

EFFECT OF QUEEN CAGING CONDITIONS ON INSEMINATION RESULTS

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S u m m a r y

The study was performed in a honeybee breeding apiary at Teodorów in 2004. Honeybee queens were caged in queenless colonies or in an incubator. Temperature was compared in spaces between frames of colonies where either non-inseminated queens were kept in Zander cages without attendant bees or instrumentally inseminated queens in "Folchron" mailing cages were attended by bees. Temperature was also measured in cages with queens that were placed in beehives or in incubators. Temperature records were also taken outside the beehives and in the incubators. During the final investigations the manner in which the queens were kept before and after insemination was compared for its impact on the filling of queen spermathecae and on the evacuation of sperm from the oviduct.

The temperature in the frame spaces of honeybee nests in which the cages with instrumentally inseminated bees together with attendant bees were placed was found to average 34.5°C and to be significantly higher than that measured in the frame spaces containing cages with non-inseminated queens and without attendant bees. In each of the nurse colonies with inseminated queens different thermal conditions prevailed, with ambient temperature having a significant impact.

New attendant bees introduced to queens upon their instrumental insemination modified the temperature inside the cage by raising it within the first several hours following re-introduction of the queen into the colony to more than 35°C.

In the incubators, the temperature in the cages was higher than outside the cages. The temperature was also higher in those cages in which the attendant bees had been introduced a day before insemination than in those in which attendant bees were introduced after the insemination (35.4°C and 34.7°C, respectively).

The highest percentage of queens with well filled spermathecae and with emptied oviducts was found in the incubator-reared group in which, following insemination, the queen was re-introduced to the same attendants with which she was kept prior to the treatment. Possibly plausibly the result can be linked to stable thermal conditions at which the queens were kept and to better care provided to the queen by the attendant bees which had already accepted the queen on the previous day and got used to being under confinement faster than by newly caged bees.

Keywords: queen bees, insemination, filling of spermathecae, stasis of sperm, temperature.

INTRODUCTION

Instrumental insemination of honeybee queens provides the only possibility to

apply individual parental selection. The insemination technique has been mastered by many inseminators and the treatment is

routinely applied in breeding apiaries. As a result of many studies, the amount of sperm as related to the filling of queens' spermathecae was precisely determined (Woyke 1960, Prabucki et al. 1987). Nevertheless, studies are being continued aimed at the improvement and simplification of the method in which the queens are caged before and after insemination to increase efficacy at which sperm passes from the oviducts to the spermatheca.

The best method to store the queens is to keep them before and after insemination as single individuals in regular colonies, nuclei or in mating nuclei from which they are removed only for the insemination to be returned to the colony immediately once the treatment is completed (Laidlaw 1981, Woyke, Jasiński 1982, Woyke, Jasiński 1985, Prabucki et al. 1987). Such a procedure most closely imitates natural conditions. However, it is expensive, troublesome, involves many colonies and maintenance persons so it is unfeasible in apiaries – mass producers of honeybee queens. Because of that, with large-scale queen insemination, queens are caged in nurse colonies without attendant bees (Mackensen and Tucker 1970, Laidlaw 1981, Harbo 1986) or are caged in incubators with several attendants (Mackensen 1955, Vesely 1971). However, it was found that when a large number of queens are kept in one colony there is a risk of them being damaged by bees (Woyke et al. 1956, Woyke 1988, Jasiński and Fliszkiwicz 1995, Jasiński et al. 1998).

For the purpose of storing the queens after insemination boxes are often used in which queens are kept at room temperature together with 350 attendant bees or at 34°C with 250 attendants. The queens kept under such conditions showed a high number of spermatozoa in their spermathecae (Woyke and Jasiński

1979, 1980; Chuda-Mickiewicz and Prabucki 1993).

Regardless of the kind of box or cage and of the number of bees attending the queen, temperature at which the queens are kept is an essential parameter. When kept at temperatures which are either too high or too low, the queens show impaired removal of sperm from oviducts or their oviducts are overheated and fail to be emptied normally resulting in the queen's death (Jasiński 1984). The effect of temperature on the filling of spermathecae was also observed by Mackensen (1969). Woyke and Jasiński (1973) found that the spermatozoa counts in the spermatheca of queens kept in incubators at 34°C following insemination averaged 808,000 more than those of the queens kept at 24°C.

The objective of the study was to investigate thermal conditions prevailing during the storage of queens in colonies and in incubators in an apiary that rears honeybee queens on a large scale.

MATERIAL AND METHODS

The study was run in a honeybee breeding apiary at Teodorów in July of 2004. It was divided into three trials:

I. Comparison of temperature in spaces between frames that contain cages containing unattended non-inseminated queens vs. attended inseminated queens

Temperature was measured in four nuclei with non-inseminated and with instrumentally inseminated queens in a two-replicate design. The nuclei established in the Wielkopolski type hives consisted of 5-6 frames: 2-3 storage frames and 3 frames with sealed brood. The sister queens, non-inseminated and without attendant bees, were confined in wooden rearing Zander cages with a 2.4 x 2.5 mm mesh screen on one side and a wooden

wall on the other. In the cages there were wax cups filled with honey to be used as food by the queens. Sister queens were instrumentally inseminated when 8 days old with sperm collected from 14 day-old drones originating from the same colony and isolated in the upper body of the hive. Immediately upon insemination, the queens were placed in two-chamber "Folchron" mailing cages with external dimensions of 7 x 4 x 1.5 cm. Queens attended by ca. 25 bees originating from the colony in which the queens had lived before insemination (anesthetized prior to caging) were introduced into the chamber 4 x 4 x 1.5 cm in size. In three walls there were cone-shaped round openings tapering towards the center of the cage which enabled contact with bees in the hive. In the smaller chamber 3 x 4 x 1.5 cm in size candy was placed to be used as food by the queens and the attending bees. In a rearing frame placed between the sealed brood frame and the storage frame there were 24 cages with the queens. Temperature was measured with an electronic gauge placed in the central site of the space between the frame with caged queens and the frame with sealed brood. The temperature was recorded at hourly intervals for 24 hours in a total of 4 nuclei.

II. Comparison of temperature in the frame space with inseminated queens in mailing cages vs. temperature outside the hive

In randomly selected colonies with instrumentally inseminated queens caged for sale temperature was measured at the central point of the gap between the frame with queens in mailing cages and the frame with sealed brood. The temperature was compared with the ambient temperature near the hive. The temperature records were taken at hourly or 2-hour intervals depending on weather conditions and bee mood for at least 24 hours from the

moment of introducing the queens to the colonies. Temperature records were discontinued when the temperature levelled off and the difference between the consecutive measurements was less than 0.2°C.

III. Temperature in cages with queens and in their vicinity, in the incubator, in the hive and outside the hive

Three queen groups were formed which were kept under different conditions prior to and after insemination.

Group 1 - (C25r/C25r) – a day before insemination sister queens were placed in "Folchron" mailing cages with 25 worker bees (25r/) from the colony in which they lived until 7 days old. Cages thus prepared were placed in an incubator (C) with a gauge set at 34°C. After insemination the queens were returned to the same cages with the same attendant bees in the incubator (/25r).

Group 2 - (R0r/C25r) – sister queens prior to insemination were kept in queenless colonies (R) in Zander rearing cages without attending bees (0r/). Immediately after insemination they were placed in mailing cages with 25 attendant bees that were originated from the same colonies in which the queens had been reared before being inseminated. The cages were put in an incubator (/25r).

Group 3 - (R0r/R25r) – sister queens prior to insemination were kept in Zander rearing cages placed in a hive without attending bees. After insemination they were placed in mailing cages with 25 bees and re-introduced in the same hive (25r).

In this experiment temperature was measured using electronic gauges:

1. in the incubator
2. in cages containing the queens and attendant bees placed in the incubator
3. in the central part of the space between the frame with queens and the frame with sealed brood
4. in mailing cages containing the queens

and attendant bees that were placed in a hive

5. outside the hive

All the queens were killed 48 hours after insemination and dissected to determine the extent to which the spermathecae were filled and to find out if any sperm remained in the oviducts. The filling of spermathecae was assessed based on their colour after the tracheas had been removed.

- milky – poorly filled
- light-creamy with poorly marked veining pattern – medium well filled
- creamy with well marked veining pattern – well filled

Remaining of sperm in the oviducts was assessed as:

- lagging (remaining of sperm in one or both oviducts)
- no lagging (oviducts emptied)

A total of 108 queens were examined in three replications.

ANOVA was used for statistical calculations. The significance of differences between means was measured using Duncan's multiple range test. The data concerning the number of queens with poorly, medium well or well filled spermathecae and the data concerning emptying of oviducts were presented as percentages. ANOVA with Bliss conversion of recorded data was applied and the

means were compared using Duncan's multiple range test.

RESULTS AND DISCUSSION

I. Comparison of temperature in nest spaces with cages containing unattended non-inseminated queens vs. attended inseminated queens

Highly significant differences in temperature were found between colonies with caged non-inseminated bees without attendants and colonies with caged inseminated bees with attendants. The average temperature in the central area of the gap between the brood frame and the frame with inseminated queens was 34.5°C and was significantly higher than that in the colonies with non-inseminated queens (Table 1). Smaller variations in temperature were also observed in colonies with instrumentally inseminated queens. The data clearly suggest that for inseminated queens the bees maintain a temperature approximating 34.5°C, a temperature that according to Woyke and Jasiński (1973) should occur in an incubator containing inseminated queens. The significantly lower temperature in the space near the cage with non-inseminated queens may have been caused by the lack of attending bees rather than by the physiological condition of the queens.

Table 1

Temperature in the space between the frame with sealed brood and the space with non-inseminated unattended queens and inseminated queens with attendant worker bees

Queens	Number of colonies	Number of records	Temperature °C average (min.-max)	Standard Deviation SD
inseminated with attendant bees	2	48	34.5 b (33.0 – 36.2)	0.88
non-inseminated without attendant bees	2	48	33.1 a (30.7 – 34.8)	0.92
Total	4	96	33.79	1.14

a,b - highly significant differences at p 0.01

Table 2

Temperature in the space between the frame with the inseminated queens placed in mailing cages with attendant bees and in the frame with sealed brood as compared to the temperature outside the hive

Hive no.	Number of records n	Temperature °C			
		frame gap		outside the hive	
		average (min. – max.)	St.Dev. SD	average (min. – max.)	St.Dev SD
96	27	33.4 b (29.4 – 36.8)	1.02	20.5 a (11.6 – 28.7)	4.25
120	17	31.4 a (29.2 – 33.8)	1.23	19.1 a (11.8 – 26.9)	4.29
132	8	33.1 b (32.4 – 33.9)	0.51	20.7 a (15.6 – 29.0)	4.18
140	44	34.3 c (31.7 – 35.6)	0.75	21.6 a (9.9 – 29.3)	3.70
153	21	35.2 d (30.1 – 35.9)	1.19	24.9 b (14.1 – 36.4)	4.61
179	30	35.0 d (33.2 – 36.2)	0.88	27.6 b (13.8 – 35.2)	5.69
Average	147	34.0 (39.2 – 36.8)	1.49	22.7 (9.9 – 36.4)	5.29

a,b,c,d – differences significant at p 0.05

II. Comparison of temperature in a frame gap containing mailing cages with inseminated queens with temperature outside the hive

As measured in the central area of the space (beeway) between the frames with attended instrumentally inseminated queens, the temperature varied significantly from colony to colony (Table 2). The lowest average temperature was 31.4°C (hive 120), the highest 35°C (hive 153). The average temperature in beeway between frames of those colonies was 34°C, with external temperature averaging 22.7°C. A highly significant correlation between those parameters ($r=0.637$; $p=0.000$) suggests that external temperature influenced the temperature as measured between frames of the colonies in which inseminated queens were caged with

attendant bees. However, the smaller variation in temperature inside the hive compared to the temperature variation outside indicates that the bees controlled the temperature by maintaining it at an average level of 34°C.

III. Temperature in the queen containing cages, in the cage vicinity, in the incubator, in the hive and outside the hive

The temperature inside the incubator came within a range of 32.4°C to 33.5°C even though the thermostat showed 34°C (Table 3). The temperature in the cages with bees attending the queens already a day before insemination (C25r/C25r) was significantly higher (35.4°C) than the temperature (34.7°C) in the cages with the queens with no attendants before the treatment (R0r/C25r).

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Table 3

Temperature in the incubator and in the cages with instrumentally inseminated queens exposed to different conditions prior to insemination

Measurement site and group	No. of replications n	Length of temperature (°C) recording interval						Average temperature °C
		4 hr	8 hr	12 hr	16 hr	20 hr	24 hr	
incubator – outside the cage	3	33,2	33,2	32,4	33,0	33,5	33,0	33,0
cage with queens group 1 - (C25r/C25r)	3	34,9	34,3	35,0	36,1	36,2	35,9	35,4 b
cage with queens group 2 - (R0r/C25r)	3	34,9	34,9	34,5	34,9	34,6	34,5	34,7 a

C-incubator; R-colony; r-worker bees; 0-25 number of worker bees; prior to insemination/after insemination, a,b-differences significant at p 0.05

colonies with instrumentally inseminated queens which, prior to insemination, were kept in colonies in Zander cages without attending bees (R0r/R25r) and, after insemination, were put into mailing cages with bee attendants and returned to the same colonies the temperature was 32.7°C and the external temperature was 20.1°C. There were significant differences among

the colonies (Table 4). Both parameters were lower than those in experiment II but also related to each other ($r = 0.39$ at $p = 0.011$). The temperature inside the cages that contained queens and attending bees, when averaged across colonies and replications, was 34.8°C and there were no differences for that parameter among the colonies (Table 4). There was a correlation

Table 4

Temperature in the beeway between frames, in the cages with attendant bees and artificially inseminated queens which, prior to insemination, were caged without attending bees (R 0r/R 25r), and outside the hive

Colony	Number of queens	Number of records	Temperature °C					
			Space between frames		Cage		Outside the hive	
			average (min. – max.)	SD	average (min. – max.)	SD	average (min. – max.)	SD
1	12	26	33.3 b (29.4 – 34.1)	1.02	34.9 a (33.1 – 38.2)	1.58	20.4 a (11.6 – 28.7)	4.2
2	12	17	31.4 a (29.2 – 33.8 0)	1.23	34.8 a (33.5 – 37.5)	1.02	19.1 a (11.8 – 26.9)	4.3
3	12	8	33.1 b (32.4 – 33.9)	0.51	34.8 a (34.1 – 36.5)	0.76	20.7 a (15.6 – 29.0)	4.2
Total	36	51	32.7 B	1.37	34.8 C	1.29	20.1 A	4.2

a,b – differences significant at p 0.05 (within columns);
A,B,C– differences significant at p 0.05 (between columns)

between the temperature outside the hive and the temperature in the cages containing queens and attendant workers ($r=0.43$ at $p=0.009$). Obviously, the bees raised the temperature inside the hive as related to the temperature outside. It was also found that the average temperature in the cages was significantly higher than the temperature in the gap between the frames. Even though no significant correlation was found between those factors it can be stated that the bee attendants modify the temperature of the ambient air around the queens which is in agreement with the data reported by other investigators (Vesely 1971, Woyke, Jasiński 1979, 1980; Chuda-Mickiewicz, Prabucki 1993).

Significant variation in temperature was observed in cages with queens and attendant bees depending on when the queens were introduced to the colonies following insemination (Table 5). Within the first two hours the average temperature in the cages was 36.5°C and after 2-12 hours it was 34.9°C . Within the next few hours the temperature levelled off and was ca. 34°C . The

temperature in the cages with the queens and their attending bees was also significantly higher than the temperature in the gap between frames.

The high temperature (over 35°C in many instances) within the first two hours of keeping the queens in the colonies may suggest that the new attendant bees, when caged and introduced into the colonies, responded with the rise of temperature.

Upon dissecting, an average of 96.1% of the queens from experiment III were found to have well filled spermathecae (Table 6). In the queen group (C25r/C25r) only one out of 36 tested queens had a poorly filled spermatheca. In the group (R0r/C25r) two queens had medium well filled spermathecae and in the group (R0r/R25r) only one queen had a poorly filled spermatheca. The percentage of the queens with well filled spermathecae was very high and did not vary significantly between the tested queen groups.

An average of 83.3% of the queens was not found to have sperm lagging in the oviducts. A significantly higher number of

Table 5

Temperature in the beeway between frames and in the cages with inseminated queens as influenced by the time during which the queens were kept in the hive (R0r/ R25)

Hours of staying in the hive	Space between frames			Cage		
	n	average °C (min. – max.)	SD	n	average °C (min. – max.)	SD
0 – 2	13	32.9 b (29.2 – 33.9)	1.42	13	36.5 c (34.3 – 38.2)	1.13
2 – 12	10	33.1 b (30.6 – 34.1)	1.24	10	34.9 b (34.0 – 35.6)	0.65
12 – 24	17	32.6 b (29.4 – 33.6)	0.89	17	33.9 a (33.1 – 35.0)	0.69
24 – 48	7	31.4 a (30.0 – 33.8)	1.61	7	34.0 a (33.2 – 35.2)	0.67
Overall average	51	32.7 A	1.30	51	34.8 B	1.29

a,b,c – differences significant at $p < 0.05$;

A,B – differences highly significant at $p < 0.01$

queens with emptied oviducts was found in the group in which worker bees attended the queens as soon as a day before insemination (C25r/C25r). In the remaining two groups the percentage of queens in which no oviductal stasis of sperm was found was significantly lower. The stasis of sperm in one or both oviducts was found in an average of 16.7% of the queens. In the group in which the queens were kept in the colonies caged but unattended by bees until inseminated and after insemination were caged with attending bees in the incubator (R0r/C25r) or in a colony (R0r/R25r) the percentage of sperm stasis was significantly higher (25.0% and 22.3%, respectively) than that in the group (C25r/C25r). In the latter group the lagging of sperm was found only in one queen and the queen had also a poorly filled spermatheca. During dissection in some of the queens intestines were found to be filled with feces.

The prerequisite for the success of insemination is the complete evacuation of sperm from the oviducts which, among other things, depends on the attendant bees and on the temperature at which the queens are kept after the treatment (Woyke, Jasiński 1973, Woyke 1983) particularly

when the dose of sperm exceeds 4 mm³. Mackensen (1969) did not find differences in the filling of spermathecae when the queens were dosed with 2 mm³ of sperm and kept at 25-35°C. However, with a higher dose of sperm and with the temperatures of 37.5 and 40.0°C the number of spermatozoa in the spermathecae was lower and queen mortality was very high, probably due to the sperm lagging in the oviducts. In this study the temperature was more than once higher than 35°C in the group where the queens, prior to insemination, were kept caged in colonies without attendant bees and after insemination were returned to the same colonies and were kept in cages with attendant bees (R0r/R25r) which could have contributed to the high percentage (ca. 22.3%) of the queens with unemptied oviducts (Table 5).

Likewise, among the queens which, prior to insemination, were caged in a colony without attendant bees and after insemination were attended by bees in a cage that was put in an incubator (R0r/C25r) the percentage of queens with unemptied oviducts was similarly high (25.0%). According to Vesely (1971) and Woyke (1979) the assistance of worker bees is necessary

Table 6

Condition of spermathecae and oviducts towards the end of the second day following insemination

Queen group	Number of queens	% of queens with spermathecae filled			% of queens with lagging of sperm in oviducts	
		poorly	medium-well	well	no lagging	lagging
group 1 (C25r/C25r)	36	2.8 a	0.00 a	97.2 a	97.2 b	2.8 a
group 2 (R0r/C25r)	36	0.00 a	5.6 a	94.0 a	75.0 a	25.0 b
group 3 (R0r/R25r)	36	2.8 a	0.00 a	97.2 a	77.7 a	22.3 b
Average	108	1.9	1.9	96.1	83.3	16.7

C – incubator, R-colony, r-worker bees, 0-25 – number of attendant workers before/after insemination, a,b – differences significant at p 0.05, statistical calculations after Bliss conversion

for the queen to have her oviducts emptied and among the queens that are prevented from direct contact with worker bees the percentage of individuals with unemptied oviducts is higher. Plausibly, the best insemination result achieved in the group (C25r/C25r) can be related to two factors: the first is that the queens stayed in the incubator (that means in more stable thermal conditions) the second is providing the queens with attendants as early as one day before the insemination that ensured the acceptance of the queen by those bees and good care right after the insemination. Such conditions are provided to the queens in mating hives and in nuclei before insemination, the queens are removed to be inseminated and returned to the same colonies immediately after the treatment. According to many investigators (Laidlaw 1981, Woyke, Jasiński 1982, Woyke, Jasiński 1985, Prabucki et al. 1987) such conditions ensure best insemination results. Instead, when new queens are added to the caged queens after the insemination they become agitated, try to leave the cage, fail to care for the queens properly, harass them and sometimes even sting them.

It is not known, however, if some influence was not exerted by the higher temperature that was generated in the cages by the attendant bees already on the second day. The problem requires further study under more strictly controlled and more uniform conditions.

CONCLUSIONS

1. Changes in ambient temperature have an impact on the thermal conditions in the nest of a honeybee colony in which instrumentally inseminated queens are reared.
2. Colonies in which instrumentally inseminated queens are reared vary for thermal conditions.

3. The average temperature in the nest in which instrumentally inseminated queens are caged with their attending bees is 34.5°C.
4. Attendant bees alter the temperature inside the cage.
5. Once the inseminated queens are returned to the colony the temperature in the cage is increased. Many a time it goes up beyond 35°C within the first two hours.
6. Storage in an incubator under stable thermal conditions after the instrumental insemination beneficially affects the filling of the spermathecae and removal of sperm from the oviducts in inseminated queens.
7. It is possible that the very attendance by worker bees of the caged queens prior to insemination beneficially affected insemination results but it requires additional study.

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WPLYW WARUNKÓW PRZETRZYMYWANIA MATEK PSZCZELICH NA WYNIKI INSEMINACJI

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S t r e s z c z e n i e

Badania prowadzono w 2004 roku w pasiece hodowlanej w Teodorowie. Matki pszczele przetrzymywano w klaticzkach w osieroconych rodzinach pszczelich lub w cieplarnie. Porównywano temperaturę w uliczkach gniazd rodzin przetrzymujących matki nieunasienione znajdujące się w klaticzkach Zandera bez pszczół oraz matki sztucznie unasienione

w klteczkach wysyłkowych typu "Folchron" z pszczołami. Badano również temperaturę w klteczkach z matkami znajdujących się w ulach lub cieplarkach. Mierzono także temperaturę na zewnątrz uli, oraz w cieplarkach. W końcowych badaniach porównywano wpływ sposobu przetrzymywania matek przed i po inseminacji na wypełnienie zbiorniczzków nasiennych matek i opróżnianie jajowodów z nasienia.

Stwierdzono wysoko istotne różnice temperatury w rodzinach przetrzymujących matki nieunasienione w klteczkach bez pszczół i unasienione sztucznie w klteczkach z pszczołami. Średnia temperatura w centralnym punkcie uliczki między plastrem z czerwiem a ramką z matkami unasienionymi wynosiła 34,5°C i była istotnie wyższa niż w rodzinach z matkami nieunasienionymi (Tab. 1). Przyczyną istotnie niższej temperatury w uliczce obok ramki z matkami nieunasienionymi był prawdopodobnie nie stan fizjologiczny matek, co brak pszczół towarzyszących w klteczkach.

Temperatura mierzona w centralnym punkcie uliczki między ramką z matkami unasienionymi sztucznie i pszczołami towarzyszącymi różniła się istotnie w różnych rodzinach (Tab. 2). Temperatura zewnętrzna miała wpływ na wysokość temperatury w uliczkach rodzin przetrzymujących matki unasienione w klteczkach z pszczołami ($r=0,637$; $p=0,000$). Jednak mniejsze wahania temperatury wewnątrz ula niż wahania temperatury zewnętrznej wskazują na to, że pszczoły regulowały temperaturę utrzymując ją w rodzinach na średnim poziomie 34°C.

Temperatura w klteczkach z matkami w cieplance, którym pszczoły towarzyszyły już na dobę przed inseminacją (C25r/C25r) była istotnie wyższa (35,4°C) od temperatury (34,7°C) w klteczkach z matkami, które przed zabiegiem były bez pszczół (R0r/C25r) (Tab. 3). Temperatura w uliczkach rodzin przetrzymujących matki sztucznie unasienione, które do inseminacji przebywały w rodzinach w klteczkach Zandera bez pszczół, a po inseminacji wracały do tych samych rodzin w klteczkach wysyłkowych z pszczołami (R0r/R25r), wynosiła 32,7°C przy średniej temperaturze zewnętrznej 20,1°C i różniła się istotnie między rodzinami (Tab. 4). Temperatura wewnątrz klteczek z unasienionymi matkami i pszczołami wynosiła średnio w trzech rodzinach 34,8°C, przy czym nie stwierdzono różnic między rodzinami (Tab. 4). W pierwszych dwóch godzinach od momentu poddania matek po inseminacji do rodzin, średnia temperatura w klteczkach wynosiła 36,5°C, a po upływie 2-12 godzin spadła do 34,9°C. W następnych godzinach temperatura jeszcze nieco spadła i wyrównała się na poziomie około 34°C (Tab. 5).

Po wypreparowaniu trzech grup badanych matek stwierdzono, że średnio 96,1% z nich miało dobrze wypełnione zbiorniczki nasienne (Tab. 6). Średnio u 83,3% matek nie stwierdzono zalegania nasienia w jajowodach. Istotnie więcej matek (97,2%) miało opróżnione jajowody gdy pszczoły robotnice towarzyszyły im już na dobę przed inseminacją (C25r/C25r). W dwu pozostałych grupach procent matek, u których nie stwierdzono zalegania nasienia w jajowodach był istotnie niższy. Zaleganie nasienia w jednym lub w obu jajowodach stwierdzono średnio u 12,9% matek. W grupach matek, które do inseminacji przebywały w rodzinach w klteczkach bez pszczół, a po inseminacji dodano nowe pszczoły i klteczki umieszczono w cieplance (R0r/C25r) lub w rodzinie (R0r/R25r), procent matek z nieopróżnionymi jajowodami był istotnie wyższy (odpowiednio 25,0% i 22,3%) niż w grupie matek które po inseminacji przebywały z tymi samymi pszczołami co przed zabiegiem (C25r/C25r). W grupie tej zaleganie nasienia w jajowodach stwierdzono zaledwie u 1 matki, która miała również słabo wypełniony zbiorniczek nasienny.

Być może najlepszy wynik inseminacji jaki uzyskano w grupie matek (C25r/C25r) wiąże się z dwoma czynnikami. Pierwszy z nich to przebywanie w cieplance (a więc w bardziej wyrównanych warunkach termicznych). Drugi czynnik to zapewnienie matkom lepszej opieki po inseminacji przez pszczoły, które już poprzedniego dnia zaakceptowały matkę i szybciej oswoiły się z zamknięciem, niż czynią to nowe pszczoły ostatnio zamknięte w klteczce.

Słowa kluczowe: matki pszczele, inseminacja, wypełnienie zbiorniczzków nasiennych, zaleganie nasienia, temperatura.