


The consequences of heatwaves for animal reproduction are timing-dependent

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Abstract

1. In light of the increased frequency of heatwaves due to climate change, it is crucial to better understand their potential effects on animal reproduction. Heat stress can affect all aspects of reproduction, including gamete development, fertilisation success, parental care and offspring survival. We may, therefore, expect these effects to be highly sensitive to the timing of a heatwave event relative to an organism's reproductive cycle.
2. Here, we use an insect study system (*Nicrophorus vespilloides*) to test whether variation in the timing of a heatwave within a short timeframe has differential effects on reproductive success and offspring fitness.
3. We found that heatwaves had little to no effect when they occurred a few days before or after mating, but they were highly detrimental for fitness if they occurred during mating. Individuals that experienced a heatwave during mating were significantly less likely to have a successful breeding bout, had a longer breeding bout, and their offspring were smaller and suffered a lower survival rate.
4. Our study shows that variation in the timing of a heatwave event over very short timescales (on the order of days) can have drastically different consequences for animal reproduction. This work provides novel insights into the vulnerability of organisms at different stages of their reproductive cycle and can improve our ability to make informed predictions about the ecological consequences of heatwaves under climate change.

KEYWORDS

climate change, fitness, mating, offspring, parental care, reproductive success, survival, temperature

1 | INTRODUCTION

Climate change is arguably the greatest threat to biodiversity in the 21st century. It has resulted in not only higher mean temperatures across the globe but also a higher likelihood of extreme weather

events, such as heatwaves (Meehl & Tebaldi, 2004). Heatwave events are also becoming more intense and longer in duration (Meehl & Tebaldi, 2004). Heat stress caused by these events can have detrimental effects on animal reproduction. A rapidly accumulating body of literature has documented the effects of high temperature

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on sperm morphology and performance, fecundity, hatching success, parental care, as well as offspring survival and condition (e.g. Chevrier et al., 2019; Fragueira et al., 2021; Martinet et al., 2021; McCowan & Griffith, 2021; Porcelli et al., 2017; Rukke et al., 2018; Vasudeva et al., 2021; Wiley & Ridley, 2016).

Nevertheless, a fundamental gap in our understanding is the role of the timing of heatwave events relative to an organism's reproductive cycle. Given that heat stress can affect all aspects of reproduction, we might expect these effects to be highly sensitive to the exact timing of exposure to high temperatures. Indeed, Cinto Mejía and Wetzel (2023) recently argued that the consequences of heatwaves and other extreme weather events will vary greatly depending on when they occur, and they highlight a pressing need for studies explicitly testing this. Previous work has typically examined the effect of heatwaves by exposing individuals to increased temperature during one particular life stage (e.g. Chevrier et al., 2019; Sidhu et al., 2022; Vasudeva et al., 2021), and a few studies have compared the effects of heatwaves during distinct life stages, for example, in juveniles versus adults (e.g. Porcelli et al., 2017; Sales et al., 2021). However, there is no prior work examining these effects at different stages of the reproductive cycle within a short timeframe.

Thus, it is still unknown whether experiencing a heatwave immediately before mating, during mating, or soon after mating can lead to differential effects on reproductive success and offspring fitness. Exposure to a heatwave shortly before mating may primarily affect spermatogenesis or oogenesis (Cameron & Blackshaw, 1980), and in turn sperm performance, fecundity and fertilisation success (Chevrier et al., 2019; Dadras et al., 2017). On the other hand, a heatwave during mating could affect sexual selection processes or copulation frequency and duration (García-Roa et al., 2020; Leith et al., 2021; Pilakouta & Ålund, 2021; Pilakouta & Baillet, 2022). Temperature has wide-ranging effects on mating interactions mediated through various physiological and sensory pathways. For

example, temperature affects metabolic rate, locomotor performance and activity levels, which can, in turn, influence the energetic costs of mate searching and assessment (e.g. Conrad et al., 2017; Jiao et al., 2009). Lastly, a heatwave at a later stage of the reproductive cycle, after offspring have hatched, may have direct effects on offspring survival and condition (Wiley & Ridley, 2016). In species with parental care, parents may increase the amount of care they provide to buffer against the detrimental effects of the heatwave on their offspring's fitness (Grew et al., 2019). Alternatively, because of the heat stress parents themselves experience, they may have a lower capacity to provide care, which could further exacerbate the direct effects of the heatwave on offspring fitness (Wiley & Ridley, 2016). In ectotherms, parental care is expected to be more energetically costly at higher temperatures because the metabolic costs of any physical activity increase with temperature.

Here, we tested whether the fitness consequences of heatwaves are timing-dependent, using the burying beetle *Nicrophorus vespilloides*. Our experimental design included (i) a control treatment where beetles were kept at a constant temperature of 18°C, (ii) beetles exposed to a simulated heatwave 3 days before being paired with a partner, (iii) beetles exposed to a heatwave immediately after being paired with a partner and (iv) beetles exposed to a heatwave 3 days after being paired with a partner (Figure 1). The simulated heatwave was 25°C for 3 days, and beetles were kept at the control temperature (18°C) before and after the heatwave. We examined how heatwave exposure at these different stages of the reproductive cycle influenced the likelihood of breeding successfully, fecundity, parental care duration, offspring size and offspring survival rate.

Since the first heatwave treatment occurred before beetles were paired up, we expected that it would primarily affect gamete development and, thus, fecundity and breeding success. The second heatwave treatment occurred during oviposition; heat stress at this stage of reproduction may affect fecundity, as well as the offspring's

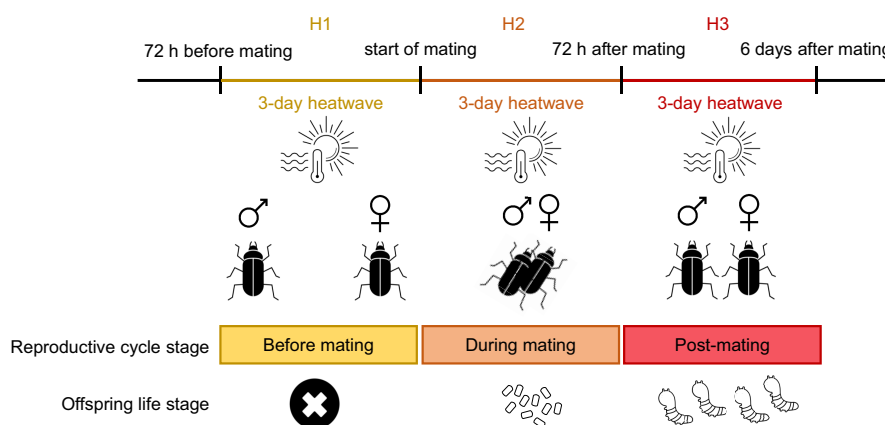


FIGURE 1 Visualisation of the experimental design of this study. In addition to a control treatment kept at a constant temperature of 18°C, we had three heatwave exposure treatments: beetles exposed to a 3-day heatwave (25°C) before being paired with a partner (H1), beetles exposed to a 3-day heatwave (25°C) immediately after being paired with a partner (H2) and beetles exposed to a 3-day heatwave (25°C) 3 days after being paired with a partner (H3). Since the H1 heatwave occurs before beetles are paired up, we expected that its effects would primarily be on spermatogenesis and oogenesis. The H2 heatwave occurs during the oviposition stage (females typically lay eggs 24–48 h after mating), so it may affect the number of eggs laid, as well as their hatching success and post-hatching survival rate. The H3 heatwave occurs during larval hatching (hatching starts 2–3 days after being laid).

hatching success and post-hatching survival rate. The third heatwave treatment occurred after the end of oviposition and during larval hatching. We hypothesised that heatwave exposure during this period of parental care might lead to a reduction in the amount of care provided due to higher energetic costs for the parents, with downstream effects on offspring fitness.

In light of the increased frequency of heatwaves due to global climate change, it is crucial to better understand their potential effects on animal reproduction and, thus, population viability. Our study can reveal whether these effects are dependent on the timing of the heatwave event relative to an organism's reproductive cycle. Resolving the role of timing will improve our ability to predict the ecological consequences of heatwaves and identify when organisms are most vulnerable to these extreme weather events (Cinto Mejía & Wetzel, 2023).

2 | MATERIALS AND METHODS

2.1 | Study species

The burying beetle *N. vespilloides* is an insect with facultative biparental care that breeds on vertebrate carcasses. Once a carcass is found, parents bury it in the soil, remove any fur or feathers and deposit antimicrobial secretions on its surface. They lay eggs in the soil around the carcass 24–48 h after mating (Eggert, 1992). Two to 3 days later, the eggs hatch and the larvae crawl to the carcass. Larvae can self-feed in a crater created by the parents at the top of the carcass, but parents also provision larvae with predigested carrion (Smiseth et al., 2003). Females typically spend more time provisioning food for the larvae and stay on the carcass for longer than males (Pilakouta et al., 2015, 2018; Pilakouta, Richardson, et al., 2016; Smiseth et al., 2005; Smiseth & Moore, 2002). Larvae disperse from the carcass 5–7 days after hatching, which corresponds to the end of the parental care period and offspring independence.

2.2 | Animal husbandry

We used second- and third-generation beetles from an outbred laboratory population maintained at the University of Aberdeen. These lines originated from wild-caught beetles collected in Tollohill Wood and Kincorth Hill in Aberdeen, UK, in July and August 2022. They were housed individually in transparent plastic containers (12 × 8 × 2 cm) filled with moist soil and kept at 18°C and a 16 h:8 h light cycle. They were fed small cubes of raw organic beef twice a week.

2.3 | Experimental design and procedures

Our experimental design included (i) a control treatment where beetles were kept at a constant temperature (18°C) throughout the experiment, (ii) beetles exposed to a heatwave (25°C) for 3 days before

being paired with a partner, (iii) beetles exposed to a heatwave (25°C) for 3 days immediately after being paired with a partner and (iv) beetles exposed to a 3-day heatwave (25°C) 3 days after being paired with a partner (Figure 1). Beetles in the heatwave treatments were kept at 18°C before and after the heatwave. To ensure that our simulated heatwave was ecologically relevant, we used 25°C for 3 days, which is representative of the typical duration and upper temperature limit of heatwaves experienced in northeast Scotland in recent years (Isotalo et al., 2022). During the simulated heatwave, beetles were placed in a cooled incubator (LMS Series 1A Model 201NP) set to a constant temperature of 25°C and a 16 h:8 h light cycle. We also used a saturated solution of magnesium chloride hexahydrate salt to maintain the humidity inside the incubator at 50% (±10%). Humidity levels were monitored using Fisherbrand™ Traceable™ Jumbo Thermo-Humidity Meters. We used the same incubator for all three heatwave treatments (H1, H2 and H3) to avoid confounding effects due to any potential differences among incubators, but this means there is some pseudoreplication in our study as is common for all research that makes use of incubators or growth chambers.

To avoid inbreeding, we only mated unrelated, virgin males and females that did not share any common ancestors for at least two generations. Each experimental pair was placed in a transparent plastic container (17 × 12 × 6 cm) filled with 1 cm of moist soil and a freshly thawed mouse carcasses of a standardised size (22–25 g). Pairs were randomly assigned to one of the four treatment groups described above (Figure 1). We set up a total of 210 matings, but this was not evenly divided across the four treatments because of differential breeding success due to the effects of the heatwave treatments (Table S1). Our sample sizes for pairs that had at least one larva surviving to independence were $n=33$ for the control treatment, $n=29$ for the heatwave exposure before mating, $n=20$ for the heatwave exposure during mating and $n=28$ for the heatwave exposure after mating (Table S1).

We counted eggs by checking the underside of each breeding box twice a day until larvae started hatching. This is the least disruptive method for estimating clutch size in this species (Bladon et al., 2020), and previous work found a strong correlation between the number of eggs counted from the underside of the breeding box and the actual number of eggs laid (Jarrett et al., 2017). To estimate the duration of care by each parent, we checked the containers daily in the morning and the afternoon to determine whether parents were present on the carcass or away from the brood in the soil. Parents that were away for more than two consecutive checks were deemed to have abandoned the brood and were removed from the boxes to prevent infanticide (Pilakouta et al., 2018). At the dispersal stage, when offspring became independent, we recorded the number of surviving larvae and measured the total brood mass to calculate average offspring mass.

2.4 | Data analysis

All analyses were performed using R version 4.2.2 (R Development Core Team, 2022), and the 'ggplot2' package was used for

generating figures (Wickham, 2016). We used a linear model for average offspring size, which was a continuous trait with normally distributed random errors. For discrete traits, we used generalised linear models fitted with a negative binomial error distribution (egg number, brood size), a Poisson error distribution (length of breeding bout) or a quasipoisson error distribution (parental care duration) to account for overdispersion. Our analysis for brood size included all breeding pairs that laid at least one egg but excluded pairs that failed to lay any eggs. For binary or proportion data (likelihood of successful breeding attempt, offspring survival rate), we used generalised linear models fitted with a quasibinomial distribution and entered these data into the models using the 'cbind' function. Treatment (control, H1, H2 or H3) was used as an explanatory variable in each of these models. For variables where treatment had a statistically significant effect, we also conducted post-hoc pairwise comparisons using the 'emmeans' package (Length, 2023) to determine which heatwave treatment was different from the control.

3 | RESULTS

We found that the effects of heatwaves on reproductive success, parental care and offspring fitness were highly dependent on the exact timing of the heatwave event (Table 1). These effects were generally more severe when parents were exposed to a heatwave during mating than when exposed before or after mating (Table 1).

3.1 | Heatwave effects on reproductive success

Exposure to a heatwave did not have an effect on the number of eggs laid ($LR \chi^2 = 0.10$, $p = 0.99$, Figure 2a). However, pairs that were exposed to a heatwave during the mating stage suffered a substantial reduction in the likelihood of a successful breeding attempt ($LR \chi^2 = 28.3$, $p < 0.0001$;

Control-H2: Estimate = 1.50 ± 0.40 , $z = 3.72$, $p = 0.001$; Figure 3a). Pairs exposed to a heatwave during mating also had smaller broods compared with the control treatment ($LR \chi^2 = 13.4$, $p = 0.004$; Control-H2: Estimate = 1.04 ± 0.32 , $z = 3.22$, $p = 0.007$, Figure 2b). Lastly, these pairs had a much longer breeding bout, from the time of mating to the stage of offspring independence ($LR \chi^2 = 198.3$, $p < 0.0001$; Control-H2: Estimate = -0.32 ± 0.2 , $z = -13.1$, $p < 0.0001$; Figure 3b).

3.2 | Heatwave effects on parental care

Heatwave exposure influenced the duration of care provided by female parents ($LR \chi^2 = 29.2$, $p < 0.0001$, Figure 4a). More specifically, in broods where the heatwave event started 3 days after mating, female parents abandoned the brood earlier compared with broods not exposed to a heatwave (Estimate = 0.20 ± 0.05 , $z = 3.89$, $p < 0.001$). Female duration of care did not differ from the control treatment for the other two heatwave treatments (Control-H1: $z = -1.37$, $p = 0.52$; Control-H2: $z = -0.33$, $p = 0.99$).

Heatwave exposure also influenced the duration of care provided by male parents ($LR \chi^2 = 21.8$, $p < 0.0001$, Figure 4b). Males in the control treatment abandoned their brood earlier than males exposed to a heatwave before mating (Estimate = -0.22 ± 0.07 , $z = -3.04$, $p = 0.013$). Males that were exposed to a heatwave at later stages (i.e. during or after mating) did not differ in parental care duration compared with males in the control treatment (Control-H2: $z = 0.71$, $p = 0.89$; Control-H3: $z = 1.47$, $p = 0.46$).

3.3 | Heatwave effects on offspring fitness

Parents that were exposed to a heatwave during or after mating had significantly smaller offspring at the stage of offspring independence ($F = 14.8$, $p < 0.0001$; Control-H2: Estimate = 0.04 ± 0.01 , $z = 4.25$, $p < 0.001$; Control-H3: Estimate = 0.03 ± 0.01 , $z = 1.47$,

	Effect of heatwave relative to control treatment		
	Before mating (H1)	During mating (H2)	After mating (H3)
Reproductive success			
Number of eggs laid	—	—	—
Brood size	—	↓	—
Likelihood of successful breeding attempt	—	↓	—
Length of breeding bout	—	↑	—
Parental care			
Female care duration	—	—	↓
Male care duration	↑	—	—
Offspring fitness			
Average offspring size	—	↓	↓
Offspring survival rate	—	↓	—

TABLE 1 Summary of the effects of heatwave events (25°C for 3 days) occurring at different stages of the reproductive cycle relative to the control treatment (constant temperature of 18°C).

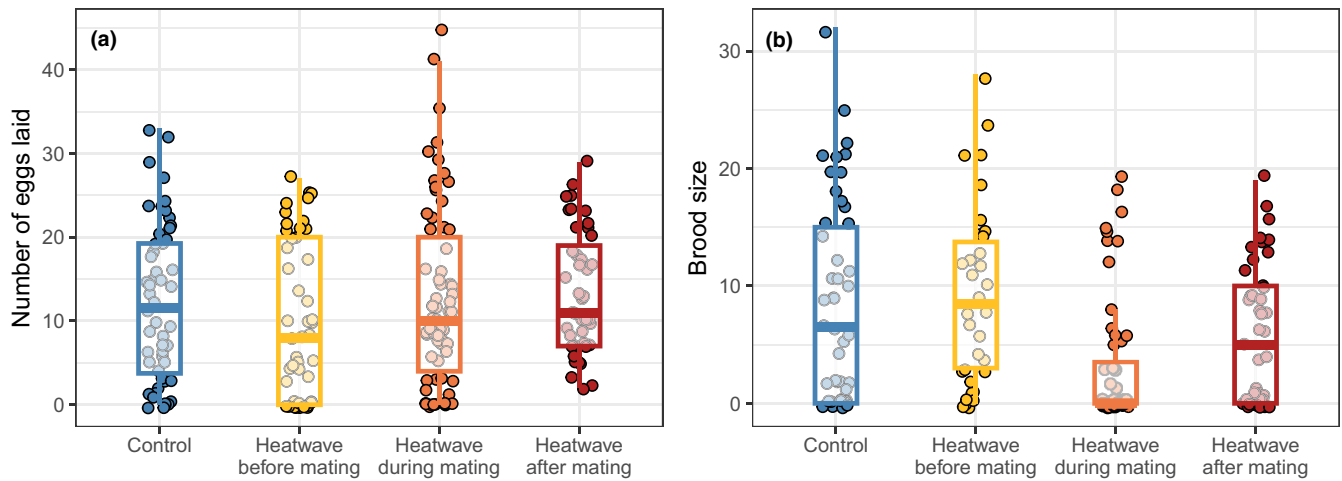


FIGURE 2 Boxplots of number of eggs laid (a) and number of surviving larvae at the dispersal stage (b) across the four treatment groups (control=blue, heatwave before mating=yellow, heatwave during mating=orange, heatwave after mating=red). The lower and upper hinges of the box correspond to the first and third quartiles, respectively. The lower and upper whiskers extend from the hinge to the smallest and largest value no further than $1.5 \times \text{IQR}$ from the hinge, respectively.

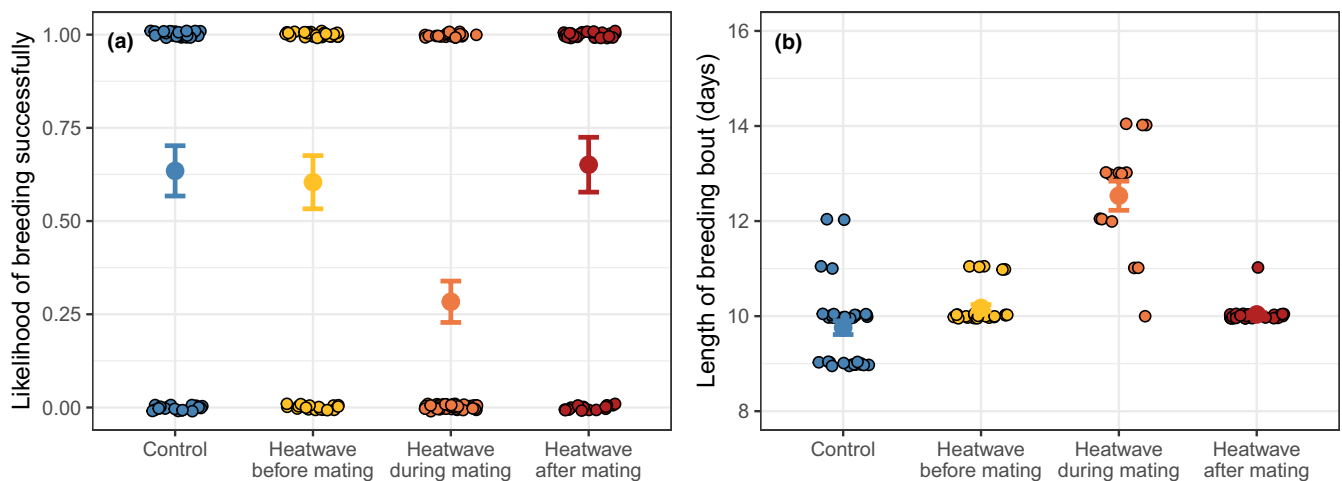


FIGURE 3 Mean (\pm SE) likelihood of a successful breeding attempt (a) and length of the breeding bout (b) across the four treatment groups (control=blue, heatwave before mating=yellow, heatwave during mating=orange, heatwave after mating=red). The former refers to the likelihood of having at least one larva surviving to independence. The latter refers to the period of time between pairing up males and females and the stage of offspring independence (dispersal of larvae into the soil).

$p=0.001$, Figure 5a). In addition, the offspring of parents exposed to a heatwave during mating suffered a significant reduction in survival rate to independence (LR $\chi^2=22.2$, $p<0.0001$; Control-H2: Estimate = 1.79 ± 0.48 , $z=3.70$, $p=0.001$, Figure 5b).

4 | DISCUSSION

A recent review by Cinto Mejía and Wetzel (2023) highlighted that ‘understanding the ecological consequences of climate change will require ecologists to adopt a temporally explicit approach that considers not just the frequency and duration of [heatwave] events but also when the events occur relative to the biological processes they impact’. Here, we provide the first empirical

evidence that variation in the timing of a heatwave event over a short timescale (on the order of days) can have drastically different consequences for reproductive success and offspring fitness. More specifically, we found that heatwaves have little to no effect when they occur a few days before or after mating, but they are detrimental when they occur during mating. Parents that experienced a heatwave during mating were significantly less likely to breed successfully, they had a longer breeding bout and their offspring were smaller and suffered a lower survival rate. By revealing the role of timing in mediating the effects of heatwaves on reproduction, this work can improve our ability to predict the ecological consequences of heatwaves and identify when organisms are most vulnerable to these extreme weather events (Cinto Mejía & Wetzel, 2023). Our findings also have major implications

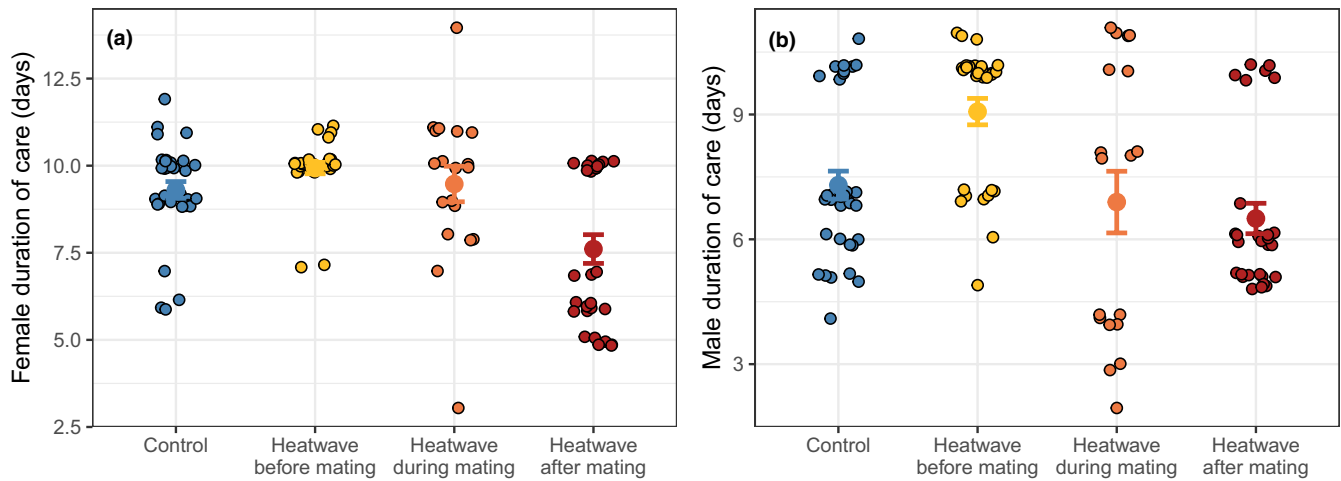


FIGURE 4 Mean (\pm SE) duration of care (days) provided by (a) female parents (left) and (b) male parents (right) across the four treatment groups (control=blue, heatwave before mating=yellow, heatwave during mating=orange, heatwave after mating=red). Filled circles represent individual data points.

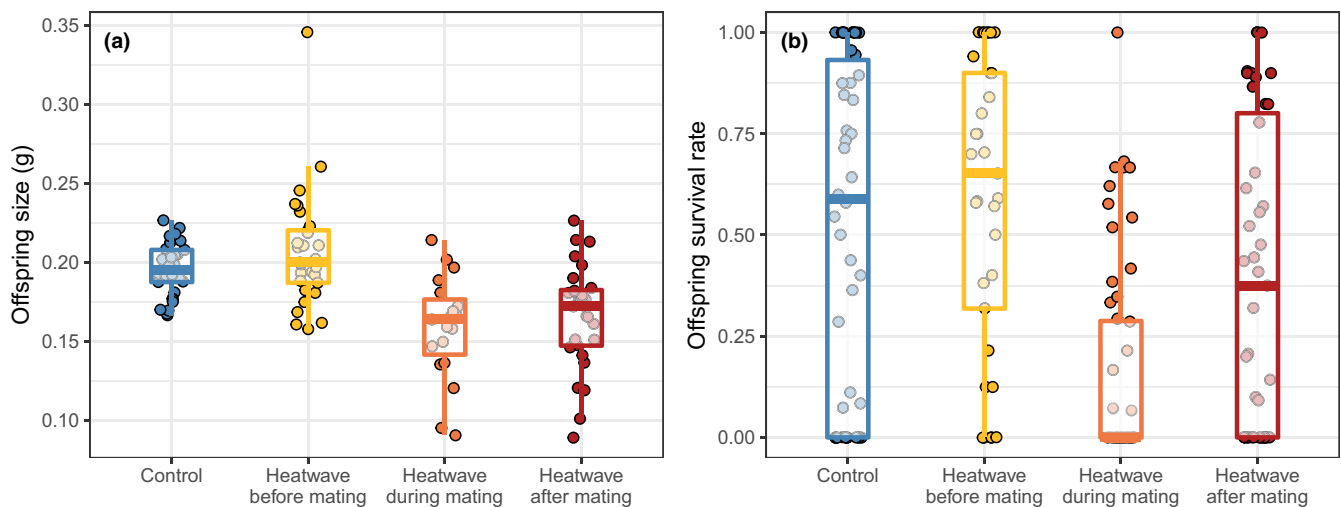


FIGURE 5 Boxplots of offspring size at independence (a) and offspring survival rate to independence (b) across the four treatment groups (control=blue, heatwave before mating=yellow, heatwave during mating=orange, heatwave after mating=red). Filled circles represent individual data points. The lower and upper hinges of the box correspond to the first and third quartiles, respectively. The lower and upper whiskers extend from the hinge to the smallest and largest value no further than $1.5 \times$ IQR from the hinge, respectively.

for how researchers design experiments to study the effects of heatwaves on reproduction or other aspects of organismal function: they demonstrate that the timepoint of experimental heatwaves should be carefully tailored to an organism's ecology and life history. Overall, our results highlight an urgent need for further studies testing the timing-dependent effects of heatwaves across a wide range of taxa.

4.1 | Effects of heatwaves occurring during mating

One of our key findings was that parents that experienced a heatwave during mating were significantly less likely to have a successful breeding bout (i.e. at least one larva surviving to independence).

There are a few potential explanations for this pattern. First, it is well established that fertility, and in particular male fertility, is highly sensitive to heat stress (David et al., 2005; Porcelli et al., 2017; Sales et al., 2018). Even though we found no evidence for an effect of the heatwave on the number of eggs laid (Figure 2a), a reduction in sperm quality or performance could have led to lower fertilisation success or lower hatching success (Breedveld et al., 2023). Second, there is evidence from other taxa that sperm characteristics can have long-term effects for offspring fitness post-hatching (Alavioun et al., 2017, 2019) even in the absence of differential fertilisation success. Thirdly, physiological stress due to heatwave exposure may have led to a reduced capacity to provide parental care (Merkling et al., 2017), although this is not reflected in our results on the duration of care (Figure 4).

Another main finding was that the length of the breeding bout was significantly longer when parents were exposed to a heatwave during mating. In these pairs, egg laying occurred a few days later compared with the control and the other two heatwave treatments (H1, H3). This is in line with a study on another *Nicrophorus* species showing that burying beetles are unlikely to initiate breeding in extreme environmental conditions (Quinby et al., 2020). Thus, the heatwave most likely delayed the onset of breeding, rather than lengthened the rate of offspring development. In natural populations, longer breeding bouts may limit the number of reproductive attempts possible over the course of a breeding season.

Despite a delay in initiating egg laying by individuals exposed to a heatwave during mating, their offspring still suffered a substantial reduction in their survival rate. This was the case even though the duration of care provided by males and females in this treatment group was unaffected by the heatwave. In addition, offspring that did survive to independence were much smaller. Offspring size at independence is a crucially important fitness component in this species. Larvae do not feed after dispersal and before eclosion, so larval mass determines the adult size (Lock et al., 2004). In turn, adult size influences lifespan, fecundity and the likelihood of acquiring a carcass for breeding (Bartlett & Ashworth, 1988; Otronen, 1988; Pilakouta, Halford, et al. 2016; Pilakouta & Smiseth, 2016). Thus, our results indicate that the timing of the heatwave event has major consequences for offspring fitness both in the short term and long term.

Such effects on offspring fitness have a potential link to population viability, although the demographic effects of heatwave timing are likely to vary among species depending on their life history. For example, for species where reproduction is highly synchronised among individuals, the timing of the heatwave may have a major impact on offspring recruitment into the breeding population, whereas in species where there is a long reproductive season, the exact timing of the heatwave could have little impact on the mean fitness of individuals in the population. Nevertheless, even in the latter scenario, if heatwaves result in longer breeding bouts as we found here, this may limit the maximum number of breeding attempts possible within the reproductive season, which could again negatively affect offspring recruitment into the population.

4.2 | Effects of heatwaves occurring before and after mating

In contrast to the findings discussed above, heatwaves of the same intensity (25°C) and duration (3 days) occurring 3 days before or 3 days after mating had little to no effect on reproductive success and offspring fitness. In the former case, when individuals were exposed to a heatwave for 3 days before being paired and allowed to mate, the only significant effect was an increase in the duration of male care (but not female care). This was not associated with any downstream consequences for offspring fitness.

When individuals were exposed to a heatwave 3–6 days after mating, there was a reduction in the duration of female care (but not male care). The period of this heatwave treatment (H3) coincides with the start of hatching and a peak in parental care 24 h after hatching (Smiseth et al., 2003). In ectotherms, the metabolic costs of any physical activity, including parental care, increase with temperature. Females typically provide more care than males in this species (Pilakouta et al., 2015, 2018; Pilakouta, Richardson, et al., 2016; Smiseth et al., 2005; Smiseth & Moore, 2002), so they may have reached their maximum capacity at an earlier stage due to the heatwave.

In addition, offspring were smaller at independence compared with the control treatment. This could have been a result of the reduced care provided by females, which includes provisioning food to the larvae, thus improving their growth rate. Alternatively, it could have been due to an accelerated decomposition rate of the carcass at a higher temperature, reducing the amount of resources available to the offspring (Arce et al., 2012). The heatwave may have also directly impaired offspring growth rate by increasing metabolic costs. These explanations are not mutually exclusive, and it is possible that any direct effects of the heatwave on offspring fitness were exacerbated by a reduction in the amount of care provided by female parents.

It is worth noting that although heatwaves occurring after mating had some effect on a few aspects of reproduction, these effects were not as severe as those of heatwaves occurring during mating (Table 1). A potential explanation for this pattern is that organisms are most vulnerable to heat stress during the copulation and fertilisation stage, and less vulnerable after fertilisation and egg-laying have taken place. Such timing-dependent effects may be even more pronounced in species with external fertilisation, where gametes are highly sensitive to changes in ambient temperature (Chirgwin et al., 2020; Levitan, 1996). Thus, we strongly encourage further work to determine to what extent the fitness consequences of heatwaves are timing-dependent across a range of externally and internally fertilising species.

5 | CONCLUSION

We show that variation in the timing of a heatwave event over very short timescales can have drastically different consequences for reproductive success and offspring fitness. These findings highlight the importance of considering when a heatwave occurs relative to the timing of the biological processes it affects (Cinto Mejía & Wetzel, 2023). It is, therefore, crucial for future studies on the organismal effects of heatwaves to carefully consider their experimental design in light of their study system's ecology and life history, as decisions about the timing of the heatwave exposure could lead to markedly different results. In sum, this work provides novel insights into the vulnerability of organisms at different stages of their reproductive cycle and can improve our ability to make informed predictions about the ecological consequences of heatwaves under global climate change.

AUTHOR CONTRIBUTIONS

Natalie Pilakouta conceived the study. Lorelei Sellers, Rebecca Barratt and Alice Ligonniere conducted the experimental work and collected the data. Natalie Pilakouta carried out the data analysis and wrote the manuscript. Lorelei Sellers, Rebecca Barratt and Alice Ligonniere reviewed and approved the manuscript draft.

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CONFLICT OF INTEREST STATEMENT

Natalie Pilakouta is an Associate Editor of Functional Ecology but took no part in the peer review and decision-making processes for this paper.

DATA AVAILABILITY STATEMENT

All relevant data are available on the Dryad Digital Repository: <https://doi.org/10.5061/dryad.mcvdnc58> (Pilakouta et al., 2023).

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

Table S1. Sample sizes for the four treatments in our experiment: (i) control treatment where beetles were kept at a constant temperature and not exposed to a heatwave, (ii) beetles exposed to a 3-day heatwave immediately before being paired with a partner, (iii) beetles exposed to a 3-day heatwave immediately after being paired with a partner and (iv) beetles exposed to a 3-day heatwave 3 days after being paired with a partner.

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