone in response to colony size, and that there was no age effect on testosterone levels despite 1- and 2-year old males having smaller testes (Brown and Brown, 2003), seems to indicate that variation in circulating levels of testosterone among breeding cliff swallows does not result from variation in testis size and that testis size alone may not fully account for the pattern in males.

Increasing evidence indicates that female birds show measurable levels of testosterone at certain times of the year (Wingfield, 1994; Wingfield et al., 2000). As with males, testosterone production by females across species appears to correlate with periods in the nesting cycle during which intrasexual competition is greatest. In cliff swallows, females show relatively high levels of testosterone; in 2001, the maximum level recorded for females was slightly higher than that for males! There was less of a seasonal peak for females than for males (Fig. 1), but the colony-size effect was as strong statistically for females as for males (Fig. 2). This suggests that females also respond hormonally to apparently greater intrasexual competition in the larger colonies. The nature of any female-female competition in cliff swallows is unclear, but it may be related to extra-pair mating. Females often seem to solicit extra-pair copulations, primarily when gathering mud for their nests away from the colony sites. Females may compete for high-quality extrapair partners, although the dynamics of extra-pair mating strategies in cliff swallows have not been studied. These results for cliff swallows do seem consistent with comparative studies showing less difference in maximum testosterone levels between males and females in socially monogamous bird species with little sexual dimorphism (Wingfield et al., 2000).

We found higher average testosterone concentrations in both males and females in 2001 than in 2000. The precise reasons for this are unclear, as the effect held when other variables (e.g., colony size) were controlled. However, because of colder and wetter weather in the 2001 season, there was greater between-colony nesting asynchrony that year. The consequence is that the timing of the peak period of testosterone levels varied more among colonies, and this may have been reflected in our sampling more birds near their peak that season than in 2000, when nesting across sites was more synchronous.

The increased levels of testosterone in residents of larger colonies may not be without cost. Some studies suggest that high concentrations of testosterone have a suppressive effect on the immune system (Folstad and Karter, 1992; Hillgarth and Wingfield, 1997; Slater and Schreck, 1993; Peters, 2000; Verhulst et al., 1999; Zuk et al., 1995), although this is not the case in all taxa (Dunlap and Schall, 1995; Hasselquist et al., 1999; Ros et al., 1997). If the high testosterone levels in cliff swallows in the larger colonies have any immunosuppressive effect, this would be especially deleterious because the birds at those sites are exposed to more blood-feeding parasites and viral pathogens (Brown and Brown, 1986, 1996, 2002, 2004a; Brown et al., 2001). We are in the process of evaluating this potential cost by estimating long-term survival of cliff swallows with different testosterone levels.

Acknowledgments – Principal contributions of the different authors were as follows: LCS, field work; SAR, field work, laboratory assays; MBB, field work, data management and analysis; JCW, laboratory direction; CRB, field work, data analysis, project direction. We thank Scott Aldridge, Kim Cornett, Jennifer Klaus, Jennifer Malfait, and Mike Shanahan for field assistance; Lynn Erckmann for laboratory assistance; the School of Biological Sciences at the University of Nebraska–Lincoln for use of the facilities at the Cedar Point Biological Station; the R. Clary, D. Knight, and L. Soper families and the Union Pacific Railroad for access to land; and the National Science Foundation (DEB-0075199, IBN-9974733) for financial support.

References

- Birkhead, T. R., Møller, A. P., 1992. Sperm Competition in Birds. Academic Press, London.
- Brown, C. R., 1998. Swallow Summer. University of Nebraska Press, Lincoln, Nebraska.
- Brown, C. R., Brown, M. B., 1986. Ectoparasitism as a cost of coloniality in cliff swallows (*Hirundo pyrrhonota*). Ecology 67, 1206–1218.
- Brown, C. R., Brown, M. B., 1995. Cliff swallow (*Hirundo pyrrhonota*). In: Poole, A., Gill, F. (Eds.), Birds of North America. Academy of Natural Sciences, Philadelphia, and American Ornithologists' Union, Washington, D.C., number 149.
- Brown, C. R., Brown, M. B., 1996. Coloniality in the Cliff Swallow: The Effect of Group Size on Social Behavior. University of Chicago Press, Chicago.
- Brown, C. R., Brown, M. B., 2002. Spleen volume varies with colony size and parasite load in a colonial bird. Proc. R. Soc. Lond., Ser. B 269, 1367–1373.
- Brown, C. R., Brown, M. B., 2003. Testis size increases with colony size in cliff swallows. Behav. Ecol. 14, 569–575.
- Brown, C. R., Brown, M. B., 2004a. Empirical measurement of parasite transmission between groups in a colonial bird. Ecology 85, 1619–1626.
- Brown, C. R., Brown, M. B., 2004b. Group size and ectoparasitism affect daily survival probability in a colonial bird. Behav. Ecol. Sociobiol. 56, 498–511.
- Brown, C. R., Stutchbury, B. J., Walsh, P. D., 1990. Choice of colony size in birds. Trends Ecol. Evol. 5, 398–403.
- Brown, C. R., Komar, N., Quick, S. B., Sethi, R. A., Panella, N. A., Brown, M. B., Pfeffer, M., 2001. Arbovirus infection increases with group size. Proc. R. Soc. Lond., Ser. B 268, 1833–1840.
- Dunlap, K. D., Schall, J. J., 1995. Hormonal alterations and reproductive inhibition in male fence lizards (*Sceloporus occidentalis*) infected with the malarial parasite *Plasmodium mexicanum*. Physiol. Zool. 68, 608–621.
- Folstad, I., Karter, A. J., 1992. Parasites, bright males, and the immunocompetence handicap. Am. Nat. 139, 603–622.
- Gladstone, D. E., 1979. Promiscuity in monogamous colonial birds. Am. Nat. 114, 545–557.
- Hasselquist, D., Marsh, J. A., Sherman, P. W., Wingfield, J. C., 1999. Is avian humoral immunocompetence suppressed by testosterone? Behav. Ecol. Sociobiol. 45, 167–175.
- Hillgarth, N., Wingfield, J. C., 1997. Parasite-mediated sexual selection: endocrine aspects. In: Clayton, D. H., Moore, J. (Eds.), Host-Parasite Evolution: General Principles and Avian Models. Oxford Univ. Press, Oxford, pp. 78–104.
- Hirschenhauser, K., Winkler, H., Oliveira, R., 2003. Comparative analysis of male androgen responsiveness to social environment in birds: the effects of mating system and paternal incubation. Horm. Behav. 43, 508–519.
- Meek, S. B., Barclay, R. M. R., 1996. Settlement patterns and nest-site selection of cliff swallows, *Hirundo pyrrhonota*: males prefer to clump but females settle randomly. Can. J. Zool. 74, 1394–1401.
- Monath, T. P., Lazuick, J. S., Cropp, C. B., Rush, W. A., Calisher, C. H., Kinney, R. M., Trent, D. W., Kemp, G. E., Bowen, G. S., Franc, D. B., 1980. Recovery of Tonate virus ("Bijou Bridge" strain), a member of the Venezuelan equine encephalomyelitis virus complex, from cliff swallow nest bugs (*Oeciacus vicarius*) and nestling birds in North America. Am. J. Trop. Med. Hyg. 29, 969–983.
- Montgomery, D. C., 2001. Design and Analysis of Experiments, 5th ed. Wiley, New York.

- Peters, A., 2000. Testosterone treatment is immunosuppressive in superb fairy-wrens, yet free-living males with high testosterone are more immunocompetent. *Proc. R. Soc. Lond., Ser. B* 267, 883–889.
- Pradel, R., Hines, J. E., Lebreton, J. D., Nichols, J. D., 1997. Capture-recapture survival models taking account of transients. *Biometrics* 53, 60–72.
- Rice, W. R., 1989. Analyzing tables of statistical tests. *Evolution* 43, 223–225.
- Ros, A. F. H., Groothuis, T. G. G., Apanius, V., 1997. The relation among gonadal steroids, immunocompetence, body mass, and behavior in young black-headed gulls (*Larus ridibundus*). *Am. Nat.* 150, 201–219.
- Ros, A. F. H., Dieleman, S. J., Groothuis, T. G. G., 2002. Social stimuli, testosterone, and aggression in gull chicks: support for the challenge hypothesis. *Horm. Behav.* 41, 334–342.
- SAS Institute, 1990. *SAS/STAT User's Guide, Version 6*. SAS Institute, Inc., Cary, NC.
- Scott, T. W., Bowen, G. S., Monath, T. P., 1984. A field study of the effects of Fort Morgan virus, an arbovirus transmitted by swallow bugs, on the reproductive success of cliff swallows and symbiotic house sparrows in Morgan County, Colorado, 1976. *Am. J. Trop. Med. Hyg.* 33, 981–991.
- Silverin, B., Wingfield, J. C., 1982. Patterns of breeding behaviour and plasma levels of hormones in a free-living population of pied flycatchers *Ficedula hypoleuca*. *J. Zool. (Lond.)* 198, 117–129.
- Slater, C. H., Schreck, C. B., 1993. Testosterone alters the immune response of chinook salmon, *Oncorhynchus tshawytscha*. *Gen. Comp. Endocrinol.* 89, 291–298.
- Verhulst, S., Dieleman, S. J., Parmentier, H. K., 1999. A tradeoff between immunocompetence and sexual ornamentation in domestic fowl. *Proc. Natl. Acad. Sci. U.S.A.* 96, 4478–4481.
- Wingfield, J. C., 1984. Androgens and mating systems: testosterone-induced polygyny in normally monogamous birds. *Auk* 101, 665–671.
- Wingfield, J. C., 1994. Hormone-behavior interactions and mating systems in male and female birds. In: Short, R. V., Balaban, E. (Eds.), *The Differences between the Sexes*. Cambridge Univ. Press, Cambridge, pp. 303–330.
- Wingfield, J. C., Hegner, R. E., Dufty Jr., A. M., Ball, G. F., 1990. The “challenge hypothesis”: theoretical implications for patterns of testosterone secretion, mating systems, and breeding strategies. *Am. Nat.* 136, 829–846.
- Wingfield, J. C., Vleck, C. M., Moore, M. C., 1992. Seasonal changes of the adrenocortical response to stress in birds of the Sonoran desert. *J. Exp. Zool.* 264, 419–428.
- Wingfield, J. C., Jacobs, J. D., Tramontin, A. D., Perfito, N., Meddle, S., Maney, D. L., Soma, K., 2000. Toward an ecological basis of hormone-behavior interactions in reproduction of birds. In: Wallen, K., Schneider, J. E. (Eds.), *Reproduction in Context: Social and Environmental Influences on Reproduction*. MIT Press, Cambridge, MA, pp. 85–128.
- Zuk, M., Johnsen, T. S., MacLarty, T., 1995. Endocrine-immune interactions, ornaments and mate choice in red jungle fowl. *Proc. R. Soc. Lond., Ser. B* 260, 205–210.