## STUDIES ON SOME FACTORS AFFECTING THE FERTILITY OF HONEYBEE APIS MELLIFERA L., QUEENS

By

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B.Sc. Agric. Sci. (Plant Protection), Fac. Agric., Cairo Univ., 2002 M.Sc. Agric. Sci. (Economic Entomology), Fac. Agric., Cairo Univ., 2006

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#### **ABSTRACT**

This work was conducted to study some factors which affecting the fertility of honeybee queens. The experiments were carried out at the apiary yard of Agric. Exp. Sta., Fac. Agric., Cairo Univ., Giza, Egypt during late winter, spring and summer seasons of 2007 & 2008.

The external and internal morphological characters which affected the fertility of newly emerged virgin queens were: acceptance % of grafted bee larvae, % of newly emerged virgin queens, queen's weight, queen cells size, queen ovarioles number and queen's spermatheca volume. The results showed that, Carniolian hybrid followed by Italian hybrid were subjected with the highest characters of the queen fertility. Carniolian race showed a decrease in all measured characters compared with the original race.

The fertility of honeybee queens maybe depend on the season of reared queens as well as their genetic origin. These factors maybe reflected the differences detected in the queen's weight, no. of ovarioles/ queen and the histological differences (densities of nurse cells and connective tissues in and between queen ovarioles) between the queens of different races and hybrids.

Concerning the concentration of ovarian total protein and no. of ovarian protein bands, the Carniolian hybrid better than the Italian one, whereas the queen ovarian total protein and no. of ovarian protein bands were higher in Carniolian race than in Egyptian race.

**Key words:** Honeybee queens, fertility, histological differences, ovarian total protein, ovarian protein bands.

## **DEDICATION**

I dedicate this work to whom my heart felt thanks to my wife Ashwaq for her patience and help, as well as to my parents and sisters for all the support they lovely offered along the period of my post graduation.

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#### INTRODUCTION

Honeybee queen rearing is considered as an essential step in the improvement of beekeeping and bee stocks. Honeybee colonies were reared queens when the colony is in the process of swarming, supersedure, or when accidentally lost or killed the queen, for these reasons the workers were reared new queens from worker larvae that are less than 48 hrs. old (Woyke, 1971).

The role of the queen in the honeybee colonies is most important as heart and mother of colony, also for her influence on worker bee activities through the queen pheromones (Al-Ghzawi and Zaitoun, 2008). The economic characteristics of the honeybee colony are dependent mainly on the quality of the queen which depends on both hereditary differences, environmental conditions and variations in care during development (Taha, 2005).

The fecundity of queen is the important factor for controlling the population of the colony and consequently the difference obtained (Nour, 1992). Various modifications concerning the age of grafted larvae and the type of artificial queen cups were affected the acceptance of grafted larvae and quality of honeybee queens in the commercial queen rearing programs (Abd-El-Aziem, 1999).

On the other hand, the fertility of honeybee queens depends on many factors such as heredity genes available and environmental conditions (Morini *et al.*, 1993). These factors can be associated with the physiological activities and thus reflect on honeybee queens productivity (Abd-Al-Fattah *et al.*, 2007). Prolific queens specialized

and very simple guide lines produce specific offspring. Bee races in Egypt exposed to inbreeding process for long periods and negative affects on honeybee queen fertility were occurred (Diab, 1986). The reproductive system and bee glands often used to describe the physiological status of honeybees that consider arbitrary scale classification have been estimated by different methods (Zeedan, 2002). The surrounded bee queens ambiences impressed on honey and pollen flows will be stronger affects on queen's quality will be better (Li, 2000).

This study was carried out to investigate the fertility of the honeybee queens through the following factors:

- 1. External and internal morphological characters which affected the fertility of reared newly emerged virgin queens (less than 24 hrs.).
- 2. Histological studies on ovaries of newly emerged virgin queens of different races and hybrid.
- 3. Biochemical differences in the ovaries of newly emerged virgin queens of different races and hybrids.

## **REVIEW OF LITERATURE**

The honeybee virgin queens were reared during seasons of the year to produce superiority queens with higher parameters to passing successfully through the active seasons (spring and summer seasons) to the dearth seasons (autumn and winter seasons) by force.

The fertility (quality) of produced virgin queens was affected by many factors (inner and outer colony factors).

## 1. Factors affecting queens production in honeybee colonies

### a. Effect of accepted bee larvae

Hanna (1963) found that the different seasons had only an insignificant effect on the acceptance of virgin queen cells, while the percentage of acceptance was some what higher in spring and summer seasons than in autumn and late winter seasons.

Abdel-Rahman (1964) found that capacity of the colony to accept cell cups averaged 18 cell cups in Alexandria and 34 cell in rants. The highest records occurred from March to September, while the number accepted in February was clearly limited.

Fotti *et al.* (1971) stated that the acceptance of honeybee queen cells was highest (about 88%) in August but lowest (about 45-77%) in May and was insignificant by their position in the hive and consequently by temperature of the nucleus.

Chang (1977) stated that the queen cell cups with diameter of 6.11 or 12 mm were not usually accepted by workers, while 7, 8, 9 and 10 mm diameter cell cups were accepted.

El-Sarrag (1979) found that queens were successfully reared in February (92%) from grafted larvae and during March-June (82%). The least favourable periods (48%) were in August-September, 28% in December-January. The mortality of embryos was significantly correlated with temperature and with pollen collection by the colony.

Ebadi and Gary (1980) recorded significant differences between the acceptance rate of queen cups made from different composition of bees wax. They attributed these differences to the presence of unknown amount of queen pheromone contained in natural beeswax from which the artificial queen cups are made.

Kither and Pichard (1983) recorded the acceptance percentage for the different designs of queen cups which were as follow: 6-mm undrawn, 10.9%; 6-mm drawn, 48.6%; 12-mm undrawn, 63.9%; 12-mm drawn, 78.6%. The overall association between larval acceptance and queen cup design was significant.

Macika (1985) grafted 1-day old larvae into wax queen cell cups, with and without royal jelly and placed them into a rearing colony. The percentage of accepted larvae varied between the top, middle and bottom bars of the grafting frame, but there was no consistent pattern from year to year. Slight differences were found between the weights of queens one day after emergence. When cups were placed 16 mm apart, the acceptance was 80.3%, but at 30 mm apart only 70.1 % was accepted. There was little difference in queen weight. Mean acceptance of larvae grafted with royal jelly was 75.6% compared with 41.3% of those without royal jelly; but means of queen weights one day after emergence were similar.

Visscher (1986) found that the bees are more selective among potential queens; when larvae are grafted into queen-cell cups acceptance is greater than when eggs are used, and there is a non-significant tendency for cells near the centre to be accepted in preference to those near the edges. The probability of a queen being reared is low at the periphery of the hive, and queens are reared more readily in the upper than in the lower part of the brood nest. He added that the location of queen cells is an important factor to be considered in the design and statistical analysis of experiments on queen rearing.

Weiss (1986) reported that the acceptance percentage decreased as did mean weights of larvae reared and of royal jelly extracted from the cell. The latter was higher in plastic than in wax cups.

Abd-AI-Fattah and EI-Shemy (1996) found that the plastic queen cups caused significant increase in the acceptance percentage (84.7%) than wax did (76%) when they grafted with young larvae and introduced into queen rearing colony. They added that non significant differences were found between plastic and wax queen cups for both percentages of ripened queen cells or queen emergence under the tested procedures of queen rearing.

Sharaf El-Din *et al.* (2000) revealed that the production and quality of queens are affected by rearing season and recorded an increase in larval acceptance during spring than summer season.

Abd-Al-Fattah *et al.* (2003) reported that the highest rate of acceptance was occurred during summer, represented 82.8%. Significant differences were found between summer and other seasons. The lowest significant percentage was appeared in winter (36.1 %)

moderate results for the accepted larvae were noticed during spring (70.0%) and autumn (72.8%) seasons with non-significant differences between them.

Taha (2005) showed that the mean percentage of accepted queen cells was affected by nectar and pollen source.

#### b. Effect of seasonal variations

Maurizio (1950) stated that the physiological conditions of summer or winter bees do not depend on the seasons of the year, but was a reaction to the living conditions in the honeybee colonies.

Avetisyan *et al.* (1967) showed that queens reared in April were superior in various biometric characters to those reared in May; queens reared in March and June were inferior. The average weight of virgin queen in 1966 was  $176.9 \pm 3.0$  mg in March,  $194.7 \pm 3.2$  in April,  $185.1 \pm 2.4$  in May;  $188.3 \pm 4.5$  in June, the number of ovarioles for these months was  $132.1 \pm 3.3$ ,  $178.8 \pm 3.2$ ,  $163.0 \pm 3.0$ ,  $160.5 \pm 3.1$ , respectively. The correlation coefficients between the volume of the queen cell and the weight and number of ovarioles of the queen emerging from it were sufficiently high to make these measurements useful in culling cells and virgin queens.

Markosyan and Akopyan (1976) reported that the newly emerged virgin queens were heavier at May, June and July than the August queens.

EsKov and Toropster (1989) found that a uniform temperature of 33-34 °C was the most favourable for the production of large prolific queens. Incubation of queen cells at 31 or 37 °C resulted in weak queens, and they were less well accepted by colonies.

Nour (1992) observed the production of queen cells in spring and early summer which may represent an index to the tendency of the colonies to prepare for swarming. He showed a positive correlation with queen cells production in spring and early summer. The number of queen cups/col. indicated a negative correlation with queen cells in spring, whereas positive correlation was recorded in summer.

Spivak *et al.* (1992) stated that when pupae were incubated at 35.5, 33.5, or 30.5 °C development times decreased significantly and colour rank of emerging queens increased with temperature.

El-Mohandes (1993) stated that the most suitable seasons for rearing queens were late summer, followed by summer and then spring. He studied the effect of temperature and rainfall on honeybee queen rearing; during May to September, he found that the grafting and development of queen cells were only slightly affected by climatic conditions.

Abd-El-Aziem (1999) stated that under Mustorod (Qalubia Governorate) environmental condition, the highest weight of virgin queens was recorded when queens were reared in late summer.

Taha (2000) found that under the environmental conditions of Kafr El-Sheikh region, the highest level of building queen cups and queen cells were during May.

Li (2000) said that when queens are reared during honey and pollen flows, they will be stronger and their quality will be better. However, when reared in the hot summer in poor honey and pollen flows, the queens will be smaller. In autumn, the queens maybe under

fertilized due to low temperatures and thus do not perform well at egg laying.

Zohairy (2001) proved that under El-Mansoura environmental conditions the mean weight of emerged queens in a descending order were in May, June, July, September, August, April and March in 1998 and in May, June, September, April, August, July and March in 1999.

El-Wasseef (2002) showed that the highest level of building queen cups was during March.

Abd-Al-Fattah *et al.* (2007) noted that the better numerous virgin queens could be successfully reared during summer and spring seasons than those occurred during autumn. The lowest percentage of queen emergence with lightest weight was recorded during winter season.

Al-Ghzawi and Zaitoun (2008) stated that the queen rearing season as well as the origin of the queens affected the queens weight, acceptance, preoviposition period, volume of spermatheca quantity and quality of sperms in the spermatheca, Italian bees were heavier than the Syrian bees at emergence.

#### c. Effect of grafted larval age

Pellett (1929) grafted 24 - 36 hours-old larvae using artificial cell cups. Grafting larvae were out-lined by Beljavsky (1933) who pointed out that the method of artificial cell-cups transferring thereto an egg was already developed during the period between 1850 and 1936.

Eckert (1934) clarified that queens reared from larvae, varying from 12 to 72 hours of age at the time of grafting, indicated no correlation between their ovarioles number and the age of grafted