

# EFFECT OF TEMPERATURE TREATMENT DURING DEVELOPMENT OF *OSMIA RUFa* L., ON MORTALITY, EMERGENCE AND LONGEVITY OF ADULTS

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## Abstract

The red mason bee (*Osmia rufa* L.) is a univoltine solitary species of the *Osmia* genus. This bee is reared on a commercial scale and used as a managed alternative pollinator. We intended that the results of our study would improve the management of this bee so as to synchronise their flying period with the blooming of crops. In the spring, we moved newly occupied nests of the red mason bee to a laboratory and placed them in incubators. Immature development was examined at three constant temperatures, 20°C, 25°C, and 30°C. Selected nests were opened to monitor the subsequent developmental stages. The remaining bees were wintered in nests stored at cool temperature (4°C). In April, we removed the insects from the nests and began incubation at 25°C to establish the emergence time of adult individuals. To determine the survival rate of adult bees, we moved the emerged specimens to cages, where they were fed and kept until death. Temperature increase generally reduced immature development time. But this tendency was not observed in the prepupal stage. During ontogeny the highest mortality rate was observed in wintering adult insects at developmental temperatures of 25°C and 30°C. Bees developing at constant temperatures emerged faster during spring incubation in comparison to those developing in natural conditions. Constant developmental temperatures decreased the survival rate of females as post-emergence adult insects. The survival rate of males was lower at the developmental temperature of 30°C.

**Keywords:** development, mason bee, rearing, temperature control

## INTRODUCTION

The red mason bee (*Osmia rufa* L.) is a solitary univoltine bee species of the *Osmia* genus, very common in Central Europe. Species of this genus are managed in many countries (Raw, 1972; Wójtowski, 1983; Parker, Batra, & Tepedino, 1987; Torchio, 1991; Bosch, 1994; Kemp, 2000; White, Son, & Park, 2009). The insects are successfully used for pollinating fruit and seed cultivations and cultivations under greenhouse shelter (Holm, 1973; Batra, 1995; Bosch & Kemp, 2002; Krunić & Stanisavljević, 2006). Since *Osmia rufa* has become commercially available in Poland, it can be used as a managed pollinator from February to July.

The red mason bee is an early spring species. The flight season of the red mason bee begins in late March and early April and ends in mid-June. Males appear approximately a week earlier

than females (protoandry), and live from 3 to 4 weeks. Females are monandrous and males are polygamous – but not all of the males mate with females (Seidelmann, 1999). While building their nests, female of red mason bees use pre-existing cavities, which are usually tube-shaped. The insects are readily attracted to holes in dry stems of hollow plants (e.g. the common reed), dead wood or even cavities in building walls (Wójtowski, 1983). Cells in the nest are arranged linearly in series with transverse partitions in between, built by the female who collects mud and mixes it with saliva (Wójtowski, 1979). In natural conditions, the female occupies cavities with a minimum diameter of 4.5 mm; the preferable diameter being 6-8 mm. The choice is determined by the female's size and the nesting resources available (Giejdasz & Wilkaniec, 2008; Maeta, 1978). *Osmia rufa* is a polylectic species, which accepts a wide range of plants as its source

of food (Raw, 1974; Wilkaniec & Warakomska, 1992). However, in the nesting period, females tend to collect pollen from several species of plants and frequently store only one species in a cell (Tasei, 1973; Free & Williams, 1970). The amount of pollen stored in a cell also depends on other factors, such as climatic conditions, availability of blooming plants, the size of the female that gathers the pollen, and the developing insect's sex (Maeta, 1978; Tasei, 1973). *Osmia* females need nearly twice the amount of food as males (Bosch & Vincens, 2005).

The development of the red mason bee is similar to that of other spring species of the *Osmia* genus. The embryonic and larval development, which is divided into five growth stages, occurs in the spring, while the prepupal and pupal stages take place in the summer (Tasei, 1973; Rust, Torchio, & Trostle, 1989; Giejdasz & Wilkaniec, 2002). In the prepupal stage, the metabolic processes slow down. The insect is then able to survive unfavourable weather conditions, such as high temperatures and low humidity, which determine the duration of this stage (Bosh & Kemp, 2000). The imago instar appears in the cocoon at the end of August or in early September. The imago is the wintering form of the red mason bee. Reproductive diapause is characterised by the cessation of ovary development, and by body fat hypertrophy (Hondelmann & Poehling, 2007). In the red mason bee, marked inhibition of ovary development and depletion of fat body resources is observed between October and January (Wasielowski et al., 2011). After this period it is possible to manipulate the duration of the wintering period by treating cocoons with high or low temperatures (Giejdasz & Wilkaniec, 1998). Numerous publications have focused on investigating relationships between pre-wintering and wintering periods and the emergence and longevity of bees (Bosch & Kemp, 2004; Kemp & Bosch, 2005; Bosch, Sgolastra, & Kemp, 2010). Findings on such relationships are used for synchronising bee emergence time with a particular crop bloom period (Bosch & Kemp, 2000; Bosch, Kemp, & Peterson, 2000; Sedivy & Dorn, 2014). There have been no laboratory studies involving

the entire immature development including the larval stage, which is crucial for gathering nutrients by the insect. Kemp and Bosch (2005) investigated the relationship between developmental temperatures during prepupal and pupal stages of *Osmia lignaria*, and emergence time and longevity of adults after wintering and incubation in the spring. Since constant and high temperature treatment during development reduces duration of activation time it may be used as a rearing method for managing flights and synchronising adult bee emergence time with a particular crop bloom period. The research is also vital from the environmental point of view, bearing in mind the current rise in temperatures, especially in the spring, i.e. the time when early spring bee species develop.

The first aim of the present paper is to determine the duration of immature development depending on temperature treatment. The second aim is to investigate the effect of temperature treatment during immature development on insect wintering survival rates, emergence time in the spring, and the longevity of adults.

## MATERIAL AND METHODS

### Experimental animals

The bees used in the study were the progeny of an *Osmia rufa* population reared following the method of Wójtowski and Wilkaniec (1978) in the dendrological garden of Poznań University of Life Sciences in 2014. Nests were obtained at the end of April when the bees were in the egg or first larval stage. Newly occupied artificial nests made from the common reed *Phragmites australis*, were selected and transported to the laboratory. The selection was determined by the appearance of the final plug. Moist, slightly soft clay or an incomplete partition on the edge of the nest tube meant that nest building had just been completed or was at its final stage. We used a total of 360 occupied nests and randomly divided them into three groups.

### Developmental monitoring and wintering

The nests of *Osmia rufa* were stored at 20°C, 25°C, and 30°C, until the emergence of imago

instars on 15 August, 30 July, and 5 August, respectively. The bees were then cooled at 15°C and wintered at 4°C from 31 October to 8 April. In order to determine the duration of particular developmental stages, we opened the brood cells with a scalpel, cutting lengthwise 20 nest tubes from each group. The tubes were attached to grooved polystyrene plates. Each cell was labelled with a number written with a marker pen on the polystyrene plate along the cut edge of the nest tube. The plates were then covered with paper to protect the pollen, eggs or larvae from falling out of the cells and drying. The prepared nests were kept in incubators at the test temperatures, only until the end of immature development and the pupation into imago.

The nests were monitored daily in the feeding larval and spinning larval stage, and every second day in the other stages. The embryonic development time was established only on the basis of those eggs in the nest which were the last to be laid by females. The eggs were monitored under stereoscopic microscope. The emergence of larvae with segmented bodies marked the beginning of the next stage – the feeding larva. In this stage, the larva feeds on its own on pollen provisions until provisions are exhausted. The emergence of the first loose cocoon threads marked the beginning of the next stage – the spinning larva, which ends with a construction of a dark brown cocoon. Cocoons with prepupae were systematically cut every second day, 5 cocoons per group. Microsurgical scissors were used to cut the end of a cocoon. The end of the cocoon was located laterally on the insects head. By creating a small hole we could determine the developmental stage of the insect. The moment when a white pupa with colourless eyes appeared was considered the end of the prepupal stage. Only such cocoons were transported and placed in cavities made in PVC plates. The cocoons were labelled according to the cell from which they had been obtained. The development of the pupae was monitored through openings in the cocoons until the adult insects emerged. The insect's sex was established on the basis of antennae length in the

pupa stage or the presence of organs for pollen collection in imago instars.

#### **Nest analysis and caged experiment with adult bees**

After the wintering period, all the nests were opened by cutting lengthwise down the tubes. The nests were then examined and the cocoons removed. At this stage of the research we created a control group of 120 nests. These insects went through their immature development and diapause in natural conditions and were cooled together with the experimental group beginning from 1 February. We determined the total number of brood cells per tube, the number of properly developed cocoons, and the number of cells with dead red mason bees at consecutive developmental stages, i.e. eggs, larvae, and ill-developed cocoons. We also noted the number of cells containing larvae or imagines of parasite species.

The cocoons obtained from each group were placed in boxes at 25°C on 8 April 2015. The emerged insects could be easily caught due to the fact they could not leave the boxes.

#### **Mortality, emergence time, and longevity**

After the first insects began to emerge, they were regularly removed from boxes and counted. From each group, a selection of 50 males and females were placed in Plexiglas rectangular-shaped cages, 22 x 22 x 40 cm in size. The obtained data were used to establish the emergence time and emergence rate of the red mason bee. To avoid excessive density of the insects in isolators, 4 isolators were used for each test group. Two of them contained males and the other two isolators contained females. Insects in the isolators were fed every second day with a sucrose solution; proportion 1:1, served on Petri dishes 5 cm in diameter with a floating wax foundation ring. This floating ring prevented the insects from drowning. The first dead insects were found in the isolators on 16 April. Starting from this date, the dead were removed every second day and their number was recorded. This was done with the aim of evaluating the survival rates of bees developing at different temperatures. The last stage of preparation involved an analysis of non-

emerged cocoons. After gently cutting them with scissors, we determined the developmental stage in which the bee died (prepupa, pupa, imago) or if parasites were found we noted their taxonomic position.

### Statistical analysis

To compare the mean duration of the developmental stages depending on temperature, we used one-way analysis of variance and Tukey's test. Estimates of survival probability and survival curves were calculated according to the Kaplan-Meier method, and survival distributions were compared using the log-rank test (chi-square statistics). The Kruskal-Wallis test by ranks (one-way ANOVA on ranks) was used for multiple comparison of survival times in the groups studied. Differences in mortality rates between the groups were noted separately for each stage, using the chi-square goodness of fit test. Analysis was conducted at the significance level  $\alpha = 0.05$ ; separately for males and females, using the Statistica v.12 software.

## RESULTS

Temperature had a significant effect on the egg development time ( $F=28.14$ ,  $p<0.001$ ). The egg stage lasted for the shortest period of time at 30°C (2 days). There were no differences in the means between the 20°C (3 days) and 25°C (3.2 days) groups (Fig. 1.A). The feeding larval stage in both sexes differed significantly depending on the temperature (males:  $F=42.46$ ,  $p<0.001$ ; females:  $F=46.91$ ,  $p<0.001$ ). An increase in the temperature decreased the mean feeding time in male larvae from 32.2 days in 20°C to 22.5

days in 30°C. Female larvae fed longer in 20°C (average 33 days) than in higher temperatures (average 24 days) (Fig. 1.B). Temperature had no influence on the time of cocoon spinning in male ( $F=1.11$ ,  $p=0.33$ ) or female ( $F=1.27$ ,  $p=0.29$ ) larvae (Fig. 1.C). Irrespective of temperature and sex, the majority of larvae completed cocoons within 3 days, with maximum duration of 6 days. The temperature significantly affected development time in the prepupal stage, both in males ( $F=6.99$ ;  $p=0.005$ ) and females ( $F=13.72$ ;  $p<0.001$ ). This stage lasted for the longest period of time at 30°C (male: 31.5 days; female: 31.2), and the shortest at 25°C (male: 23.1 days; female: 24.0), (Fig. 1.D). Temperature also significantly affected development time in the pupal stage, in both males ( $F=3.8$ ,  $p=0.05$ ) and females ( $F=7.38$ ,  $p=0.003$ ). An increase in temperature decreased the duration of the pupal stage in females from 32.0 days in 20°C to 23.1 days in 30°C. The pupal stage in males lasted much longer (average 32 days) at 20°C than at 25°C and 30°C (average 27.0 and 26.1 days, respectively), (Fig. 1.E). When based on the means for the subsequent stages of development, it was estimated that the total development time of the red mason bee was the shortest at the temperature of 25°C: 80 days for males and nearly 83 days for females. The longest development time was noted at 20°C: 97.2 days for males and 98.8 days for females (Fig. 1.F).

The highest developmental mortality rate was observed in the group of insects which developed at 25°C, and in natural conditions in the egg stage ( $\chi^2=18.053$ ;  $df=3$ ;  $p=0.0004$ ), (Tab. 1.). The mortality rate at other develop-

Table 1  
Developmental and wintering mortality expressed as the total percentage of cells built by *Osmia rufa* treated with various temperatures

Treatment	n	egg	feeding larva	spinning larva	prepupa & pupa	adult *	parasites
Nature	983	20.3	10.5	2.1	0.4	6.2	8.8
20°C	1260	6.3	6.1	0.3	0.1	0.6	7.0
25°C	951	24.7	13.0	1.4	1.5	9.4	1.4
30°C	1349	6.9	8.9	0.8	0.9	17.4	2.1

\* pre-wintering and wintering adult bees

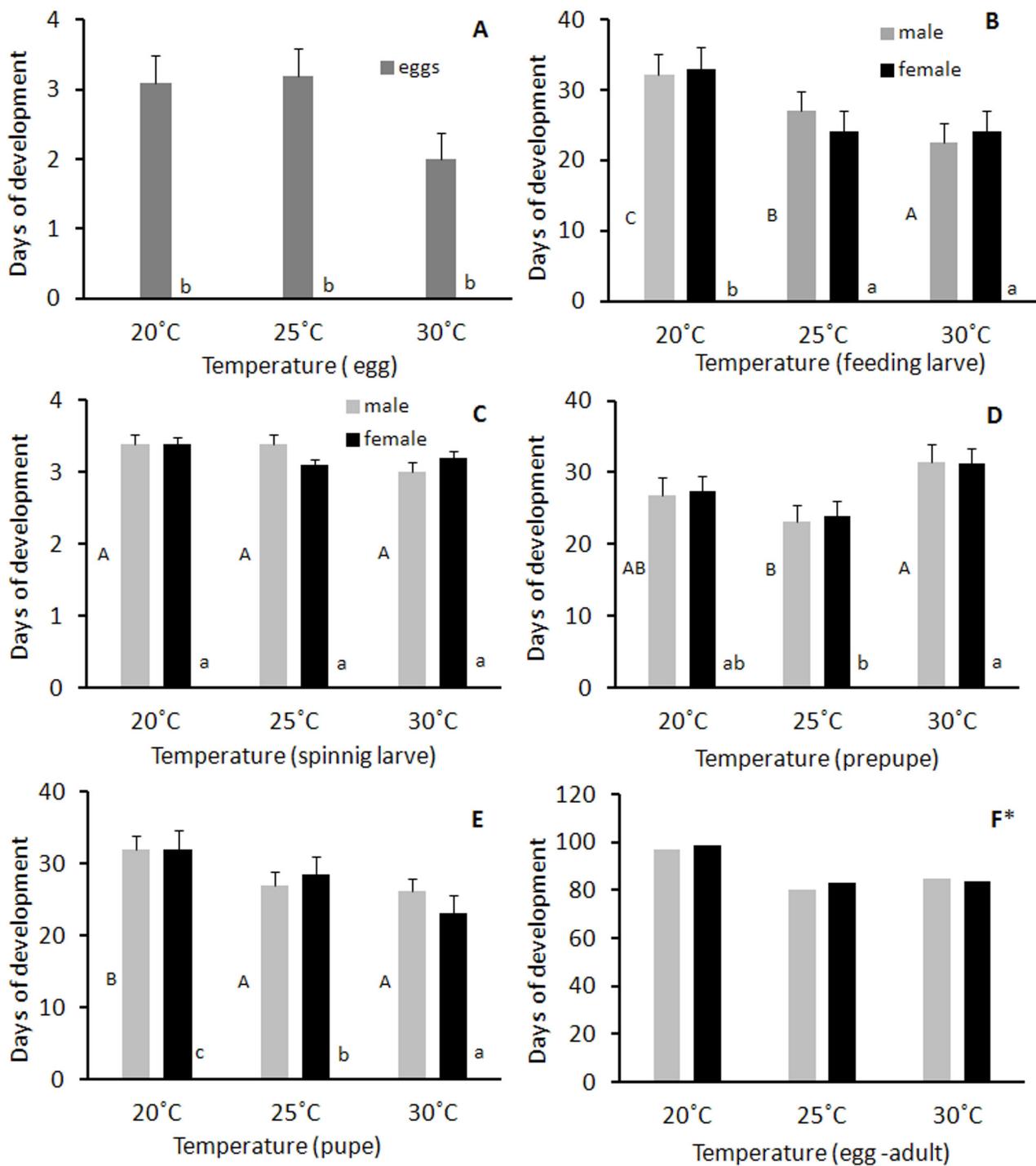


Fig. 1. The mean duration of the developmental stages (A-E) and total development time (F) depending on the temperature (°C) in which the red mason bee developed

\*sum of duration calculated based on the means for subsequent stages

mental stages was not related to temperature, although the insects were more prone to die in the feeding larval stage. During wintering the highest number of adult insects died in the 30°C and 25°C groups ( $\chi^2=13.581$ ;  $df=3$ ;  $p=0.0005$ ). The highest percentage of cells infected by parasites was observed in bees developing in natural conditions and at 20°C ( $\chi^2=8.225$ ;  $df=3$ ;

$p=0.041$ ). Parasitism was caused mainly by one cleptoparasite: the sapygid wasp *Sapyga quinquepunctata* (Hymenoptera: Sapygidae), and two parasites: the chalcid wasp *Monodontomerus obscurus* (Hymenoptera: Torymidae) and the bee fly *Hemipenthes morio* (Diptera; Bombyliidae). The effect of developmental temperature on the

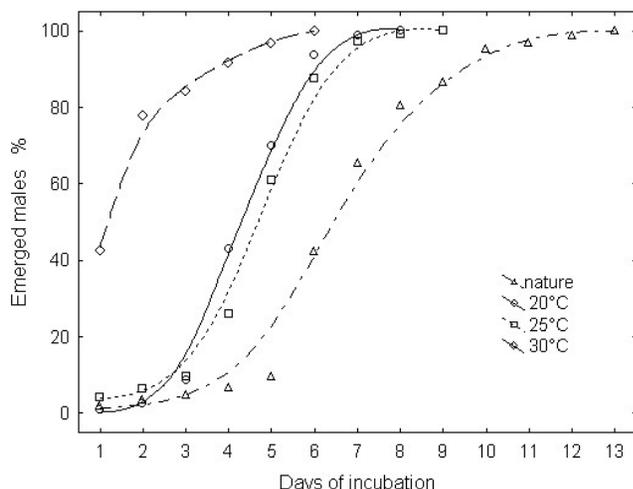


Fig. 2. Cumulative percentage of emerged male adults treated with various developmental temperatures as a function of the number of days after the beginning of incubation

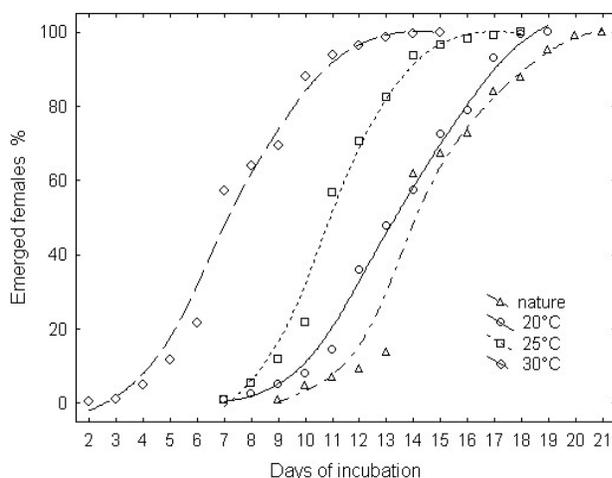


Fig. 3. Cumulative percentage of emerged female adults treated with various developmental temperatures as a function of the number of days after the beginning of incubation

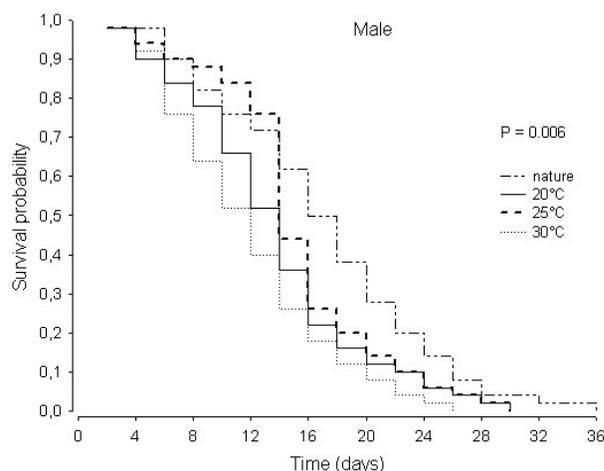


Fig. 4. Kaplan-Meier survival curve for *O. rufa* male adults developed at various temperatures in laboratory conditions

emergence of insects is illustrated by line charts using the distance-weighted least squares smoothing procedure (Fig. 2. and Fig. 3.). A calculation of the cumulative percentage of emerged bees was based on only living, emerged bees. The emergence time was longest in insects which developed in natural conditions, and shortest in those which developed at 30°C. Males already appeared on the first day of incubation in all groups, though with a different intensity. Within 6 days of incubation, all males of the 30°C group, over 80% of the 20°C and 25°C groups, and a little over 40% of the nature group, left the cocoons. The emergence period was measured as the time between the emergence of the first and last individual depending on the developmental temperature. This period lasted from 6 to 13 days in males (Fig. 2.).

The first females to emerge were bees of the 30°C group on the second day of incubation. Females of the 20°C and 25°C groups began to emerge on the seventh day, and those developing in natural conditions on the ninth day. Within 15 days of incubation, all females of the 30°C group, over 90% of the 25°C group, and about 70% of the 20°C and nature groups, left their cocoons (Fig. 3.). The emergence period of females was 14 days in the 30°C group, 12 days in the 25°C group, and 13 days for the 20°C and nature group.

There was a statistically significant difference ( $\chi^2=17.491$ ;  $df=3$ ;  $p=0.006$ ) among the groups of males treated with various temperatures with respect to the survival probability estimates (Fig. 4.). The median survival time (MST) in male adults developed in natural conditions and in the constant temperatures of 20°C, 25°C, and 30°C was 17, 14, 14, and 12 days, respectively. The overall median for males was 14. The Kruskal-Wallis one-way analysis of variance and multiple comparison tests showed noticeable differences of survival times among the groups of males. The median for males developed at 30°C, differed significantly from medians in the group of bees developed in natural conditions ( $p<0.001$ ) and in the 25°C group ( $p=0.037$ ). The 14-day survival rate of males developed in natural conditions, in the constant tempera-

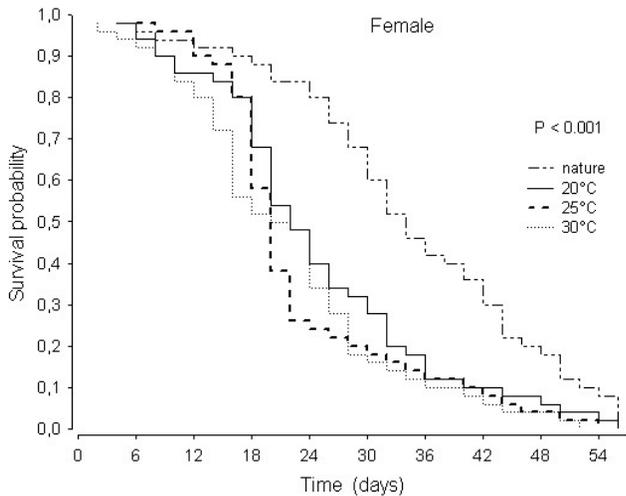


Fig. 5. Kaplan-Meier survival curve for *O. rufa* female adults developed at various temperatures in laboratory conditions

tures of 20°C, 25°C, and 30°C, was 62%, 36%, 44%, and 26%, respectively.

There was a statistically significant difference ( $\chi^2=31.842$ ;  $df=3$ ;  $p<0.001$ ) among the groups of females treated with various temperatures with respect to the survival probability estimates (Fig. 5.) The median survival time in female adults which developed in natural conditions and in the constant temperatures of 20°C, 25°C, and 30°C was 34, 22, 20, and 21 days, respectively. The overall median for females was 24. The Kruskal-Wallis one-way analysis of variance and multiple comparison tests showed noticeable differences of survival times among the groups of females. The median in the group of females which developed in natural conditions differed significantly from the medians in groups of bees developed at temperatures 20°C ( $p=0,002$ ) and 25°C, and 30°C ( $p<0.001$ ). There were no differences between medians in the female groups developed at constant temperatures. The 24-day survival rate of females which developed in natural conditions and in the constant temperatures of 20°C, 25°C, and 30°C, was 80%, 40%, 24%, and 34%, respectively.

## DISCUSSION

The conducted experiment involved the controlled development of *O. rufa* at constant temperatures in laboratory conditions. In nature, the rate of development depends on air temperature, which fluctuates during the day. In the

spring, air temperature may reach physiological zero, which ranges from 9.5 to 14.2°C in insects of the *Osmia* genus, depending on the species and stage of development (Maeta, 1978). At a constant temperature of 22°C, the overall development of the red mason bee lasts for 97.6 days (Tasei, 1973). In Polish climate conditions, females lay their first eggs in April and imagines appear in early September, which means that the development period lasts for over 120 days (Wójtowski, 1979). On average, the egg stage lasts for 7.1 days at a constant temperature of 22°C, 4 days at 23°C, and 3 days at 28°C (Tasei, 1973; Rust, Torchio, & Trostle, 1989; Giejdasz & Wilkaniec, 2002). It is worth noting, that in Tasei's (1973) study the egg stage was measured together with the larval stage until the first moulting of the larva. In our experiment, the egg stage at 20°C and 25°C ended after about 3 days. The egg stage was one day shorter when the temperature was raised to 30°C. In the course of daily monitoring, larva segmentation was considered to mark the end of the egg stage. Thus, the frequency of control might have been insufficient with regard to the duration of this stage. Temperature accelerates numerous physiological processes in insects, such as metabolism and oxygen consumption (Neven, 2000). An increase in temperature shortened the duration of all developmental stages of *O. lignaria* except for the prepupal stage (Bosch, Kemp, & Peterson, 2000; Kemp & Bosch, 2005). Our results show an evident linear relationship between temperature and development time in the feeding larval stage in males and the pupal stage in females. In contrast, treating bees with a temperature of 30°C did not shorten the development time of female larvae and male pupae in comparison with the 25°C group. In our study, we also distinguished the spinning larval stage, but its duration was not affected by temperature. This stage is mainly related to secretion from silk glands, which are responsible for synthesis of proteins forming the silk fibroin structure (Li et al., 2001). The cocoon is created by intensive movements of the larva, which are not affected by temperature.

The duration of the prepupal stage did not decrease as a result of an increase in temper-

ature; on the contrary, it was longer at 30°C than at 25°C. This is similar to the results of studies on *O. lignaria*, whose prepupal stage was shortest at 26°C in both sexes, and longest at the minimum and maximum temperatures applied in the experiment (Kemp & Bosch, 2005). It is probable, therefore, that the U-shape relationship between developmental temperature and length of the prepupal stage is typical of univoltine species, which winter as adult insects. The prepupal stage, which lasts for about 4 weeks and takes place during the warmest days of summer (July), is characterised by a slowdown in the life processes. This period is also described as summer diapause. During this period, the insect is able to survive unfavourable weather conditions in the summer (high temperature, low absolute humidity of the air), while changes in temperature are considered to be a stimulating factor for the termination of the prepupal stage (Bosch & Kemp, 2000).

In our study, the reasons for offspring reduction were grouped into the following categories: death during the developmental stages (egg, feeding larva, prepupa, and pupa), death during the pre-wintering and wintering imago stage, and parasitism. The highest number of eggs died at the temperature of 25°C, and in natural conditions. In another species, *O. lignaria*, the total developmental mortality rate varied from 7.6% to 36.5% and the percentage of dead eggs was between 6% and 31%. Differences at this stage for *O. lignaria* did not depend on the treatment temperature (Bosch, Kemp, & Peterson, 2000). In *O. cornuta* aggregation, the average mortality rate was 15%. It was mainly caused by developmental disorders (12%) and the presence of parasites (about 3%) (Bosh & Vicens, 2005). Our research showed an increase in the mortality rate of adult insects developing at 25° or 30°C in the wintering period or in the autumn. The survival rate for *O. lignaria* was significantly lower when the treatment temperature was 29°C and 32°C in the prepupal and pupal stages; this was observed mainly in females (Kemp & Bosch, 2005). It is probable that at higher temperatures, insects which develop more rapidly use more energy for basic

metabolic processes. As food resources of a larva are limited, metabolic processes happen at the cost of energy reserves needed for fat body synthesis by insect.

The most frequently observed parasite species, *Monodontomerus obscurus* and *Hemipenthes morio*, occurring in Northern and Western Europe respectively, are parasitoids frequently found in nests of solitary bees. *Sapyga quinquepunctata* are classified as cleptoparasites due to the fact that their larvae feed on pollen provision in cells (Krunić et al., 2005). All these parasites were also recorded in this study. These two parasitoid species are characteristic of managed mason bees of the *Osmia* genus since they develop in bee cocoons and are difficult to eliminate. As our research material was obtained from the same place at the same time, all the nests were equally likely to have been infected by parasites. The fact that fewer parasites were found in nests of bees developing at higher temperatures might be an effect of parasite developmental forms being less tolerant to temperature.

In Polish climatic conditions, real diapause in *Osmia rufa* begins in early October (Wasielewski et al., 2011), which means that the emergence on 8 April occurred after 180 days of wintering. This date was adopted as the beginning of the second phase of the experiment, since in nature *Osmia rufa* appears in late March and early April (Wójtowski, 1979). The duration of the wintering period and wintering temperature are important factors influencing the emergence time (Bosch & Blas, 1994; Bosch & Kemp, 2003; Sgolastra et al., 2010). Wintering temperature ceases to be significant when wintering lasts longer than 120 days (Bosch & Kemp, 2004). Wintering in controlled conditions eliminates these factors and creates comparable conditions for insects in all test groups. Emergence time of insects may be manipulated with incubation temperature due to the fact that temperature increase reduces emergence time (White, Son, & Park, 2009). Temperature in the pre-wintering period and duration of this period also influence the emergence time of insects (Sgolastra et al., 2011). The shortest emergence time was observed when the duration of the pre-winter-

ing period was average, preventing excessive reduction of the fat body. This suggests that emergence time depends not only on the duration of the wintering period but also on the insect's condition, which is related to body weight and reduction of the fat body reserves (Bosch, Sgolastra, & Kemp, 2010).

After the initial cooling at 15°C, bees from all groups were placed simultaneously at the temperature of 4°C. However, insects which completed their development 10-15 days earlier at 25°C and 30°C spent more time at the transitional temperature. This temperature might also have caused more rapid reduction of the fat body in the pre-wintering period, which leads to a shorter wintering period and shorter longevity of adult insects in the spring. On the whole, earlier emergence of insects in the spring is caused by temperature, which accelerates immature development, as well as by a long wintering period. The emergence time of insects depends on the developmental temperature in the pre-wintering and wintering periods and on the duration of wintering. Both factors are related to the metabolism of reserves (Bosch & Kemp, 2003, 2004; Bosch, Kemp, & Peterson, 2000; Sgolastra et al., 2010; Radmacher & Strohm, 2011).

In *Osmia lignaria*, increasing the temperature to 26°C in the prepupal and pupal stages shortened the emergence time of imagines in the spring (Kemp & Bosch, 2005). There were no further changes at 29°C, whilst at 32°C the trend reversed (Kemp & Bosch, 2005). In the experiment, reduced incubation and emergence time in bees were obtained in all developmental temperatures within the 20-30°C range, in comparison with natural conditions. The effect was not pronounced only in the case of females developed at 20°C. Bees raised at constant temperatures from the egg stage, react differently than those treated with temperature only in later stages. It may be concluded, therefore, that temperature has an effect on the emergence time of adult insects throughout the developmental period. A prolonged post-wintering period in species of the *Osmia* genus results in reduced fat body, which leads to

shorter longevity in adult insects in the spring (Bosch, Kemp, & Peterson, 2000). Insects in the prepupal and pupal stages exhibit a slight tendency for increased longevity at higher temperatures, but this is true only for females (Kemp & Bosch, 2005). This is caused by the fact that in controlled conditions insects completed their development more quickly and entered the period of low activity earlier. Field studies show, though, that bees which develop earlier lose a considerable amount of body weight, their fat body is reduced and their longevity in the spring is shorter (Bosch, Sgolastra, & Kemp, 2010). The parameter used most often in the cited works was longevity without feeding. It describes the survival rate of bees which used only their own energy reserves. In our study, we used semi-field cage tests, in which bees were kept and fed until their death. Developmental temperature (20-30°) significantly lowered the survival rate of females. In males, though, the effect was pronounced only when they developed at 30°C. This means that maintaining energy reserves after the winter is of greater importance for the development and survival of females than males. The reduction in adult bee longevity may have an effect on reproduction, but should not limit the possibility of using these bees for pollination since survival rates in the first two weeks are very high.

The development of *Osmia rufa* in controlled conditions offers the possibility of shifting the emergence time of adult insects. Consequently, constant developmental temperature may be used for rearing bee populations with shorter wintering period. Such populations could be used for pollination of early blooming crops. The possibility of reducing incubation and emergence time due to accelerated development also facilitates synchronising bee flights with crop bloom periods.

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