The Condor 99:966–969

REPRODUCTIVE ANATOMY OF THE REED BUNTING: A SPECIES WHICH EXHIBITS A HIGH DEGREE OF SPERM COMPETITION THROUGH EXTRA-PAIR COPULATIONS[1]

A. DIXON[2]
Department of Zoology, University of Leicester, Leicester, LE1 7RH, UK

T. R. BIRKHEAD
Department of Animal and Plant Sciences, University of Sheffield,
Sheffield, S10 2TN, UK

Abstract. The morphology of the reproductive organs of both male and female Reed Buntings (*Emberiza schoeniclus*) are described in relation to the high level of sperm competition exhibited by the species as a result of extra-pair copulation behavior. In males, the volume of the cloacal protuberance in relation to body weight was 26.7 mm^3 g^{-1}, which is relatively large for a socially monogamous passerine. The weight of the testes was 23% greater than that expected for a bird of comparable size, and the spermatozoa were among the longest recorded for any avian species (mean length = 291 μm). The morphology of the female tract also showed adaptations associated with a high level of sperm competition: the sperm storage tubules were extremely long (mean length = 865 μm).

Key words: Reed Bunting, Emberiza schoeniclus, *reproductive anatomy, sperm competition, sperm storage, extra-pair copulation.*

The Reed Bunting *Emberiza schoeniclus* is a predominantly socially monogamous passerine which exhibits a high degree of sperm competition through extra-pair copulation behavior: 97% of females had at least one extra-pair offspring; 55% (118/216) of young were extra-pair, and 86% (50/58) of broods had at least one extra-pair young (Dixon et al. 1994).

Recent comparative analyses indicate that physiological adaptations of the male reproductive organs evolved in tandem with the degree of sperm competition found in a variety of avian species (Møller 1991, Briskie 1993). In species exhibiting a high degree of sperm competition, selection should favor males that produce and deliver relatively more and better quality sperm than competing males (Møller 1988). In addition to quantitative differences in the production and delivery of sperm, direct sperm competition within the female reproductive tract also is expected to lead to qualitative differences in spermatozoa (Briskie and Montgomerie 1992, Birkhead et al. 1993b).

Female birds store sperm following copulation, prior to fertilization of their eggs (Birkhead 1988). The storage of sperm further increases the possibility of sperm competition through direct competition between sperm within the female reproductive tract (Parker 1970, Birkhead and Møller 1992a). The primary storage site is at the utero-vaginal junction, where sperm are held in sperm storage tubules (Bakst 1987). Variation in the number of sperm storage tubules between individuals of the same species is small, but across a range of species the number of sperm storage tubules varies by nearly two orders of magnitude (Birkhead and Hunter 1990, Birkhead and Møller 1992b). However, comparative analyses have not revealed any relationship between female sperm storage anatomy and the intensity of sperm competition (Birkhead and Møller 1992a, Briskie and Montgomerie 1993).

Here we describe the reproductive organs of both male and female Reed Buntings and relate their anatomical characteristics to the high degree of sperm competition exhibited in this species.

METHODS

The study was carried out at Rutland Water in central England between 1990 and 1993. Males were caught during the breeding season and the dimensions of their cloacal protuberance were measured to the nearest 0.1 mm. The volume of the protuberance was calculated by the equation $\pi r^2 \times$ height (the protuberance being approximately cylindrical in shape). The volume of the protuberance increased to a maximum as the breeding season progressed. In order to control for this growth, only measurements taken on 11 males after 1 May were used in the analysis of mean protuberance size as this date is approximate to the full development of the cloacal protuberance (A. Dixon, unpubl. data). Semen samples were obtained from four males by cloacal massage (Wolfson 1952).

Two males, both road-kill casualties, found freshly killed during the breeding season were stored at −70°C before examination. The corpses were thawed at room temperature prior to dissection. The testes were re-

---

[1] Received 29 July 1996. Accepted 8 July 1997.

[2] Present address: Ecology Centre, University of Sunderland, Sunderland, SR1 3SD, UK, e-mail: andrew.dixon@sunderland.ac.uk

moved, measured to the nearest 0.1 mm, and weighed to the nearest 0.001 g. The seminal glomera were removed by dissection, measured and weighed in a similar manner and the number of sperm they contained counted (Birkhead et al. 1991).

A single female Reed Bunting was killed but not eaten by a predator at its nest on the morning before the third and final egg of her clutch was laid (as evidenced from the dissection). This bird was subsequently stored at −70°C, and gradually thawed at room temperature for the dissection. The utero-vaginal junction region of the oviduct was removed and examined as described by Birkhead and Hunter (1990).

RESULTS

MALE REPRODUCTIVE TRACT

The mean volume of the cloacal protuberance was 410 mm^3 (range = 320 to 530 mm^3), whereas the mean height was 6.98 mm and the mean width 8.65 mm. The mean volume index in proportion to body weight was 26.7 mm^3 g^{-1} (following Birkhead et al. 1991). Dissection of two males showed that the protuberance comprised mainly the seminal glomera, extensively coiled tubules of the ductus deferens.

The dimensions of the seminal glomera from one male were 7.7 × 4.6 mm (left) and 8.0 × 4.4 mm (right). The mean mass (g) of both seminal glomera combined was: male A = 0.098 and male B = 0.127 (Table 1), which represents 0.50% and 0.64% of mean male body mass, respectively.

The mass (g) of both testes combined was: male A = 0.553 and male B = 0.490 (Table 1), which represents 2.8% and 2.5% of the mean male body weight, respectively. In both males, the left testis was larger than the right.

The mean length (μm) ± SD of 30 spermatozoa from each of four males was (male 1) 292.4 ± 4.7, (2) 291.5 ± 4.77, (3) 288.9 ± 8.0, and (4) 290.6 ± 4.5. The difference between males was not significant (ANOVA; $F_{3,119} = 0.73$, $P > 0.05$).

FEMALE REPRODUCTIVE TRACT

Some tissue degradation had taken place in the specimen examined and the mucosal folds of the utero-vaginal junction were extremely delicate, resulting in the disintegration of many sperm storage tubules. However, where there was no tissue degradation it was possible to examine the characteristics of the sperm storage tubules. No branched tubules were noted; all were straight blind-ending sacs. The most striking characteristic of the tubules was their large size; of 26 complete tubules measured, the mean length (± SE) was 865 (± 200) μm. The largest tubule measured was 1,270 μm in length. The width of the tubules was quite uniform, being between 40 and 60 μm in diameter. The proportion of undamaged sperm storage tubules containing sperm was very low, 4.8% (7/146 tubules examined). It is not possible to say whether this result was due to the condition of the tissues or the copulatory behavior of the female. The orientation of sperm in the tubules was similar for all those examined: in every case the sperm heads were found to face the blind-end of the tubule.

TABLE 1. Data relating to the testes and seminal glomera from the dissection of two male Reed Buntings.

		Male A	Male B
Testes			
size (mm)	left	10.6 × 8.7	10.2 × 6.2
	right	9.3 × 7.6	7.6 × 5.8
mass (g)	left	0.328	0.269
	right	0.225	0.221
Seminal glomera			
mass (g)	left	0.052	0.066
	right	0.046	0.061
total sperm		1.78×10^6	17.88×10^6

DISCUSSION

MALE REPRODUCTIVE TRACT

The mean height of the cloacal protuberance (CP) was 6.98 mm, which is only 5.8% greater than that predicted for a bird the size of a Reed Bunting (i.e., log CP height = 0.63 log male mass; Birkhead et al. 1993a). The volume index of the cloacal protuberance in relation to body size was similar to that found in the Chaffinch *Fringilla coelebs* (28.4 mm^3 g^{-1}; Sheldon and Birkhead 1994) and larger than that found in the Dunnock *Prunella modularis* (22.9 mm^3 g^{-1}; Birkhead et al. 1991). Much larger volume indices have been recorded in the Alpine Accentor *Prunella collaris* (56.5 mm^3 g^{-1}; Nakamura 1990), Smith's Longspur *Calcarius pictus* (45.9 mm^3 g^{-1}; Briskie 1993), and Bearded Tit *Panurus biarmicus* (72.2 mm^3 g^{-1}; Birkhead and Hoi 1994). Smith's Longspurs and Alpine Accentors have complex social mating systems with a high degree of sperm competition (Briskie 1993, Hartley et al. 1995), whereas in the Bearded Tit, the cloacal protuberance possibly serves as a copulatory organ in which the seminal glomera occupy only a small part, so direct comparison is inappropriate (Birkhead and Hoi 1994). Comparative analysis has shown that cloacal protuberance size is positively related to copulation frequency (Birkhead et al. 1993a).

The mean mass of the seminal glomera represented 0.57% of male body weight, which again is less than that found in species with complex social mating systems, i.e., Dunnock, 0.82% (Birkhead et al. 1991), Smith's Longspur, 1.13% (Briskie 1993), and Aquatic Warbler *Acrocephalus paludicola,* 1.0% (Schulze-Hagen et al. 1995), and also less than that found in the monogamous Chaffinch, 0.74% (Sheldon and Birkhead 1994). Similarly, the mean number of spermatozoa within the seminal glomera was very low in comparison to that found in other species, but this value can vary greatly in relation to the daily copulatory activity of the male (Birkhead et al. 1995). These measures of male sperm reserves suggest that copulation frequency in the Reed Bunting is not very high, a finding consistent with field observations (A. Dixon, unpubl. data).

The testes of the two males examined were 23% larger than the predicted testes mass of 0.40 g for a bird the size of a Reed Bunting (Møller 1991). Using museum specimens, the calculated testes mass from at least ten males obtained during the breeding season was 0.64 g (Møller 1991), which is larger still than the fresh testes mass measured during this study. This difference is possibly due to the method by which testes mass was calculated for the museum specimens, but nevertheless it does indicate that the testes from the fresh males used in this study were not exceptionally large for the species. Møller (1988) showed that sperm production is positively related to testes mass, indicating that the Reed Bunting is anatomically adapted for high levels of sperm production. An extensive comparative analysis of bird species supported the hypothesis that sperm competition, but not male copulation frequency, promotes the evolution of large relative sperm production rates (Møller 1991).

The length of the sperm in the Reed Bunting are among the highest recorded for any bird species, with most previous recorded lengths ranging from 50–300 μm (Briskie and Montgomerie 1992). In mammals and Coleoptera, sperm length appears to be positively associated with a high degree of sperm competition (Dybas and Dybas 1981, Gomendio and Roldan 1991), and a similar conclusion was reached by Briskie and Montgomerie (1992) from their study of 20 species of North American passerine birds.

FEMALE REPRODUCTIVE TRACT

An interesting feature of the female reproductive tract was the extreme size of the sperm storage tubules. The mean length was longer than that recorded for any other avian species (Birkhead and Møller 1992b, Briskie and Montgomerie 1992). Briskie and Montgomerie (1993) found that sperm storage tubules in most passerines could accommodate two layers of sperm. In this respect the Reed Bunting, a passerine bird, stands out as being different because the sperm storage tubules are nearly three times as long as the sperm (mean sperm length 291 μm with mean sperm storage tubule length 865 μm). However, a comparative study across a wide range of bird species by Birkhead and Møller (1992a) indicated that, on average, sperm are about one third the length of the sperm storage tubules.

Briskie and Montgomerie (1992) found a positive correlation between sperm length and the length of the female sperm storage tubules and a negative correlation between sperm length and number of sperm storage tubules. They concluded that sperm competition does influence sperm size in birds, possibly because larger sperm swim faster and selection would favor long sperm when sperm storage tubules are in short supply; sperm long enough to fill a storage tubule also might prevent the storage of sperm from other males. If spermatozoan length is a reflection of the degree of sperm competition in the Reed Bunting, then a corollary of this male morphological adaptation has been the co-evolution of larger sperm storage tubules in the female.

DNA fingerprinting has shown that, through the pursuit of extra-pair copulations, female Reed Buntings are highly polyandrous, resulting in intense sperm competition. Here we show that male Reed Buntings have relatively large testes but do not exhibit concomitantly large sperm stores, suggesting that it is the timing rather than the frequency of copulations that determines male success at obtaining paternity. A low copulation frequency may result in greater numbers of sperm per ejaculate (Birkhead and Fletcher 1995) and the delivery of more motile sperm (Birkhead et al. 1995).

We thank Jayne Pellat for carrying out the sperm counts and Douglas Ross and Sean O'Malley for assistance in the field. AD was supported by a NERC studentship grant and TRB by a grant from BBSRC.

LITERATURE CITED

BAKST, M. R. 1987. Anatomical basis of sperm storage in the avian oviduct. Scan. Microsc. 1:1257–1266.

BIRKHEAD, T. R. 1988. Behavioral aspects of sperm competition in birds. Adv. Stud. Behav. 18:35–72.

BIRKHEAD, T. R., J. V. BRISKIE, AND A. P. MØLLER. 1993a. Male sperm reserves and copulation frequency in birds. Behav. Ecol. Sociobiol. 32:85–93.

BIRKHEAD, T. R., AND F. FLETCHER. 1995. Depletion determines sperm numbers in male Zebra Finches. Anim. Behav. 49:451–456.

BIRKHEAD, T. R., F. FLETCHER, E. J PELLAT, AND A. STAPLES. 1995. Ejaculate quality and the success of extra-pair copulations in the Zebra Finch. Nature 377:422–423.

BIRKHEAD, T. R, B. J. HATCHWELL, AND N. B. DAVIES. 1991. Sperm competition and the reproductive organs of the male and female Dunnock *Prunella modularis*. Ibis 133:306–311.

BIRKHEAD, T. R., AND H. HOI. 1994. Reproductive organs and mating strategies of the Bearded Tit *Panurus biarmicus*. Ibis 136:356–360.

BIRKHEAD, T. R., AND F. M. HUNTER. 1990. Numbers of sperm storage tubules in the Zebra Finch (*Poephila guttata*) and the Bengalese Finch (*Lonchura striata*). Auk 107:193–197.

BIRKHEAD, T. R., AND A. P. MØLLER. 1992a. Numbers and size of sperm storage tubules and the duration of sperm storage in birds: a comparative study. Biol. J. Linn. Soc. 45:363–372.

BIRKHEAD, T. R., AND A. P. MØLLER. 1992b. Sperm competition in birds: evolutionary causes and consequences. Academic Press, London.

BIRKHEAD, T. R, A. P. MØLLER, AND W. J. SUTHERLAND. 1993b. Why do females make it so difficult for males to fertilize their eggs? J. theor. Biol. 161: 51–60.

BRISKIE, J. V. 1993. Anatomical adaptations to sperm competition in Smith's Longspurs and other polygynandrous passerines. Auk 110:875–888.

BRISKIE, J. V., AND R. MONTGOMERIE. 1992. Sperm size and sperm competition in birds. Proc. R. Soc. Lond. B 247:89–95.

BRISKIE, J. V., AND R. MONTGOMERIE. 1993. Patterns of sperm storage in relation to sperm competition in passerine birds. Condor 95:442–454.

DIXON, A., D. R. ROSS, S. L. C. O'MALLEY, AND T. A. BURKE. 1994. Paternal investment inversely related to degree of extra-pair paternity in the Reed

Bunting (*Emberiza schoeniclus*). Nature 371:698–700.

DYBAS, L. K., AND H. S. DYBAS. 1981. Coadaptation and taxonomic differentiation of sperm and spermathecae in featherwing beetles. Evolution 35: 168–174.

GOMENDIO, M., AND E. R. S. ROLDAN. 1991. Sperm size and sperm competition in mammals. Proc. R. Soc. Lond. B 243:181–185.

HARTLEY, I. R., N. B. DAVIES, B. J. HATCHWELL, A. DESROCHERS, D. NEBEL, D., AND T. BURKE. 1995. The polygynandrous mating system of the Alpine Accentor, *Prunella collaris*. II. Multiple paternity and parental effort. Anim. Behav. 49:789–803.

MØLLER, A. P. 1988. Testes size, ejaculate quality, and sperm competition in birds. Biol. J. Linn. Soc. 33: 273-283.

MØLLER, A. P. 1991. Sperm competition, sperm depletion, paternal care and relative testes size in birds. Am. Nat. 137:882–906.

NAKAMURA, M. 1990. Cloacal protuberance and copulatory behaviour of the Alpine Accentor (*Prunella collaris*). Auk 107:284–295.

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

SHELDON, B. C., AND T. R. BIRKHEAD. 1994. Reproductive anatomy of the Chaffinch in relation to sperm competition. Condor 96:1099–1103.

SCHULZE-HAGEN, K., B. LEISLER, T. R. BIRKHEAD, AND A. DYRCZ. 1995. Prolonged copulation, sperm reserves and sperm competition in the aquatic warbler *Acrocephalus paludicola*. Ibis 137:85–91.

WOLFSON, A. 1952. The cloaca protuberance—a means for determining breeding condition in live male passerines. Bird-Banding 23:159–165.

The Condor 99:969–971

HIGHER WINTER MORTALITY OF THE BARN OWL COMPARED TO THE LONG-EARED OWL AND THE TAWNY OWL: INFLUENCE OF LIPID RESERVES AND INSULATION?[1]

SYLVIE MASSEMIN AND YVES HANDRICH
Centre d'Ecologie et Physiologie Energétiques, CNRS, 23 rue Becquerel, 67087
Strasbourg, France, e-mail: massemin@c-strasbourg.fr

Abstract. The role of lipid reserves in winter mortality of the European Barn Owl (*Tyto alba*) was investigated by comparing its adiposity to that of two sympatric nocturnal raptors, the Tawny Owl (*Strix aluco*) and the Long-eared Owl (*Asio otus*), considered as less affected by winter mortality. Adiposity was similar for the three species. Feather insulation, diet, and hunting behavior may explain winter mortality of the Barn Owl.

Key words: Barn Owl, Long-eared Owl, Tawny Owl, adiposity, insulation, winter mortality.

The Barn Owl (*Tyto alba*), Long-eared Owl (*Asio otus*), and Tawny Owl (*Strix aluco*), all three middle-sized nocturnal raptors, are sympatric species in Europe. The Barn Owl and Long-eared Owl inhabit grasslands, whereas the heavier Tawny Owl inhabits forest (Mikkola 1983). In contrast to these other raptors, the Barn Owl in Europe is already at the northern limit of its geographical distribution (Taylor 1994). Harsh winters in Europe are responsible for the death of many Barn Owls (Schönfeld et al. 1977, de Bruijn 1994, Taylor 1994), and the number of Barn Owls found dead in winter is greater than that of Long-eared or Tawny Owls (Piechocki 1960, Guichon 1966).

The explanation usually given for the higher Barn Owl mortality is that the amount of adipose tissue, where major energy reserves (lipids) are stored (Blem 1990), is lower for Barn Owls compared to Long-eared Owls and Tawny Owls (Schönfeld et al. 1977, Marti and Wagner 1985, Baudvin et al. 1991). This assumption apparently is based on a single study in which some individuals that suffered a severe food scarcity were used (Piechocki 1960). To determine whether the Barn Owl should be considered as a "lean" species with little lipid reserve, we compared the adiposity of Barn Owls, Long-eared Owls, and Tawny Owls, none of which were emaciated or breeding. We also studied the insulative value of the plumage in the three species.

METHODS

Barn Owls, Long-eared Owls, and Tawny Owls killed by cars on motorways were collected in 1992–1994 in Alsace-Lorraine, northeastern France. We used intact specimens for which the sex was determined by gonad identification. Some birds reached a critical phase of starvation as evidenced by the absence of abdominal adipose tissue and were therefore excluded from this study. Only nine Long-eared Owls (five males, four females) and eight Tawny Owls (six males, two fe-

---

[1] Received 31 October 1996. Accepted 11 June 1997.