

# THE EFFECT OF OPEN BROOD AND COLONY STRENGTH ON THE ONSET OF OVIPOSITION BY QUEEN BEES

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## Abstract

In bee colonies without open brood, e.g., after swarming, there is no need for royal jelly, and nurse bees thus do not produce it. According to many beekeepers, adding combs with open brood restarts the production of royal jelly by nurse bees, and the virgin queens then are better fed and start earlier oviposition. The purpose of this study was to investigate whether the presence of open brood and the strength of the colonies affect the onset of oviposition by queen bees. Open brood in colonies with virgins before and during mating flights did not accelerate the initiation of oviposition by the queens. In addition, no differences were identified in starting oviposition by queens in strong colonies of more than 30,000 worker bees, or in weak colonies with up to 1,000 workers. Overall, the results showed that neither open brood in the nests, nor the strength of the colonies affects the onset of oviposition by queen bees.

**Keywords:** honey bee, initiation of oviposition, open brood, queen bee.

## INTRODUCTION

Early onset of oviposition by queen bees mated both naturally or instrumentally reduces the cost of their production. Additionally, a break in oviposition, occurring between the removal of the old queen and the start of oviposition by the young one, has a negative impact on subsequent nectar collection because of the reduced number of foragers. According to many beekeepers, the presence of open brood in the colonies accelerates the onset of oviposition (OofO) by the queen bees. The presence of open brood reactivates the secretory activity of the royal jelly glands in nurse bees (Haydak, 1970; Hrassing and Crailsheim, 1998), and better nourishment of the queens results in an earlier initiation of oviposition. Meteorological conditions affect the mating flights of virgin queens, which do not perform mating flights in temperatures below 19°C or above 31°C (Soczek, 1958). According to various authors (Kaftanoglu and Peng, 1982; Wilde, 1994; Woyke et al., 2001, 2008; Gerula et al., 2011), naturally mated queens begin egg laying at the age of 7 to 29 days, mostly between age 9 and 15 days.

The purpose of this study was to investigate whether the presence of open brood and the strength of the colonies affect the onset of oviposition by queen bees.

## MATERIAL AND METHODS

The study was conducted in May 2009 and 2010. In total, 174 *Apis mellifera ligustica* queen bees were studied. Three experiments were performed, each at different times.

In the first experiment, we investigated whether open brood present in bee colonies when the virgins are up to 6 days old accelerates the onset of oviposition (OofO) by the queens. During this time, virgin queens mature sexually before starting mating flights. We established 40 three-comb nuclei in Dadant hives. Into half of them, one comb with open brood was inserted and in the other half, there was no brood. Virgin queens, which emerged the previous day in an incubator, were introduced into these nuclei. When the virgins reached the age of 6 days, the brood comb was removed. From the next day, OofO was checked daily.

In the second experiment, we tested whether open brood present in the nuclei when the queens are older than 6 days accelerates OofO. At that time, the queens perform mating flights. We created 68 three-comb nuclei without brood. Virgin queens that emerged the previous day in an incubator were introduced. After the queens reached the age of 6 days, a comb with open brood was introduced into every other nucleus. Starting the next day, OofO was monitored daily. In the nuclei in which the queens did not start oviposition within the next 8 days, the combs with brood that already was covered were replaced by new ones with open brood.

In the third experiment, we investigated whether the strength of the colonies affects OofO by the queen bees. Virgin queens were introduced into 36 strong colonies with more than 30,000 worker bees, and into 30 trapezoidal mating nuclei with fewer than 1,000 workers. None of these colonies had open brood.

The Kolmogorov-Smirnov test was applied to determine whether the distribution of the dates concerning OofO significantly differed from normal values. When the distribution was not uniform, the

nonparametric Mann-Whitney test was applied for statistical comparisons. When the distribution was normal, one-way ANOVA was used.

## RESULTS

### Onset of oviposition by queens in nuclei with or without open brood that is present during the first 6 days of the virgins' lives

The queens started oviposition in all 40 nuclei with or without brood. The Kolmogorov-Smirnov test showed that the distribution of the age of queens starting oviposition did not differ significantly from normal in group 1A with open brood ( $Z_{19} = 1.247$ ,  $p = 0.089$ ) or in group 1B without open brood ( $Z_{19} = 0.814$ ,  $p = 0.522$ ).

The distribution of the age at which the queens started oviposition in nuclei with open brood was moderately skewed (below 1) while it was almost symmetrical in nuclei without brood (below 0.5) (Tab. 1). Standard skewness for both the groups with and without open brood was below 2; however, it was above 2 overall for the colonies, indicating that the skewness was significant.

Table 1.

Effect of presence or absence of open brood and strength of colonies on the age (days) of queens starting oviposition

Exp/group	Colonies	No queens	Min - Max	Mean	Median	Mode	Skewness	Standard skewness
1	3 comb Dadant nuclei with queens up to the age of 6 days							
1A	With open brood	20	12 - 19	14.7	14	14	0.8	1.5
1B	Without open brood	20	11 - 17	13.6	13.5	13	0.3	0.5
	Overall	40	11 - 19	14.2	14	14	0.9	2.3
2	3 comb Dadant nuclei with queens older than 6 days							
2A	With open brood	30	8 - 20	10.9	9.5	9	1.7	3.7
2B	Without open brood	33	9 - 19	11.5	10	9	1.4	3.3
	Overall	63	8 - 20	11.2	10	9	1.5	4.7
3	Colonies with different number of worker bees without open brood							
3A	More than 30,000	34	8 - 21	12.1	10	9	1.0	2.4
3B	Fewer than 1,000	27	7 - 22	11.1	10	8	1.4	3.0
	Overall	61	7 - 22	11.7	10	9	1.1	3.6

The age range of OofO was one day longer (8 days, 12 - 19 days) in colonies with brood than in those without brood (7 days, 11 - 17). The average age of queens starting oviposition was one day older in colonies with open brood (14.7) than in those without brood. The modal age of OofO was also one day greater in colonies with brood (14 days) than in those without brood. The median ages of OofO by queens in the nuclei with or without open brood were very similar (14 and 13.5 days, respectively) (Fig 1., groups 1A and 1B). One-factor ANOVA did not show significant effect of the presence or absence of brood on the OofO ( $F_{1, 39} = 3.25, p = 0.08$ ).

The range of the age of OofO was two days greater (13 days, 8 - 20) in colonies with brood than in those without brood (11 days, 9 - 19). The average age of queens starting oviposition was almost one day less in colonies with open brood (10.9) than in those without brood. The modal age of OofO by queens in both groups was identical (9 days), (Fig. 1., groups 2A and 2B).

No significant difference was found between the medians of the age of OofO by queens in nuclei with open brood (9 days) or without it (10) present after the virgins were older than 6 days (Mann-Whitney test:  $U = 400, N_1 = 30, N_2 = 33, p = 0.178$ ).

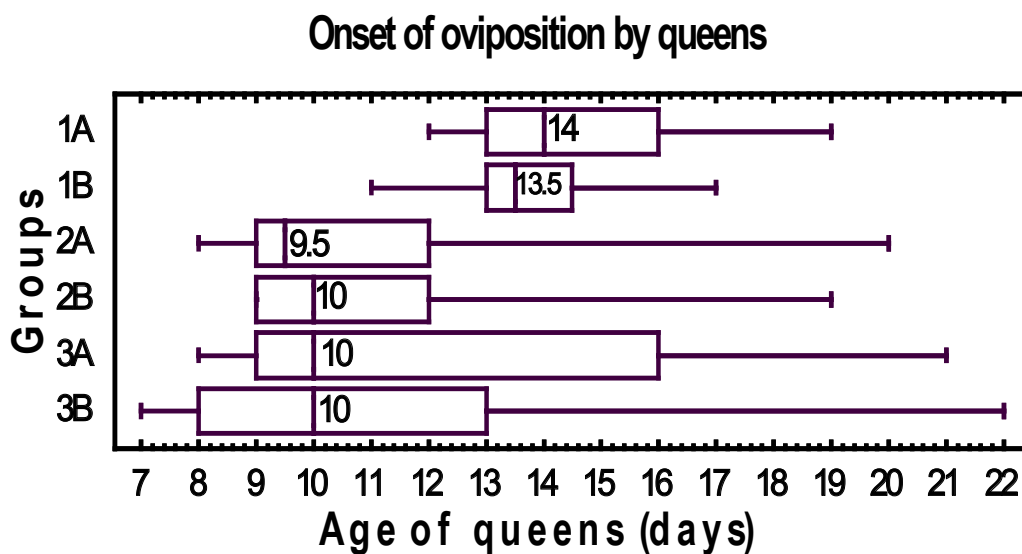


Fig. 1. The quartile (0, 25, 50, 75, and 100%) distribution of the onset of oviposition by queens in successive experiments. 1. The effect of open brood in colonies during the first 6 days of the virgins' lives (1A, with brood; 1B, without brood). 2. The effect of open brood in nuclei with queens older than 6 days (2A, with brood; 2B, without brood). 3. The effect of colony strength (3A, strong; 3B, weak). Middle lines in the boxes indicate the medians; numbers indicate the values of the median.

#### Onset of oviposition by queens in nuclei with or without open brood present after the queens were older than 6 days

The queens started oviposition in the 30 colonies with open brood and in the 33 without brood. The distribution of the age of OofO significantly differed from normal in nuclei with brood (Kolmogorov-Smirnov test:  $Z_{29} = 1.408, p = 0.038$ ) as well as in nuclei without brood (Kolmogorov-Smirnov test:  $Z_{32} = 1.528, p = 0.019$ ).

Table 1 shows that the distribution of the age of OofO by queens in both nuclei with or without brood was highly skewed (above 1). The standard skewness was  $>2$  in each of the two groups concerning presence or absence of brood, as well as in all nuclei (overall). This result indicates a significant departure from normality.

#### Onset of oviposition by queens in colonies of different strength

The queens started oviposition in 34 strong colonies and in 27 mating nuclei. The Kolmogorov-Smirnov test showed that the distribution of the age of queens starting oviposition in strong colonies differed significantly from normal ( $Z_{33} = 1.638, p = 0.009$ ) while it did not differ significantly from normal in weak colonies ( $Z_{26} = 1.258, p = 0.084$ ).

Table 1 shows that the distribution of the age of OofO by queens in strong and weak colonies was highly skewed (1 and above 1, respectively). The standard skewness was  $>2$  in each of the two groups of colonies of different strengths, indicating a significant departure from normality.

The range of the age of OofO was two days shorter (14 days, 8 - 21) in strong colonies than in weak

ones (16 days, 7 - 22). The queens started oviposition on average one day later in colonies with more than 30,000 workers (12.1 days) compared to those with up to a 1,000 bees. The modal age of OofO was also one day greater (9 and 8 days, respectively), (Fig. 1., groups 3A and 3B).

No statistically significant differences were identified between both medians of the age of 10 days of the OofO by queens in the strong colonies (of more than 30,000 workers) and in the small mating nuclei (up to a 1,000 workers) (Mann-Whitney test:  $U = 374$ ,  $N_1 = 34$ ,  $N_2 = 27$ ,  $p = 0.21$ ).

### Onset of oviposition by all queens in all three experiments

Altogether, 164 queens out of 174 started oviposition. The remaining animals perished during mating flights. The distribution of OofO for the 164 queens highly significantly diverged from normal (Kolmogorov-Smirnov test:  $Z_{163} = 2.275$ ,  $N = 164$ ,  $p < 0.001$ ) (Fig. 2). The skewness in the distribution of OofO was 0.8, and the standard skewness was 4.3. Because it was  $> 2$ , this result indicates a significant departure from normality.

All the queens started oviposition between the ages of 7 and 22 days. The average age of OofO by queens (12.1) was higher than the median (11) and the modal (9). Among all the queens, 25%, 50%, 75%, and 100% started oviposition at the age of 9, 11, 14, and 22 days, respectively.

### DISCUSSION

Bee colonies without open brood, e.g., following swarming, have no requirement for royal jelly, and nurse bees cease to produce it. According to many beekeepers, adding combs with open brood reactivates the secretory function of the royal jelly glands of nurse bees, which results in better nourishment of virgin queens and earlier oviposition. In our study, the introduction of combs with open brood into the nuclei prior to the mating flights of virgin queens did not accelerate OofO by the queens. This outcome can be explained by the fact that virgin queens are not fed by nurse bees with royal jelly. Instead, they receive honey (carbohydrate diet), which provides energy for the mating flights. If they were fed with royal jelly, their ovaries would increase, which would disturb the flights. Also, the introduction of combs with open brood after the queens started mating flights did not accelerate OofO. Skowronek et al. (2002) reported that the presence of differently aged larvae did not significantly affect OofO by instrumentally inseminated queen bees; however, the presence of eggs significantly delayed it.

We did not identify any statistically significant differences in OofO by queens in strong colonies of more than 30,000 workers and in weak nuclei with up to 1,000 workers. This observation suggests that queens in mating nuclei are fed in a similar fashion to those in normal colonies.

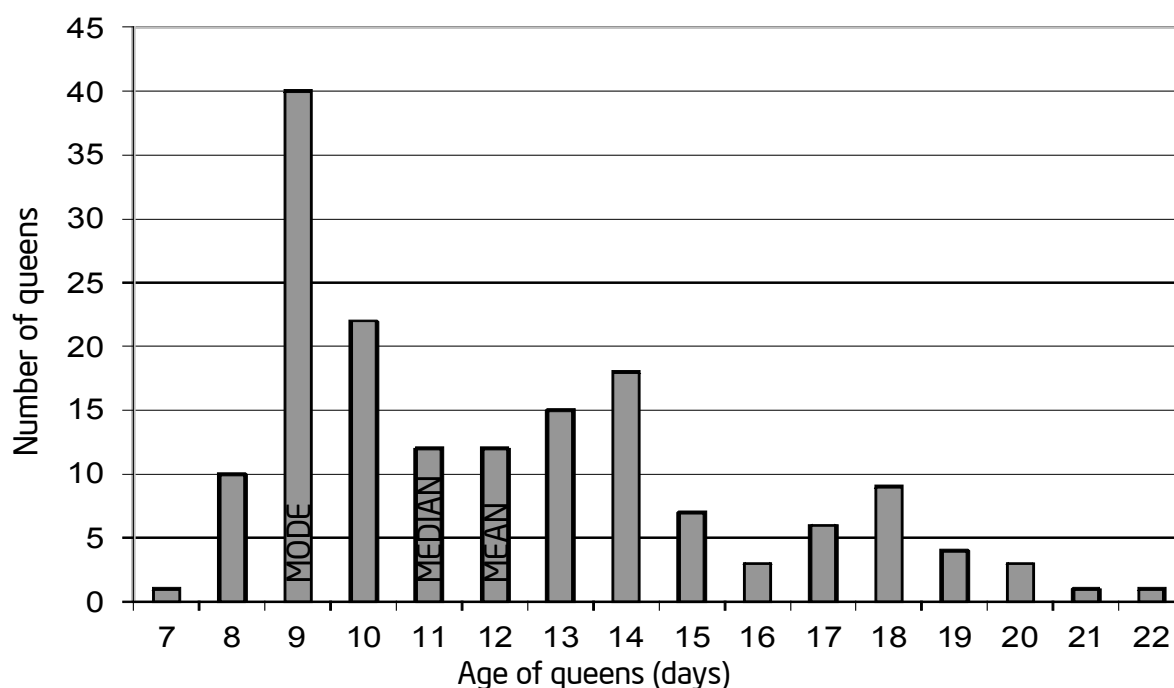


Fig. 2. Onset of oviposition by all 164 queens.

The weather conditions were worse during the first experiment (addressing the open brood effect within the first 6 days of the virgins' lives on OofO) than during the two other experiments. This distinction is probably why the queens performed their mating flights later and started OofO later (at the age of 14.2 days, on average) than in the successive two experiments (11.2 and 11.7 days, respectively). Similar results were reported by Wilde (1994), Czekońska et al. (2003), Woyke et al. (2008), and Gerula et al. (2011) (15.1, 13.7, 13.1, and 13 days, respectively). According to Kaftanoglu and Peng (1982) and Woyke et al. (2001), queens started oviposition slightly earlier (at 10.3 and 9 days, respectively).

Our study confirms the findings of Woyke et al. (2008) that the distribution of OofO by queens is skewed. As a result, the mean age of onset of oviposition by queens is higher than the median and the modal.

## CONCLUSION

The presence of open brood in bee colonies with virgin queens up to the age of 6 days, during the sexual maturation period and before performing the mating flights, does not affect significantly the onset of oviposition.

The presence of open brood in bee colonies with queens older than 6 days, i.e., when they already perform mating flights, does not affect significantly the onset of oviposition.

The strength of bee colonies does not affect significantly the onset of oviposition by the queen bees.

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