ORIGINAL ARTICLE

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Testes mass in megachiropteran bats varies in accordance with sperm competition theory

Abstract Sperm competition is a widespread phenomenon influencing a range of characters, including investment in gonadal tissue. Conspecific proximity is one factor which can influence the risk of sperm competition and hence testicular investment, and decreased confidence of paternity may be one cost of group living. Aspects of female biology may also influence spermatogenic investment and sperm morphology. This study examines the associations between relative testes mass and roost-group size across 17 species of Megachiroptera. Associations between breeding season duration and investment in spermatogenesis are also examined, as are associations between female reproductive tract dimensions and testes mass and dimensions of spermatozoa across all bats. Relative testes mass was significantly positively associated with roost-group size at a species level and after appropriate phylogenetic control (pairwise comparisons and comparison of independent contrasts). There were no significant relationships between breeding season duration and relative testes mass. Across all bats, neither testes mass nor sperm length were significantly related to dimensions of the female tract. The results are discussed in the context of sperm competition.

Key words Testes mass · Sperm length · Sperm competition risk · Bats

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Introduction

Sperm competition (Parker 1970) is now recognized as a widespread phenomenon, influencing behaviour, physiology and anatomy (e.g. Birkhead and Møller 1992; Ginsberg and Huck 1989; Simmons et al. 1993; Smith 1984). Parker (1970, 1982, 1990a,b) argued that the mechanism of sperm competition in mammals is fundamentally analogous to a raffle, with males entering the greatest number of sperm into a competition likely to win most fertilizations, a situation which should select for increased investment in spermatogenesis. However, since sperm production is costly (Dewsbury 1982; Nakatsuru and Kramer 1982), males should optimize their investment in ejaculates according to sperm competition risk (Parker 1982, 1990a,b). As predicted, investment in spermatogenic relative to somatic tissue is positively related to sperm competition risk across many groups (e.g. primates, Harcourt et al. 1981; butterflies, Gage 1994; fish, Stockley et al. 1997; birds, Møller 1988a,b, 1991; mammals, Kenagy and Trombulak 1986), and large testes generally produce more sperm (e.g. Amann 1970; Gage 1994; Møller 1988b, 1989; Stockley et al. 1997).

Population density may also influence investment in spermatogenic tissue, since large group size may increase the likelihood of sperm competition (Kenagy and Trombulak 1986), and relative testis mass is positively associated with group size in microchiropteran bats and social dispersion in birds (Hosken 1997a; Møller 1991). Moreover, comparative studies indicate that sperm competition risk is positively related to group density in insects (e.g. Gage 1995), molluscs (Baur et al., in press) and mammals (Møller and Birkhead 1989; and see Komers and Brotherton 1997). The effects of density on sperm competition risk within bird species is also generally positive (Birkhead and Møller 1992; Hoi and Hoi-Leitner 1997; Westneat and Sherman 1997), but across species the evidence is equivocal, depending in part on the measure of risk (e.g. extra-pair copulations or extra-

pair fertilizations) (Westneat and Sherman 1997, but see Møller and Tegelström 1997). Arguably, extra-pair copulations more accurately reflect sperm competition risk, in which case risk also increases with group size in birds (Birkhead and Møller 1992). However, it seems clear that reduced confidence of paternity typically represents one cost of group living (Alexander 1974; Thornhill and Alcock 1983).

Factors other than sperm competition may also influence investment in spermatogenic tissue (Gomendio and Roldan 1993; Stockley et al. 1996). In mammals, breeding season duration potentially influences investment in spermatogenesis (Kenagy and Trombulak 1986; Stockley and Purvis 1993). Kenagy and Trombulak (1986) predicted that in species with shorter mating seasons, males should invest relatively more in spermatogenic tissue because of higher demand for sperm production. However, factors such as mating system and population structure may have confounding effects (Kenagy and Trombulak 1986).

While sperm number is predicted to vary with sperm competition risk, sperm size is predicted to evolve independently of it, except under certain conditions, as when the competitive benefits of size vary with sperm density (Parker 1993; and see e.g. Oppliger et al. 1998). Resource investment in individual spermatozoa may also be influenced by factors other than the risk of sperm competition, with some sperm features possibly representing adaptations to the female reproductive tract (Briskie et al. 1997; Cummins 1990). Moreover, it has been argued that longer sperm swim faster (Gomendio and Roldan 1991), although this study did not control for phylogenetic associations and included data from only five species. However, if this is a general relationship, selection could favour larger size when the distance sperm must swim is longer (Stockley et al. 1997), and Cohen (1977) reported swim distance effects on sperm morphology.

While sperm competition has been investigated in a range of mammals (e.g. Dewsbury 1984; Ginsberg and Huck 1989), there have been few investigations of sperm competition in bats (but see Hosken 1997a, 1998; Mayer 1995), and no studies of evolutionary responses to sperm competition in megachiropteran bats, in spite of the occurrence of promiscuous matings in some species (e.g. Hill and Smith 1984; Mickleburgh et al. 1992). Furthermore, roost-group size varies widely across this taxon potentially leading to tremendous variation in sperm competition risk. In addition, recent investigations of sperm length in birds indicate female reproductive tract dimensions may be a key to understanding sperm form (Briskie et al. 1997), but there has been no examination of relationships between female tract and sperm dimensions across bats.

Here, I examine the relationship between testes mass, roost-group size and duration of the breeding season across megachiropteran bats, both at a species level and using appropriate phylogenetic controls. I also examine associations between sperm length and female reproductive tract dimensions across all bats.

Methods

Data on mean testes size, mean body mass, mean group size, female reproductive tract dimensions and duration of the breeding season were collected from published studies of megachiropteran bats (Table 1).

Mean testes size during the period of spermatogenesis was used in analyses, and included volume, length and width, and mass measures. When length and width measures were both available, they were used to calculate testes volumes ($0.523 \times L \times W^2$; Abbott and Hearns 1978). Testes volume and mass were highly correlated in species for which both measures were available ($\log_{10}$ testes mass $= 0.799$ $\log_{10}$ volume $+ 0.1$; $F = 85.5$; $P = 0.0001$; $r^2 = 0.93$; $n = 9$), and volume data were converted to mass using the equation describing this relationship. This allometry analysis included some microchiropteran bats (Table 2). When only testes length data were available they were converted directly to mass using the equation $\log_{10}$ testes mass $= 0.073$ length-1.079 (the relationship between testes mass and length was highly significant in species for which both measures were available: $r^2 = 0.93$, $F = 105.3$, $P = 0.001$, $n = 10$).

Testes size data includes fixed and fresh material (5 fixed, 11 fresh, 1 not stipulated). Fixing has no detectable effect on testis size in at least one bat species, although only three specimens were examined (D.J. Hosken, unpublished data). However, the fixed data included in this study were spread evenly throughout the data set (one of five species with group size less than 50, one of five species with group size between 50 and 1000, and three of six with group size greater than 1000) so there should be no systematic errors introduced into the analysis by combining both types of data. In addition, comparisons of residuals of species-level regression of relative testes mass against group size revealed no differences related to whether testes were fixed or fresh ($t = -0.04$, $P = 0.70$).

Where possible, body mass data used were from animals from which testes size data were obtained. In most instances this was not possible and body mass data were median or mean male mass for the species. In analyses that include female tract dimensions (see below), body mass data were mean female mass for the species.

Because the probability of sperm competition can be positively associated with the proximity of conspecifics (e.g. Møller and Birkhead 1989), roost-group size was used as a potential indicator of sperm competition risk (cf. Hosken 1997a). Since group size may vary within a species, where possible I used the size of roost groups from which testes data were obtained (e.g. *Rousettus amplexicaudatus*, *Acerodon mackloti*). Alternatively, data were obtained by averaging reports of roost-group size from published observations. In the second instance there was often a wide range of values available, but bats roosting in large groups were generally always reported in large groups (e.g. *Eidolon helvum*), while bats in small groups were always in small groups (e.g. *Macroglossus minimus*) (Table 1). Therefore the mean group size values used in this study should provide a reasonably accurate measure of relative sperm competition risk (c.f. Dixson 1995; Hosken 1997a). Moreover, the use of pairwise analysis (see below) is not dependent on absolute roost-group size per se but on relative group size, so, for example, if bats in small groups are typically always found in relatively smaller groups (see above) this comparison essentially controls for potential effects that result from using a particular group size value.

Associations between the duration of the breeding season and relative testes mass were also examined, as were those between testes mass, sperm length and female tract dimensions across both mega- and microchiropteran bats (Tables 1,3). The female tract dimension used was length of the uterine horn of non-pregnant

Table 1 Megachiropteran data used in the analysis (*NA* not available)

Species	Body mass (g)	Group size (range)	Uterine horn length (mm)	Testes mass (g)	Breeding season (months)	References
Pteropus giganteus	1021	900 (800–1,000)	23	8.4	4	Marshall 1947, 1949; Neuweiler 1969
Pteropus tonganus	850	3,020 (60–7,000)	NA	3.8	NA	Baker and Baker 1936; Flannery 1995; Marshall 1947
Pteropus oetinus	470	NA	NA	5.5	1	Baker and Baker 1936; Koopman 1984
Pteropus poliocephalus	677	50,000 (10,000–220,000)	NA	6.75	1.5	Nelson 1965; Ratcliffe 1931; Richards 1991a; Silva and Downing 1995, Tidemann 1995
Pteropus alecto	800	1,200 (400–100,000, but typically 400–2,000)	NA	5.1	2	O'Brien 1993; Hall 1991; Silva and Downing 1995; Thompson 1991
Pteropus scapulatus	400	50,000 (3,000–100,000)	11	5.3	1.5	Nelson 1965; O'Brien et al. 1993; Reardon and Flavel 1987; Richards 1991b
Eidolon helvum	325	50,000 (3,000–1,000,000)	NA	5.5	3	Kingdon 1974; Mutere 1965, 1967; DeFrees and Wilson 1988
Rousettus aegyptiacus	142	3,000 (1,000–9,000)	8.1	3.5	4	Bernard 1988; Mutere 1968, Nowak 1994, pp. 51–53; Skinner and Smithers 1990
Rousettus amplexicaudatus	91.8	650 (500–800)	NA	0.74	3	Goodwin 1979; Silva and Downing 1995
Micropteropus pusillus	32.5	1.5 (1–2)	NA	0.23	6	Jones 1972; Nowak 1994, pp. 67–88; Thomas and Marshall 1984
Cynopterus sphinx	35	4 (1–20, but typically 3–8)	9	0.36	4	Balasingh et al. 1995; Krishna and Domenic 1984; Sandhu 1984; Sreenivisan et al. 1974; Vamburkar 1958
Epomophorus anurus	80	65 (60–70)	NA	0.9	2	Kingdon 1974; Okia 1974
Epomops buettikoferi	160	2 (1–3)	NA	0.299	6	Nowak 1994, pp. 64–65; Thomas and Marshall 1984
Acerodon mackloti	518	325 (300–350)	NA	3.25	NA	Goodwin 1979
Dobsonia peroni	224	147.5 (5–300)	NA	1.64	NA	Goodwin 1979
Macroglossus minimus	15.9	3 (1–5)	NA	0.287	12	Gunnell et al. 1996; Novak 1994, pp. 82–84; Silva and Downing 1995; Start 1974; Thomson 1991
Macroglossus sobrinus	23	3 (1–5/10)	NA	0.317	12	Novak 1994, pp. 82–84; Start 1974
Eonycteris spelaea	60	3,000	NA	0.798	12	Nowak 1994, pp 80–81; Silva and Downing 1995; Start 1974
Haplonycteris fischeri	18	NA	NA	0.81	12	Heidemann 1989

individuals (this was the most widely available measure), and breeding season duration was defined as the period during which copulations occur.

Sperm length data were obtained from Cummins and Woodall (1985). Body mass was not used as a covariate in CAIC (see below) sperm length analysis because previous work indicated that after appropriate phylogenetic control, sperm length does not vary with body mass (Hosken 1997a). However, body mass was controlled for in species-level analysis of spermatozoa since Cummins and Woodall (1985) found a positive relationship between sperm length and body mass across bat species.

Sample sizes presented vary depending on the availability of data. After controlling for phylogeny, sample sizes represent the number of independent contrasts and not the number of species used in the comparisons or, alternatively, the number of pairwise comparisons (see below).

Associations were analysed at the species level (using least-squares linear regression), and where possible as independent contrasts and pairwise comparisons (Møller and Birkhead 1992; Purvis and Rambaut 1994) to minimize problems associated with taxonomic relatedness and phylogenetic inertia (Harvey and Pagel 1991). Data from a maximum of 25 species in 16 genera (including

Table 2 Species and data used in testes mass/testis volume allometry analysis. (Data obtained from references in Table 1 for megachiropteran bats and from references in Hosken 1997a for microchiropteran bats)

Species	Testes mass (g)	Testis volume (cm^3)
Pteropus tonganus	3.8	2.6
Pteropus poliocephalus	6.75	8.7
Pteropus scapulatus	5.3	6.5
Cynopterus sphinx	0.36	1.01
Nyctalus noctule	0.543	0.188
Miniopterus schreibersii	0.08	0.038
Chalinolobus gouldii	0.03	0.01
Tadarida condylura	0.2	0.075
Eidolon helvum	5.5	3.389

microchiropteran bats, see below) were available for analysis. For the contrast analysis I used the Comparative Analysis by Independent Contrasts (CAIC) program (Purvis and Rambaut 1994) to estimate contrasts for each node in the phylogeny for which there was variation in the independent variable. The phylogeny used (Fig. 1) was based on a majority-rule consensus tree generated using anatomical characters (Springer et al. 1995); this phylogeny was chosen since it contained all the genera included in the present study with the exception of *Haplonycteris* (which for this reason was excluded from CAIC analysis of associations between breeding season duration and testes mass). Contrasts were analysed using least-squares regression analysis forced through the origin (Harvey and Pagel 1991). In CAIC analyses, branch lengths were assumed to be equal, thereby assuming a punctuated model of evolution. Equal branch lengths were used because a subsample of the total number of bat species was used in the phylogeny, and some of the phylogenetic relationships are debatable (see e.g. Springer et al. 1995). However, by using two comparative methods, conclusions should be robust.

In the pairwise comparisons (based on comparisons between pairs of closely related species that differ in the dependent variable; Møller and Birkhead 1992), if more than two species were available for comparison, average values for all possible combinations were used. The pairwise comparisons were analysed using one-tailed paired Student's *t*-tests. Pairs were chosen based on the Springer et al. (1995) phylogeny, and Koopman's (1984) phylogeny [in Hosken (1997a)] in analyses that included some microchiropteran bats. However, note that *Pipistrellus pipistrellus* data from Hosken (1997a) were not considered for pairwise comparisons as data may have come from two species (see Barratt et al. 1997). Since organ size usually scales allometrically with body size, testes mass, body mass and female tract dimensions were log$_{10}$ transformed. Group size and sperm length data were log$_{10}$ transformed to improve normality. Power analysis was performed following Zar (1996).

Table 3 Species and data used in analysis of associations between sperm length and female reproductive tract dimensions (all sperm length data from Cummins and Woodall 1985)

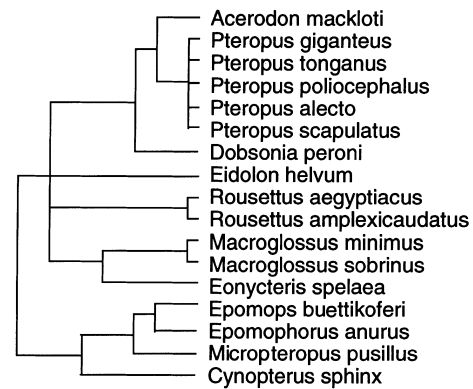| Species	Sperm length (μm)	Length of uterine horn (mm)
Nyctophilus major	52	2.1 (Hosken 1997b)
Nyctophilus geoffroyi	53	1.6 (Hosken 1997b)
Chalinolobus morio	45	2 (Kitchener and Costa 1981)
Chalinolobus gouldii	58	3.5 (Kitchener 1975)
Myotis lucifugus	51	1.5 (Reeder 1939)
Mormopterus planiceps	66	2.5 (Krutzsch and Crichton 1987)
Pteropus scapulatus	101.2	11 (Pow and Martin 1994)

Fig. 1 Phylogeny of Megachiroptera used in CAIC and pairwise analysis. Based on a majority-rule concensus tree generated using anatomical data (from Springer et al. 1995)

Results

At a species level, testes mass was significantly related to body mass ($F = 45.1$, $P = 0.0001$, $r^2 = 0.71$, $n = 19$). Therefore to remove body mass effects, residual testes mass was calculated, from a regression of testes mass against body mass, and regressed against roost-group size. Residual testes mass was significantly positively related to roost-group size at the species level (Fig. 2; $F = 16.3$, $P = 0.001$, $r^2 = 0.49$, $n = 17$).

Using pairwise comparisons, species with relatively larger testes belonged to larger roost groups than species with relatively small testes ($df = 4$, paired t-value = -2.194, $P = 0.047$; Table 4). When data from microchiropteran bats were included in this analysis (Table 5), the relationship was even stronger ($df = 12$, paired t-value = -2.388, $P = 0.017$) [note that in this analysis, testis mass data from Hosken (1997a) were doubled to
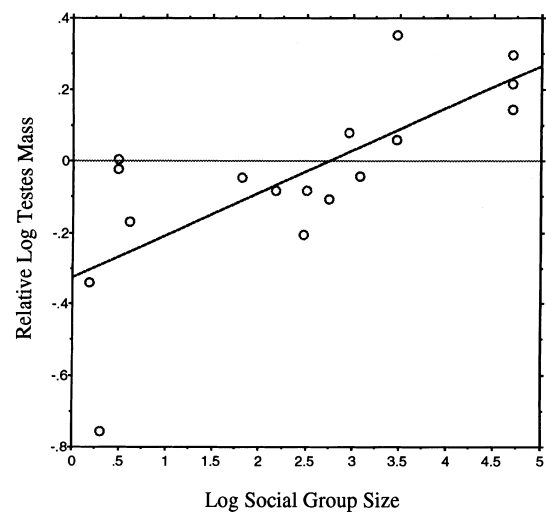

Fig. 2 Plot of relative testes mass (residuals from a least-squares linear regression of testes mass against body mass) against roost-group size at a species level for megachiropteran bats

Table 4 Data used in pairwise comparison of testes mass variation with group size in megachiropteran bats; relative testes mass (residuals from a linear regression of testes mass against body mass) in relation to group size. When more than two species were available for comparison, mean values of all possible combinations were used

Smaller group size		Larger group size	
Dobsonia peroni	−0.103	*Acerodon mackloti*	−0.1
Pteropus mean	−0.08	*Pteropus* mean	0.105
Rousettus amplexicaudatus	−0.132	*Rousettus aegyptiacus*	0.386
Epomops buettikoferi	−0.724	*Epomophorus anurus*	−0.003
Macroglossus/ Eonycteris mean	−0.032	*Macroglossus/ Eonycteris* mean	0.045

Fig. 3 Plot of residual contrasts in testes mass (residuals generated from a least-squares linear regression of contrasts in testes mass against contrasts in body mass) against contrasts in roost-group size for megachiropteran bats. Contrasts were generated using CAIC

give testes mass, and body mass effects were removed as above].

Comparing independent contrasts gave a similar result. Because contrasts in testes mass were positively associated with contrasts in body mass ($df = 11$, $r^2 = 0.44$, $\beta = 0.79$, $F = 7.936$, $P = 0.018$), residual contrasts in testes mass were calculated. A significant and positive relationship was found between residual contrasts in testes mass and contrasts in roost-group size ($df = 11$, $r^2 = 0.4$, $\beta = 0.137$, $F = 6.67$, $P = 0.027$; Fig. 3). This indicates a significant evolutionary association between testes mass and roost-group size (Grafen 1989).

At a species level, there was no association between residual testes mass and duration of the breeding season ($n = 16$, $r^2 = -0.07$, $F = 0.04$, $P = 0.95$, and power = 0.95), but when seasonal breeders were considered alone, residual testes mass was significantly negatively related to the duration of the breeding season ($n = 12$, $r^2 = 0.38$, $F = 6.23$, $P = 0.03$). However, there was no significant relationship between residual contrasts in testes mass and contrasts in breeding season duration, either when including continuous breeders ($df = 9$, $r^2 = 0.25$, $\beta = -0.07$, $F = 2.73$, $P = 0.13$) or considering seasonal breeders alone ($df = 7$, $r^2 = 0.39$, $\beta = -0.12$, $F = 3.921$, $P = 0.095$), although a weak trend for a difference was noted in the second comparison. The powers of these tests were calculated as 0.69 and 0.64, respectively.
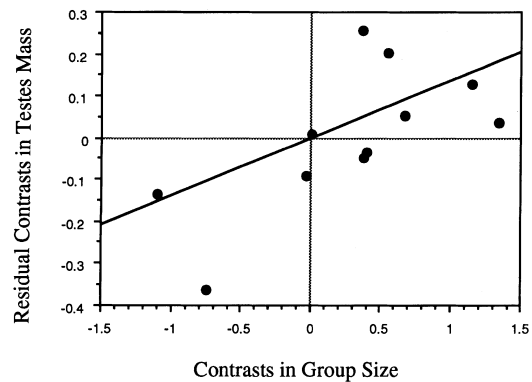
Similarly in a pairwise comparison, there was no significant relationship between residual testes mass and breeding season duration, although the number of comparisons was small ($df = 3$, paired t-value = −0.466, $P = 0.34$; Table 6). The power of detecting a difference equal to the mean difference between the two groups in this analysis was calculated to be between 0.5 and 0.6.

Across all bats (Micro- and Megachiroptera), uterine horn length was significantly positively related to body mass at a species level ($n = 10$, $r^2 = 0.89$, $F = 67.3$, $P = 0.0001$). When mass effects were removed (as above), there was no significant relationship between residual testes mass and residual uterine horn length ($n = 7$, $r^2 = 0.01$, $F = 0.05$, $P = 0.94$) or residual sperm length and residual uterine horn length ($n = 7$, $r^2 = 0.11$, $F = 0.6$, $P = 0.47$), both at a species level. The power of these tests was calculated to be 0.84 and 0.67, respectively. Independent contrasts in uterine horn length were significantly positively related to contrasts in body mass ($n = 8$, $r^2 = 0.701$, $\beta = 0.49$, $F = 16.386$, $P = 0.049$). Therefore mass effects were removed (as above). There was no significant relationship between residual contrasts in testes mass and residual contrasts in uterine horn length ($n = 6$, $r^2 = 0.10$, $\beta = -0.79$, $F = 0.71$,

Table 5 Data used in pairwise comparison of testes mass variation with group size across all bats; relative testes mass (residuals from a linear regression of testes mass against body mass) in relation to group size. When more than two species were available for comparison, mean values of all possible combinations were used

Smaller group size		Larger group size	
Dobsonia peroni	−0.103	*Acerodon mackloti*	−0.1
Pteropus mean	−0.08	*Pteropus* mean	0.105
Rousettus amplexicaudatus	−0.132	*Rousettus aegyptiacus*	0.386
Epomops buettikoferi	−0.724	*Epomophorus anurus*	−0.003
Macroglossus/Eonycteris mean	−0.032	*Macroglossus/Eonycteris* mean	0.045
Chalinolubus gouldii	−1.206	*Myotis lucifugus*	0.495
Pipistrellus subflavus	0.021	*Pipistrellus minimus*	0.1
Tyloncteris pachypus	0.997	*Tyloncteris robustula*	1.021
Miniopterus minor	−0.329	*Miniopterus schreibersi*	−0.152
Tadarida pumila	0.291	*Tadarida aegyptiaca*	0.276
Taphozous longimanus	−0.732	*Taphozous georgianus*	−0.648
Hipposideros galeritus	−0.071	*Hipposideros speoris*	−0.012
Rhinolophus hipposideros	−0.567	*Rhinolophus clivosis*	1.218

Table 6 Data used in pairwise analysis of variation in relative testes mass (residuals from a linear regression of testes mass against body mass) in relation to breeding season duration

Longer season duration		Shorter season duration	
Pteropus giganteus	0.074	Pteropus oetinus	0.162
Pteropus alecto	−0.059	Pteropus poliocephalus	0.123
Rousettus aegyptiacus	0.386	Rousettus amplexicaudatus	−0.132
Epomops buettikoferi	−0.724	Epomophorus anurus	−0.003

$P = 0.48$, power $= 0.81$) or between sperm length and residual contrasts in uterine horn length ($n = 5$, $r^2 = 0.15$, $\beta = 0.35$, $F = 0.71$, $P = 0.45$, power $= 0.5$). Associations between testes mass, sperm length and uterine horn length were not undertaken using pairwise analysis as too few data were available.

Discussion

In all analyses there was a significant and positive relationship between roost-group size and testes mass across megachiropteran bats. Testes mass is known to be positively associated with the likelihood of sperm competition in a range of taxa, including mammals (e.g. Ginsberg and Rubenstein 1990; Harcourt et al. 1981), and sperm competition risk increases with group size/density in at least some taxa (e.g. Gage 1995; Møller and Birkhead 1989). Thus the most likely explanation for the association is that sperm competition risk increases with group size across Megachiroptera, selecting for increased spermatogenic investment. Alternatively, copulation frequency, which also affects gonadal investment, may be higher in large groups, but if so why? Again the most parsimonious explanation relates to sperm competition as copulation rates are generally positively associated with sperm competition risk (e.g. Dixson 1995; Ginsberg and Rubenstein 1990; Hogg 1988; and see Møller 1991). Since male megachiropteran bats often copulate throughout a colony (Hill and Smith 1984), and promiscuous matings are reported for a number of gregarious species (Hill and Smith 1984; Mickleburgh et al. 1992), it seems likely that sperm competition risk would increase with roost-group size. An identical relationship was found across microchiropteran bats, and a similar explanation suggested (Hosken 1997a), and across mammals generally the risk of sperm competition is related to the proximity of conspecific males (Møller and Birkhead 1989). Similarly, across birds, Møller (1991) defined sperm competition risk as the number of neighbouring males and found a positive association between sperm competition risk and relative testes mass. Thus the results of this bat study are consistent with predictions of sperm competition theory (Parker 1970, 1982, 1990a,b); as the risk of sperm competition (group size) increases, males invest proportionally more in spermatogenic tissue. Moreover, pairwise comparisons including microchiropteran bats were highly significant, indicating that the conclusions of an earlier study, which employed independent contrast analysis alone to control for phylogenetic associations, are likely to be robust (Hosken 1997a). Note however that less than half the variation in relative testes mass was explained by roost-group size. Clearly, additional factors will influence spermatogenic investment, especially mating patterns (Kenagy and Trombulak 1986). In addition, while little information is available on bat mating systems, some bats do not mate within roost groups. For example, the hammer-headed bat, *Hypsignathus monstrosus* forms leks, with lekking thought to be the result of wide, even female distribution (Bradbury 1977a). Although this bat was not included in the study, calling behaviour of this nature is widespread in epomophorine bats. However, there is some congruence between calling-group size and roost-group size in at least some species (e.g. *Epomops franqueti*, Bradbury 1977b), and the positive relationship between testes mass and group size is strong in spite of confounding influences such as mating patterns.

Dilution of ejaculates within the female reproductive tract may also select for larger testes and ejaculates (Short 1980). However, in the current study there was no association between testes mass and uterine horn length across all bats. The sample sizes in these analyses were small, but high *P*-values and power indicate that the lack of association is unlikely to be the result of type II error. While larger samples would be beneficial, a similar result was reported across microchiropteran bats alone: no significant relationship between uterine horn width and testis mass was detected (Hosken 1997a). Assuming sperm production and ejaculate size correlate with testes mass in bats (as in many other taxa; e.g. Gage 1994; Møller 1988a,b, 1989; Stockley et al. 1997; and see review in Birkhead 1995), it appears that ejaculate dilution may have had little influence on the evolution of testes mass across bats, although as noted, the sample sizes in these analyses were small. In contrast, ejaculate size and female tract dimensions were positively associated across muriod rodents and primates with similar sample sizes to those employed here (species $n = 11$, contrasts $n = 8$; Gomendio and Roldan 1993). Bats may differ from other eutherian mammals in this regard, possibly due to prolonged female sperm storage; females of some mega- and microchiropteran bat species store sperm (e.g. Hood and Smith 1989; Hosken et al. 1996), and female sperm storage affects ejaculate size in marsupials (e.g. Bedford et al. 1984).

Under conditions of sperm competition, increased female tract size or complexity may be selectively favoured to grant females greater control over fertilization (Ward 1993; Birkhead 1995). Therefore, an association between testes mass and female tract dimensions may also be predicted on this basis. However, as stated, this was not the case across bats, although tract complexity was not directly examined.

There was no significant association between testes mass and duration of the breeding season in analyses

that controlled for phylogeny. Stockley and Purvis (1993) reported differences in testes/body mass relationships between continuous and seasonal breeders, and Kenagy and Trombulak (1986) predicted relative testes mass would vary depending on breeding season duration. In the current study, the relationship detected when aseasonal breeders were considered alone appears to be a phylogenetic effect rather than an effect of breeding season duration per se, since after controlling for phylogeny the association was non-significant. However, the low power of these tests together with the relatively low P-values render the conclusions somewhat equivocal.

There was no association between sperm length and female tract dimensions. However, the power of one of these tests was very low (0.5). Positive relationships between sperm length and female tract dimensions have been found in other taxa (e.g. birds, Briskie et al. 1997; insects, Pitnick and Markow 1994), including mammals (Gomendio and Roldan 1993). Since sperm movement within the mammalian uterus is largely passive but sperm swim in the oviducts (Katz et al. 1989), perhaps oviduct length would be a more informative dimension to use when looking for such associations, especially as across a sample of mammalian taxa, sperm length and oviduct length were associated (Gomendio and Roldan 1993). However, insufficient data were available to carry out analysis with this dimension, but female tract length can account for at least some ejaculate features (e.g. pigs, R.V. Short, personal communication).

Analyses of sperm dimensions included a number of sperm-storing bats (e.g. *Chalinolobus gouldii*, Hosken et al. 1996; *Nyctophilus geoffroyi*, Hosken 1997b, 1998). Aspects of sperm morphology other than length (e.g. mitochondrial number) possibly represent adaptations to sperm competition and female reproductive tract dimensions in sperm-storing species. However, female physiological support of stored spermatozoa may negate the need for specfic specializations (and see Gage 1997). More data from a wide range of bat taxa are needed to further investigate the significance of sperm size variation.

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