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Body mass and mate choice in bumblebees (*Bombus terrestris*) under climate heating

Min Su Park [a,1], Ji Hyun Woo [a,1], Hyung Joo Yoon [a], Bo Yeon Kim [a], Kyeong Yong Lee [b], Steven A. Trewick [c], Kwang Sik Lee [a,*], Byung Rae Jin [a,**]

[a] *College of Natural Resources and Life Science, Dong-A University, Busan 49315, Republic of Korea*
[b] *Department of Agricultural Biology, National Academy of Agricultural Science, Wanju 55365, Republic of Korea*
[c] *Wildlife & Ecology Group, SFTNS, Massey University Manawatu, Palmerston North 4410, New Zealand*

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ABSTRACT

Climate warming influences the population size, body size, and diversity of bumblebees, important pollinators in agriculture and ecosystems. Increases in developmental temperatures due to climate warming lead to reduction in bumblebee body size, particularly among the worker caste, resulting in reduced pollination activity. However, how reduced body mass resulting from warm conditions influences mate choice between new queens and males remains unclear. This interaction has a major role in viability of subsequent generations, and we investigated male-male competition and queen mate choice in *Bombus terrestris* at optimal and elevated mating temperatures (23 °C and 32 °C, respectively). At both temperatures, large males had a significantly higher mating rate than small males. Small males exhibited slightly prolonged mating, while large males transferred more sperm to queens' spermathecae owing to their higher sperm count. In mating experiments with queens of different body weights, no significant difference was observed in the mating rate between large and small queens at 23 °C, but at 32 °C, large queens had a significantly higher mating rate than small queens, indicating strong male preferences for large queens. Additionally, the survival of mated queens after hibernation was positively correlated with their body weight prior to hibernation. These results demonstrate the changes in the mating preferences of queens and males under warm conditions, suggesting that mate choice shapes subsequent generations in such environments.

1. Introduction

Climate change is a global environmental issue, with one of its major detriments being its threat to biodiversity. The increasing frequency and duration of extreme climatic events, such as heatwaves, severely endanger the populations and diversity of cold-adapted bumblebees (Maebe et al., 2021). These bees are crucial pollinators in agricultural and natural ecosystems in temperate regions. Bumblebees are sensitive to changes in ambient temperature, and elevated developmental temperatures have been shown to negatively affect their morphology and behavior (Gérard et al., 2022a; Perl et al., 2022; Kim et al., 2024a, 2025). Although various traits of bumblebees may show plasticity and adaptive responses to climatic stressors (Maebe et al., 2021), climate change has caused declines in their biodiversity and abundance (Martinet et al., 2015; Sirois-Delisle and Kerr, 2018; Fourcade et al., 2019; Soroye et al., 2020; White and Dillon, 2023).

The optimal nest temperature for the development of bumblebee colonies is approximately 27–29 °C (Röseler, 1985; Vogt, 1986; Yoon et al., 2005; Sepúlveda et al., 2024). Ambient temperatures at or above 32 °C cause thermal stress in bumblebees (Gérard et al., 2018, 2022a, 2022b, 2023; Guiraud et al., 2021; Perl et al., 2022), so they thermoregulate their nests via wing fanning (Röseler, 1985; Vogt, 1986; Yoon et al., 2005; Sepúlveda et al., 2024). In experimental studies considering climate heating on bumblebees, temperatures of 30–32 °C were indicative of a heatwave, while 34–36 °C characterized an extreme heatwave (White and Dillon, 2023; Sepúlveda et al., 2024). Studies on the effects of heat exposure on bumblebees use variation in body size as a key indicator of thermal stress (Guiraud et al., 2021; Gérard et al., 2023; White

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* Corresponding author.
** Corresponding author.
E-mail addresses: leeks@dau.ac.kr (K.S. Lee), brjin@dau.ac.kr (B.R. Jin).
[1] These authors contributed equally.

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and Dillon, 2023) with higher temperatures yielding reduced body size, especially in worker bees, and this decreased pollination activity (Vanderplanck et al., 2019; Guiraud et al., 2021; Kenna et al., 2021; Gérard et al., 2022b, 2023; Kuo et al., 2023; Naumchik and Youngsteadt, 2023; Kim et al., 2024a). Trade-offs have been found between colony size and body size in bumblebees, with reduced body size being associated with increases in colony size (del Castillo et al., 2015; Kim et al., 2025). Higher temperatures have been shown to negatively affect oviposition and mating behavior in bumblebee queens (Kim et al., 2025), impair learning and memory (Gérard et al., 2022a), and disrupt behavioral responses to critical sensory stimuli (Perl et al., 2022). Thus, climate heating is explicitly linked to changes in the morphology and performance of bumblebees, but mate selection—a key process for sustaining subsequent generations—remains unclear.

Mating in bumblebees is generally monoandrous, although some species exhibit polyandry (Estoup et al., 1995; Brown et al., 2002). Queens of the bumblebee *Bombus terrestris* typically mate singly with males and receive a mating plug that prevents re-mating (Baer et al., 2001). They overwinter underground during winter after mating and begin the life cycle independently in spring, so mating decisions directly impact subsequent generations. Furthermore, *B. terrestris* queens mate with only one male, but males can mate multiple times, which influences the importance of male weight for remating success (Gosterit and Gurel, 2016). Body size is a factor in mate choice, with larger partners preferred in many animal species including insects (Byrne and Rice, 2006; Mattle and Wilson, 2009; Amin et al., 2012; Walzer and Schausberger, 2015; Zhang et al., 2021; Zhao et al., 2021). Such a mating preference, as an outcome of competition, represents a reproductive strategy in which genetics linked to large body size is beneficial (Andersson, 1994; Blanckenhorn, 2005; Gençer and Firatli, 2005; Gençer and Kahya, 2011). However, mating behavior is also influenced by other factors, including sex ratio, age, ambient temperature, and body size (Boomsma and Ratnieks, 1996; Amin et al., 2010; Treanore et al., 2021; Kim et al., 2025).

In bumblebees, simulated climate heating results in reduction in body size, with significantly smaller offspring emerging across various castes, and mating avoidance behavior (Kim et al., 2024a, 2025). Here, we focused on how male body mass affected male-male competition and male-female mate choice. Our findings provide evidence of shifts in the mate choice of bumblebee queens and males under experimental warming conditions. Results could serve as a model for predicting the potential negative impacts of future climate warming on bumblebees in the wild.

2. Materials and methods

2.1. Bumblebee rearing

Colonies of *Bombus terrestris* bumblebees were previously reared in an indoor system (Yoon et al., 2005) at the Department of Applied Biology, Dong-A University. Bumblebees (after three generations of indoor rearing at 32 °C) designated for warming experiments were maintained in a room at a constant temperature and relative humidity of 32 °C and 65 %, respectively, under continuous dark conditions. They were fed ad libitum with a mixture of pollen and a 40 % sucrose solution. To prevent inbreeding, bumblebees were mated with more than 10 newly emerged queens (5–6 days after emergence) and 20–30 newly emerged males (7–8 days after emergence), all of which were collected from different colonies (Treanore et al., 2021; Kim et al., 2025). Mating was conducted in wooden mating boxes with steel mesh sides (60 × 50 × 65 cm) at 23 °C for one week, under lighting conditions of 1000 lux and a 14L:10D photoperiod, with pollen bread and a 40 % sucrose solution provided (Kim et al., 2025). Mated queens were then separated and placed in ventilated plastic boxes at 10–15 °C for one day. The queens were weighed, placed in ventilated tubes, and stored in incubators at 2.5 °C and 70 % relative humidity under constant darkness for 12 weeks to induce hibernation (Yoon et al., 2010). After hibernation, the queens were weighed and placed in flight boxes (60.0 cm × 50.0 cm × 65.0 cm) at 23–25 °C for three days to induce flight orientation (Kim et al., 2025). Subsequently, the hibernated bumblebee queens were transferred to ventilated wooden nesting boxes (9.5 cm × 15.0 cm × 10.5 cm) to initiate oviposition. After 5–10 workers emerged, the oviposition plates were transferred to ventilated plastic nesting boxes (15.5 cm × 16.5 cm × 10.5 cm). Once an additional 40–50 workers had emerged, the nests were moved to even larger boxes (22.0 cm × 28.0 cm × 14.0 cm) for colony development. Newly emerged queens and males from over 30 colonies were subsequently used for further experiments.

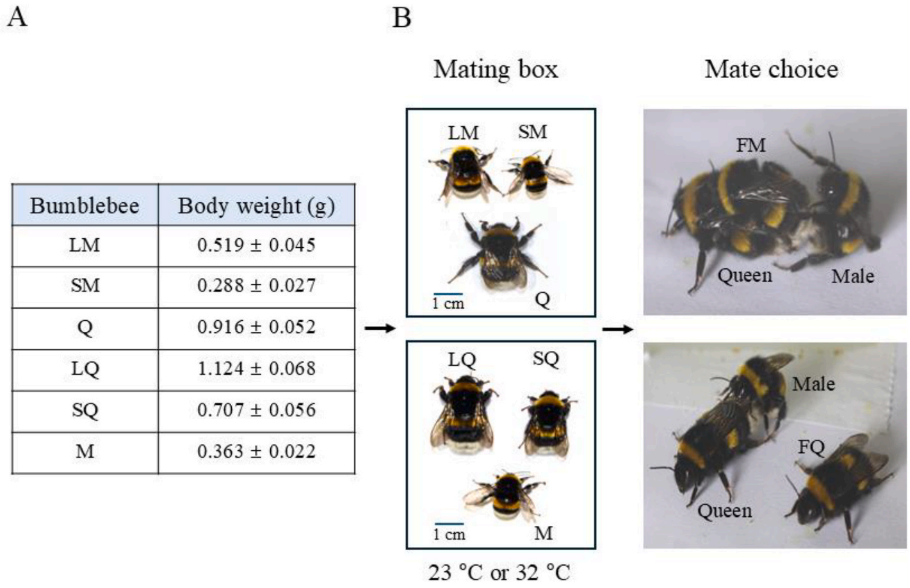
2.2. Mate choice experiment

For mate choice experiments, new queens (6–8 d post-emergence) and males (6–8 d post-emergence) were collected from different colonies to avoid inbreeding (Treanore et al., 2021; Kim et al., 2025). The body weights of the newly emerged queens and males were measured using an electronic scale (AE260 Delta Range, Mettler Toledo, Columbus, Ohio, USA) while each bumblebee was individually secured in a transparent tube cap, without anesthesia. Considering the changes in morphological traits due to higher temperatures in *B. terrestris* castes (Kim et al., 2024a), the newly emerged experimental queens and males were divided into three groups based on body weight (Fig. 1A). Newly emerged males reared indoors at 32 °C were categorized by body weight as large (LM, average 0.519 ± 0.045 g), medium (M, average 0.363 ± 0.022 g), or small (SM, average 0.288 ± 0.027 g). Similarly, newly emerged queens obtained from indoor rearing at 32 °C were classified into large-size (LQ, average 1.124 ± 0.068 g), medium-size (Q, average 0.916 ± 0.052 g), or small-size (SQ, average 0.707 ± 0.056 g) groups. For mate choice experiments (Fig. 1B), males and queens were organised in two permutations: LM + SM + Q ($n = 9–11$, four replicates) or LQ + SQ + M ($n = 10–11$, three replicates). In each replicate, equal numbers of LM, SM, and Q, or LQ, SQ, and M, were placed in a wooden mating box (46 cm × 46 cm × 47 cm) with steel mesh sides, along with a mixture containing pollen bread and a 40 % sucrose solution. Mating experiments were performed under optimal (23 °C) and high (32 °C) temperatures in humidity-controlled incubators with illuminance of 1000 lux. During mating, the queens and males, while in separate incubators with the same conditions, were moved to ventilated wooden nesting boxes (9.5 cm × 15.0 cm × 10.5 cm) to estimate mating duration. The mating rate, timing of mating occurrence, and mating duration for newly emerged queens and males were recorded at 23 °C and 32 °C over a period of 120 min. Mating rate was expressed either as the cumulative mating rate (i.e., the percentage of mated individuals relative to the total) or as the individual mating rates of LM and SM, or of LQ and SQ, normalized to 100 % of the cumulative mating rate. Moreover, mating experiments without competition were conducted using males (FLM or FSM) that had failed to mate during the initial competitive experiment. For these experiments, equal numbers of males and queens (FLM + Q or FSM + Q) were placed in wooden mating boxes at both 23 °C and 32 °C. The mating rate, timing of mating occurrence, and mating duration were recorded as described above.

Queens that had mated at 23 °C were weighed, placed in ventilated tubes, and stored in darkness for over 12 weeks in incubators maintained at 2.5 °C with a relative humidity above 70 % (Yoon et al., 2010). After hibernation, the survival rates of queens from each batch were analyzed in relation to their body weight prior to hibernation ($n = 382$, 1172, and 719 for the three replicates, respectively).

2.3. Sperm counting

Sperm from the spermathecae and seminal vesicles of queens and males, respectively, were collected after mating at different temperatures (23 °C and 32 °C) during mate choice experiments and were



Bumblebee	Body weight (g)
LM	0.519 ± 0.045
SM	0.288 ± 0.027
Q	0.916 ± 0.052
LQ	1.124 ± 0.068
SQ	0.707 ± 0.056
M	0.363 ± 0.022

Fig. 1. Mate choice experiments. (A) Selection of males and queens with different body weights. Large and small queens (LQ and SQ, respectively) and males (LM and SM, respectively) were selected based on body weights measured 6–8 d after emergence at a constant high temperature of 32 °C. Q and M denote medium-weight queens and males, respectively. (B) For male-male competition and queen mate choice experiments, the treatment subgroups were as follows: LM, SM, and Q at 23 °C (optimal mating temperature) or 32 °C (high mating temperature); and LQ, SQ, and M at 23 °C or 32 °C. Experiments for male-male competition were conducted in a mating box with equal numbers of LM, SM, and Q, while those for queen mate choice had equal numbers of LQ, SQ, and M. FM and FQ represent males and queens that did not mate during mating experiment, respectively. Representative images of males and queens are provided.

counted. Seminal vesicles from mated males were dissected individually 6 h after mating, and the seminal fluid was extracted by pressing the dissected vesicles. Similarly, spermathecae from mated queens were dissected individually 24 h after mating, and the spermathecal fluid was extracted by pressing the spermathecae. Subsequently, the seminal fluid or spermathecal fluid was diluted individually in a separate Eppendorf tube with phosphate-buffered saline (pH 7.4). Sperm counts from the seminal and spermathecal fluids were determined using a hemocytometer under a light microscope (Optinity 4K HD Camera KCX-80LA, Olympus, Tokyo, Japan) (Kim et al., 2024b). Sperm quantities were estimated based on the remaining sperm count in seminal fluids after mating and the transferred sperm count in spermathecal fluids following mating.

2.4. Statistical analysis

Statistical analyses were conducted using SPSS 22.0 (IBM Inc., Chicago, IL, USA), and the data were expressed as means ± standard deviation. Statistically significant differences were analyzed using one-way ANOVA, followed by Tukey's HSD post-hoc test. Normality of the data was determined using the Shapiro–Wilk test, while statistically significant differences between two samples were analyzed using Wilcoxon rank-sum test, followed by a two-sample *t*-test.

3. Results

3.1. Male body mass and mating competition in bumblebees

In the first mating experiment, conducted using males of different body weights, male-male competition for queens between LM and SM was investigated at optimal and elevated temperatures of 23 °C and 32 °C. The cumulative mating rate of males was higher at 23 °C than at 32 °C, but the difference was not statistically significant (two-sample *t*-test, df = 5, $P = 0.368$; Fig. 2A). When this cumulative mating rate (Fig. 2A) was normalized to 100 % and divided into LM and SM groups, the mating rate was significantly higher in LM than in SM at both temperatures (one-way ANOVA, $F_{3,12} = 15.76$, $P = 0.0001$; Fig. 2B). At 32 °C, the mating rate of SM increased slightly compared to that at 23 °C, but the difference was not statistically significant (two-sample *t*-test, df = 4, $P = 0.210$; Fig. 2B). The majority of males mated within 10 min at both temperatures, with the highest mating occurrence observed in LM at 23 °C, followed by LM at 32 °C (one-way ANOVA, $F_{3,12} = 4.86$, $P = 0.019$; Fig. 2C). Mating duration was, on average, approximately 1.3 times longer for SM at both temperatures (one-way ANOVA, $F_{3,66} = 7.24$, $P = 0.0001$; Fig. 2D). However, LM exhibited a higher sperm count than SM (one-way ANOVA, $F_{3,407} = 28.99$, $P = 0.0001$; Fig. 3A) and transferred more sperm to queens' spermathecae (one-way ANOVA, $F_{3,376} = 5.38$, $P = 0.001$; Fig. 3B). Averaged across both temperatures, the remaining sperm count in the seminal vesicles after mating was approximately 585,775 ± 10,925 for LM and 415,425 ± 20,125 for SM (Fig. 3A), while the number of sperm transferred to the spermathecae following mating with LM or SM was 42,665 ± 20 and 36,735 ± 365, respectively (Fig. 3B).

During competition, males made repeated attempts to mate with a queen (Fig. S1). Males that had failed to mate during competitive trials were successfully mated in large numbers during mating experiments without competition (Fig. S2).

3.2. Queen body mass and mate choice in bumblebees

In the second mating experiment, male mate choice between LQ and SQ was assessed at 23 °C and 32 °C. The cumulative mating rate of queens was higher at 23 °C than at 32 °C, but the difference was not statistically significant (two-sample *t*-test, df = 2, $P = 0.294$; Fig. 4A). When their cumulative mating rate (Fig. 4A) was normalized to 100 % and divided into LQ and SQ groups, interestingly, there was no significant difference in the mating rate between LQ and SQ at 23 °C (two-sample *t*-test, df = 3, $P = 0.069$). However, at 32 °C, LQ exhibited a significantly higher mating rate than SQ (one-way ANOVA, $F_{3,8} = 64.29$, $P = 0.0001$; Fig. 4B). At both temperatures, the majority of queens mated within 10 min, with LQ at 32 °C exhibiting the highest occurrence, followed by LQ and SQ at 23 °C (one-way ANOVA, $F_{3,8} = 20.59$, $P = 0.001$; Fig. 4C). Mating duration did not differ significantly between LQ and SQ at either temperature (one-way ANOVA, $F_{3,52} = 0.36$, $P =$

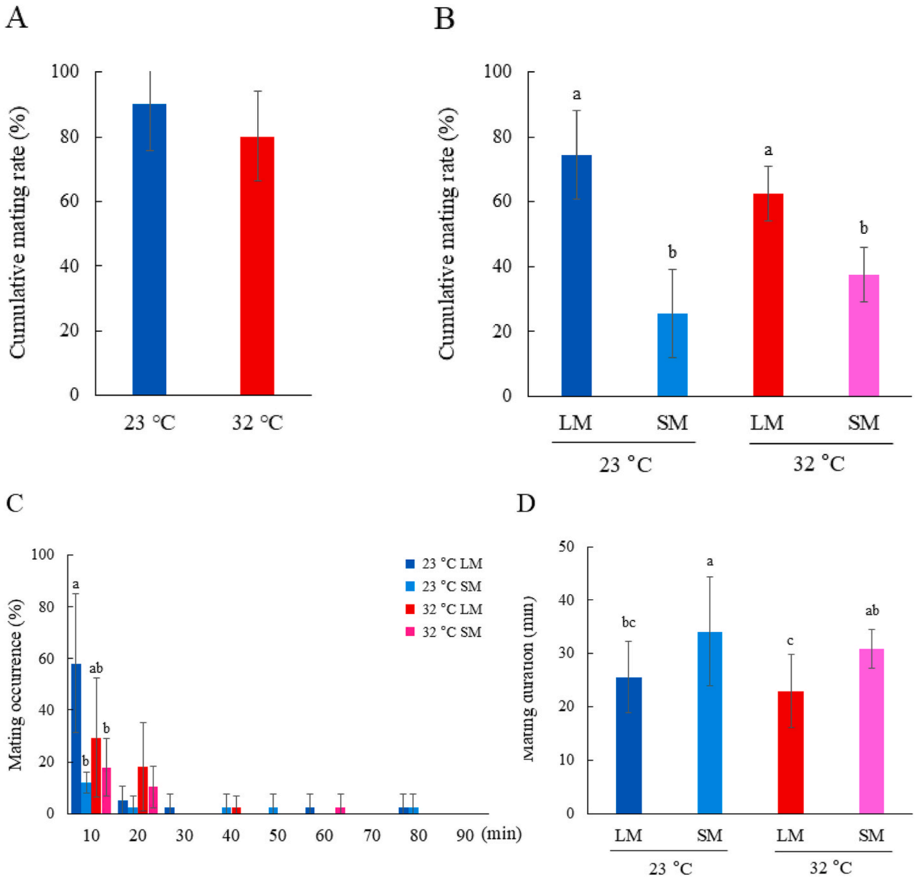


Fig. 2. Male body mass and mating competition. Equal numbers of LM, SM, and Q were placed in a mating box for a male-male competition experiment. (A) Cumulative mating rates (%) at 23 °C or 32 °C (n = 9–11, four replicates; *t*-test, *P* = 0.368). (B) Mating rates (%) of LM and SM at 23 °C or 32 °C (n = 9–11, four replicates). Different letters indicate significant differences among treatments (one-way ANOVA, *P* = 0.0001). (C) Timing of mating occurrences for LM and SM at 23 °C or 32 °C (n = 29 and 9, respectively, at 23 °C, and 20 and 12, respectively, at 32 °C). The timing of mating occurrence was marked at 10-min intervals. Different letters indicate significant differences among treatments in the proportion of mating occurrences within 10 min (one-way ANOVA, *P* = 0.019). (D) Mating durations for LM and SM at 23 °C or 32 °C (n = 29 and 9, respectively, at 23 °C, and 20 and 12, respectively, at 32 °C). Different letters indicate significant differences among treatments (one-way ANOVA, *P* = 0.0001).

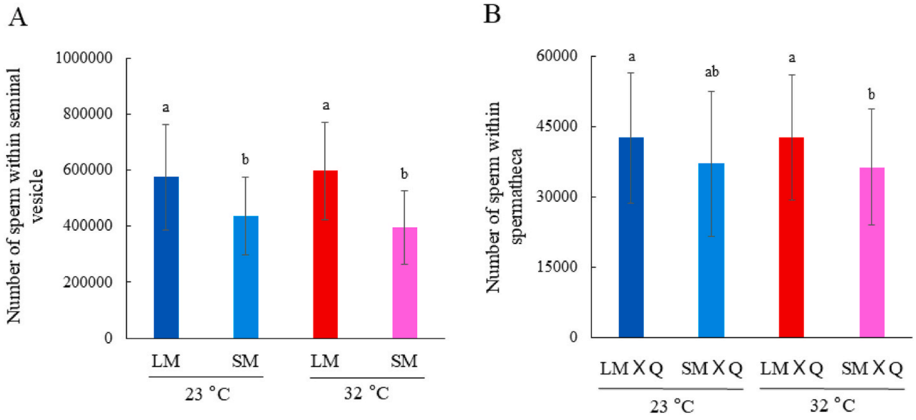


Fig. 3. Transfer of sperm from males to queens' spermathecae through mating. (A) Number of sperm in the seminal vesicles of LM or SM after mating at 23 °C or 32 °C (n = 29 and 8, respectively, at 23 °C, and 20 and 12, respectively, at 32 °C). Different letters indicate significant differences among treatments (one-way ANOVA, *P* = 0.0001). (B) Number of sperm in queens' spermathecae 24 h after mating with LM or SM at 23 °C or 32 °C (n = 24 LM × Q or 8 SM × Q at 23 °C, and 18 LM × Q or 11 SM × Q at 32 °C). Different letters indicate significant differences among treatments (one-way ANOVA, *P* = 0.001).

0.780; Fig. 4D). The survival of mated queens after hibernation decreased with body weight, indicating a dependence on body mass prior to hibernation (one-way ANOVA, $F_{5,12}$ = 77.16, *P* = 0.0001; Fig. 5).

4. Discussion

Ambient temperature is known to influence many aspects of bumblebee ecology (Amin et al., 2010; Cornelissen, 2011; Maebe et al., 2021; Vasiliev and Greenwood, 2021; Guiraud et al., 2021; Kenna et al.,

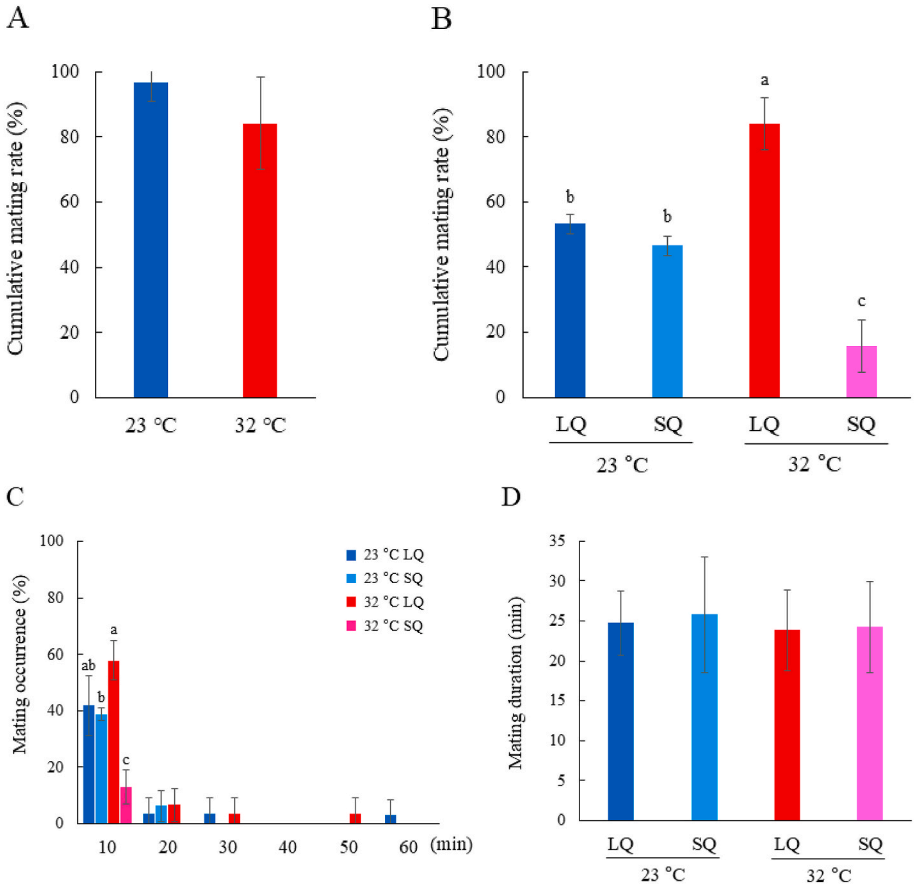


Fig. 4. Queen body mass and mate choice. Equal numbers of LQ, SQ, and M were placed in a mating box for a mate choice experiment. (A) Cumulative mating rates (%) at 23 °C or 32 °C (n = 10–11, triplicates; *t*-test, $P = 0.294$). (B) Mating rates (%) of LQ and SQ at 23 °C or 32 °C (n = 10–11, triplicates). Different letters indicate significant differences among treatments (one-way ANOVA, $P = 0.0001$). (C) Timing of mating occurrences for LQ and SQ at 23 °C or 32 °C (n = 26 and 14, respectively, at 23 °C, and 22 and 4, respectively, at 32 °C). The timing of mating occurrence was marked at 10-min intervals. Different letters indicate significant differences among treatments in the proportion of mating occurrences within 10 min (one-way ANOVA, $P = 0.001$). (D) Mating durations for LQ and SQ at 23 °C or 32 °C (n = 26 and 14, respectively, at 23 °C, and 22 and 4, respectively, at 32 °C; one-way ANOVA, $P = 0.780$).

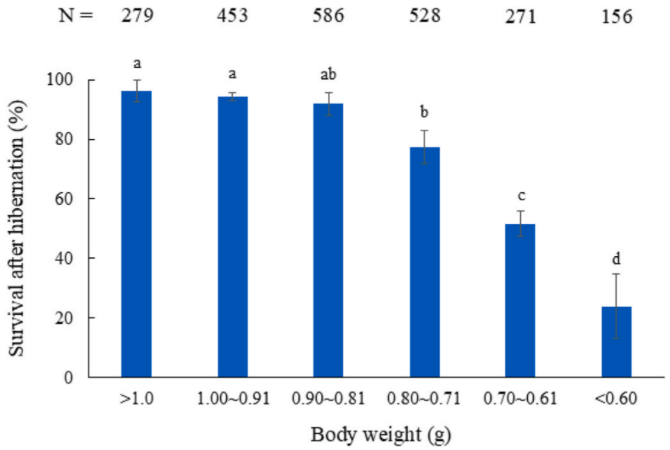


Fig. 5. Survival of *Bombus terrestris* queens after hibernation. The body weights of mated queens were measured before hibernation, and their survival was assessed after hibernation. Different letters indicate significant differences among treatments (one-way ANOVA, $P = 0.0001$). The total sample size (n) of each group is shown in the figure.

2021; Gérard et al., 2022a, 2022b, 2023; Kuo et al., 2023; Naumchik and Youngsteadt, 2023; Kim et al., 2024a, 2025), including reproduction (Campion et al., 2023). However, little is known about the impact of body mass on mate choice in bumblebees under warm conditions. Since virgin queens typically mate with a single male before undergoing winter hibernation (Baer et al., 2001), successful mating is crucial for the survival of subsequent generations.

In our male-male competition experiments with bumblebees of different body weights, large males exhibited a significantly higher mating rate than small males at both optimal (23 °C) and elevated (32 °C) mating temperatures. This suggest that male body mass in bumblebees influences mating success, which is consistent with the male–male competition and female mate choice observed in numerous other animals including insect species (Blanckenhorn, 2005; Byrne and Rice, 2006; Mattle and Wilson, 2009; Amin et al., 2012; Walzer and Schausberger, 2015). Small males appear to compensate for dominance of larger males in securing matings by displaying hyperaggressive behavior toward large male competitors during mating, as observed in other animals (Moretz, 2003; Jenssen et al., 2005; Morrell et al., 2005). However, our results indicate that in the bumblebee *B. terrestris*, large males outperformed small males in mating competition at both optimal and elevated temperatures.

During mating, bumblebee males transfer sperm, along with other substances, into the reproductive tracts of queens (Chapman et al., 1995; Baer et al., 2001). In the present study, the mate choice for large males may represent an important strategy for ensuring reproductive success in queens (Evans and Garcia-Gonzalez, 2016). Compared to small males, larger males initiate mating more quickly and have a shorter mating duration. Although small males mate slightly longer than large males at both temperatures, the large males exhibited a higher sperm count, enabling them to successfully transfer more sperm to the queens' spermathecae. Ejaculate volume is influenced by male body size, and queens may benefit from receiving more substances, as well as a higher quantity of sperm in the seminal fluid, from large males during mating (Ebbert, 1998; Harari et al., 1999; Schlüns et al., 2003; Gençer and Firatli, 2005; Gençer and Kahya, 2011; Crean et al., 2016; Hopkins et al., 2017; Macartney et al., 2019). Therefore, a queen's preference for large males may enhance sperm viability through male substances including seminal fluid proteins, thereby enhancing reproductive success (den Boer et al., 2015; Zhang et al., 2021; Kim et al., 2024b). Thus, our findings may suggest a mating preference for large males as a fecundity benefit of mate choice in which a higher quantity of sperm is potentially favored.

Body size influences mating preference—both queens and males have a preference for large mating partners (Andersson, 1994; Blanckenhorn, 2005), which represents a behavioral evolution aimed at optimizing reproduction (Evans and Garcia-Gonzalez, 2016). Mating behaviors are also influenced by the sex ratio, age, and ambient temperature (Boomsma and Ratnieks, 1996; Amin et al., 2010; Treanore et al., 2021; Kim et al., 2025). In a mate choice experiment with bumblebee queens of different body weights, no significant difference existed in the mating rates between large and small queens at 23 °C but there was a strong shift in male preference towards mating with large queen at 32 °C. The significantly higher male preference for large queens at 32 °C was attributed to temperature effects. This suggests a mate choice shift in response to thermal stress.

In the annual life cycle of bumblebees, mated queens hibernate underground during winter and initiate new colonies in spring. Sufficient energy reserves are vital for queens to survive the hibernation period (Hahn and Denlinger, 2007, 2011), as reflected by their higher body weights (Vesterlund et al., 2014). We found the expected positive relationship between the survival and body mass of mated queens during hibernation, indicating that mated queens with a smaller body size were less effective at surviving hibernation (Vesterlund et al., 2014; Keaveny and Dillon, 2022). This further confirms that the body mass of bumblebee males and queens may be a key factor influencing reproduction, including both sperm quantity and survival during hibernation.

In conclusion, our findings reveal the relationships between body mass and success in mate choice in bumblebees, offering novel insights into the shifts in mate choice occurring under warm conditions. We also developed a model (Fig. 6) illustrating how warm conditions and the body mass of bumblebee queens and males influence reproductive processes. This model shows that a larger male body mass influences male-male competition, resulting in the transfer of a higher quantity of sperm to queens' spermathecae. Furthermore, a preference for large queens was observed, particularly at elevated temperatures, possibly representing a form of mate choice in response to thermal stress, ensuring a sufficient sperm quantity and high survival after hibernation. The results obtained regarding the mating behavior of bumblebees under warm conditions provide new insights into their reproductive strategies and have important implications for the survival of wild bumblebee populations amidst ongoing and future climate change.

CRediT authorship contribution statement

Min Su Park: Validation, Investigation, Formal analysis. **Ji Hyun Woo:** Validation, Investigation, Formal analysis. **Hyung Joo Yoon:** Visualization, Data curation. **Bo Yeon Kim:** Visualization, Data curation. **Kyeong Yong Lee:** Software, Formal analysis. **Steven A. Trewick:** Writing – review & editing, Data curation. **Kwang Sik Lee:** Writing – review & editing, Writing – original draft, Supervision, Conceptualization. **Byung Rae Jin:** Writing – review & editing, Writing – original draft, Supervision, Conceptualization.

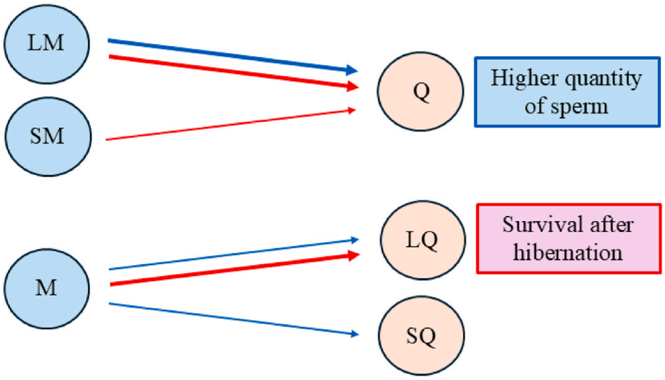


Fig. 6. Summary of the relationships between body mass and success in mate choice in bumblebees. Thick blue and red arrows indicate significant associations at mating temperatures of 23 °C and 32 °C, respectively. The thin red arrow represents a nonsignificant increasing trend at 32 °C, while thin blue arrows indicate nonsignificant differences at 23 °C.

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Declaration of competing interest

There are no competing interests to declare.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jtherbio.2025.104210.

Data availability

Data will be made available on request.

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