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Male age does not affect female fitness in a polyandrous beetle, *Callosobruchus maculatus*

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Males in different taxa are likely to suffer from a reduction in the quantity and/or quality of their sperm with age. This predicts age-related direct and indirect effects on female fitness. Hence, females may be selected to avoid matings with old males, or to employ alternative mating tactics, such as polyandry, to avoid fertilization by sperm of older males. In contrast, 'viability indicator' models of mate choice predict female preference for old males that have proven their survival ability and signal more reliably. We used a polygamous seed beetle, *Callosobruchus maculatus*, to test for the effects of male age on male mating success and examine the relationship between male age and female fitness, measured as female life span, lifetime fecundity, hatching success of eggs, larval development rate and egg-to-adult survival of offspring. Furthermore, we tested the hypothesis that polyandry may protect females against low numbers of functional sperm produced by old males. We report, contrary to previous findings, that male mating success indeed decreases with male age in this species. However, mating with older males did not in any way compromise female fitness and, consequently, we found no support for the idea that polyandry helps females reduce any costs of mating with older males.

Keywords: *Callosobruchus maculatus*; mating success; multiple mating; seed beetle; senescence; sexual selection

The effect of male age on female fitness and mate choice is a long-standing controversy in the evolutionary studies of mating behaviour (Trivers 1972; Manning 1985; Price & Hansen 1998; Beck & Powell 2000; Jones et al. 2000; Brooks & Kemp 2001; Radwan 2003; Jones & Elgar 2004). Age-related models of mate choice maintain that females should preferentially mate with older males because they have proven survival ability (Trivers 1972; Manning 1985; Kokko & Lindström 1996) and because the reliability of male signalling increases with age (Proulx et al. 2002). However, the quality of male gametes is likely to decline with age as the number of germ cell divisions, which lead to de novo mutations in the male germ line, increases with age (Crow 1993, 1997; Drost & Lee 1995; Hansen & Price 1995; Radwan 2003). Such effects may be particularly important in highly promiscuous species, which produce larger numbers of sperm (Bartosch-Härlid et al. 2003). Thus, females that mate with older males may have a higher chance of being fertilized by gametes of decreased genetic quality, resulting in lower fitness of their offspring. There is some evidence that progeny of older males indeed show reduced survival (neonatal death rate in Norway rats, *Rattus norvegicus*: Serre & Robaire 1998; larval viability in *Drosophila melanogaster*: Price & Hansen 1998; hatching rate in sandflies *Lutzomyia longipalpis*: Jones et al. 2000; see Radwan 2003 for review). Furthermore, older males may suffer from sperm depletion (Wedell et al. 2002; Wedell & Ritchie 2004), especially if the ability to replenish sperm decreases with age (e.g. Radwan & Bogacz 2000). Additionally, in species where sperm is stored for a considerable amount of time, older males will possess older sperm, which may have negative effects on zygote viability (Crow 1997; Siva-Jothy 2000; Reinhardt & Siva-Jothy 2005). Collectively, these arguments suggest that females may be selected to avoid matings

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with older males (Hansen & Price 1995; Jones et al. 2000), or to employ alternative mating tactics, such as polyandry (Radwan 2003; Radwan et al. 2005), to avoid fertilization by sperm of older males.

We examined the effect of male age on female fitness and male mating success in a polyandrous beetle, *Callosobruchus maculatus*. This species has been widely used in recent years to investigate different aspects of male–female coevolution and the evolution of mating systems (e.g. Fox 1993; Savalli & Fox 1999; Crudgington & Siva-Jothy 2000; Eady et al. 2000; Arnqvist et al. 2005; Rönn et al. 2006). Mating in *C. maculatus* is costly to females due to internal injuries inflicted by spiky male genitalia (Crudgington & Siva-Jothy 2000). Such injuries reduce female life span (Crudgington & Siva-Jothy 2000) and lower lifetime fecundity (Edvardsson & Tregenza 2005). Additional costs of mating probably come from toxic seminal compounds, which have been documented in a related bruchid beetle, *Acanthoscelides obtectus* (Das et al. 1980). Jointly, these studies suggest that polyandry can be disadvantageous to females due to the cumulative costs of mating. However, *C. maculatus* females are also likely to enjoy direct benefits of multiple mating. *C. maculatus* males produce large ejaculates that constitute up to 10% of a male's body mass (Savalli & Fox 1998). The size of the spermatophore is known to affect female fecundity (Savalli & Fox 1999; Eady & Brown 2000), and the direction of this effect may depend on male age at mating (Eady & Brown 2000). Females can obtain water and, possibly, nutrients from male ejaculates (Savalli & Fox 1999) and a single mating can increase female life span (Rönn et al. 2006). Another recent study has shown that females are more likely to remate when dehydrated (Edvardsson 2005). Consequently, the relationship between mating rate and female fitness in *C. maculatus* is complex and nonlinear as was shown by Arnqvist et al. (2005).

Several hypotheses suggest that *C. maculatus* females may benefit from, or at least mitigate the costs of, multiple mating by avoiding age-related decline in ejaculate quality (Radwan 2003). First, old males may suffer from sperm depletion, since in this species, like in many other taxa, ejaculate size decreases with the number of matings (Savalli & Fox 1999). The size of the ejaculate, however, is dependent on the time interval between matings (Fox et al. 1995), which suggests that males produce sperm throughout their life. The effect of a decrease in ejaculate size can be amplified if there is an age-related decrease in sperm replenishment (e.g. Radwan & Bogacz 2000). Second, older males may produce subfertile sperm even when capable of mating, if spermatogenic tissue is ageing faster than somatic tissue (Radwan 2003), leading to decreased hatching success. *C. maculatus* is a short-lived but promiscuous species, with both males and females mating multiply during their reproductive cycle. As this species is a short-lived one, a decline in genetic quality of sperm due to mutagenesis may not be severe; however, promiscuity may accelerate ageing of spermatogenic tissue. Two studies suggest that male mating history may have an effect on female life span (Paukku & Kotiaho 2005) as well as female fecundity (Ofuya 1995) in *C. maculatus*. Furthermore, Jones et al. (2000) found reduced hatching success of eggs produced from old males in a similarly short-lived sandfly, *L. longipalpis*. When genetic quality of male gametes declines with age, females may benefit from multiple mating if sperm competitiveness also declines with age. Recent studies indicate that older males indeed lose out to younger males in sperm competition (Schäfer & Uhl 2002; Radwan et al. 2005; but see LaMunyon 2001).

The goal of this study was three-fold: (1) we examined the effect of male age on female fitness, measured as female life span, lifetime fecundity, hatching success, larval development rate, egg-to-adult survival of offspring and total offspring production; (2) we tested the hypothesis that polyandry may protect females against the infertility of older males; and (3) we tested for the effect of male age on male mating success.

METHODS

We used a strain of the seed beetle *C. maculatus* originating from Mali. Beetles were obtained from Peter Credland (University of London) and cultured on black-eyed beans, *Vigna unguiculata*, at 30 ± 0.5°C and 45% (±10%) RH with a 12:12 h light:dark cycle prior to and during the experiment. These beetles are capital breeders, that is they complete their life cycle using only resources acquired during their larval stage (Fox 1993). They are cosmopolitan pests of legume storages, such that laboratory conditions are a good approximation of their natural environment (Fox et al. 2003). The beetle culture used in this study was maintained in our laboratory for 30 generations prior to the experiments. All females used in this experiment were virgin and 1 day post-eclosion. Males were derived from two groups, the young males, which were also virgin and 1 day post-eclosion, and the old males, which were nonvirgin and 5 days post-eclosion. The old males were kept in groups together with nonexperimental females for 4 days (five males and five females in 90-mm petri vials) and as males and females of *C. maculatus* mate multiply during their life span (Ofuya 1995; Arnqvist et al. 2005; Rönn et al. 2006), we expected these males to mate multiply during this period. After 4 days, males were separated and kept individually in 30-mm petri vials for 24 h prior to the experiment. Male mortality after 5 days was between 50% and 70%. We thus ensured that old males used in our experiment were: (1) at the very end of their reproductive cycle; (2) mated multiply to several different females so that any potential effects due to sperm depletion or age-related infertility would be revealed; and (3) passed through viability selection. Such a set-up did not allow testing for the effects of sperm age, but such effects are unlikely to play a role in this system, since males are extremely promiscuous and emerge ready to copulate (Fox et al. 2003). This set-up, however, imitates the natural situation when a newly matured virgin female can be mated either by an old multiply mated male or a recently hatched virgin male.

We used 60 old males and 50 young males to examine the effect of male age on male mating success. Young and old males were treated as described above and randomly paired with virgin females. Pairs were introduced to each other in 30-mm petri vials and their behaviour was monitored for 30 min. We recorded whether mating occurred or not, how fast males were able to achieve mating after being introduced to a female (latency to mating) and for how long the pairs copulated. Males and females were kept individually for 24 h in 30-mm petri vials prior to the experiment.

We used the following design to test for the effect of male age on female fitness. We randomly assigned beetles into six different treatment groups with 20 females each, where females were mated either once or twice to young and/or old males in the following combinations (sample size): young ($N = 20$), old ($N = 20$), young–young ($N = 23$), old–old ($N = 23$), old–young ($N = 20$) and young–old ($N = 20$). Each male was used only once; consequently, we used 212 males and 126 females in these experiments. We observed all matings to control for female mating rate and males were discarded after termination of a mating. In the double-mating groups we allowed for 24 h between matings. Not all females in the double-mating treatment remated and those were discarded and excluded from the analysis. After a successful first mating (single-mating treatment) or a successful second mating (double-mating treatment), females were transferred into 90-mm petri vials with 10 g of beans and kept individually until their natural death. Thus, only males that succeeded in mating with a female were used in this experiment. By comparing the fitness and offspring performance (survival and development rate) of females mated to old males that passed viability and mating selection to that of females mated to young virgin males, we increase the chance to distinguish between the positive and negative effects related to genetic and/or phenotypic quality, which could be conferred by old males. The underlying idea is that the quality of sperm from old males is independent from the effects of genes resulting in high viability. According to this logic, if negative effects through reduced quantity of adequate sperm or other ejaculate components exist, we expect direct effects on female fitness through decreased fertility or fecundity. In contrast, if old males confer indirect beneficial effects on offspring through enhanced viability, we would find this result reflected in higher hatching success or egg-to-adult survival.

This design allows us to test the effect of male age on female and offspring fitness both in single- and double-mating situations. Furthermore, we directly test whether polyandry is involved in mitigating fitness costs of male age effects inflicted on females. According to the hypothesis put forward by Radwan (2003), polyandry may help females to avoid costs caused by a mating with an old male. These costs may be due to either indirect (i.e. increased mutagenesis) or direct accelerated ageing of spermatogenic tissue (Radwan 2003). Under this scenario, polyandry is predicted to equally rescue female fitness in mixed young–old and old–young treatments if reduction in sperm quality correlates with a reduced success in sperm competition (cf. Tregenza & Wedell 2002). If no such correlation exists, the rescue will be incomplete, and will depend on mating order, since second males have a fertilization advantage in this species (Eady 1991, 1994).

We recorded female life span (number of days), female lifetime fecundity and the number of hatched and unhatched eggs. The number of eclosing offspring was counted on two occasions (23 and 35 days after the first mating) to obtain a relative measure of the development rate. All offspring hatched after 35 days (the first beetles hatch after 21 days at 30°C). We then estimated the hatching rate, egg-to-adult survival (total number of offspring/hatched eggs) and larval development rate (number of eclosed offspring on day 23/total number of offspring).

The effects of treatment on hatching rate, development rate and egg-to-adult survival were analysed in GENSTAT 7 using EXTRABINOMIAL procedure for generalized linear models with binomial error distribution and logit link function with Williams correction for overdispersion (Crawley 1993). Other statistical analyses of female fitness data were performed in JMP 6 (SAS Institute) using general linear models. The assumptions of the linear models were checked by plotting model residuals against values predicted by the normal distribution in JMP 6 and by Levene's test of the equality of variances. To compensate for multiple comparisons, we controlled for False Discovery Rate (FDR) using R-based QVALUE software (Storey 2002) with $\pi_0 = 1$ and consequently report adjusted P values (i.e. Q values; Storey 2003). The FDR procedure is commonly used in different areas of biology and was strongly recommended for use in studies of animal behaviour instead of the traditional Bonferroni correction (Nakagawa 2004). Additionally, we also report effect sizes for our data, and we believe that the combination of our statistical approach, effect sizes and powerful FDR correction, together with the graphical illustration based on box plots, enables us to draw biologically meaningful conclusions from nonsignificant parts of our results. We used nonparametric statistics to analyse the data on male mating behaviour.

RESULTS

There was no significant difference in latency to mating between old and young males (Mann–Whitney U test: $U = 1405$, $N_1 = 49$, $N_2 = 48$, $P = 0.098$) (old males: median = 1.24 min, lower quartile = 0.46 min, upper

Table 1. The effect of the single mating treatment (male age; see text for treatment details) on female life span, lifetime fecundity, total offspring production, hatching rate, egg-to-adult survival of offspring and larval development rate

Effect	*F*/LLR	*df*	Effect size	*P*	*Q*
General linear model					
Life span	0.058	1, 39	0.038	0.811	0.811
Fecundity	0.344	1, 39	0.094	0.561	0.673
Offspring	1.624	1, 39	0.203	0.210	0.55
General linearized model					
Hatching rate	5.02	1, 38	0.351	0.025	0.15
Survival	1.19	1, 38	0.174	0.275	0.55
Development	0.54	1, 38	0.117	0.464	0.637

We report *F*/LLR ratios and degrees of freedom from general linear and linearized models with *P* and *Q* values (adjustment for False Discovery Rate), as well as effect sizes (Fisher's *z*) for each of the variables.

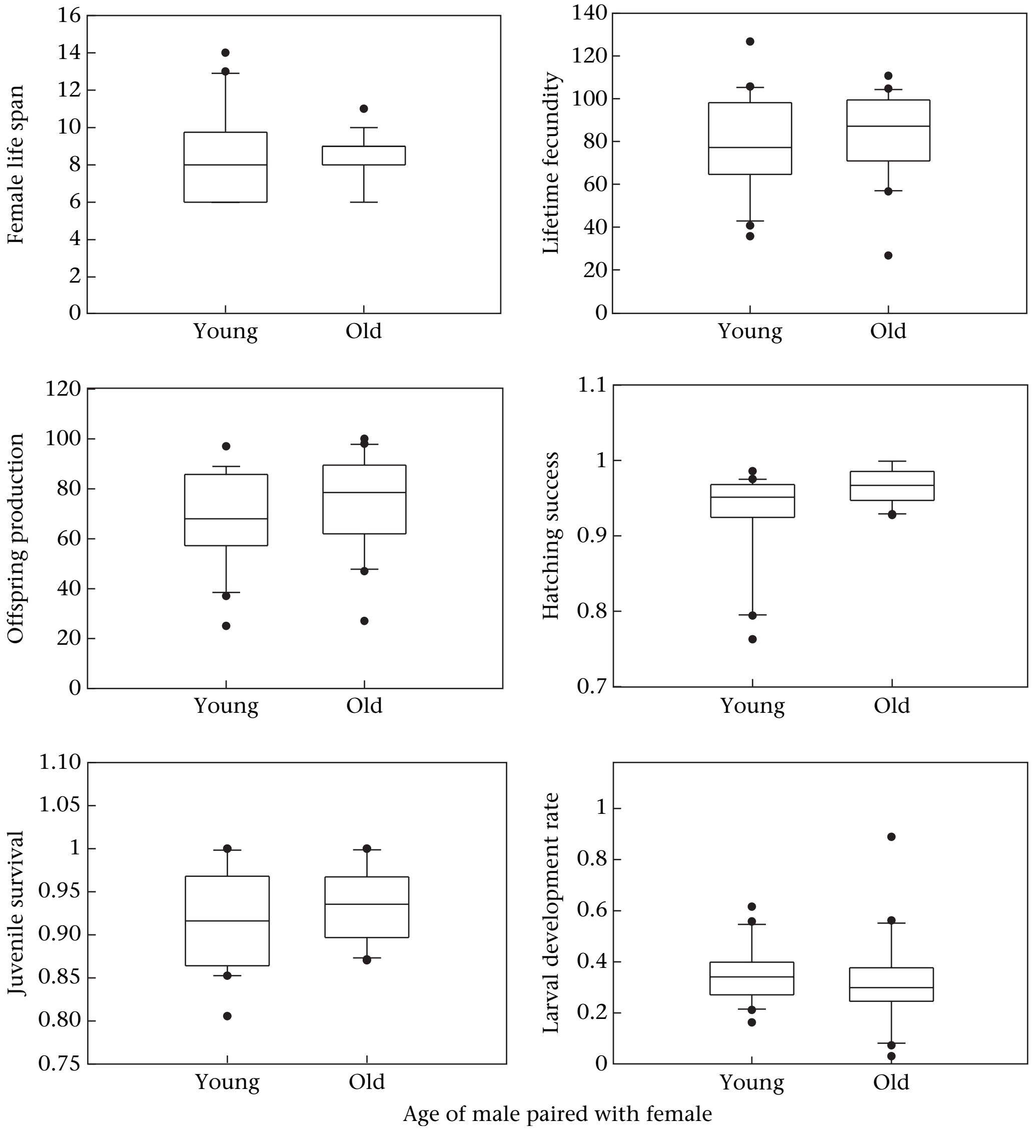

Figure 1. The effect of a single mating with an old or a young male on female fitness, with each individual figure depicting box plots representing the median and the 25% and 75% quartiles for one female fitness variable. Outliers beyond the 10th and 90th percentiles (whiskers) are presented. Female life span was measured in days, juvenile survival as the rate of the total number of offspring/hatched eggs, lifetime fecundity as the total number of eggs laid, hatching success as the rate of the number of hatched eggs/total number of eggs produced, and larval development as the number of eclosed offspring on day 23/total number of offspring.

quartile = 3.24 min; young males: median = 0.56 min, lower quartile = 0.26 min, upper quartile = 2.16 min). Old males, however, copulated longer than young males (old males: median = 10.75 min, lower quartile = 6.84 min, upper quartile = 14.9 min; young males: 7.22 min, lower quartile = 5.89 min, upper quartile = 9.13 min; Mann–Whitney U test: $U = 1922$, $N_1 = 49$, $N_2 = 48$, $P < 0.001$). Males of different ages were also significantly different in their ability to achieve copulations (Pearson chi-square: $\chi^2_1 = 6.123$, $P = 0.013$), with old males more often failing in achieving a mating (20.97%) than young ones (4%).

There was no effect of male age on female life span, lifetime fecundity, total number of offspring produced, hatching success, larval development rate and egg-to-adult survival of offspring (see Table 1 for F and deviance ratios, effect sizes as well as P and Q values) after a single mating (see Fig. 1). Moreover, there was no effect of our double-mating treatment on any of the female fitness variables measured (see Table 2, Fig. 2). Hatching success was significant in single-mated females, suggesting that females mated to old males had higher hatching success (Fig. 1) but this effect was not robust and disappeared after the FDR correction was applied (see Table 1). Still we decided to further test for this effect by directly comparing young–young and old–old treatments from double-mated females. This comparison did not support a trend for mates of old males to have higher hatching success ($LLR_{1,43} = 0.01$, $P = 0.915$; see also Fig. 2), thus confirming our conclusion based on the FDR correction that there is no effect of male age on hatching success. There was a significant effect of mating rate (mated once or twice) on female

Table 2. The individual effects of the first (1. ♂) and the second male (2. ♂) (see Methods for details on the order of presentation of males) in the double-mating treatment on female life span, lifetime fecundity, total offspring production, hatching rate, egg-to-adult survival of offspring and larval development rate

Effect	1. ♂	2. ♂	1. ♂×2. ♂	1. ♂	2. ♂	1. ♂×2. ♂	1. ♂	2. ♂	1. ♂×2. ♂
ANOVA									
Source		Life span			Lifetime fecundity			Total offspring production	
F	3.066	0.393	0.048	3.745	1.510	0.606	2.629	1.843	0.020
df	1, 82	1, 82	1, 82	1, 82	1, 82	1, 82	1, 82	1, 82	1, 82
Effect size	0.187	0.068	0.024	0.208	0.133	0.084	0.175	0.147	0.015
P value	0.084	0.533	0.828	0.056	0.223	0.439	0.109	0.178	0.889
Q value	0.164	0.709	0.889	0.164	0.446	0.658	0.164	0.446	0.889
General linearized model									
Source		Hatching rate			Juvenile survival			Larval development rate	
LLR	0.09	0.14	0.76	2.94	0.22	1.51	0.28	4.09	1.61
df	1, 82	1, 82	1, 82	1, 82	1, 82	1, 82	1, 82	1, 82	1, 82
Effect size	0.032	0.041	0.094	0.185	0.051	0.133	0.057	0.217	0.137
P value	0.767	0.709	0.382	0.086	0.642	0.220	0.595	0.043	0.205
Q value	0.767	0.709	0.658	0.174	0.709	0.658	0.714	0.258	0.658

We report the results of an analysis of variance and the results of a linearized model with binomial errors and a logit link function with *P* and *Q* values (adjustment for False Discovery Rate), as well as effect sizes (Fisher's *z*) for each of the variables.

lifetime fecundity, when life span was used as a covariate ($F_{1,123} = 6.021$, $P = 0.016$). Single-mated females laid on average 81.40 (SE ± 3.30) eggs, while double-mated females laid 89.37 (SE ± 2.47) eggs. However, this effect was not found to be significant for the total number of offspring produced ($F_{1,123} = 3.028$, $P = 0.084$). This lack of an effect of mating rate on net offspring production was due to the lower survival rate of the offspring of double-mated females (single-mated females: mean ± SE = 0.93 ± 0.008; double-mated females: 0.91 ± 0.006, $F_{1,123} = 5.803$, $P = 0.017$), rather than the difference in hatching success ($F_{1,123} = 0.011$, $P = 0.916$).

DISCUSSION

Ageing can reduce male persistence and hence decrease the ability of males to overcome female resistance to mating. Likewise, females may actively discriminate against older males. We found that older males are more likely to fail in mating with virgin females. Savalli & Fox (1999) tested the effect of male age on male mating success in a different strain of *C. maculatus* and found no effect of male age on the probability of mating in a competitive setting. Such a difference between the studies can be attributed to differences between populations and experimental protocols (e.g. Eady & Brown 2000). In the study carried out by Savalli & Fox (1999), both young (1 day posteclosion) and old (7 days posteclosion) males were virgin. Hence, old males in their study were in a better condition compared with the old males in our experiment, where males had the opportunity to mate multiply for 4 days, causing higher water and energy loss. Since it is unlikely that males ever survive unmated to the age of 7 days in this system, we argue that our design was a closer approximation of the natural conditions.

Female preference, defined broadly as any female trait that biases mating success towards males of certain phenotypes (Maynard-Smith 1987; Pomiankowski 1988), may evolve via different evolutionary routes (Eberhard 1996; Arnqvist & Rowe 2005). Females may actively bias matings or fertilizations towards particular male phenotypes (Eberhard 2002), and such a bias may lead to behavioural discrimination against older males (Jones et al. 2000). The same pattern may occur, however, if female resistance is indiscriminate, yet males exhibit phenotypic variation in persistence (Arnqvist & Rowe 2005). Under such a scenario, older senescent males may be selected against because they invest less in sexual advertisement (e.g. Hansen & Price 1995). Interestingly, Jones et al. (2000) suggested that female discrimination against younger males in sandflies may occur because such males are yet incapable of pheromone production. Thus, reduction in sexual advertisement by senescent, as well as newly hatched, males may be a general explanation for the pattern of female preference for middle-aged partners (Jones et al. 2000). Finally, older males can lose out in intrasexual competition, which can, in theory, explain the variation in age-related mating success (Jones & Elgar 2004). This explanation, however, is not applicable to our study because we observe mating bias against older males in the absence of male–male competition.

Hansen & Price (1995) argued that studies that report female preference for older males often use males that are, in practice, of intermediate age. In line with this suggestion, two recent studies reported increased mating success of intermediate-age males compared with old males (sandflies: Jones et al. 2000; hide beetles: Jones & Elgar 2004). These studies, however, also emphasized female discrimination against young males. We did not explicitly test for the female preference for mates of intermediate age in our study. Male *C. maculatus* hatch ready to copulate yet their seminal vesicles are not completely filled (Fox et al. 1995). Thus, it is possible that mating with newly hatched males could have detrimental direct effects on female fitness. Nevertheless, sandfly females do not suffer from decreased fitness when mated to young males and female discrimination against young males may occur simply because young males are unable to signal properly (Jones et al. 2000; see above).

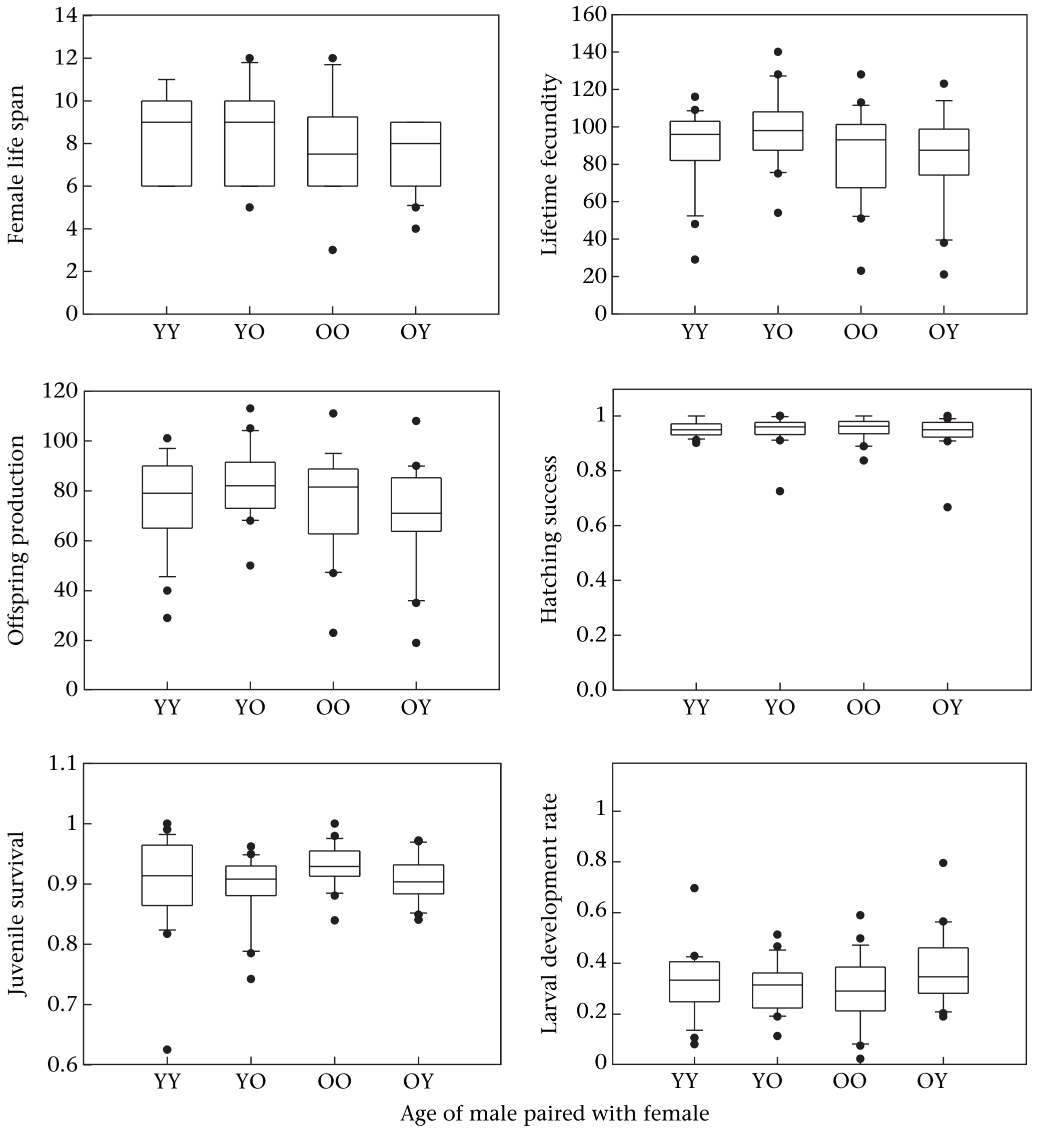

Figure 2. The effects of the double-mating treatment on female fitness, with each individual figure depicting box plots representing the median and the 25% and 75% quartiles for one female fitness variable. Outliers beyond the 10th and 90th percentiles (whiskers) are presented. (Y = young male and O = old male). Female life span was measured in days, juvenile survival as the rate of the total number of offspring/hatched eggs, lifetime fecundity as the total number of eggs laid, hatching success as the rate of the number of hatched eggs/total number of eggs produced, and larval development as the number of eclosed offspring on day 23/total number of offspring.

Old males copulated longer than young males, which could be explained by age-dependent decline in the ability to transfer sperm. Alternatively, increased copulation duration may result from increased investment in reproduction by older males, due to the reduction of residual reproductive success with age (Williams 1966). We, however, found no evidence for increased investment by old males in terms of female fitness. Yet it is possible that males increase their investment in sperm competition, as such an investment could not be detected in our experiment, and there is some evidence that longer copulations may indeed play a role in sperm defence (Edvardsson 2005). Age-dependent investment in sperm competition requires further study as the current evidence is mixed (LaMunyon 2001; Schäfer & Uhl 2002; Radwan et al. 2005).

Theory suggests that old males may suffer from a reduction in the quantity and/or quality of their sperm (Radwan 2003) and other ejaculate components. Surprisingly, we found no direct or indirect effects of male age on female fitness correlates, including female lifetime reproductive success and offspring survival. In *C. maculatus*, the size of the ejaculate has been repeatedly shown to affect female fitness (Savalli & Fox 1999; Eady & Brown 2000; Edvardsson 2005) and the quantity of the ejaculate declines dramatically with consecutive matings (Savalli & Fox 1999) and with age (Fox et al. 1995). This predicts strong age-related direct effects on female fitness in this system. In our experimental design, we allowed old males to replenish their sperm for 24 h prior to mating, thus potentially enabling them to transfer a sufficiently large ejaculate to females. This may account for the fact that we found no direct effects of male age on female fitness despite the results of previous studies suggesting such costs to females. Thus, our results suggest the length of the interval between copulations can be more important in determining the size of ejaculate than male age per se.

Old males used in our experiment passed through strong viability selection (i.e. differential mortality) and mating selection, which could result in potential indirect genetic benefits to females through increased viability of their offspring. However, there was also no effect of male age on offspring egg-to-adult survival and development rate, suggesting that old males do not confer advantage in terms of indirect benefits as measured in this study.

We note that the scope of our experiment and the effect sizes observed strongly suggest that the lack of significant effects does not result from low statistical power. This implies that paternal age does not have immediate fitness consequences to females in this system, and, therefore, may not select for female pre- or postcopulatory preference. It is likely that the number of germ cell divisions in this system is not sufficiently high to increase the number of de novo mutations in the male germ line with age. The lack of the age-related direct effects on female longevity and fecundity is however more striking. These results suggest that males maintain the ability to produce fully functional ejaculates even late in life (cf. Fox et al. 1995). This result is in contrast with the data from *D. melanogaster*, where all senescent live males were sterile when only ca. 20% of males were dead (Prowse & Partridge 1997).

Additionally, we would like to note that two out of 24 *P* values changed their value from significant to nonsignificant following adjustment for multiple comparisons. Noticeably, these results were not supported by corresponding findings in another part of the results, that is the marginally nonsignificant effect for single mating was not supported by the data from multiple matings and vice versa.

Female avoidance of fertilizations by old males was proposed as a potential explanation for the maintenance of polyandry (Radwan 2003). We found no evidence for the role of multiple mating in indirect avoidance of fertilizations by old males in a polyandrous seed beetle with well-documented costs of mating. If anything, doubly mated females had reduced offspring survival. Eady et al. (2000) found that *C. maculatus* females suffered from lower egg-to-adult survival of offspring when mated to different males than when mated repeatedly to the same male. Our data thus provide additional support for the cost of multiple mating in *C. maculatus*. Obviously, we did not measure all possible components of offspring fitness, for example offspring reproductive success in a competitive setting. We note, however, that the larval stage represents around 80% of the total life span of this beetle (Devereau et al. 2003), and that the development rate is likely to be a key factor in a scramble competition for mates. It is likely that the differential male mating success observed in our study resulted from male behaviour rather than from active discrimination against older males by females. We suggest that age-related decline in male mating success results from a senescent decrease in male performance.

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