Development Times of Male and Female Eggs of the Honey Bee[1]

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ABSTRACT

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The development times of female eggs of the honey bee, *Apis mellifera* L., were significantly shorter ($P < 0.01$) than development times of male eggs. Throughout their developmental period, the eggs were kept in an incubator (34.8° C, 60 to 80% RH) without adult bees. Haploid and diploid male eggs required about 3 h longer than female (always diploid) eggs. It is not known whether the development times of haploid and diploid male eggs are significantly different. Female eggs from 20 unrelated queens had mean development times that ranged from 68.8 to 74.2 h at 34.8° C; the 20 means averaged 71.4 ± 1.2 h (SD). The development times were altered by very slight changes in temperature. Female eggs kept at 34.8° C hatched about 1.4 h sooner than those kept at 34.3° C. Only 1% of the eggs hatched when kept at 29.8° C.

The development time of the honey bee (*Apis mellifera* L.) egg is a variable that can be readily measured in an incubator without the influence of adult bees. Harbo et al. (in press) found that the eggs of Africanized bees in Venezuela had development times that were about 3.7 h shorter than the eggs of European bees. Reinhardt (1960) reported that female eggs develop more rapidly than haploid male eggs. In addition, environmental variables such as temperature also affect the rate of egg development.

The objective of this study was to measure the effects of temperature and sex on the development time of the egg. Also included in this study were measurements of egg development times from 20 unrelated queens in the Baton Rouge, La., area. Such measurements were indicators of the amount of variability in the development times of eggs from a small population of queen bees in the United States.

Materials and Methods

Comparing Diploid Male and Female Eggs

Sister queens were each mated to a single male; these males and queens were all progeny of the same queen. This type of mating was termed "mother-daughter" by Polhemus et al. (1950) and results in half of the queens producing only female (always diploid) eggs and the other half producing equal numbers of diploid male and female eggs (Mackensen 1951).

We identified the queens that produced diploid males by observing brood. Knowing that worker bees remove diploid males from their cells in the early larval stage (Woyke 1962), we caged the queens after they had filled an area of comb with eggs, identified 66 to 96 centrally located cells that contained one egg each, and then checked these cells 5 days later. Those combs that had only about half of their cells (51, 56, 61%) occupied by larvae had contained diploid males, and the queens producing this kind of brood were thus producers of diploid males. Those combs that contained larvae in over 90% of the cells 5 days after oviposition apparently had not contained diploid males, and the queens producing this brood were laying only female eggs.

Eggs were collected from five queens (three produced diploid males) by confining each queen to a section (8 by 8 cm) of comb in her colony for 2 h. (Harbo et al., in press). The combs with eggs were then put into an incubator (34.8° C, 60 to 80% RH) without adult bees present.

The development time of each egg was calculated by means of hourly observations which began 68 h after the midpoint of the caging period. The time assigned to each egg was the midpoint of the hour interval in which it hatched. For example, an egg that was first seen to be hatched at the 71-h observation was designated a 70.5-h development time.

The mean development times of groups of eggs was simple to calculate as long as the queen produced only female eggs. However, if a queen also produced diploid male eggs and if diploid male eggs have development times that differ from female eggs, a bimodal egg-hatching distribution would be expected. One of the modes should correspond in time to the single hatching mode of the eggs of sister queens that produced only females. This would be the mode of the female eggs; the remaining mode must then represent the diploid males.

The mean and SD for the diploid male peak were calculated by first removing values for the female eggs from the distribution. This was done by first calculating the mean and SD of the single mode produced by the groups of eggs that were entirely female.

The mean and SD of the single female mode (Fig. 1A) was assigned to the corresponding mode in Fig. 1B, and the bimodal distribution in Fig. 1B was then separated by removing all the observations within 2 SD of the mean for the female eggs. The few remaining observations that were thereby separated from the remaining mode were also removed. A mean and SD were then calculated on the remaining population.

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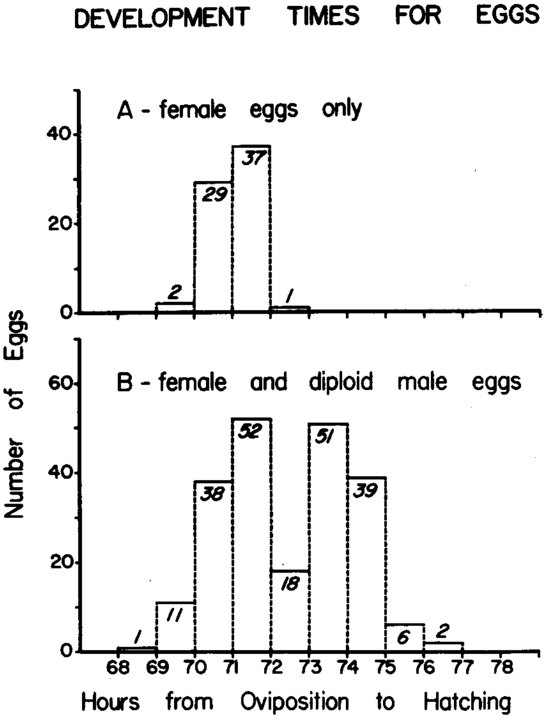

FIG. 1.—Development times of eggs produced by five sister queens that were each mated to a single "brother" drone. Two queens produced only female eggs (A), and three queens produced diploid male and female eggs (B).

Comparing Haploid Male and Female (Always Diploid) Eggs

Miscellaneous unrelated queens were confined, as described above, to a section of comb. However, in this case, each queen was confined to a comb having worker-sized cells for 2 h and then moved to a section having drone-sized (i.e., larger) cells for 2 h. In some cases the queens were confined to the drone-sized cells first. Data were collected as described for the diploid male and female eggs.

Effect of Temperature on Egg Development Times

The queens used in this experiment were three of the sister queens used to compare diploid male and female eggs; the eggs were collected and counted as in that experiment. Only the development times of the female eggs, those in the first hatching peak, were measured and compared.

Development times of eggs were compared at 29.8, 31.3, 34.3, and 34.8° C. Combs with eggs were placed close together in the incubator, and temperatures varied only ±0.1° C at any point. Temperatures were measured with thermister probes.

Development Times for Eggs of 20 Unrelated Queens

This study was conducted last, so we utilized information gained during the other experiments. For example, since very slight changes in temperature were so important to egg development times, the combs of eggs were placed close to one another and moved about six times during the 3-day developmental period to promote equal temperatures. The only other difference between this and previous experiments was that each group of eggs was laid during a 1-h period rather than a 2-h period.

Only female eggs were compared. When only a few eggs were laid in a worker comb, some or all of them were often males, so we looked for two hatching modes. In cases where one or two eggs in a group hatched and then a few more hatched 2 to 5 h later, we considered the early hatching mode to be female eggs and the late mode to be male eggs.

Results and Discussion

Comparing Diploid Male and Female Eggs

Diploid male eggs hatched about 2.9 h later than females when kept at 34.8° C (Fig. 1). The development times of 69 female eggs produced by two queens in a 2-h period averaged 71.0 ± 0.6 h (mean ± SD). The three queens that laid both diploid male and female eggs (Fig. 1B) produced 218 eggs in the same 2-h period; about 109 should be diploid males. Since the first mode in Fig. 1B corresponded to the mode in Fig. 1A, we concluded that the first mode consisted of females. We assigned the females in Fig. 1B the mean and SD from the females in Fig. 1A (71.0 ± 0.6 h), and we separated the bimodal distribution 2 SD above that mean, at 72.2 h. Based on this separation, we identified 106 eggs as females and 112 as diploid males. The egg development times of those 112 diploid males averaged 73.9 ± 0.8 h (mean ± SD).

Comparing Haploid Male and Female (Always Diploid) Eggs

Eggs laid in the drone-sized (larger) cells required 3.6 ± 1.0 h (mean ± SD) longer to develop than eggs laid in worker-sized cells (Table 1). If there had been no difference, the results would have been inconclusive because the sex of newly hatched larvae was not determined, and it is not uncommon for queens to lay female eggs when forced to lay in drone-sized cells. Because the eggs in drone-sized cells had development times that were consistently different from those in worker-sized cells, we con-

Table 1.—Comparison of development times of haploid male and diploid female eggs[a]

	Development time (h) in:				
	Worker-sized cells		Drone-sized cells		Difference (h) between eggs in worker and drone cells
♀ no.	($\bar{x}$ ± SD)	*n*	($\bar{x}$ ± SD)	*n*	
1	68.0 ± 1.1	6	73.0 ± 0.7	36	5.0
2	71.4 ± 0.7	53	74.2 ± 0.9	30	2.8
3	70.7 ± 0.9	88	74.5 ± 0.8	74	3.8
4	71.6 ± 1.1	91	74.2 ± 0.9	58	2.6
5	71.6[b]	3	75.3 ± 1.0	31	3.7

[a] Eggs in drone cells were assumed to be haploid, and those in worker cells were assumed to be female. $\bar{x}$, Mean.
[b] All three hatched during the same interval, so SD was not calculated.

Table 2.—Development times of female eggs from 20 unrelated queens

Queen no.	No. of eggs	Time (h) from midpoint of oviposition period to hatching ($\bar{x} \pm$ SD)[a]
1	47	68.8 ± 0.60
2	2	69.8
3	17	70.0 ± 0.56
4	12	70.3 ± 0.56
5[b]	1	70.8
6	1	70.8
7	28	70.9 ± 0.73
8[b]	3	71.1 ± 0.29
9	16	71.1 ± 0.72
10	35	71.2 ± 0.40
11[b]	1	71.3
12	53	71.5 ± 0.49
13	30	71.7 ± 0.64
14	16	71.9 ± 0.65
15	8	72.0 ± 0.46
16	34	72.4 ± 0.61
17	42	72.6 ± 0.55
18[b]	1	72.8
19	11	73.0 ± 0.52
20	8	74.2 ± 0.50
Analysis of the means		71.4 ± 1.25

[a] $\bar{x}$, Mean.
[b] Eggs from these queens were laid within a 2-h period; all others were laid within a 1-h period.

cluded that the queens had laid haploid male eggs in the drone-sized cells and female eggs in the worker-sized cells. Thus, we concluded that haploid male eggs required 3.6 ± 1.0 h (mean ± SD) longer to develop than female eggs.

A comparison of development times between haploid and diploid male eggs was not made. Both have longer egg development times than female eggs. The difference between the development times of haploid and diploid male eggs, if a difference truly exists, is small.

Effect of Temperature on Egg Development Time

The development times of eggs were sensitive to slight temperature differences. The time from oviposition to hatching was 71.0 ± 0.7 (mean ± SD) h when kept at 34.8° C (n = 125) and 72.4 ± 0.7 h when kept at 34.3° C (n = 157). The development times at 31.3° C were not measured as precisely, but were about 99 h. Thus, there was a difference of 1.4 h per 0.5° C at 34.3 to 34.8° C, but a greater difference at lower temperatures (an average of 4.4 h/ 0.5° C between 31.3 and 34.3° C).

The lower temperature limit for complete development of 50% of the eggs was between 29.8 and 31.3° C. Only 1% (4/341) of the eggs hatched when kept at 29.8° C; 67% (180/267) hatched when kept at 31.3° C.

Development Times for Eggs of 20 Unrelated Queens

Mean development times of eggs from each of the 20 queens in the survey ranged from 68.8 to 74.2 h (Table 2). These means averaged 71.4 h. Thus, considerable variability existed in the development times of eggs from a group of only 20 queens in Baton Rouge, La.

REFERENCES CITED

Harbo, J. R., A. B. Bolten, T. E. Rinderer, and A. M. Collins. 1981. Development periods for eggs of Africanized and European honey bees. J. Apic. Res. In press.

Mackensen, O. 1951. Viability and sex determination in the honey bee (*Apis mellifera* L.). Genetics 36: 500–509.

Polhemus, M. S., J. L. Lush, and W. C. Rothenbuhler. 1950. Mating systems in honey bees. J. Hered. 41: 151–155.

Reinhardt, E. 1960. Kernverhaltnisse, Eisystem und Entwicklungsweise von Drohnen-und Arbeiterinneneiern der Honigbiene (*Apis mellifera*) Zool. Jahrb. Abt. Anat. Ontog. Tiere 78: 167–234.

Woyke, J. 1962. The hatchability of 'lethal' eggs in a 2 sex allele fraternity of honey bees. J. Apic. Res. 1: 6–13.