Original Research Article

Divergent survival costs in homosexual and heterosexual interactions among coffee white stem borers, *Xylotrechusquadripes* (Coleoptera: Cerambycidae)

Abstract

In our comprehensive study on coffee white stem borers (CWSBs), we conducted a series of experiments to investigate the influence of social and ecological factors on beetle survivorship and life spans. Our experiments involved various scenarios, including isolated virgin males, isolated virgin females, males in group settings, females in group settings, and mixed-gender groups. We made several noteworthy observations. First, survivorship reached 100% within the initial 15 days for all groups except isolated virgin males, which had a survival period of only 5 days. In contrast, treatments involving solely virgin females and males consistently exhibited higher survivorship rates ranging from 80% to 90%. Notably, in scenarios with males in groups or mixed-gender groups, survival dropped to zero percent after 45 days. Our study also highlighted the comparable costs incurred by males engaging in both homosexual and heterosexual interactions. Males interacting with females and those interacting with other males exhibited similar survival curves, both displaying shorter median life spans compared to isolated virgin males. This suggests that both courting and mounting behaviors, regardless of the recipient's sex, exact a substantial toll on male beetles. Furthermore, our research revealed that female-female interactions, while less costly than male-male interactions, still led to reduced female survival in group settings. This emphasizes the importance of quantifying the costs associated with same-sex interactions, shedding light on the dynamics of such behaviors in CWSBs. In conclusion, our findings underscore the significance of studying behavioral and life history traits in different environmental contexts. Social circumstances and ecological factors significantly influence sexual dimorphism in lifespan and highlight the complex interplay between reproductive strategies, survival rates, and behavioral costs in CWSBs.

Keywords: Coffee white stem borer, survival, coffee, behaviour

Introduction

A fundamental concept within the framework of life cycle theory revolves around the notion of reproductive costs (Stearns 1992; Roff & Fairbairn 2007). Reproduction, due to its inherent impact on future survival and reproductive potential, emerges as a costly endeavor. The burden of these costs predominantly falls upon females, encompassing expenses related to mating, offspring development, and caregiving. Conversely, the realm of male reproduction involves a spectrum of expenditures, encompassing both material resources and energy investments in gamete production, courtship rituals, competitive encounters, and mating pursuits (Cordts & Partridge 1996; Kotiaho 2001; Martin & Hosken 2004).

Historically, the majority of research concerning the costs of sexual behaviors and interactions has centered on heterosexual encounters. However, in numerous species, both sexes exhibit the potential for aggressive behaviors directed towards individuals of the same sex, including courtship and mounting attempts (Aiken 1981; Serrano et al. 1991; Srivastava et al. 1991; Andersson 1994; Vasey 1995; Bagemihl 1999; Fang & Clemens 1999; Harari et al. 2000; Sommer & Vasey 2006; Vasey et al. 2008). These interactions may manifest as elements of intrasexual competition, but distinguishing them from mere perception errors can be challenging within real-world contexts.

Consequently, we use the term "homosexual relationships" to encompass all interactions between individuals of the same sex. Despite their pivotal significance in models elucidating the evolution and sustenance of sexual behaviors, the costs associated with homosexual relationships have been quantified in only a limited number of species, particularly among females. Engaging in homosexual behaviors can entail substantial costs. In many species, males partake in scramble and/or combat encounters with rivals to gain access to females or territories, often expending considerable time and energy while risking substantial harm (Thornhill & Alcock 1983; Andersson 1994; Kotiaho 2001). Males may also engage in courting and mounting other males, with courtship itself incurring significant expenses (Cordts & Partridge 1996).

Homosexual mounting has been observed across a wide range of taxa, including mammals, birds, reptiles, and insects (Aiken 1981; Thornhill & Alcock 1983; Bagemihl 1999; Harari et al. 2000; Switzer et al. 2004; Sommer & Vasey 2006). Notably, in insects, homosexual mounting may be more prevalent, accounting for up to 50% of reported mating attempts in some cases (Aiken

1981; Serrano et al. 2000). While research on homosexual interactions in mammals has garnered significant attention, it is essential to acknowledge that females exhibit homosexual behaviors in numerous species, even though much of the research has focused on male-to-male relationships (Srivastava et al. 1991; Vasey 1995; Fang & Clemens 1999; Harari et al. 2000; Sommer & Vasey 2006; Gastal et al. 2007).

Homosexual mounting and courtship (or courtship-like) behaviors may serve various sexual functions, ranging from establishing dominance over rivals to acting as practice for heterosexual interactions (Vasey 1995). However, it is worth noting that these actions are often attributed to sex recognition failures or perception errors (Parker 1968; Aiken 1981; Thornhill & Alcock 1983; Serrano et al. 1991; Harari et al. 2000; Switzer et al. 2004). For instance, research on homosexual mounting in the flour beetle Triboliumcastaneum suggests the absence of sex recognition in this species, with the frequencies of intra- and intersexual mounting aligning with a random null model (Serrano et al. 2000). Although sex recognition exists in many other species, it may be relatively weak. In the water insect Palmacorixa nana, males exhibit a preference for mounting larger individuals, likely driven by the size disparity between males and females (Aiken 1981). This preference results in a notable rate of errors (Aiken 1980). Additionally, several studies (Savalli & Fox 1999; Harari et al. 2000; Serrano et al. 2000; Switzer et al. 2004; Van Gossum et al. 2005) have provided evidence in support of the perception error hypothesis.

Thornhill & Alcock (1983) propose that homosexual mounting in insects may evolve through male selection if the advantages of increased mating success through rapid, indiscriminate mating attempts outweigh the drawbacks of discrimination. A key prediction stemming from this theory posits that males would tolerate higher costs associated with homosexual mounting because only males stand to benefit from heightened mounting frequency in terms of fitness. Conversely, given that females are less likely to gain from such interactions, selection is expected to act against the evolution of costly female-female interactions (though also Harari et al. 2000).

Materials and methods

Xylotrechusquadripes, commonly known as the coffee white stem borer, served as the focal species in a series of experimental investigations aimed at comparing the costs associated with homosexual interactions vis-à-vis heterosexual interactions.

To create a controlled environment for our experiments, we utilized a nylon net enclosure measuring 3 x 3 x 3 meters. This enclosure was specifically designed to house coffee stems infested with the borer, and it was situated within the field coordinates of 13.1365° N, 75.6403° E, at an elevation of 970 meters above sea level. The area was kept open yet shaded to mimic natural conditions. Freshly emerged adult beetles were segregated by gender and housed in a laboratory setting, maintaining a controlled temperature of 23°C and relative humidity at 70%. They were provided with a cotton wad saturated in a 10% sugar water solution.

For our investigation, we exclusively utilized newly emerged virgin males and females. Six distinct experimental treatments were established: virgin males in isolation, virgin females in isolation, virgin males in group settings, virgin females in group settings, mixed-sex groups of males and females, and mixed-sex groups of females and males.

Virgin beetles were collected from the infected coffee stems at 24-hour intervals and housed individually in 25ml plastic vials containing cotton wads saturated with 10% honey. In each "isolated" treatment, ten containers were designated for males and ten for females, totaling 10 individuals of each sex. Within the "grouped" treatments, individuals of the same sex were grouped into five containers, each containing ten beetles. To distinguish individual beetles, five individuals in each "grouped" container were marked with white, non-toxic face paint. These marked beetles served as a reference group to assess mortality among unmarked focal individuals. Whenever a marked beetle perished, it was promptly replaced from the available pool of newly hatched beetles, ensuring that there were consistently five marked beetles in each container at any given time during the experiment. Mortality records were exclusively maintained for the unmarked focal insects, with a total of 25 individuals for each sex.

To establish "mixed-sex groups" for both males and females, we employed similar procedures. However, in this case, instead of using marked beetles of the same sex, we employed marked beetles of the opposite sex (i.e., marked females for the "male mixed-sex" group, and vice versa). For each of the two "mixed-sex" treatments, we created five containers, each containing five males and five females, resulting in a total of 25 individuals. In the "male mixed-sex" treatment, only male deaths were recorded, and any deceased females were replaced from the available stock. The order was reversed for the "female mixed-sex" treatment, where only female deaths were recorded, and deceased males were replaced from the stock. Mortality counts were conducted once daily for all treatment types.

To assess potential variations in life expectancy among the different treatments, we conducted survival analysis, utilizing block-mean life duration as the unit of replication for treatments involving groups of animals. However, the results remained qualitatively consistent when individual lifespan served as the unit of replication. We conducted the survival analysis using Excel.

Results

In our experimental treatments, which included scenarios featuring isolated virgin males, isolated virgin females, males in group settings, females in group settings, and mixed-gender groups, we observed a remarkable trend in beetle survivorship. Specifically, we noted that survivorship reached 100% within the initial 15 days for all groups except for the treatment involving isolated virgin males, where survival extended only up to 5 days.

Furthermore, when examining treatments comprising males in groups, females in groups, males in mixed-gender groups, and females in mixed-gender groups over a 20 to 30-day timeframe, we observed a substantial disparity in survivorship. In these scenarios, beetle survivorship ranged between 40%, compared to a significantly higher 80% to 90% in treatments involving solely virgin females and males.

Notably, in treatments featuring males in groups and males in mixed-gender groups, beetle survival diminished to zero percent over a 45-day duration. Conversely, in the remaining treatments, some beetles displayed remarkable resilience, surviving for up to an impressive 50-day period (Figure 1).

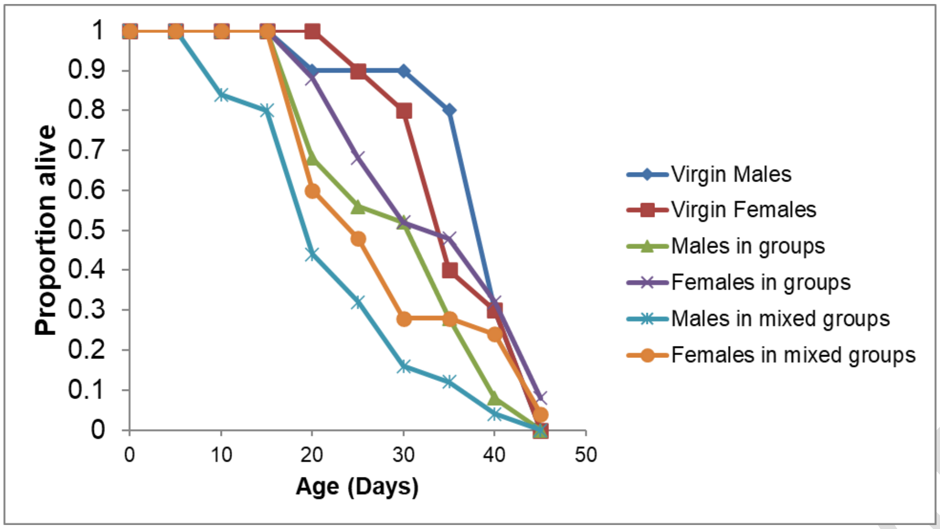

Figure 1. Survival curves of *Xylotrechusquadripes*in different experimental treatments.

In our study, we delved into the intriguing realm of the coffee white stem borer (CWSB), focusing on the adult females and males, and examining the nuances of their life spans under various experimental conditions.

We observed that adult female CWSBs exhibited their longest mean life span in treatments where they were housed alone (31±2.34 days), followed by treatments where females were placed in groups (19.68±2.92 days), and their shortest life span was recorded in treatments involving females in mixed-gender groups (16.84±3.25 days). Conversely, adult male CWSBs displayed their lengthiest mean life span in treatments where they were isolated, i.e., males alone (24.5±3.7 days), followed by treatments featuring males in mixed-gender groups (14.24±3.23 days), and their shortest life span occurred in treatments with males in group settings (10.8±3.38 days) (Figure 2).

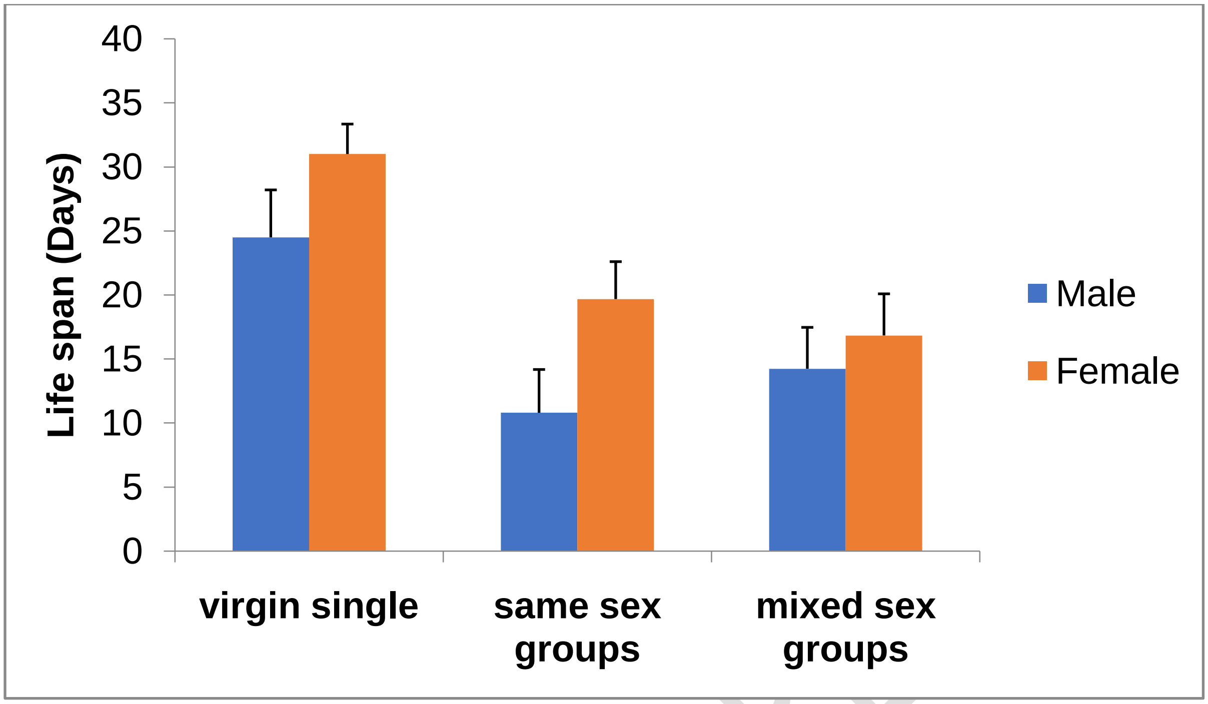

Figure 2. Mean life span ± SE of *Xylotrechusquadripes*in different experimental treatments.

One remarkable finding was the comparable costs incurred by males engaged in both homosexual and heterosexual interactions, as depicted in Figures 1 and 2. We observed that males interacting with females and those interacting with other males exhibited analogous survival curves, and both scenarios displayed shorter median life spans when contrasted with virgin males kept in isolation. This underscores the substantial expenses associated not only with courting but also mounting same-sex conspecifics. It's worth emphasizing that these same-sex courtship and mounting behaviors are likely to exact a toll on both the initiator and the recipient.

Notably, male CWSBs exhibit vigorous pursuit and mounting of other males (Mangalgikar, P., personal observations), which is believed to demand considerable energy and hydration resources. Furthermore, during such interactions, there's a potential risk of injuries or even rapid mortality if one male's genitalia become entangled beneath their elytra (Mangalgikar, P., personal observations). It's worth noting that CWSB males do not engage in physical combat.

Our principal discovery hinges on the observation that males experience both homosexual and heterosexual interactions at roughly equivalent costs, whereas this parity is not evident among females. Additionally, our study revealed that beetles maintained in groups of ten individuals, with no separation and prompt replacement of deceased individuals, experienced a notable reduction in the life expectancy of females in group settings by 11.2% when compared to solitary females (Figure 2). This reduction is suggestive of potential costs associated with female-female interactions, even though they do not involve overt agonistic behaviors such as fighting. In CWSB, females do indeed exhibit mounting behaviors toward other females (Mangalgikar, P., personal observations). In contrast, our analysis indicated that female survival in CWSB was considerably less affected by homosexual interactions when compared to their male counterparts (Figure 2).

Furthermore, it is noteworthy that male homosexual interactions substantially diminish survival, to an extent comparable to the overall costs of reproduction, as our findings elucidate. This highlights the importance of quantifying the costs associated with homosexual interactions, a facet of animal behavior that has often been presumed to be modest despite its long-standing interest in the context of evolutionary biology (Aiken 1981). Our study underscores the necessity of systematically assessing these costs, akin to how researchers routinely scrutinize the development of mating strategies, as indicated by our empirical data. Furthermore, our results align with the hypothesis that females exhibit less frequent and less costly homosexual behavior in comparison to males, shedding light on the intriguing dynamics of same-sex interactions in this species.

Discussion

Our research findings underscore the paramount significance of investigating behavioral and life history traits within the context of their developmental environments. Our study suggests that the costs associated with sexual behaviors such as courtship and mounting, often meticulously quantified within heterosexual interactions, exhibit substantial variation contingent upon social and ecological factors, including the adult sex ratio. This variability arises from a significant portion of the net cost being attributed to behaviors directed towards individuals of the same sex. While certain behaviors aimed at conspecifics of the same sex may play a role in sexual competition, others, exemplified by gay mounting in C. maculatus, appear to manifest as nonadaptive consequences of highly indiscriminate male responses. Consequently, the relative prevalence of such behaviors can markedly influence their net costs and benefits.

Furthermore, our findings unveil intriguing implications for understanding the emergence of sex-based disparities in life expectancy. The prevalent phenomenon of sexual dimorphism in lifespan is often attributed to differential reproductive costs between the sexes (Trivers 1972; Liker & Szekely 2005). Given that sexual dimorphism in lifespan is a recurring pattern in C. maculatus (Fox et al. 2003, 2004; Maklakov et al. 2007), we purposefully manipulated reproductive opportunities in our study. Remarkably, when individuals were housed in same-sex cohorts, the contrast in lifespan between virgin males and virgin females in C. maculatus surged to 42%. In contrast, when males and females were allowed to mate (i.e., maintained in mixed-sex environments), we detected no discernible sexual dimorphism in lifespan.

Therefore, it becomes evident that social circumstances exert a profound influence on sexual dimorphism in lifespan: in isolation, virgin females outlived males significantly. However, as soon as individuals of both sexes were provided with opportunities for interaction and reproduction, sexual dimorphism in lifespan virtually dissipated. This observation indicates that despite their intrinsic potential for a longer lifespan, females actually exhibit mortality rates on par with those of males under conventional reproductive conditions, thereby implying that selection acts comparably on survival (and conceivably, the rate of aging) in both sexes. Consequently, sex-based disparities in various other life history traits (e.g., resource acquisition) and sexual behaviors, which are deeply rooted in sex-specific reproductive strategies, may potentially emanate as indirect by-products of the sexual dimorphism in lifespan frequently documented in studies involving virgin cohorts (Bilde et al. 2009).

COMPETING INTERESTS DISCLAIMER:

Authors have declared that they have no known competing financial interests OR non-financial interests OR personal relationships that could have appeared to influence the work reported in this paper.

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