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Original article

Mating propensity of *Bombus terrestris* reared in different photoperiodic regimes*

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Abstract – The study investigated the mating propensity of *Bombus terrestris* by altering the photoperiod (L0:D24, L8:D16, L16:D8 and L24:D0) experienced by different age groups of bees. Three day old queens showed the lowest mating propensity under all photoperiod regimes. Queen mating propensity increased with age. Among the constant-dark treated bumblebees, the highest frequency of mating (68.33%) was observed for 9 day old queens. In contrast, 7 day old queens showed the highest mating propensities at L8:D16, L16:D8 and L24:D0 (88.33, 93.33 and 77.92%, respectively). The highest mating propensities under both short and long day conditions occurred with a queen-to-male ratio of 1:2. The maximum post-mating queen survival rate (96.67%) was found in L8:D16 conditions, and the lowest (60.83%) in L24:D0 condition. These different response patterns indicate that pre-mating light regimes greatly influence mating propensity of *B. terrestris*.

***Bombus terrestris* / photoperiod / mating**

1. INTRODUCTION

In nature, *Bombus terrestris* L. queens usually mate with males in late summer and the mated queens leave the nest and seek out suitable sites for hibernation. Commercial companies rear colonies throughout the year and mate young queens with males from other colonies. But in the closed breeding systems of commercial breeders there might be a low level of mating success. Copulation itself is a lengthy affair, allowing females to further evaluate their chosen mate's quality before accepting and storing his sperm (Field et al., 1999). The dynamics of sperm descent and the extent of sperm storage might limit the possibilities for a male to mate successfully with a newly encountered female. Bergström (1982) reported that volatile secretions from the cephalic labial glands of bumblebee males and the mandibular glands of queens played a role in producing sex pheromones which strongly influenced mating. Other authors reported that sperm content of the spermatheca (Röseler, 1973) or vas-deferentia (Duchateau and Marien, 1995) were responsible for mating in *B. terrestris.* There are many specific reciprocal activities between sexes in insect mating behavior, which are usually initiated by one sex and influenced by environmental, chemical, visual, tactile or other cues. Photoperiod is known to be an important factor controlling the sexual maturation of insects. In migratory locust, *Locusta migratoria* L., sexual maturation proceeds normally at a short photoperiod, but is inhibited by a longer photoperiod (Takana, 1994). Circadian rhythms also influence behavior by transmitting intracellular signals to organize cellular metabolism, and intercellular signals to organize multicellular physiology and behavior (Jackson et al., 1998). Photoperiodism

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had significant effects on the oviposition and colony development of *B. terrestris* (Tasei et al., 1998; Yoon et al., 2003). Philogene and McNeil (1984) stated that sperm migration and mating of insects was influenced by artificial light. Duchateau (1985) reported data on the impact of temperature, light and time of day for bumblebee mating. Despite abundant literature on bumblebee sexuality there is still missing information on factors eliciting mating behavior. In the present study, we examined four photoperiodic regimes to evaluate: (a) How day length and bee age affect mating propensity, (b) How mating propensity is affected by the sex ratio in the mating cage, and (c) How photoperiod regime affects pre and post mating survival.

2. MATERIALS AND METHODS

2.1. Origin of bumblebees and rearing conditions

For mass rearing of *Bombus terrestris dalmatinus* (Dalla Torre), hibernated queens were obtained from the mass rearing system of the Department of Agricultural Biology, Kyungpook National University, Daegu, Korea. Colonies were maintained in a growth chamber at 28 °C, 60% RH and the bees were provided with pollen and sugar solutions (1.5:1, w/v) as a diet. For observation of the colonies, red light of 10 lux was used. Following repeated observations, newly emerged males and queens which were whitish in colour were collected daily by using forceps. Mating experiments were observed with a total of 5280 queens. They were collected from 218 colonies and were transferred in small transparent plastic boxes measuring 16 × 11 × 7 cm. Ten queens were kept in each box and provided with fresh pollen in a small plastic petridish and sugar solutions (1.5:1, w/v) in a small tube. Males were reared in illuminated flight cages (40 × 30 × 30 cm). Fifty males were put in each cage and supplied with fresh pollen and sugar solutions (1.5:1, w/v). The boxes of queens and cages of males were held in four separate rooms maintained at 25 °C, 50% RH and photoperiodic regimes of L0:D24, L8:D16, L16:D8 and L24:D0 until mating time. Photoperiodic treatments were administered with fluorescent white (L36/18-860, 300 Hz, λ = 460 nm) light at an intensity of ~700 lux on the floor of the male cages and queen boxes. The intensity of light was measured with a digital lux meter and the photophase was regulated with an appliance timer. In our two experiments the males were 2 days older than the queens in their respective age groups. Number of mating couples was observed in 40 × 40 × 60 cm plastic cages. Sugar solutions and fresh pollen were supplied inside the cages. Mating was recorded in a room maintained at 25 °C and 50% RH. During mating, the cages were illuminated with 200 W mercury bulbs (GGY200, 60 Hz, λ = 580 nm) positioned 0.5 m above the mating cages. The intensity of light on the floor of the mating cages was 2300 lux.

2.2. Observations

Before mating, the number of surviving queens in each queen rearing box were counted. The occurrence of copulation was recorded for one hour and the mating couples were withdrawn every 15 minutes. After mating sessions the mated queens were reared in small transparent plastic boxes measuring 16 × 11 × 7 cm. Ten queens were kept in each box. The boxes were provided with fresh pollen in a small petridish and sugar solution (1.5:1 w/v) in a tube. The boxes were kept in the respective photoperiodic room (L0:D24, L8:D16, L16:D8 and L24:D0). After 15 days the number of post-mating queen surviving were recorded.

2.3. Influence of age of sexuals and photoperiod on mating propensity

To observe the number of matings in each photoperiodic regime, five different age groups of queens were selected, including: 3, 5, 7, 9 and 11 days old. For mating twenty queens and 40 males were used in each replication, and each age group reared in the same photoperiodic regime was replicated 12 times.

2.4. Influence of queen: male ratio on mating propensity

Queen:male ratios of 1:1, 1:1.5 and 1:2 were used to observe the mating propensities of 7 day old queens with 9 day old males reared in short day, L8:D16 and long day, L16:D8 conditions.

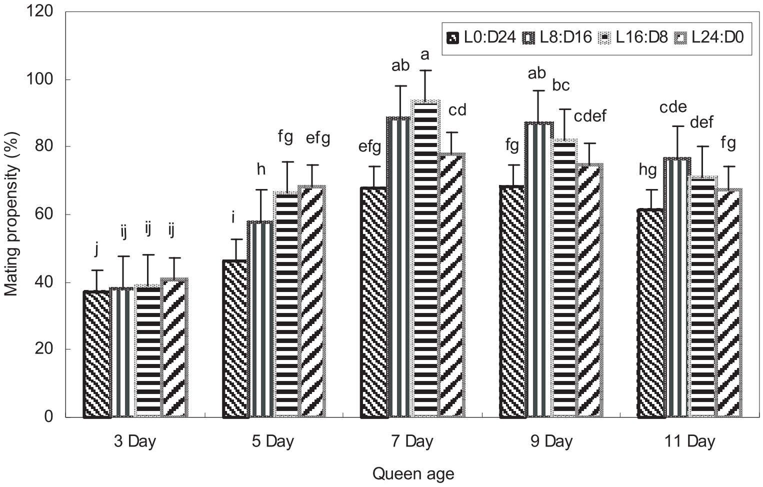

Figure 1. Mating propensity (% mean ± S.E.) of *B. terrestris* in relation to photoperiodic regimes and ages of bees. Bars with no common letter are significantly different ($P \leq 0.05$, Duncan's multiple range test). N = 240 for each photoperiodic regime and age group of queens.

2.5. Statistics

Percent mating and survival data were analyzed by analysis of variance (ANOVA) after arc-sine transformation. The mean values were then separated by Duncan's multiple range test (Duncan, 1951). Means are reported as ± one standard error (S.E.).

3. RESULTS

3.1. Photoperiod and age-specific mating propensity

A total of 4800 *B. terrestris* queens belonging to 5 age groups were used to observe the mating propensity in relation to 4 photoperiodic regimes. Photoperiodic regimes and age of sexuals had significant effects on the mating propensity ($F_{3,220} = 27.91$, $P < 0.0001$, $F_{4,220} = 130.27$, $P < 0.0001$, respectively). However, photoperiod regime and age showed a significant interaction ($F_{12,220} = 5.26$, $P < 0.0001$). Figure 1 shows that 3 day old queens reared in all photoperiodic regimes achieved the lowest mating propensities ranging from 37% to 40% for the 4 photoperiods. The higher percentage of mating propensities were observed for the 7 day old queens reared in long day and short day (93.33 ± 5.77 and 88.33 ± 9.85%, respectively). However, in L0:D24 and L24:D0 conditions, the highest copulation rates (68.33 ± 10.30 and 77.92 ± 10.54%, respectively) were found in 9 and 7 day old queens with 11 and 9 day old males, respectively.

3.2. Queen: male ratio and mating propensity

When examining mating propensities for queen: male ratios of 1:1, 1:1.5 and 1:2 reared in short and long day condition, significant differences were found ($F_{2,33} = 8.47$, $P = 0.001$ and $F_{2,33} = 20.83$, $P < 0.0001$ respectively). Figure 2 A shows that in L8:D16 photoperiod the queen: male ratios of 1:2 and 1:1.5 resulted in higher mating propensities (88.33 ± 9.85, 83.75 ± 9.08%, respectively) than the ratio of 1:1 (71.67 ± 7.78%). The results of queen: male ratios of 1:2, 1:1.5 and 1:1 (Fig. 2 B) shows that in long day treated bumblebees mating propensities (93.33 ± 5.77%) with a 1:2 queen: male ratio was significantly higher than with the other two ratios (86.67 ± 6.51 and 73.75 ± 8.56%, respectively).

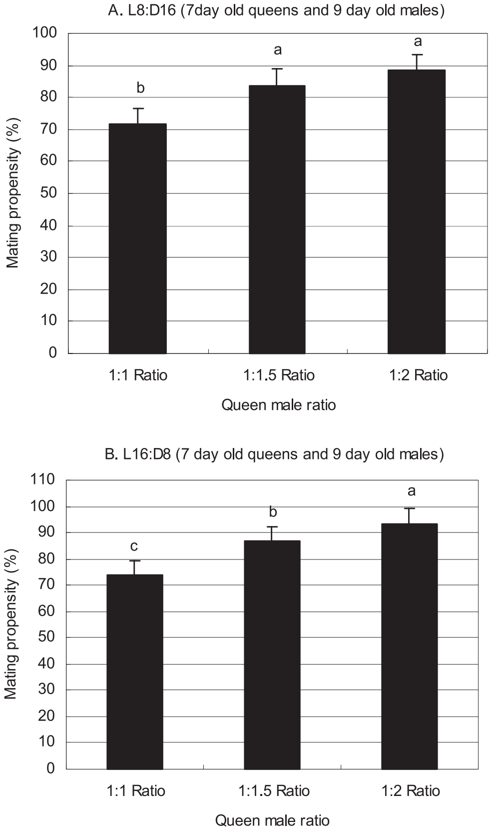

Figure 2. Mating propensity (% mean ± S.E.) of *B. terrestris* in relation to (A) L8:D16 and (B) L16:D8 photoperiodic regimes using different ratio of queens and males. Bars with no common letter are significantly different ($P \leq 0.05$, Duncan's multiple range test). N = 240 queens for each ratio.

3.3. Pre-mating and post-mating survival of queen

The different photoperiodic regimes resulted in significant effects on the pre-mating and post-mating survival (Fig. 3) of *B. terrestris* ($F_{3,44} = 8.54$, $P = 0.0001$, and $F_{3,44} = 27.29$, $P < 0.0001$, respectively). Short and long day treated bumblebees had the highest pre-mating survival rates (97.50 ± 6.22 and 95.83 ± 7.93%, respectively), whereas constant light and constant dark treated bumblebees displayed significantly lower pre-mating survival rates (76.67 ± 15.57 and 84.17 ± 14.43%, respectively). On the other hand, post mating queen survival rates were higher in the constant dark, short-day and long-day treatments i.e.: 92.5 ± 10.55, 96.67 ± 6.51, and 95.83 ± 7.93%, respectively. The minimum survival rate was 60.83 ± 16.76% in the constant light treatment.

4. DISCUSSION

Different light regimes showed a strong effect on the mating propensity of *B. terrestris*. The higher rates of copulation success (93.33 ± 5.77 and 88.33 ± 9.85%, respectively) occurred in the L16:D8 and L8:D16 pre-mating conditions, when queens were 7 day old and males 9 days old. Thus, the effects of photoperiod on bumblebee mating were age dependent. These results were in agreement with Tasei et al. (1998) who reported that 6 day old queens were effective for mating. Tasei et al. (1998) also defined the age groups for mating as ranging between 6 and 27 days for males and between 2 and 11 days for queens, and showed that the chances of the queens mating dropped dramatically after 11 days. Duvoisin et al. (1999) stated that the mean value of the age of mating was 6.1 days for queens and 12.1 days for males, and the sperm content of their spermathecae increased from emergence until 6 days. It was suggested by Gretenkord (1997) that the readiness of queens to mate seemed to be the greatest at around 6 days of age. Our findings are in agreement of Duchateau (1985), who reported that aging queens become gradually less receptive to mating. Duchateau (1985) suggested that this might be due to a decrease in queen's sensitivity to the male pheromones. She suggested that other factors are also important in eliciting mating behavior, for example, temperature, light or time of day. In the present investigation, all the photoperiodic regimes using 7–9 day old queens allowed mating. As the age increased the receptivity decreased significantly. Nevertheless the ages 7 and 9 days for queens and males, respectively, were close to those reported as the more favourable to mating by Duchateau, 1985 and Djegham et al., 1994. These results

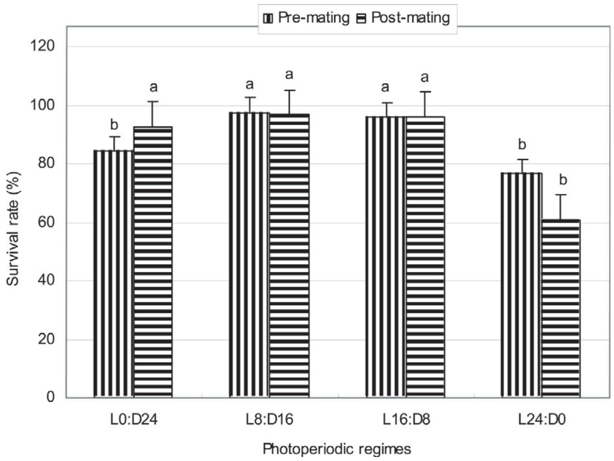

Figure 3. Pre-mating and post-mating queen survival rate (% mean ± S.E.) of *B. terrestris* in relation to different photoperiodic regimes. Bars with no common letter are significantly different ($P \leq 0.05$, Duncan's multiple range test). N = 120 queens for each photoperiodic treatment.

are consistent with those of Tasei et al. (1998), who reported that 14–18 day old males exhibit less sexual activity. Jung et al. (2001) recorded the highest mating rate of *B. ardens* by 7 day old queens. They found that 80% of queens mated in 4 hours at L14:D10 photoperiodic regime. Lee et al. (2002) conducted a study of bumblebee, *B. ignitus* mating in the field and in indoor conditions. They observed that the mating success in the field were 80%, and 60% in indoors. Our study suggests that 7 day old queens with 9 day old males living in long day or short days have significantly higher successful rate of mating compared to bumblebees of the same age, kept in constant light or dark. A queen: male ratio of 1:2 showed significantly higher mating propensity because the queens had more chances to select the best partner. When newly emerged queens were kept in L24:D0 conditions, they showed high sensitivity to light and a tendency to fight and kill each other in the confined spaces and also made wax structures and laid eggs. This, in turn, may have contributed to lower mating success. Similar conflicts were observed in the case of post-mated queens in the same photoperiodic treatment. As a result queen post-mating survival rate was significantly lower in L24:D0 than in L8:D16 and L16:D8 treated bumblebees.

Photoperiod is an abiotic factor that influences insect behaviour (Leather et al., 1993). Light regimes change neurotransmitter concentrations in *Acheta*'s brain (Germ and Kral, 1995) or the hormonal state in *Tribolium* (Hirashima et al., 1995) and influence the sexual maturity and pheromone production of a Lepidoptera species (Gemeno and Haynes, 2001). Photoperiod influences ovarian maturation of *Locusta* (Tanaka et al., 1993). Barker and Herman (1976) showed that Lepidoptera reproduction was controlled by the neuroendocrine system. Wang and Millar (2000) and Hel et al. (2004) reported that photoperiod reduced the pre-mating period in Heteroptera. Barth et al. (1997) reared *Drosophila melanogaster* in different photoperiodic regimes and found that light regimes had a positive effect on mating. Inappropriate lighting adversely affected male fertility in moths (Bebas and Cymborowski, 1999) and photoperiod had a significant role in the mating success in moths (Seth et al., 2002). The effects of light regimes caused, at least in part, interference with sperm release and descent (Bebas and Cymborowski, 1999). So far, the discussion had concentrated on the effects of light regimes on mating behaviour. The present results showed that mating status of *B. terrestris* was affected by the combined effect of photoperiodism and the age of bees. Queens of 7 days old and 9 days old males reared in short day and long day photoperiodic regimes reached the best receptivity to mating and attained higher rate of mating than in constant dark or light conditions. So, we believe that our results will contribute information for the effective rearing of bumblebees to the commercial breeders who usually rear new sexuals in dark conditions.

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Résumé – **Propension de *Bombus terrestris* à s'accoupler en fonction du régime photopériodique durant l'élevage.** La photopériode influence la production de phéromone, la maturation sexuelle et le comportement d'accouplement des insectes. Dans cette étude nous avons examiné l'action de 4 régimes photopériodiques ((L0:D24, L8:D16, L16:D8 et L24:D0) sur la propension du bourdon *Bombus terrestris* L. à s'accoupler. Au total 5280 reines appartenant à 5 groupes d'âge (3, 5, 7, 9 et 11 j) et des mâles plus vieux de 2 j pour chaque groupe d'âge ont été observés et élevés jusqu'au moment de l'accouplement dans 4 enceintes de régulation de la photopériode éclairées avec une lumière blanche fluorescente d'environ 700 lux. La chambre d'accouplement était maintenue à 25 °C et 50% d'HR et les cages d'accouplement éclairées avec des ampoules à mercure de 200 watts et d'une intensité de 2300 lux. Après l'accouplement les reines ont été élevées dans l'enceinte de régulation de la photopériode durant 2 semaines. Avant l'accouplement et durant 15 j après, on a enregistré les données des reines survivantes. Les résultats concernant l'accouplement montrent que les régimes photopériodiques et l'âge des reines ont eu un effet significatif sur la propension à s'accoupler (Fig. 1). Les propensions les plus fortes se sont produites pour les reines âgées de 7 j élevées en jours longs et jours courts (93,33 ± 5,77 et 88,33 ± 9,85 % respectivement). Pourtant, en conditions L0:D24, le taux de copulation le plus fort (68,33 ± 10,30 %) a été trouvé pour les reines âgées de 9 j et les mâles de 11 j. Avec des ratios reine/mâle de 1:1, 1:1,5 et 1:2 dans des conditions de jours courts et de jours longs, les différences dans la propension à s'accoupler étaient significatives (Fig. 2A, B). En jours courts et en jours longs, la plus forte propension à s'accoupler a été trouvée pour le ratio reine/mâle de 1:2 (88,33 ± 9,85, et 93,33 ± 5,77 %, respectivement) et la plus faible propension pour le ratio 1:1 (71,67 ± 7,78 et 73,75 ± 8,56 % respectivement). Parmi les divers régimes photopériodiques, les reines traitées par jours courts (Fig. 3D) ont montré le plus fort taux de survie avant l'accouplement (97,50 ± 6,22 %) et celles traitées par lumière constante le plus faible taux de survie (76,67 ± 15,57 %). Par contre, le taux de survie maximum après l'accouplement a été enregistré pour la photopériode L8:D16 (96,67 ± 6,51 %) et le plus faible (60,83 ± 16,76 %) pour la photopériode L24:D0. Nos résultats montrent que les reines élevées sous les régimes L8:D16 et L16:D8 étaient plus enclines à s'accoupler que les reines élevées dans des conditions de lumière ou d'obscurité constante. Dans le ratio reine/mâle de 1:2 les reines avaient plus d'occasions de choisir le meilleur partenaire et la propension à s'accoupler a été la plus forte. La réceptivité à l'accouplement et la survie après l'accouplement, moindres dans le régime L24:D0, peuvent être dues à la forte sensibilité des reines à la lumière et à des conflits dans un espace confiné. Les résultats suggèrent que l'effet combiné du photopériodisme et de l'âge des insectes ont influencé la propension de *B. terrestris* à s'accoupler. Ces informations peuvent avoir d'importantes implications dans l'élevage en masse des bourdons.

***Bombus terrestris* / accouplement / photopériode / âge**

Zusammenfassung – **Paarungsverhalten von *Bombus terrestris* bei unterschiedlichen Fotoperioden.** Die Fotoperiode hat bei Insekten Einfluss auf die Pheromonproduktion, den sexuellen Reifungsprozess und das Paarungsverhalten. In der vorliegenden Arbeit haben wir den Einfluss von 4 verschiedenen fotoperiodischen Arrangements (Licht-Dunkel-Perioden von L0:D24, L8:D16, L16:D8 und L24:D0) auf das Paarungsverhalten von *Bombus terrestris* untersucht. Insgesamt wurden 5280 Königinnen in 5 Altersgruppen mit 3, 5, 7, 9 und 11 Tagen getestet und bis zur Paarung in 4 getrennten Räumen mit unterschiedlichen Licht-Dunkel-Perioden (weißes Fluoreszenzlicht, Intensität 700 Lux) gehalten. Die Männchen wurden bis zur Paarung in den entsprechenden Räumen in separaten Flugkäfigen aufgezogen. Sie waren zum Zeitpunkt der Paarung jeweils 2 Tage älter als die Weibchen aus den entsprechenden Altersgruppen. Die Paarungsbox wurde bei 25 °C und einer RF von 50 % mit einer 200W Mercury Lampe mit 2300 Lux beleuchtet. Nach der Paarung wurden die Königinnen unter den jeweiligen fotoperiodischen Bedingungen für weitere zwei Wochen gehalten. Vor der Paarung und 15 Tage danach wurde jeweils der Anteil an überlebenden Königinnen erfasst.

Sowohl die fotoperiodischen Bedingungen als auch das Alter der Königin hatten einen signifikanten Effekt auf die Paarungsneigung

(Abb. 1). Die höchste Paarungsneigung zeigten 7 Tage alte Königinnen, die unter Kurztag- bzw. Langtagbedingungen aufgezogen wurden (93,33 ± 5,77 bzw. 88,33 ± 9,85 %). Allerdings wurde unter L0:D24-Bedingungen die höchste Kopulationsrate (68,33 ± 10,30 %) bei 9 Tage alten Königinnen mit 11 Tage alten Männchen gefunden. Das Verhältnis von Königinnen zu Männchen (1:1, 1:1,5 und 1:2), die unter Kurztag- bzw. Langtagbedingungen aufgezogen wurden, hatte einen signifikanten Einfluss auf die Paarungsneigung (Abb. 2 A und B). Bei beiden Aufzuchtbedingungen war die Paarungsneigung bei einem Geschlechterverhältnis von 1:2 am höchsten (88,33 ± 9,85 bzw. 93,33 ± 5,77 %) und bei 1:1 am niedrigsten (71,67 ± 7,78 bzw. 73,75 ± 8,56 %). Die Überlebensrate vor der Paarung (Abb. 3) war bei Hummeln unter Kurztagbedingungen am höchsten (97,50 ± 6,22 %) während die unter Dauerlicht gehaltenen Hummeln die niedrigsten Überlebensraten aufwiesen (76,67 ± 15,57 %). Nach der Paarung war dagegen die Überlebensrate bei den unter L8:D16-Bedingungen gehaltenen Hummeln am höchsten (96,67 ± 6,51 %) und bei den L24:D0-Bedingungen am niedrigsten (60,83 ± 16,76 %).

Unsere Ergebnisse zeigen, dass Königinnen die unter den fotoperiodischen Bedingungen von L8:D16 und L16:D8 aufgezogen wurden eher zur Paarung neigen als Königinnen die unter Dauerlicht oder Dauerdunkel aufgezogen wurden. Bei einem Verhältnis Männchen zu Königinnen von 2:1 haben die Königinnen mehr Möglichkeiten zur Auswahl des besten Partners und zeigen somit die höchste Paarungsneigung. Die geringere Paarungsbereitschaft und Überlebensrate nach der Paarung unter den L24:D0-Bedingungen könnten durch die hohe Lichtempfindlichkeit der Königin und Konflikte innerhalb der engen Aufzuchtbox bedingt sein. Die Ergebnisse deuten darauf hin, dass die Paarungsneigung bei *B. terrestris* durch einen kombinierten Effekt aus Fotoperiodik und Alter der Hummeln beeinflusst wird. Diese Befunde könnten für die Massenaufzucht von Hummeln von großer praktischer Bedeutung sein.

***Bombus* / Fotoperiode / Paarung**

REFERENCES

Barker J.F., Herman W.S. (1976) Effect of photoperiod and temperature on reproduction of the monarch butterfly, *Danus plexippus*, J. Insect. Physiol. 22, 1565−1568.

Barth M., Hirsch H.V.B., Heisenberg M. (1997) Rearing in different light regime affects courtship behaviour in *Drosophila melanogaster*, Anim. Behav. 53, 25−38.

Bebas P., Cymborowski B. (1999) Effect of constant light on male sterility in the cotton leaf worm, *Spodoptera littoralis*, Physiol. Entomol. 24, 165−170.

Bergström G. (1982) Chemical composition, similarity and dissimilarity in volatile secretions: examples of indications of biological function, in: Les médiateurs chimiques agissant sur le comportement des insectes, Les colloques de I'INRA n° 7, 16–20 nov. 1981, publ. 1982, pp. 289−296.

Djegham Y., Verhaeghe J.C., Rasmont P. (1994) Copulation of *Bombus terrestris* L, J. Apic. Res. 33, 15−20.

Duchateau M.J. (1985) Analysis of some methods for rearing bumblebee colonies, Apidologie 16, 225−227.

Duchateau M.J., Marien J. (1995) Sexual biology of haploid and diploid males in the bumble bee, *Bombus terrestris,* Insectes Soc. 42, 255−266.

Duncan D.B. (1951) A significance test for differences between ranked treatments in an analysis of variance, Virginia J. Sci. 2, 171–189.

Duvoisin N., Baer B., Schmid-Hempel P. (1999) Sperm transfer and male competition in bumble bees, Anim. Behav. 58, 743−749.

Field S.A., Taylor P.W., Yuval B. (1999) Sources of variability in copulation duration of Mediterranean fruit flies, Entomol. Exp. Appl. 92, 271–276.

Gemeno C., Haynes K.F. (2001) Impact of photoperiod on the sexual behaviour of black cutworm moth (Lepidoptera: Noctuidae), Environ. Entomol. 30, 189−195.

Germ M., Kral K. (1995) Influence of visual deprivation on levels of dopamine and serotonin in the visual system of house crickets, *Acheta domesticus*, J. Insect Physiol. 41, 57−63.

Gretenkord C. (1997) Laborzucht der Dunklen Erdhummel *Bombus terrestris* L. (Hymenoptera: Apidae) und toxikologische Untersuchungen unter Labor- und Halbfreilandbedingungen, Ph.D. Thesis, Rheinische Friedrich-Wilhelms University, Bonn.

Hel X.Z., Wang Q., Carpenter A. (2004) Effect of day length on development and reproductive diapause in *Nysius huttoni* White (Heteroptera: Lygaeidae), J. Appl. Entomol. 128, 528−532.

Hirashima A., Takeya R., Taniguchi E., Eto M. (1995) Metamorphosis, activity of Juvenile-hormone esterase and alteration of ecdysteroid titres; effects of larval density and various stress on the red flour beetle, *Tribolium freemani* Hinton (Coleoptera: Tenebrionidae), J. Insect Physiol. 41, 383−388.

Jackson F.R., Zhang X., McNeil G.P. (1998) Oscillating molecules and circadian clock output mechanisms, Mol. Physiol. 3, 381–385.

Jung J.K., Mah Y.I., Kim J.S. (2001) Mating rate of *Bombus ardens* (Hymenoptera: Apidae) queens with day time and age, Korean J. Apic. 16, 37−44 [in Korean].

Leather S.R., Walters K.F.A., Bale J.S. (1993) The ecology of insect overwintering, Cambridge Univ. Press.

Lee S., Mah Y., Yoon H., Park I., Bae T. (2002) The mating in the field and the indoor condition of Korea indigenous bumble bee, *Bombus ignitus*, queen, Korean J. Apic. 17, 103−112 [in Korean].

Philogene B., McNeil J.N. (1984) The influence of light on the nondiapause related aspects of development and reproduction in insects, Photochem. Photobiol. 40, 753–761.

Röseler P.F. (1973) Die Anzahl der Spermien im Receptaculum seminis von Hummel Königinnen (Hym. Apoidea, Bombinae), Apidologie 4, 267−274.

Seth R.K., Rao D.K., Reynolds S.E. (2002) Movement of spermatozoa in the reproductive tract of adult male, *Spodoptera litura*: daily rhythm of sperm descent and the effect of light regime on male reproduction, J. Insect Physiol. 48, 119−131.

Takana H. (1994) Geographic variation of embryonic diapause in the migratory locust, *Locusta migratoria* L. (Orthoptera: Acrididae) in Japan, Jpn J. Entomol. 62, 629–639.

Tanaka S., Hakomori T., Hasegawa E. (1993) Effect of day length and hopper density on reproductive traits in a Japanese population of the migratory locust, *Locusta migratoria* L, J. Insect Physiol. 39, 571−580.

Tasei J.N., Moinard C., Moreau L., Himpens B., Guyonnaud S. (1998) Relationship between aging, mating and sperm production in captive *Bombus terrestris*, J. Apic. Res. 37, 107−113.

Wang Q., Millar J.G. (2000) Mating behaviour and evidence for male produced sex pheromones in *Leptoglossus clypealis* (Heteroptera: Coreidae), Ann. Entomol. Soc. Am. 93, 972−976.

Yoon H.J., Lee S.B., Park I.G., Sim H.S., Kim S.E. (2003) Effect of photoperiod on oviposition and colony development of the bumblebee, *Bombus terrestris* queens, Korean J. Apic. 18, 49–54 [in Korean].