Neither Mating Rate Nor Spermatophore Feeding Influences Longevity in a Ladybird Beetle

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Abstract

Females of many species experience costs associated with mating. Seminal products, including nuptial gifts, may mitigate these mating costs or exacerbate them. For example, nuptial gifts derived from male accessory glands may transfer nutrition or potentially harmful seminal proteins to females. In this study, we assay the costs of multiple mating and the consumption of seminal products in a ladybird beetle. We compared longevity in females mated singly or multiply, while allowing or preventing spermatophore consumption at each mating. In order to distinguish a cost of mating *per se* from a cost of elevated reproduction, we prevented reproduction by using nutrient-stressed females. Mating singly or multiply had no effect on female longevity, nor did spermatophore feeding influence longevity. The results imply, first, that intermediate mating rates do not directly harm females, though females may experience other indirect costs of mating (e.g. reduced foraging efficiency) or costs of reproduction; and second, that spermatophores transfer neither food nor directly harmful substances to female ladybirds.

Introduction

Much current research in sexual selection is centred on the conflicting evolutionary interests of males and females (Arnqvist & Rowe 2005; Andersson & Simmons 2006; Kokko et al. 2006). Sexual conflict is defined as a conflict between the evolutionary interests of individuals of the two sexes, and this conflict may often result in sexually antagonistic selection on traits that are part of the phenotypes of both sexes (Parker 1979, 2006; Rowe & Day 2006). For example, increasing mating rate may often be favoured in males but disfavoured in females (Arnqvist & Rowe 2005). Several reviews have highlighted the need for more empirical investigation of the economics of shared traits, such as mating rate, so that we can evaluate the extent to which these traits are shaped by sexually antagonistic selection (Chapman et al. 2003; Zeh & Zeh 2003; Arnqvist & Rowe 2005; Kokko et al. 2006; Rowe & Day 2006; Vahed 2007).

Costs of mating to females have been documented in a number of taxa (reviewed in Arnqvist & Rowe 2005), and may include elevated predation and parasitism, reduced foraging success, genital damage and a variety of life history adjustments caused by signals from male seminal products (e.g. Rowe 1994; Wolfner 1997; Crudgington & Siva-Jothy 2000; Baer et al. 2001; Blanckenhorn et al. 2002). A recent meta-analysis of insect polyandry suggests that there is generally an intermediate optimal mating rate for females, resulting from increased offspring production and decreased longevity with multiple mating, and furthermore that observed mating rates may often exceed the optimum (Arnqvist & Nilsson 2000). However, in those cases where mating includes nuptial gifts, increased mating rates tend to increase reproductive success while having little or a weak positive effect on longevity (Arnqvist & Nilsson 2000). A reasonable inference is that consumption of these seminal products tends to offset any longevity

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costs associated with mating, and is on average a net benefit to females.

In spite of this broad pattern, another line of evidence suggests that nuptial gifts may impose costs on females. Nuptial gifts are often produced by male accessory glands and there is reason to believe that accessory gland products may not always be beneficial to females. For example, there is increasing evidence that some seminal products delivered to the female reproductive tract reduce female fitness. In *Drosophila melanogaster*, components of male accessory gland proteins (Acps) in the seminal fluid (e.g. sex peptide) reduce female lifetime reproductive success (Chapman et al. 1995; Wigby & Chapman 2005). Likewise, in species with nuptial gifts derived from male accessory glands, transferred products may carry some costs to females (Simmons & Parker 1989; Sakaluk 2000; Bonduriansky et al. 2005; Sakaluk et al. 2006; Engqvist 2007b). This interesting possibility has led to calls for more study of the fitness consequences of nuptial feeding for females (Vahed 1998, 2007; Arnqvist & Rowe 2005; Gwynne 2008).

Although there is a large number of studies of nuptial feeding on components of female fitness (for reviews, see Vahed 1998, 2007; Gwynne 2008), these studies typically manipulate mating rate and thereby nuptial feeding, rather than nuptial feeding alone. As discussed above, there is a generally positive effect of an increased mating rate in these studies (Arnqvist & Nilsson 2000). There are fewer studies that manipulate nuptial feeding independently of mating, and the results of these do not always lead to the conclusion of a net benefit to females. Although several report positive effects of nuptial feeding on female reproductive parameters (e.g. orthopterans, Gwynne 1984, 1988; Simmons 1988, 1990; Kasuya & Sato 1998; Reinhold 1999; Ono et al. 2004; scorpionflies, Engqvist 2007a), several other studies report no effect on parameters such as fecundity and egg mass (in orthopterans, Gwynne et al. 1984; Wedell & Arak 1989; Will & Sakaluk 1994; Vahed & Gilbert 1997; Vahed 2003; piophilid flies, Bonduriansky et al. 2005; cockroaches, Mondet et al. 2006; coccinellid beetles, Perry & Rowe, in press). One study found a positive effect on female longevity (tree cricket, Brown 1997), while others detected no effect (wartbiters, Wedell & Arak 1989; piophilid flies, Bonduriansky et al. 2005; cockroaches, Mondet et al. 2006; coccinellid beetles, Perry & Rowe, in press). Only one of these studies independently compared the effects of mating and nuptial feeding rates on female fitness (Vahed 2003).

We examined the effect of multiple mating and spermatophore feeding on female longevity in the two-spot ladybird beetle (*Adalia bipunctata*). Males of the two-spot ladybird beetle transfer a proteinaceous spermatophore during copulation, which females eject and consume after mating. Data for two other ladybird species that do not ingest spermatophores suggest a longevity cost of increased number of matings, possibly due to increased oviposition with increased mating (Omkar & Srivastava 2002; Omkar & Mishra 2005; Omkar et al. 2006; but see Haddrill et al. 2007). Our previous work on *A. bipunctata* indicates no long-term benefit to females of repeated spermatophore feeding independent of mating rate (own data). However, that study, like most previous studies of the effect of nuptial feeding on females, was conducted under relatively benign laboratory conditions of sufficient food. There is good reason to expect that any costs (or benefits) of mating and nuptial feeding be exaggerated under stressful conditions. For example, a nutritional effect of nuptial feeding may be detectable only when females are food-limited, when the marginal benefit would be higher. To address this shortcoming, we conducted a study of the economics of mating and nuptial feeding in a highly food-stressed environment. Because we deprived females of food during the experiment, we expected to see costs or benefits of nuptial feeding that might be masked in more benign environments. Moreover, because food deprivation prevented egg production, any observed depression in longevity can be attributed to harmful substances in the gift or ejaculate, or other interactions with the male at mating, rather than to an increase in oviposition induced by mating.

Methods

Species

Adalia bipunctata (Coleoptera: Coccinellidae) is an aphidophagous predator broadly distributed across temperate habitats. Following most copulations (>90%), females eject a hollow spermatophore and most females (>90%) immediately ingest the spermatophore.

We obtained several hundred first and second instar *A. bipunctata* (Natural Insect Control, Stevensville, ON, Canada) and reared them to adulthood on a combined diet of pea aphids (*Acyrthosiphon pisum* reared on broad bean, *Vicia faba*) and UV-sterilized *Ephestia kuehniella* eggs (Beneficial Insectary, Redding, CA, USA). Larvae and adults were housed in an

environmental chamber on a 16:8 h dark:light cycle at 23°C ± 1°C. Adults used in the experiment were at least 12 d post-emergence. Adults were fed aphids *ad libitum* until 1 d prior to the initial mating trial; on that day each female was fed four adult aphids and males continued on the *ad libitum* diet. Throughout the experiment, females were housed individually in petri dishes (50 mm × 12 mm) and transferred to new petri dishes daily.

Experimental procedure

Females were randomly assigned to the two crossed experimental treatments: number of matings (one, three or five), and spermatophore feeding (allowed or prevented at each mating). Females were mated once or twice per day until the assigned number of matings was attained, which occurred within 5 d. Although females may mate many more times over a lifetime in a natural setting, the mating rates we used are within the range of rates observed in the field (0.1–0.5 copulations/d, Webberley et al. 2006; see also Brakefield 1984). The males used in the experiment had not mated for at least 2 d to ensure time to replenish ejaculate stores. If a male failed to mount a female within 1 h, it was discarded and a new male introduced. We ignored copulations that lasted <30 min (19 occurrences out of 433) because we have seldom observed spermatophore transfer in such brief matings. After each copulation ended, we monitored females for spermatophore ejection and, according to the assigned treatment either removed the spermatophore immediately or allowed females to ingest it. For females in the latter treatment, we simulated removing a spermatophore shortly after females finished eating the spermatophore to control for any slight disturbance to females caused by the removal.

After mating, and between matings, females were housed in the environmental chamber under the conditions described and provided with cotton dampened with 50 μl of water daily. Females were deprived of food from the first day of mating, except on day 3, when we fed each female a large pea aphid to stimulate mating receptivity. Beginning on the first day of matings, we checked for survival at 09:00, 13:00 and 17:00 h daily until all females perished. We also noted and removed any eggs laid. Depriving females of food stopped virtually all oviposition by day 3 of the mating portion of the experiment. Forty-eight females oviposited in the first 2 d, whereas only four females oviposited from day 3 onward and only two laid eggs after mating was completed on day 5.

Analyses

To test for an effect of the number of matings, spermatophore feeding, and the interaction between these two factors on female survival, we used a proportional hazards model, with the response variable being hours survived since the beginning of the experiment.

We initially excluded females based on three criteria: (1) failure to mate the number of times assigned; (2) failure to eject spermatophores and (3) failure to eat all of the ejected spermatophore when assigned to the 'allow' treatment. The initial sample sizes for the mating treatment levels of 1, 3 and 5 were 29, 31 and 31 females for the 'allow' spermatophore feeding treatment and 20, 24 and 20 for the 'prevent' treatment. After excluding females, the final sample sizes for those respective treatment groups were 10, 11 and 4 females vs. 14, 24 and 16 females. The sample size was reduced to four for females mating five times and feeding on spermatophores because females rarely ejected and ate five entire spermatophores with five matings. The small sample size in this group reduced our ability to meaningfully test the hypotheses, and for this reason we performed a second analysis in which we grouped females into those that mated singly vs. multiply. This allowed us to include additional females that mated two or four times, and the sample sizes for these pooled data were, for females mating singly vs. multiply, 10 vs. 23 for females allowed to eat the spermatophore and 14 vs. 48 for prevented females. The mean number of matings in the multiply mated group was 3.7 ± 0.1 SE. We report statistics from the first analysis because this was the planned approach, but focus on results from the second test.

Least squares means are reported with SE. Analyses were conducted using JMP 5.0 (SAS Institute).

Results

One female survived much longer than the others (720 h vs. x = 161 h ± 28). To avoid having an extreme outlier, we set survival for this individual to the next longest survival time (280 h).

When we analysed survival for females that mated one, three or five times, neither feeding on spermatophores nor the number of matings influenced female survival (spermatophore feeding: $\chi^2_1 = 0.03$, p = 0.70; number of mates: $\chi^2_2 = 0.84$, p = 0.66; interaction: $\chi^2_2 = 0.9$, p = 0.64). The 95% confidence intervals for each treatment group overlap broadly

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and thus give no indication of a trend (spermatophore feeding allowed, one mating: 139–174 h; three matings: 145–178 h; five matings: 141–196 h; spermatophore feeding prevented, one mating: 148–178 h; three matings: 152–175 h; five matings: 144–172 h).

When we grouped females that mated multiply and compared their survival to that of females mated singly, there was again no effect from spermatophore feeding ($\chi^2_1 = 0.75$, $p = 0.39$) or the number of matings ($\chi^2_1 = 0.05$, $p = 0.82$), or their interaction ($\chi^2_1 = 0.11$, $p = 0.74$; Fig. 1, Table 1; results were similar from a reduced model dropping the non-significant interaction term). Because these results were null, it is of interest to estimate the magnitude of treatment effect that we can confidently exclude at the $\alpha = 0.05$ level, given our data (e.g. Hoenig & Heisey 2001). We did so by constructing confidence intervals for the differences between groups (Table 2). For example, at $\alpha = 0.05$, our experiment could have detected a decrease in longevity larger than 18% or an increase larger than 11%, between singly and multiply mated females that ingested spermatophores (Table 2).

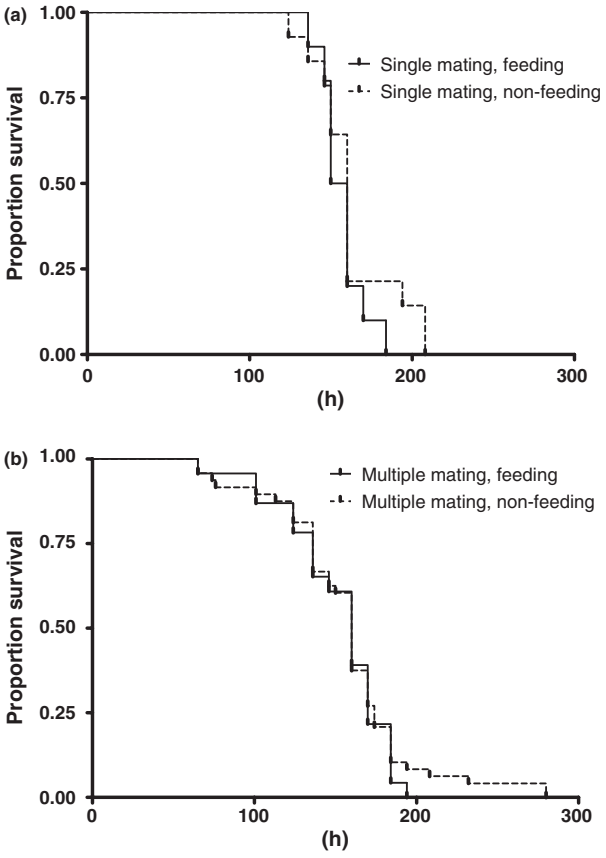

Fig. 1: Survival of females mated singly (a) or multiply (b), after eating a spermatophore at each mating (solid line) or not (dotted line). Because the lines overlap extensively, the confidence intervals are not presented for visual clarity.

Table 1: Survival of females mated singly or multiply and either allowed to or prevented from eating a spermatophore at each mating

Treatment combination	Survival (h)	SE
Single mating, spermatophore eaten	156.7	11.5
Single mating, no spermatophore	162.6	9.7
Multiple matings, spermatophores eaten	151.2	7.6
Multiple matings, no spermatophore	156.3	5.2

The survival values are least squares means from a 2 × 2 ANOVA.

All females mated at least once, but some females refused subsequent matings (six females originally assigned to five matings). If mating refusal is governed by female condition and there is a relationship between condition and longevity, then excluding these females could bias the outcome (Rönn et al. 2006). To test for this possibility, we conducted a Student's t-test on survival times based on whether females completed their assigned number of matings. There was no difference in survival between the groups for females assigned to five matings ($x = 160.1$ h $\pm$ 6.0 for females that completed the mating treatment; $x = 160.0$ h $\pm$ 26.1 for females that refused matings; $F_{1,18} = 0.0$, $p = 0.99$).

Discussion

Our results suggest that there are no detectable costs of either multiple mating or nuptial feeding for female longevity in this ladybird beetle, even when beetles are nutritionally stressed. We deprived females of food during the experiment, an approach that offered two advantages. First, females deprived of food quickly cease oviposition and oogenesis, thus nullifying a potential trade-off between reproduction and longevity. Our earlier studies demonstrated that consumption of spermatophores advanced oviposition in time (Perry & Rowe, in press), and this may itself elevate mortality (Stearns 1992). Second, the chance of detecting either a longevity benefit or cost of nuptial feeding or mating should be maximized under conditions of nutritional stress, and nutritional stress is a common feature of these beetles' environment (Sloggett & Majerus 2000; Evans 2003). We discuss this puzzling behaviour of nuptial feeding in coccinellid beetles in the context of other studies of nuptial feeding and mating costs.

Table 2: The difference in longevity between females subjected to spermatophore feeding and mating rate treatments, given that neither treatment had a statistically significant effect on longevity (least squares means; negative values indicate a lower mean for the spermatophore feeding and multiple mating groups, compared to non-feeding and singly mating females)

	Comparison			
	Spermatophore feeding (SF)	No SF	Single mating	Multiple matings
	Single vs. multiple mating	Single vs. multiple mating	SF vs. no SF	SF vs. no SF
Difference in means (h)	−5.5	−6.2	−5.9	−5.1
Confidence interval	−18%, 11%	−15%, 7.3%	−19%, 12%	−13%, 6%

Any treatment effect outside the confidence interval can be ruled out at the $\alpha = 0.05$ level (Hoenig & Heisey 2001).

Given the extent to which nuptial gifts have been characterized as gifts of nutrition from males to females (Vahed 1998, 2007; Gwynne 2008), it is initially surprising that female ladybirds should gain no apparent nutritional benefit from nuptial feeding, even under conditions of extreme nutritional duress. Our results suggest that any benefit for female longevity from spermatophore ingestion must be smaller than 12% for a single spermatophore or smaller than 6% for multiple spermatophores (see Table 2). Yet, despite the overall weakly positive relation between female longevity and mating rate in nuptial feeding arthropods (Arnqvist & Nilsson 2000), the finding of no apparent nutritional benefit from nuptial feeding is in fact common for studies that manipulate nuptial feeding independent of mating rate (reviewed by Boggs 1995; Vahed 1998, 2007; Arnqvist & Rowe 2005; Gwynne 2008). What does this imply about the fitness value of nuptial feeding for females?

One possible explanation is that nuptial feeding offers minute benefits not detected by experimenters but favoured by selection. With several studies that fail to detect any benefit (reviewed by Vahed 1998, 2007; but see Gwynne 2008), however, this hypothesis appears increasingly unlikely to be general. Sakaluk (2000) proposed an alternative explanation: females are selected in a non-mating context to eat items with certain chemical or physical properties, and nuptial gifts that exploits this 'gustatory response' are favoured by selection. The nuptial gift of a decorated cricket appears designed for this purpose: it has a distinct amino acid profile high in free amino acids, which can be tasted, and low in essential amino acids (Warwick 1999), making the spermatophylax nuptial gift an irresistibly delicious, albeit worthless, food item (Sakaluk et al. 2006). Likewise there is evidence that females' foraging instinct maintains nuptial feeding in a spider (Bilde et al. 2007). Currently, we have no detailed data on spermatophore composition in coccinellid beetles, and the hypothesis that the ladybird spermatophore has evolved to exploit a gustatory response remains untested.

We have earlier raised the alternative that spermatophores might contain signals that are detrimental to females, such that nuptial feeding decreases longevity. Our data do not support this hypothesis. The possibility remains, though, that nuptial gifts transfer signalling proteins that affect female reproduction or re-mating behaviour, and any costs or benefits of such effects would not have been detected in this study. Our previous work has shown that eating a single spermatophore advances oviposition by approx. 1 d, compared to females prevented from eating a spermatophore, but that overall fecundity does not increase from spermatophore feeding (Perry & Rowe, in press). It is not known whether this shift in reproductive schedule is beneficial or costly to females in the wild.

We have also found that females become more resistant to re-mating in the first day after spermatophore feeding, though the effect disappears within 3 d (Perry & Rowe, in press). Males are likely to benefit from increased re-mating resistance in their mates if it leads to reduced sperm competition. The current study suggests that a decreased mating rate will have minimal direct costs or benefits to female longevity.

Although we found no indication of a longevity cost for females due to additional copulations (any such cost must have been smaller than 18% for spermatophore-feeding females or 15% for non-feeding females; see Table 2), our experiment was designed to detect only direct costs arising from mating. It remains possible that female *A. bipunctata* do experience other costs of mating that our study was not designed to detect, such as reduced foraging success or increased predation rate during mating (e.g. Rowe 1994), increased transmission of pathogens (e.g. Hurst et al. 1995) or the cost of replacing stored

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sperm with sperm from a less desirable mate (Arnqvist & Rowe 2005). Costs like these could account for the fact that females often resist mating vigorously and at length (Majerus 1994). Finally, because females were deprived of food after the mating treatment, costs arising from a trade-off between reproduction and longevity were removed; there is, in fact, evidence of such a trade-off in two coccinellid species (Omkar & Mishra 2005). If Acps transferred from males induce increased or accelerated oogenesis in females, females may experience decreased longevity as a result, and our study would not detect this type of indirect mating cost.

A recent study suggested that spermatophore feeding might mitigate mating costs in another ladybird. Omkar & Mishra (2005) manipulated mating rate in three ladybirds and found increased fecundity and decreased longevity with increased mating in two species that do not eject spermatophores (*Propylea dissecta* and *Cheilomenes sexmaculata*) but no effect in the third species (*Coccinella septempunctata*). Although the authors propose that spermatophore feeding in *C. septempunctata* might explain the absence of costs from multiple mating, we think this unlikely for two reasons. First, spermatophore feeding is infrequent (Obata & Johki 1991) or absent (Omkar & Srivastava 2002; own data) in this species. Second, both female and male *P. dissecta* and *C. sexmaculata* had depressed longevity from multiple mating; in contrast, longevity was not affected by multiple mating in either male or female *C. septempunctata*, but the hypothesis that spermatophore feeding accounts for the absence of reduced longevity addresses females only.

An important question is whether the mating rates we tested were high enough to educe mating costs. Although there is no data on the optimum mating rate for female *A. bipunctata*, we note that the intermediate and elevated mating rates we tested are within the range of rates reported from field observations (Brakefield 1984; Webberley et al. 2006). Furthermore, many features of the laboratory environment in this study were similar to the natural conditions experienced by *A. bipunctata*: temperature, humidity and the aphid diet provided before the mating treatment. Thus, the experimental setting should have detected realistic direct fitness costs to females from additional copulations.

Nuptial gifts have often been described as male donations with direct benefits to females; in contrast to this view, our results contribute to a growing appreciation of the possibility of little or no direct benefit of nuptial feeding to females in some species. In contrast to related coccinellid species, *A. bipunctata* females appear to experience no direct longevity costs of mating for moderate mating rates. A next step for understanding nuptial feeding in ladybird beetles is to address the maintenance of spermatophore feeding in spite of its minimal contribution to female fitness.

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Literature Cited

Andersson, M. & Simmons, L. W. 2006: Sexual selection and mate choice. Trends Ecol. Evol. **21**, 296—302.

Arnqvist, G. & Nilsson, T. 2000: The evolution of polyandry: multiple mating and female fitness in insects. Anim. Behav. **60**, 145—164.

Arnqvist, G. & Rowe, L.2005: Sexual Conflict. Princeton Univ. Press, Princeton, NJ.

Baer, B., Morgan, E. D. & Schmid-Hempel, P. 2001: A nonspecific fatty acid within the bumblebee mating plug prevents females from remating. Proc. Natl Acad. Sci. USA **98**, 3926—3928.

Bilde, T., Tuni, C., Elsayed, R., Pekar, S. & Toft, S. 2007: Nuptial gifts of male spiders: sensory exploitation of the female's maternal care instinct or foraging motivation? Anim. Behav. **73**, 267—273.

Blanckenhorn, W. U., Hosken, D. J., Martin, O. Y., Reim, C., Teuschl, Y. & Ward, P. I. 2002: The costs of copulating in the dung fly *Sepsis cynipsea*. Behav. Ecol. **13**, 353—358.

Boggs, C. L. 1995: Male nuptial gifts: phenotypic consequences and evolutionary implications. In: Insect Reproduction (Leather, S. R. & Hardie, J., eds). CRC Press, Boca Raton, FL, pp. 215—242.

Bonduriansky, R., Wheeler, J. & Rowe, L. 2005: Ejaculate feeding and female fitness in the sexually dimorphic fly *Prochyliza xanthostoma* (Diptera: Piophilidae). Anim. Behav. **69**, 489—497.

Brakefield, P. M. 1984: Ecological studies on the polymorphic ladybird *Adalia bipunctata* in the Netherlands. 1. Population biology and geographical variation of melanism. J. Anim. Ecol. **53**, 761—774.

Brown, W. D. 1997: Courtship feeding in tree crickets increases insemination and female reproductive lifespan. Anim. Behav. **54**, 1369—1382.

Chapman, T., Liddle, L. F., Kalb, J. M., Wolfner, M. F. & Partridge, L. 1995: Cost of mating in *Drosophila melanogaster* females is mediated by male accessory-gland products. Nature **373**, 241—244.

Chapman, T., Arnqvist, G., Bangham, J. & Rowe, L. 2003: Response to Eberhard and Cordero, and Córdoba-Aguilar and Contreras-Garduño: sexual conflict and female choice. Trends Ecol. Evol. **18**, 440—441.

Crudgington, H. S. & Siva-Jothy, M. T. 2000: Genital damage, kicking and early death – the battle of the sexes takes a sinister turn in the bean weevil. Nature **407**, 855—856.

Engqvist, L. 2007a: Nuptial food gifts influence female egg production in the scorpionfly *Panorpa cognata*. Ecol. Entomol. **32**, 327—332.

Engqvist, L. 2007b: Nuptial gift consumption influences female remating in a scorpionfly: male or female control of mating rate? Evol. Ecol. **21**, 49—61.

Evans, E. W. 2003: Searching and reproductive behaviour of female aphidophagous ladybirds (Coleoptera: Coccinellidae): a review. Eur. J. Entomol. **100**, 1—10.

Gwynne, D. T. 1984: Courtship feeding increases female reproductive success in bush crickets. Nature **307**, 361—363.

Gwynne, D. T. 1988: Courtship feeding and the fitness of female katydids (Orthoptera: Tettigoniidae). Evolution **42**, 545—555.

Gwynne, D. T. 2008: Sexual conflict over nuptial gifts in insects. Annu. Rev. Entomol. **53**, 5.1—5.19.

Gwynne, D. T., Bowen, B. J. & Codd, C. G. 1984: The function of the katydid spermatophore and its role in fecundity and insemination (Orthoptera: Tettigoniidae). Aust. J. Zool. **32**, 15—22.

Haddrill, P. R., Shuker, D. M., Mayes, S. & Majerus, M. 2007: Temporal effects of multiple mating on components of fitness in the two-spot ladybird, *Adalia bipunctata* (Coleoptera: Coccinellidae). Eur. J. Entomol. **104**, 393—398.

Hoenig, J. M. & Heisey, D. M. 2001: The abuse of power: the pervasive fallacy of power calculations for data analysis. Am. Statist. **55**, 19—24.

Hurst, G. D. D., Sharpe, R. G., Broomfield, A. H., Walker, L. E., Majerus, T. M. O., Zakharov, I. A. & Majerus, M. E. N.1995: Sexually transmitted disease in a promiscuous insect, *Adalia bipunctata*. Ecol. Entomol. **20**, 230—236.

Kasuya, E. & Sato, N. 1998: Effects of the consumption of male spermatophylax on the oviposition schedule of females in the decorated cricket, *Gryllodes sigillatus*. Zool. Sci. **15**, 127—130.

Kokko, H., Jennions, M. D. & Brooks, R. 2006: Unifying and testing models of sexual selection. Annu. Rev. Ecol. Syst. **37**, 43—66.

Majerus, M. E. N.1994: Ladybirds. HarperCollins, London.

Mondet, C., Gautier, P., Abed-Vieillard, D., Chaudy, S., Brossut, R. & Farine, J.-P.2006: Could male tergal secretions be considered as nuptial gifts in the Madeira Cockroach, *Leucophaea maderae* (Blaberidae: Oxyhaloinae)? Paper presented at the ISBE 2006: 11th Congress of the International Society for Behavioural Ecology, Tours, France.

Obata, S. & Johki, Y. 1991: Comparative study on copulatory behaviour in four species of aphidophagous ladybirds. In: Behaviour and Impact of Aphidophaga (Polgár, L., Chambers, R. J., Dixon, A. F. G. & Hodek, I., eds). SPB Academic Publishing, The Hague, pp. 207—212.

Omkar & Mishra, G. 2005: Mating in aphidophagous ladybirds: costs and benefits. J. Appl. Entomol. **129**, 432—436.

Omkar & Srivastava, S. 2002: The reproductive behaviour of an aphidophagous ladybeetle, *Coccinella septempunctata* (Coleoptera: Coccinellidae). Eur. J. Entomol. **99**, 465—470.

Omkar, Mishra, G. & Singh, S. K. 2006: Optimal number of matings in two aphidophagous ladybirds. Ecol. Entomol. **31**, 1—4.

Ono, T., Ando, C. & Kondo, Y. 2004: Effect of metanotal secretion ingestion on oviposition in a tree cricket, *Truljalia hibinonis* (Orthoptera: Gryllidae). Entomol. Sci. **7**, 9—13.

Parker, G. A. 1979: Sexual selection and sexual conflict. In: Sexual Selection and Reproductive Competition in Insects (Blum, M. S. & Blum, N. A., eds). Academic Press, New York, pp. 123—166.

Parker, G. A. 2006: Sexual conflict over mating and fertilization: an overview. Philos. Trans. R. Soc. Lond. B **361**, 235—259.

Perry, J.C. & Rowe, L. (in press): Ingested spermatophores accelerate reproduction and increase mating resistance but are not a source of sexual conflict. Anim. Behav., in press.

Reinhold, K. 1999: Paternal investment in *Poecilimon veluchianus* bushcrickets: beneficial effects of nuptial feeding on offspring viability. Behav. Ecol. Sociobiol. **45**, 293—299.

Rönn, J., Katvala, M. & Arnqvist, G. 2006: The costs of mating and egg production in *Callosobruchus* seed beetles. Anim. Behav. **72**, 335—342.

Rowe, L. 1994: The cost of mating and mate choice in water striders. Anim. Behav. **48**, 1049—1056.

Rowe, L. & Day, T. 2006: Detecting sexual conflict and sexually antagonistic coevolution. Philos. Trans. R. Soc. Lond. B **361**, 277—285.

Ethology **114** (2008) 504–511

Sakaluk, S. K. 2000: Sensory exploitation as an evolutionary origin to nuptial food gifts in insects. Proc. R. Soc. Lond. B **267**, 339—343.

Sakaluk, S. K., Avery, R. L. & Weddle, C. B. 2006: Cryptic sexual conflict in gift-giving insects: chasing the chase-away. Am. Nat. **167**, 94—104.

Simmons, L. W. 1988: The contribution of multiple mating and spermatophore consumption to the lifetime reproductive success of female field crickets (*Gryllus bimaculatus*). Ecol. Entomol. **13**, 57—69.

Simmons, L. W. 1990: Nuptial feeding in tettigoniids: male costs and the rates of fecundity increase. Behav. Ecol. Sociobiol. **27**, 43—47.

Simmons, L. W. & Parker, G. A. 1989: Nuptial feeding in insects: mating effort versus paternal investment. Ethology **81**, 332—343.

Sloggett, J. J. & Majerus, M. E. N. 2000: Habitat preferences and diet in the predatory Coccinellidae (Coleoptera): an evolutionary perspective. Biol. J. Linn. Soc. **70**, 63—88.

Stearns, S. C. 1992: The Evolution of Life Histories. Oxford Univ. Press, Oxford.

Vahed, K. 1998: The function of nuptial feeding in insects: a review of empirical studies. Biol. Rev. **73**, 43—78.

Vahed, K. 2003: Increases in egg production in multiply mated female bushcrickets *Leptophyes punctatissima* are not due to substances in the nuptial gift. Ecol. Entomol. **28**, 124—128.

Vahed, K. 2007: All that glisters is not gold: sensory bias, sexual conflict and nuptial feeding in insects and spiders. Ethology **113**, 105—127.

Vahed, K. & Gilbert, F. S. 1997: No effect of nuptial gift consumption on female reproductive output in the bushcricket *Leptophyes laticauda* Friv. Ecol. Entomol. **22**, 479—482.

Warwick, S.1999: Nutritional regulation and spermatophylax donation in the mating system of Gryllodes sigillatus (Orthoptera: Gryllidae). PhD Thesis, Univ. of Oxford, Oxford.

Webberley, K. M., Buszko, J., Isham, V. & Hurst, G. D. D. 2006: Sexually transmitted disease epidemics in a natural insect population. J. Anim. Ecol. **75**, 33—43.

Wedell, N. & Arak, A. 1989: The wartbiter spermatophore and its effect on female reproductive output (Orthoptera: Tettigoniidae, *Decticus verrucivorus*). Behav. Ecol. Sociobiol. **24**, 117—125.

Wigby, S. & Chapman, T. 2005: Sex peptide causes mating costs in female *Drosophila melanogaster*. Curr. Biol. **15**, 316—321.

Will, M. W. & Sakaluk, S. K. 1994: Courtship feeding in decorated crickets: is the spermatophylax a sham? Anim. Behav. **48**, 1309—1315.

Wolfner, M. F. 1997: Tokens of love: functions and regulation of *Drosophila* male accessory gland products. Insect Biochem. Mol. Biol. **27**, 179—192.

Zeh, J. A. & Zeh, D. W. 2003: Toward a new sexual selection paradigm: polyandry, conflict and incompatibility. Ethology **109**, 929—950.