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Kin selection and sexual conflict: male relatedness and

familiarity do not affect female fitness in seed beetles

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Summary

Theory maintains that kin selection can mediate sexual conflict because within-group male relatedness should reduce male-male competition, thereby reducing collateral harm to females. We tested whether male relatedness and familiarity can lessen female harm in the seed beetle *Callosobruchus maculatus*. Neither male relatedness nor familiarity influenced female lifetime reproductive success or individual fitness. However, male relatedness, but not familiarity, marginally improved female survival. Because male relatedness improved female survival in late life when *C. maculatus* females are no longer producing offspring, our results do not provide support for the role of kin selection in mediating sexual conflict. The fact that male relatedness improves the post-reproductive part of female life cycle strongly suggests that the effect is non-adaptive. We discuss adaptive and non-adaptive mechanisms that could result in reduced female harm in this and previous studies and suggest that cognitive error is a likely explanation.

Keywords: sexual conflict, kin selection, mate harm, *Callosobruchus maculatus*

1. INTRODUCTION

Males and females have different routes to successful reproduction [1], and this can lead to evolutionary conflict between the sexes [2-4]. One extreme form of this conflict is mate harm, when one sex (usually the male) physically injures the opposite sex (usually the female) [4, 5]. Male mate harm occurs in many animals and has been especially well studied in insects, including the bed bug *Cimex lectularius*, the seed beetle *Callosobruchus maculatus*, and the fruit fly *Drosophila melanogaster*. Male bed bugs stab females with their genitalia, inseminating females directly into the abdominal cavity [6, 7]. Male seed beetles have spines on their intromittent organs that pierce holes in the female's genital tract, reducing female longevity [8-10]. In the fruit fly, males may harass females during courtship [11-14] or physically harm them during the mating process [11, 15]. *Drosophila* and *C. maculatus* ejaculates also contain accessory gland proteins that modulate female reproductive behaviour [5, 16-18]. Mate harm can evolve for two reasons. One possibility is that the harm itself increases male fitness by causing females to allocate more resources into current reproduction and away from future reproductive attempts with other males [19]. A better-supported alternative in most cases is that mate harm is simply a deleterious side effect of male-male competition over fertilization [4, 20, 21].

Kin selection theory [22, 23] suggests that the level of genetic relatedness among competing males can moderate sexual conflict in viscous populations [24-27]. In populations in which close adult relatives are likely to interact, males would gain indirect fitness benefits by helping their close male kin to reproduce. Such cooperation might take the form of reducing mate harm

71   to facilitate sexual access to those females. As Chippindale *et al.* [28] point

72   out, for this kind of kin selection to occur, three conditions must be met: 1)

73   males must harm their mates in some way, thereby reducing female

74   reproductive success; 2) there must be some mechanism in place for reliably

75   recognizing kin; and 3) groups of related males must have a reasonable

76   chance of encountering each other during the reproductive period.

77   A series of recent studies on *Drosophila melanogaster* has investigated

78   this possible role of kin selection in moderating mate harm, with conflicting

79   results [28-33]. In support of the kin selection hypothesis, Carazo *et al.* [28]

80   found that females exposed to groups of three full-sib brothers did indeed

81   have higher lifetime reproductive success and slower reproductive ageing

82   than females exposed to trios of unrelated males. Groups of brothers were

83   less aggressive towards each other, courted females less vigorously, and

84   lived longer. Finally, Carazo *et al.* [28] found that when two brothers were

85   housed with an unrelated male, that unrelated male sired more than one-third

86   of the offspring, suggesting that groups of cooperating relatives are vulnerable

87   to invasion by non-cooperative non-relatives.

88   One important point to consider in the Carazo *et al.* [28] study is that it

89   confounded familiarity and relatedness. Brothers used in the study had also

90   been reared together in the same vial, whereas the unrelated males had been

91   raised in separate vials. Hollis *et al.* [30] conducted a follow-up study on a

92   different population of *Drosophila* in which they controlled for familiarity by

93   testing the effect of brothers raised together versus apart. Females exposed

94   to brothers raised together had higher lifetime reproductive success, but this

95   effect disappeared when females were raised with brothers raised apart.

96   Hollis *et al.* [30] concluded that familiarity and not relatedness *per se* was

97   likely driving the patterns Carazo *et al.* [28] observed.

98   However, one weakness of Hollis *et al.*'s [30] study is that they did not

99   include a treatment with unrelated males that had been reared together.

100   Without this treatment it is impossible to conclude whether familiarity alone

101   would be sufficient for cooperation. To address this issue, Chippindale *et al.*

102   [29] performed a fully crossed experiment in which they exposed females to

103   brothers that had been raised together, brothers that had been raised apart,

104   and unrelated males that had been raised apart. Unlike Carazo *et al*. [28],

105   they found no evidence that either familiarity or relatedness among males had

106   any effect on female lifespan or reproductive success, a result corroborated in

107   a separate study by Martin and Long [31]. Further, Chippindale *et al.* [29]

108   found no evidence that unrelated or unfamiliar males sired a disproportionate

109   number of offspring when introduced to pairs of brothers or males raised in

110   the same environment. They conclude that cooperation among males does

111   not appear to lower mate harm in this system, at least not in the populations

112   they examined. Finally, a later study by Le Page *et al*. [32] suggested that

113   both relatedness and familiarity are required for reduced female harm in *D.*

114   *melanogaster* in the population used by Carazo *et al.* [28, 33].

115   These conflicting results suggest that, at least in *Drosophila*, it remains

116   unclear what role kin selection plays in mediating male-male cooperation and

117   mate harm. Moreover, if we are to understand whether inclusive fitness

118   benefits mediate sexual conflict in the animal kingdom, we need to expand our

119   research focus into other model systems. One excellent candidate is the seed

120   beetle *C. maculatus*. As described above, male seed beetles inflict physical

121 harm on their mates [9, 10] and this species has been used routinely as a

122 model system to study the economics and genetics of sexual conflict over

123 lifespan and reproduction [34-36].

124   A recent study by Lymbery *et al.* [37] generally supported the

125 importance of male relatedness in mediating male harm to females. They

126 found that females housed with familiar brothers produced more offspring,

127 suggesting that relatedness and familiarity among males act together to

128 reduce male-induced harm to females. However, the beetles in the Lymbery

129 *et al.* [37] study were provided with Baker's yeast, which is rather unusual for

130 a species that inhabits human grain storages and is commonly kept in the

131 laboratory as a capital breeder that is aphagous in the adult stage.

132 Furthermore, while *C. maculatus* beetles can technically ingest yeast, yeast

133 consumption *per se* does not necessarily have a positive effect on longevity,

134 fecundity or offspring production [38] suggesting that access to yeast is not a

135 part of a normal life cycle of this species. Therefore, we investigated the effect

136 of male relatedness and familiarity in a large outbred and well-described

137 population of *C. maculatus* (SI USA) that was not provided with yeast in the

138 adult stage, which is in line with the recent evolutionary history of this species

139 and this population. We conducted a fully crossed experiment with respect to

140 male relatedness and familiarity, quantifying the lifetime reproductive success

141 and lifespan of virgin females exposed to four different trios of males: 1)

142 brothers raised together, 2) brothers raised apart, 3) unrelated males raised

143 together, and 4) unrelated males raised apart. If kin selection is indeed

144 mediating sexual conflict in this system, then brothers raised together should

145   exhibit less mate-harming behaviour than other groups, resulting in higher

146   relative fitness of females.

147

148   **2. METHODS**

149   **(a) *Study system***

150   Seed beetles are common pest of stored legumes indigenous to Asia and

151   Africa. Females lay their eggs on the surface of dried beans. Once the larvae

152   hatch, they burrow into the bean and eclose as reproductively mature adults

153   approximately 23-27 days later. *C. maculatus* are facultatively aphagous,

154   obtaining all the nutrients they require for survival and reproduction during the

155   larval stage [39]. Adult feeding increases fecundity and longevity [39]. Early

156   studies used a combination of yeast and sugar solutions, so it was difficult to

157   disentangle the effect of the separate components in fitness-related traits.

158   Ursprung *et al.* [38] found that sugar solution and water do increase fecundity

159   and longevity, but there was no effect of yeast on these key life-history traits.

160   Lymbery *et al.* [37] provided their study beetles with *ad lib* access to yeast, but

161   there was no obvious benefit in terms of fecundity or longevity, although the

162   direct comparison is not possible because their study did not include standard

163   aphagous conditions.

164   The study population was derived from an outbred South Indian stock

165   population ("SI USA") of *C. maculatus* originally obtained from C. W. Fox at

166   the University of Kentucky, USA, and then subsequently moved to Uppsala

167   University and finally to The American University of Paris three months prior to

168   the first block of the experiment. The original SI USA stock population was

169   collected from infested mung beans (*Vigna radiata*) in Tirunelveli, India in

170   1979 [40]. Both prior to and during the experiment, beetles were cultured

171   exclusively on mung beans and kept at aphagy (no food or water) in climate

172   chambers at 29°C, 50% relative humidity and a 12:12 h light:dark cycle. One

173   great advantage of this system is that the laboratory conditions closely

174   resemble natural conditions, because these beetles have associated with

175   dried legumes for thousands of years and their life history is adapted to life in

176   a storage environment [41, 42].

177

178   **(b) *Establishing the four treatment groups***

179   The experiment was carried out in two blocks. The first block was completed

180   in 2015. In 2018 the experiment was replicated and expanded to provide

181   additional data on daily fecundity of females. During both blocks, base

182   populations of beetles were kept in 1L jars with 150g of mung beans, and

183   approximately 250 newly hatched beetles were transferred to new jars with

184   fresh beans every 24 days on a continual basis. From this base population,

185   we established four different treatment groups (with a goal of approximately N

186   = 75 each in each block, 150 total): **1) related familiar (RF), 2) related**

187   **unfamiliar (RU), 3) non-related familiar (NF),** and **4) non-related unfamiliar**

188   **(NU)**. In the RF treatment, three full-sib brothers were housed in a Petri dish

189   together for 24 hours before being added to a dish with a (non-related)

190   female. In the RU treatment, three brothers with no prior experience with each

191   other were added all at once to a dish with a female. In the NF treatment,

192   three non-related males were housed together for 24 hours before being

193   added to a dish with a female. Finally, in the NU treatment, three non-related

194   and unfamiliar males were added to a dish with a female.

195   To generate full sibling brothers for the RF and RU treatments, we

196   transferred a random subset of beans with developing larvae into virgin

197   chambers (aerated plastic culture plates with a separate well for each

198   individual) and monitored the virgin chambers daily. Approximately one day

199   after hatch, we randomly paired 180 males and females and placed them into

200   180 60-mm Petri dishes with 75 beans each. We then removed the males and

201   females after 48 hours and allowed the eggs to develop. Since females can

202   lay up to 65 eggs per day (Berg, unpublished data), we wanted to provide

203   enough beans that females would lay only one egg on each bean. Before the

204   offspring hatched, we transferred the fertilized beans from the Petri dishes to

205   virgin chambers, carefully marking which beans came from which parents,

206   and monitoring hatch daily. Once these offspring hatched, we set up the four

207   different treatment groups above.

208   For both "familiar" treatments, trios of males were introduced to each

209   other on the same day that they hatched. For both "unfamiliar" treatments,

210   males were housed in their separate virgin cells until one day post hatch and

211   then introduced together with the female without any time to acclimatize to

212   each other. In all treatments, females were randomly selected from the base

213   population one day after hatch and were unrelated to the males. Males from

214   the non-related treatments were randomly selected from the base population

215   as well. All males and females used in this study were maintained as virgins

216   prior to the pairing. The Petri dishes in which males and females were housed

217   measured 100-mm and contained 150 beans. This number is sufficient to

218   allow females to lay just one egg per bean, reducing any larval competition

219   that might affect data on reproductive success. All sets of brothers used in this

220   study came from different parents, thus obviating the need to control for

221   parental identity in the analyses.

222   Three days after the trios of males were introduced to females, we

223   swapped out all the males for new males. This was done to reduce the

224   variance caused by male condition or behaviour on female reproductive

225   success or lifespan. In preparation for this, we set up new trios of freshly

226   hatched related familiar and non-related familiar males one day before. For all

227   the "related" dishes, we used brothers of the previous trio. Since fewer males

228   were eclosing this late in the hatch cycle, we had to use slightly older males in

229   some cases. We excluded few females that escaped/died from unnatural

230   causes resulting in slight deviations from the initial sample size (N = 75 for

231   RF, N = 75 for RU, N = 71 for NF, and N = 76 for NU in the first block; N = 75

232   for all treatments in the second block).

233

234   **(c) *Lifespan and fitness assays***

235   During both blocks of the experiment, we conducted both lifespan and fitness

236   assays for each female within each treatment group. For lifespan assays, we

237   monitored the Petri dishes daily and recorded the date of death of each

238   female. Once all adults were dead, we removed them from the dishes. We

239   collected two kinds of fitness data. During the first block of the experiment, we

240   measured total offspring production only. We did this by counting the number

241   of eclosed young per female, a standard measure of lifetime reproductive

242   success in this system. To facilitate the counting of offspring, we froze the

243   dishes 37 days after the initial pairing, well after all the offspring had eclosed

244   but before a subsequent generation could develop.

245   During the second block, we also measured daily offspring production

246   for each female. To do this we moved the female and males to new Petri

247   dishes with new beans every 24 hours until the female died (maximum of 9

248   sets of Petri dishes per female). Approximately 37 days later, we froze the

249   dishes and counted number of eclosed offspring per day per female.

250

251   **(d) *Statistical analyses***

252   Before analysis, we excluded all individuals that did not reproduce (NF = 2,

253   NU = 4, RF = 4, RU = 10). We analysed the lifetime offspring production as

254   well as age-specific reproduction using a generalized mixed effect model with

255   a Poisson error structure implemented in the *lme4* package in *R 3.3.3.* ,

256   treating Relatedness and Familiarity as crossed fixed factors. We tested for

257   overdispersion using the *dispersion_glmer* function in the *blmeco* package,

258   and if above 1.4, we controlled for overdispersion by adding a subject-level

259   random effect. For total reproduction, we used Block as a random factor.

260   Age-specific reproduction and individual fitness was only analysed for

261   Block 2, as this was the only block where age-specific fecundity data was

262   collected. For age-specific reproduction, we included Relatednes and

263   Familiarity as crossed fixed factors, as well as all interactions with Age and

264   Age^2. In addition, we also included Age at last reproduction (ALR) as a

265   crossed covariate. Age and ALR were scaled and centered before analysis

266   (mean = 0, s.d. = 1) and we used the *bobyqa* optimizer as well as increased

267   the default number of iterations to 10.000 in order to obtain good model

268   convergence. For all mixed-effect models, chi-square tests of fixed effects

269   were performed using the *car* package.

270   Individual fitness (λ_{ind}) was calculated from the life-table of age-specific

271   reproduction [43, 44], with a development time of 23 days, by solving the

272   Euler-Lotka equation for each individual using the *lambda* function in the

273   *popbio* package. λ_{ind} was then analysed in a linear model using Relatedness

274   and Familiarity as crossed fixed factors.

275   Survival was analysed in a Cox proportional hazard model using the

276   *coxme* package, with Relatedness and Familiarity as crossed fixed factors,

277   and Block as a random effect.

278

3. RESULTS

280   We measured lifetime reproductive success (total number of eclosed

281   offspring) of individual females introduced to one of four different groups of

282   male trios: brothers raised together (related familiar, or RF, N = 146), brothers

283   raised separately (related unfamiliar, RU, N = 140), non-related males raised

284   together (non-related familiar, NF, N = 144), or non-related males raised

285   separately (non-related unfamiliar, NU, N = 147). There was no significant

286   difference in female lifetime reproductive success between the four treatments

287   (Relatedness: χ^2 = 1.37, df = 1, p = 0.392; Familiarity: χ^2 = 0.00, df = 1, p =

288   0.999; Relatedness × Familiarity: χ^2 = 0.0015, df = 1, p = 0.969; Figure 1). If

289   anything, mean number of eclosed young was slightly higher for the non-

290   related treatments. If the non-reproducing females are included in the dataset,

291   we actually find higher reproduction in the non-related treatment group

292   (Relatedness: χ^2 = 4.30, df = 1, p = 0.038; Familiarity: χ^2 = 1.33, df = 1, p =

293   0.248; Relatedness × Familiarity: χ^2 = 0.240, df = 1, p = 0.624, Supplementary

294   figure 1). We did find different shapes of the age-specific fecundity, illustrated

295   by the significant interaction Relatedness × Familiarity × Age^2 (Table 1, Figure

296   2). However, we found no effect on individual fitness λ_{ind} (Relatedness: F =

297   0.026, df = 1, p = 0.872, Familiarity: F = 0.244, df = 1, p = 0.622, Relatedness

298   × Familiarity: F = 0.072, df = 1, p = 0.789).

299   When we measured the lifespan of the females introduced to the

300   different treatment groups, we found that male relatedness, but not familiarity,

301   improved female survival (Relatedness: z = -2.57, df = 1, p = 0.01; Familiarity:

302   z 0 0.34, df = 1, p = 0.73; Relatedness × Familiarity: z = 0.06, df = 1, p = 0.96;

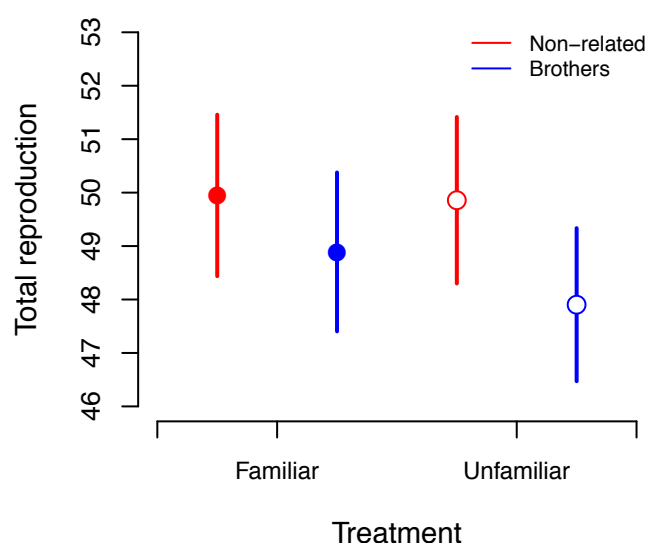303   Figure 3).

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308   **Figure 1.** Lifetime reproductive success LRS (number of adult offspring) by
309   treatment group: brothers (blue), non-related males (red), familiar individuals
310   (solid symbols) and unfamiliar individuals (open symbols). Symbols represent
311   mean ± SE.

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Table 1. Age-specific reproduction, result from generalized linear model with poisson error structure.

| Parameter	χ^2	df	p
Relatedness	0.2719	1	0.60204
Familiarity	0.1325	1	0.71588
ALR	52.8875	1	< 0.001
Age	1459.7816	1	< 0.001
Age^2	623.6481	1	< 0.001
Relatedness × Familiarity	0.0653	1	0.79830
Relatedness × ALR	0.2163	1	0.64190
Familiarity × ALR	0.0061	1	0.93791
Relatedness × Age	1.3663	1	0.24246
Familiarity × Age	1.0830	1	0.29802
ALR × Age	214.4389	1	< 0.001
Relatedness × Age^2	1.6252	1	0.20237
Familiarity × Age^2	1.8441	1	0.17448
ALR × Age^2	64.3620	1	< 0.001
Relatedness × Familiarity × ALR	1.6448	1	0.19967
Relatedness × Familiarity × Age	0.9137	1	0.33914
Relatedness × ALR × Age	0.0013	1	0.97119
Familiarity × ALR × Age	2.4446	1	0.11793
Relatedness × Familiarity × Age^2	4.7754	1	0.02887
Relatedness × ALR × Age^2	0.5634	1	0.45291
Familiarity × ALR × Age^2	2.1438	1	0.14315
Relatedness × Familiarity × ALR × Age	1.8233	1	0.17692
Relatedness × Familiarity × ALR × Age^2	3.3245	1	0.06825
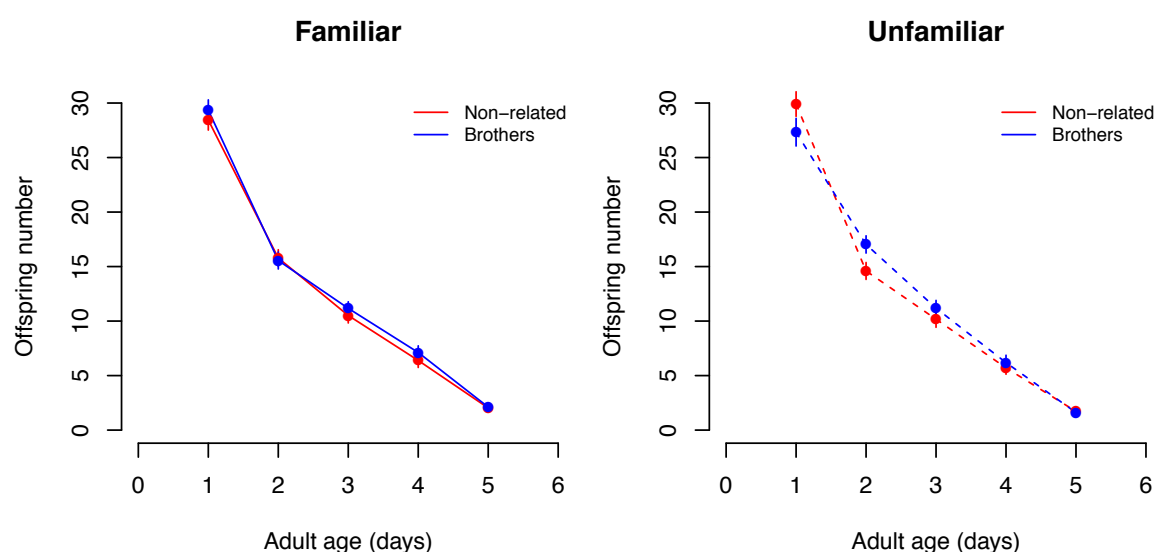
Figure 2. Age-specific reproduction for females mated with (A) familiar and (B) non-familiar males. Brothers are shown as blue, and non-related males as red.
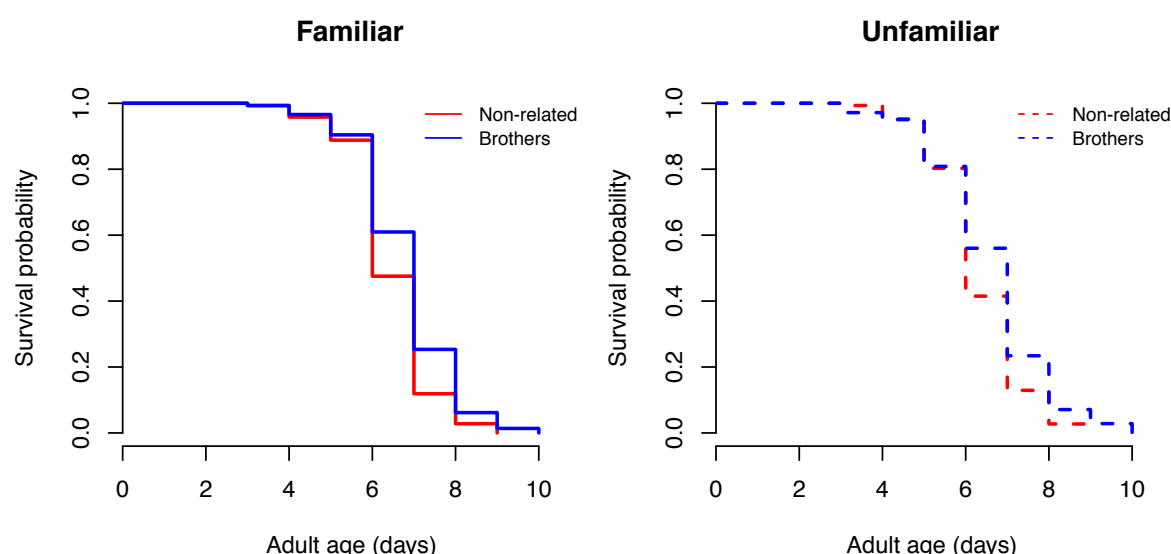
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Figure 3 Survival probability for females mated to (A) familiar and (B) unfamiliar males. Blue represents brothers and red represents non-related males.

4. DISCUSSION

Adaptive reduction in mate harm can only evolve if 1) there are reliable

mechanisms for recognizing kin, and 2) populations are sufficiently viscous

(i.e. genetically structured) for relatives to have a reasonable chance of

encountering each other while they are reproductively active. The population

genetic structure is one challenge facing the hypothesis that kin selection can

mitigate the evolution of male harm via interlocus sexual conflict. For example,

while kin recognition mechanisms may exist in *Drosophila* (e.g. through

cuticular hydrocarbons [45, 46]), Chippindale *et al.* [29] point out that both

natural and laboratory populations of *Drosophila* are unlikely to be sufficiently

structured to promote kin-selected reduction in male-female conflict. Simply

344   put, adults emerge and fly off and are unlikely to remain in or disperse into

345   genetically structured populations. Le Page *et al.* [32] countered this point by

346   suggesting that genetic structure may occur in fruit flies during colonization of

347   new patches by a small group of females, such that male relatedness-driven

348   reduction of female harm in the established populations are a relic of "the

349   foundation past". Future work will test whether selection during the foundation

350   of a new population in the natural environment is sufficiently strong to

351   generate long-lasting effects on male reproductive behaviour.

352   In contrast, *C. maculatus* beetles may, in theory, meet the necessary

353   pre-conditions without difficulty. Female seed beetles lay eggs in clusters, and

354   will deposit all of their eggs in close proximity to each other provided there is

355   sufficient supply of unoccupied beans. Upon emergence from the bean, male

356   seed beetles aggressively court females and begin mating immediately, which

357   increases the probability of encountering relatives. *C. maculatus* is a pest

358   species that infests supplies of stored legumes – under those conditions, it is

359   likely that many beetles will emerge and mate simultaneously providing

360   sufficient variation in the relatedness of competitors. At the same time, we

361   note that *C. maculatus* males are relatively indiscriminate in their mating

362   behaviour, likely because of the high "missing opportunity" cost that is

363   associated with living in high density populations, and commonly mount other

364   males because of perception errors [47, 48], which would complicate selection

365   for fine-tuned kin recognition mechanism that could lead, in theory, to reduced

366   female harm.

367   In this study, similar to Chippindale *et al.*'s [29] *Drosophila* study, we

368   found that neither male relatedness nor familiarity influenced female lifetime

369   reproductive success. However, in contrast to Chippindale *et al.* [29], male

370   relatedness, but not familiarity, improved female survival. This is also in

371   contrast to Lymbery *et al.* [37], who found that both familiarity and relatedness

372   increase reproductive success, but not survival. In our study, since the effect

373   on survival occurred only in late life, around day six of age when females

374   already stopped producing eggs, it failed to increase female lifetime

375   reproductive success. Therefore, our results do not provide support for the

376   role of kin selection in mitigating the effects of male harm.

377   Seed beetles are facultatively aphagous – that is, eclosed adults do not

378   require food or water to breed and survive [49]. While in many bruchid beetles

379   adults commonly consume pollen, nectar or fungi [50], *Callosobruchus* beetles

380   do not usually feed as adults. In the current study, we opted to keep the

381   beetles under the aphagous conditions in which they have evolved for over

382   500 generations since they were first brought into the laboratory in 1979.

383   Thus, our schedule reflects not only the original conditions of the human grain

384   storage, but also the recent evolutionary history of this large outbred

385   population.

386   Nevertheless, it is interesting to consider how male relatedness could

387   affect the fitness of female beetles in different environments. One may

388   hypothesize that increased survival could result in increased fecundity when

389   beetles have access to additional resources to continue reproduction in late

390   life. Yet, in a recent study by Lymbery *et al.* [37], beetles were raised with the

391   access to yeast but neither lived longer, nor produced more offspring

392   compared to normal non-feeding conditions. This finding seems concordant

393   with earlier reports that did not find positive effects of yeast consumption on

394   fitness in *C. maculatus* [38]. However, despite the lack of positive effects of

395   access to yeast on life-history traits, females kept with related familiar males

396   produced more offspring in Lymbery *et al.* [37] study. This suggests that the

397   presence of yeast interacts with male behaviour in an unknown way to affect

398   female reproductive performance. In our study, we found a three-way

399   interaction between relatedness, familiarity and the shape of age-specific

400   reproduction curve, stemming from the increased early-life reproduction and

401   steeper age-specific decline of females mated to unrelated males then

402   females mated to groups of brothers when both males were raised in a way

403   that precluded familiarity. Increased reproductive performance in early-life

404   could translate into increased individual fitness, but this was not the case. On

405   the other hand, when we included females that failed to produce viable

406   offspring in the analysis, we found that females kept with groups of unrelated

407   males had higher reproductive success, because most of failed reproductive

408   attempts were among females kept with groups of brothers. This finding is in

409   line with the idea that multiple mating increases female fitness when it

410   increases the genetic diversity of partners [51-54].

411   To summarize, there is little conclusive evidence to date for the role of

412   kin selection in mediating sexual conflict, and, specifically, in reducing male-

413   induced harm to females. More importantly, it is not always easy to see how

414   the selection for such an effect can operate in the natural environment

415   because it requires many opportunities for sib-sib interaction that may not be

416   very common in wild populations of invertebrates [29]. Some organisms, such

417   as bulb mites (*Rhizoglyphus robini*), maybe more prone to the evolution of kin-

418   selected benefits of reduced male harm because of the metapopulation

419   structure characterized by rapid population growth and colonization of the new

420   patches [55]. Indeed, recent work on bulb mites suggest that there is standing

421   genetic variation for male harm that evolves rapidly under kin selection [55], in

422   line with theoretical models [24, 27, 56]. Le Page *et al.* [32] discussed several

423   possible explanations for the fact that reduction in male-induced harm to

424   females is observed in some populations. One likely non-adaptive explanation

425   is a perception error. Indeed, as suggested by Le Page *et al.* [32], males can

426   use cuticular hydrocarbon profiles, or gut microbiota, as a measure of male-

427   male competition, and they may underestimate the level of competition when

428   surrounded exclusively by related males with similar odours, thereby investing

429   less in sperm competition. Such a non-adaptive hypothesis fits squarely with

430   the results of our study, because females housed with groups of related males

431   did enjoy improved survival, suggesting reduced male harm, but this effect

432   was entirely limited to post-reproductive part of their life cycle and had no

433   effect on their individual fitness. We suggest that more work is needed to

434   evaluate the importance of kin selection in the evolution of mating systems.

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