

## **Impact of Different Workers Population in Queenless Rearing Colonies on the Quality of Produced *Apis mellifera carnica* Queen in Manzala Region**

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### **ABSTRACT**

In the current study, The variations exist in acceptance percentage of grafted larvae, emergence percentage and weight of virgin queens were investigated in queenless rearing colonies of local carniolan race (*Apis mellifera carnica*) with different worker populations in Manzala region as isolated area. The accepted percentage of grafted larvae in different worker populations was varying from 52.4 to 100.0% and the variations were significant by different between higher and lower worker population. The higher value of accepted percentage showed in May, 14, 2016, while the lower value appeared in February,16, 2016. The variation in the emerged percentage between lower and higher worker population was also significant. The emerged percentage of virgin queens varying from 84.1 to 100%. The mean weight of virgin queens was 159.63 and 160.58 mg. for lower and higher worker population, respectively with no significant difference between them. The minimum, maximum and mean temperatures had a significant and positive effect on the studied parameters when the rearing occurred in colonies with lower worker. It could be concluded that the preparing queen rearing colonies with worker populations equal 1.5 kilogram or more is very satisfactory in producing good queens.

### **INTRODUCTION**

Rearing pure Carniolan queens (*Apis mellifera carnica*) in Egypt was conducted in two isolated regions. The first was located in the north in Manzala region; the second was established in the new valley, south of Egypt. The two regions produce thousands of virgin and mated queens every year, and these queens are usually used for developing the beekeeping (Mazeed, 1992).

The quality of honeybee queen is a major current concern depends on her genotype and the environmental conditions of the grafted larvae and the dequeened rearing colonies. The suitable larvae for queen rearing is also affected the increase of acceptance honeybee queen cells (Sahinler and Kaftanolu, 2005 and Cengiz *et al.*, 2009).

Weight at emergence and other characters were used to describe the queen reproductive potential (Woyke,1971 and Tapy *et al.*,2000). The quality of queens produced by queen rearing colony depends on various factors among them the strength of colony and time of season (Abd Al-Fattah, 1983; Abd Al-Fattah *et al.*, 2003; Zeedan, 2002; Tapy *et al.*, 2004; Al-Ghzawi and Zaitoun, 2008; Abd Al-Fattah *et al.*, 2011; Sharaf El-Din, 2016).

Nowadays, many queen breeders in Egypt do not consider the colony strength in their production of queens. This research may provide an explanation for the problems of acceptance larvae and poor quality of queens reported by those commercial beekeeper.

### **MATERIALS AND METHODS**

This study was carried out in a private apiary at Meet Salseel, Manzala region, Dakahlia governorate which is considered as isolated area for rearing Carniolan race (*Apis mellifera carnica*), during the period from February,16,2016 to May,14,2016, to Compare the effect of the different queenless workers population on honeybee queen rearing. The following procedure was carried out; six queenless colonies of the Carniolan honeybee race were used for this study. Three queenless colonies contained one kilogram of young workers (lower population) and the others contained one and half kilogram of young workers (higher population). Each colony contained two brood combs (sealed brood). The rearing colonies were provided with sugar syrup (1:1) during the experimental period.

Young larvae less than 36 hours old (depending on the size) were collected from combs which were selected from the center of brood nest. Grafted larvae were transferred from the edge area of the brood comb into wax queen cups containing a drop of a 50% aqueous solution of royal jelly (Laidlaw and Page, 1997). Each grafted frame was constructed of three removable bars and each bar carried 25 queen cups. The grafted operation was undertaken in a room maintained at 25°C and 60% RH.

Rearing colonies were supported with two combs of capped brood weekly and were fed continuously with sugar syrup to stimulate both the production of royal jelly and secretion of wax.

#### **Measurements**

The number of accepted queen cells was counted after 24 hours of inserted into queen rearing colonies and the acceptance percentages / bar / colony was calculated. The number of emerged virgin queens / bar/ colony was counted, individually weighed on balance 0.01 grams and the percentages of emergence were calculated.

#### **Statistical analysis**

Data were subjected to analysis of variance (ANOVA) using “SPSS” computer statistical program. The randomized complete Block Design was used and the mean values were contrasted using Duncan’s multiple range test (Duncan, 1955).

### **RESULTS AND DISCUSSION**

#### **1. Effect of different queenless workers population on queen rearing.**

##### **1. The effect on acceptance percentages:**

Clear significant differences in the acceptance percentage of grafted queen cups between the lower and higher worker populations of the tested colonies (Table 1 and Fig. 1). The mean acceptance percentage for the lower population was 81.5% with a range from 52.3% to 100 %. The lowest mean was showed in February, 16, 2016, while the highest mean values were recorded in May, 14, 2016. The mean acceptance percentage of the higher population colonies were 95.6% with a range from 89.9% to 100 %. The lowest mean was showed in February, 16, 2016. While the highest ones were recorded in May, 14, 2016.

The minimum mean of accepted larvae was recorded in the lower worker population (39.3 cells) in

February, 16, 2016, and the maximum mean of accepted larvae was noticed in the lower (May,14,2016) and higher (April, 11, 2016) worker population colonies (75 cells) respectively.

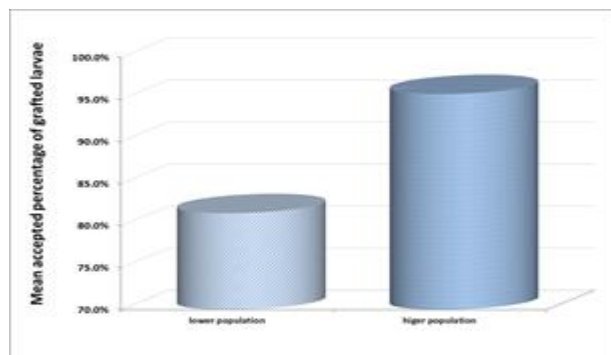
Many authors have discussed this point and found that strong queenless colonies had a positive effect upon their acceptance and early rearing during spring and summer seasons. Shawer *et al.* (1980) registered a percentage of acceptances of larvae 90%. Dodologlu *et al.* (2004) reported that the average acceptance rate in queenless colonies was 95% and Cengiz *et al.* (2009) noticed that the average acceptance rate in queenless colonies in June, July and August were 95, 86.6 and 78.3%.

Zohairy 2001, under the environmental conditions of El-Manzala region found that, the mean percentages of accepted queen cells in a descending order were in May, June, July, April, September, August and March in 1998.

**Table 1. Effect of different queenless workers population on the acceptance of grafted larvae at various dates during the period from Feb.,16 to May,14/2016.**

Date	No. of grafted cups/col.	Lower population		Higher population		Mean / date
		Mean	%	Mean	%	
16/02/2016	75	39.3	52.4	67.4	89.9	71.1 e
27/02/2016	75	42.3	56.4	70.0	93.3	74.9 e
09/03/2016	75	61.1	81.5	69.3	92.4	86.9 d
20/03/2016	75	62.4	83.2	71.3	95.1	89.1 cd
11/04/2016	75	64.3	85.7	75.0	100.0	92.9 bc
22/04/2016	75	71.3	95.1	71.3	95.1	95.1ab
03/05/2016	75	73.1	97.5	74.4	99.0	98.3 a
14/05/2016	75	75.0	100.0	75.0	100.0	100.0 a
Mean / treat.	75.0	61.1 ± 4.784	81.5 B	71.7 ± 1.005	95.6 A	88.5%

Mean in each factor designated by the same letter are not significantly different at 5 % level using Duncan's Multiple Range Test.



**Fig. 1. Mean accepted percentage of grafted larvae in queenless colonies with different worker population.**

**2. The effect on emergence percentages of virgin queens.**

Clear significant differences in the emergence percentage of virgin queens between the lower and higher worker populations of the tested colonies (Table 2 and Fig. 2). The mean of emergence percentage of the lower population colonies were 90.1% with a range from 85.8% to 97.3 %. The lowest mean was showed in February, 16, 2016. While the highest mean values were recorded in May, 14, 2016. The mean emergence percentage of the higher population colonies were 93.7% with a range from 88.6% to 100 %. The lowest mean was showed in April, 22, 2016. While the highest mean values were recorded in May, 14, 2016.

The minimum mean of emergence virgin queens was recorded in the lower worker population colonies (33.7 queens) in February,16,2016 and the maximum mean was noticed in the higher worker population colonies (75 queens) in May,14,2016.

While they were in June, July, August, May, September, April and March in 1999.

Under the environmental conditions of North Sinai region, Abd Al-Fattah *et al.* (2003) determined that the highest rate of acceptance occurred during summer. Significant differences were found between summer and other seasons. The lowest significant percentage appeared in winter while moderate results for the acceptance were noticed during spring and autumn seasons with insignificant differences between them .While Hamada (2012) produced the virgin queens from Carniolan colonies at Meet Salseel, Manzala region, Dakahlia governorate. He found that the highest value (97%) of acceptance was noticed by the 4<sup>th</sup> grafted patch (March, 1, 2010). While the acceptance percentage was clearly decreased to 64% with grafted patch no. 2 (March, 17, 2010). On the other hand, the lowest values (60%) of acceptance was observed in the patch no. 1 (February, 13, 2010).

The results in table, 2 show also, that the emerged queens were significantly higher in May followed by April, March and lastly in February.

Under environmental conditions of Giza region, Hammad (2007) reported that the mean number of queen cells produced/colony during the spring season was higher than in the summer one.

**Table 2. Effect of different queenless workers population on emerged of virgin queens at various dates during the period from Feb.,16 to May,14/2016.**

Date	Lower population		Higher population		Mean / [date]
	Mean	%	Mean	%	
16/02/2016	33.7	85.8	65.4	97.0	91.4 f
27/02/2016	38.4	90.8	66.3	94.7	92.7 f
09/03/2016	51.4	84.1	60.4	87.2	85.6 e
20/03/2016	54.3	87.0	66.3	93.0	90.0 d
11/04/2016	55.4	86.2	70.1	93.5	89.8 cd
22/04/2016	67.0	94.0	63.2	88.6	91.3 c
03/05/2016	70.2	96.0	71.1	95.6	95.8 b
14/05/2016	73.0	97.3	75.0	100.0	98.7 a
Mean / treat.	55.4 ± 5.072	90.1 B	67.2 ± 1.645	93.7 A	91.9%

Mean in each factor designated by the same letter are not significantly different at 5 % level using Duncan's Multiple Range Test.

**3. The effect on weight of emerged virgin queens.**

The obtained results showed no significant difference in the weight of virgin queens (mg./queen) between the different worker populations of the tested colonies (Table 3 and Fig. 3). The mean weight of virgin queens for the lower worker populations colonies was 159.63 mg./queen with a range 151.67 to 174.0 mg./queen. The mean weight of virgin queens (mg./queen) for the higher worker populations colonies was 160.58 mg./queen with a range 150.33 to 175.33 mg./queen. The lowest (150.33 mg./queen) and the

highest (175.33mg./queen) mean weight of virgin queens were recorded in colonies with higher worker populations in April,22,2016 and May,14,2016 respectively.

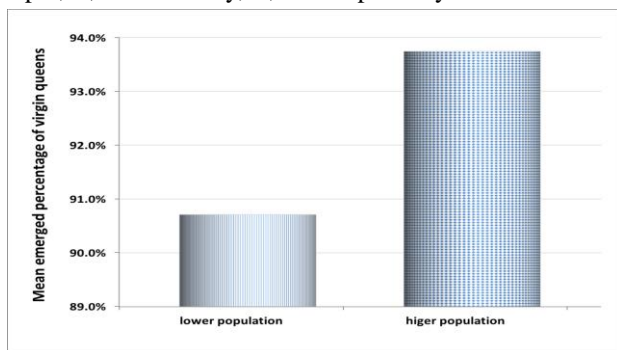


Fig. 2. Mean emerged percentage of virgin queens in queenless colonies with different worker population.

Table 3. Effect of different queenless workers population on weight of virgin queens (mg.) at various dates during the period from Feb., 16 to May,14/2016.

Date	Lower population		Higher population		Mean / date
	No. of weighed queens	Mean	No. of weighed queens	Mean	
16/02/2016	33	151.67	65	155.67	153.7 f
27/02/2016	38	157.67	66	158.33	158.0 c
09/03/2016	51	155.67	60	154.00	154.8 ef
20/03/2016	54	153.00	66	158.67	155.8 de
11/04/2016	55	153.67	70	161.67	157.7 c
22/04/2016	67	163.00	63	150.33	156.7 cd
03/05/2016	70	168.33	71	170.67	169.5 b
14/05/2016	73	174.00	75	175.33	174.7 a
Mean / treat.		159.63 A ± 2.8539		160.58 A ± 2.9932	160.1 ± 2.7058

Mean in each factor designated by the same letter are not significantly different at 5 % level using Duncan's Multiple Range Test.

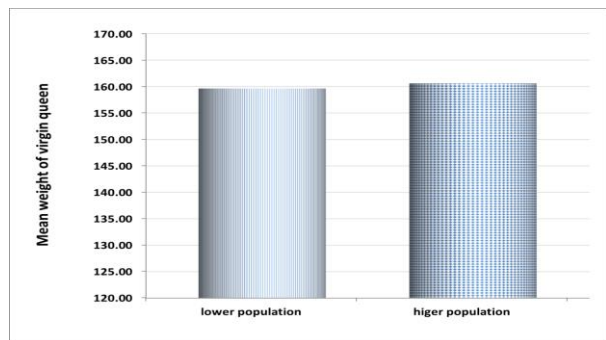


Fig. 3. Mean weight of virgin queens in queenless colonies with different worker population.

The finding results agreed with those found by Abdellatif (1967) who reported that the average weights of the queen in Marsh, April and May were 153.8, 156.6, 153.5 mg, respectively, as there was a positive correlation between the queen weight and its cell size in March ( $r=0.7$ ), but no relationship existed in April and May ( $r = 0.4$  and  $0.3$ , respectively). Shower *et al.* (1980) indicated that the body weight of virgin queens differed significantly according to the rearing season. The heaviest queens were produced in May and August. Queens reared in February, March and July had an intermediate weight, while those reared in April, June and October had the least body weight. Zeedan (2002) mentioned that under Giza environmental conditions, the heaviest queens were produced during summer (178.7 mg. / queen)

and this value was significantly higher than the other seasons which were 175.6, 171.8 and 168.2 mg./queen in spring, autumn and winter, respectively. Zohairy (2007) realized that the mean weight of emerged queens in a descending order were in May, June, September, August, April and March in 1998 and in May, June, September, April, August, July and Marsh in 1999. Abdulaziz *et al.* (2013) shows that the general mean body weight of newly emerged virgin queens (less than 24 h) of *A. m. carnica* queens reared by the grafting method during spring season of 2010 and 2011 in Riyadh, were  $165.87 \pm 9.79$  ( $n=20$ ) and ranged between 153.0-195.0. Moustafa *et al.* (2014) studied the quality of virgin queens of honey bee that produced commercially from four large queen producers in Egypt, results indicated that the mean body weight was  $127.6 \pm 0.017$  mg. in spring season and  $144.17 \pm 0.020$  mg. in summer season. However, (Gregorc and Škerl 2015) reported that for *Apis mellifera carnica*, the average weights of queens were  $208.40 \pm 15.31$ mg,  $209.49 \pm 9.82$ mg and  $201.83 \pm 15.85$  mg, during 2006, 2008 and 2010 respectively.

## 2. The effect of interaction between ambient temperature and workers population on quality of produced queens.

The obtained results showed that no significant effect on the acceptance percentage of grafted larvae for the interaction between worker populations and minimum, maximum and mean temperatures (Table 4). However, clear significant impact on the emergence percentage of virgin queens between the lower worker populations and the tested temperatures. The mean weights of virgin queens were significantly followed the change of temperatures in the lower worker populations only

Table 4. Simple correlation coefficients between ambient temperatures and both of acceptance of grafted larvae, emergence of virgin queens and weight of virgin queens in honeybee queenless colonies with different populations during the period from Feb.,16 to May,14/2016.

Temp. °c	Lower population		Higher population		weight	weight
	accep	emerg	accep	emerg		
Min. temp. Corr.(r)	0.643	0.882*	0.869*	0.470	0.280	0.576
Max. temp. Corr.(r)	0.433	0.774*	0.777*	0.073	0.210	0.486
Mean temp. Corr.(r)	0.482	0.825*	0.823*	0.149	0.274	0.545

\* Correlation is significant at the 0.05 level  $r = .707$

The obtained results are agreement with those found by various authors who reported that the thermoregulation within honeybee colony is mainly depend on the population density of workers. They also, added that rearing brood need a constant nest temperature (Secley and Heinirich, 1981; Eskov, 1990; Degrandi-Hoffman *et al.*, 1993; Jones *et al.*, 2004; Chuda-Mickiewicz and Samborski, 2015)

## REFERENCES

Abd Al-Fattah, M. A. (1983). Some Ecological Studies of Honeybee Colonies (*Apis mellifera* L.) Under the Environmental Conditions of Giza Region. M. Sc. Thesis, Fac. Agric, Cairo Univ., Egypt, 190p.

- Abd Al-Fattah, M. A.; El-Basiony, M. N. and Mahfouz, H. M. (2003). Some environmental factors affecting the quality of artificially reared queens, (*Apis mellifera* L.) in North Sinai region. Egypt. J. Agric. Sc. Mansoura Univ., 28 (8):6407-6417.
- Abd Al-Fattah, M. A.; Mazed, A.M., Al-Hady, N.A. (2011). Quality and quantity of honeybee queens as affected by the number and distribution of queen cells within queen rearing colonies. J. Apic. Sci., 55(2), 31-43.
- Abdellatif, M. A. (1967). Some studies on queen honey bee rearing in the Alexandria region of Egypt. Am. Bee J., 107:88-89.
- Abdulaziz S. A.; Hassan M. B. and Ayman A. O. (2013). Queen morphometric and reproductive characters of *Apis mellifera jemenitica*, a native honey bee to Saudi Arabia, Bulletin of Insectology 66 (2): 239-244.
- AL-Ghzawi, A. and Zaitoun, S. (2008). Origin and rearing season of honeybee queens affect some of their physiological and reproductive characteristics. Entomol. Res., 38:139-148.
- Cengiz, M.; Emsen, B. and Dodologlu, A. (2009). Some characteristics of queen bees (*Apis mellifera* L.) rearing in queenright and queenless colonies. J. Anim. Vet. Adv., 8:1083-1085.
- Chuda-Mickiewicz, B. (1994). Wpływ temperatury otoczenia na zachowanie kłebu zimującej rodziny [Ambient temperature effect on wintering bee colony cluster behaviour]. Rozprawy nr 162, AR Szczecin [in Polish].
- Degrandi-Hoffman, G., Spivak, M., Martin, J.H. (1993). Role of thermoregulation by nestmates on the development time of honey bee (Hymenoptera: Apidae) queens. Ann. Entomol. Soc. Amer., 86(2), 165-172.
- Dodologlu, A.; Emsen, B. and Genc, F. (2004). Comparison of some characteristics of queen honey bees (*Apis mellifera* L.) reared by using Doolittle method and natural queen cell. J. Appl. Anim. Res., 26:113-115. 162.
- Duncan, D. B. (1955). Multiple range and multiple F tests. Biometrics 11, 1-42.
- Eskov, E.K. (1990). Termofaktor. V: Ekologija medonosnoj pceli [The thermal factor. In:Ecology of honey-bee], Moscow, Rasogropomizdat, 39-96.[in Russian]
- Gregorc A. and Smodiš Škerl M. (2015): Characteristics Of Honey Bee (*Apis Mellifera Carnica*, Pollman (1879) Queens Reared In Slovenian Commercial Breeding Stations (J. Apic. Sci. Vol. 59 No. 2, p. 5-12).
- Hamada M.A.I. (2012). Morphometrical and biological studies of local carniolan honeybee colonies in manzala region . M. Sc. Thesis, Fac. Agric., Cairo Univ., Egypt, , 207p.
- Hammad, H. M. A. (2007). Effect of Simulative Feeding with Pollen Substitutes on the Development and Production of Honeybee Colonies. M. Sc. Thesis, Fac. Agric., Cairo Univ., Egypt, 226 p.
- Jones, J.C., Myerscough, M.R., Graham, S., Oldroyd, B.P. (2004). Honey bee nest thermoregulation: diversity promotes stability. Science, 305, 402-404.
- Laidlaw H. H., Page R. E. (1997) Queen rearing and bee breeding. Wicwas Press. Connecticut, USA. 224 pp.
- Mazed, A. (1992): Morphometrical and biological studies on some races and hybrids of honeybees (*Apis mellifera* L.) in Egypt by instrumental insemination. M.Sc. Thesis, Faculty of Agriculture, Cairo University, pp164.
- Şahinler, N. and Kaftanoğlu, O. (2005). The Effects of season and honeybee (*Apis mellifera* L.) genotype on acceptance rates and royal jelly production. Turk. J. Vet. Anim. Sci., 29:499-503.
- Sharaf El -Din , H . A . (2016) : honeybee queens performance in relation to their long period storage in queen – right colonies . ph.D. Thesis , fac . Agric . Cairo univ , 158 p .
- Shawer, M. B.; Abd El – Rahim, W. A. and Abd El – Rahman, I. (1980). Effect of rearing season on certain characters of carniolan honey bee queen at Kafr El – Sheikh Governorate. J. Agric. Res. Tanta Univ., 6(1):195-204.
- Secley, T.D., Heinrich, B. (1981). Regulation of temperature in nests of social bee. In:Henrich B. (ed) Insect thermoregulation. Wiley, New York, 159-234.
- Tarpy, D. R.; Hatch, S. and Fletcher, D. J. C. (2000). The influence of queen age and quality during queen replacement in honeybee colonies. Anim. Behav., 59:97-101. 171
- Tarpy, D. R.; Gilly, D. C. and Seeley, T. D. (2004). Levels of selection in a social insect: a review of conflict and cooperation during honey bee (*Apis mellifera*) queen replacement. Behav. Ecol. Sociobiol., 55:513-523.
- Woyke, J. (1971). Correlation between the age at which honeybee brood was grafted, characteristics of the resultant queens, and results of insemination. J. Apic. Res., 10:45-55.
- Zeedan, E. W. M. (2002). Studies on Certain Factors Affecting Production and Quality of Queen Honeybees (*Apis mellifera* L.) in Giza Region. M.Sc. Thesis, Fac. Agric., Cairo Univ., 134 p.
- Zohairy, A. M. E. (2001). Studies on Queen Rearing using Cell Punch and New other Methods on *Apis mellifera* L. M. Sc. Thesis, Fac. Agric., Mansoura Univ., Egypt, 137 p.
- Zohairy, A. M. E. (2007). Studies on Queen Rearing. Ph.D. Thesis, Fac. Agric., Mansoura Univ., 122 p.

## تأثير التعداد المختلف للشغالات داخل طوائف تربية الملكات اليتيمة على جودة الملكات الناتجة في منطقة المنزلة حسن محمد فتحي<sup>1</sup>، أحمد محمد زهيرى<sup>2</sup> و محمد عبد الوهاب حماده<sup>2</sup> <sup>1</sup>قسم الحشرات الاقتصادية، كلية الزراعة، جامعة المنصورة <sup>2</sup>قسم بحوث النحل، معهد وقاية النباتات، مركز البحوث الزراعية

في الدراسة الحالية تم اختبار النسبة المئوية لقبول اليرقات المطعومه، النسبة المئوية لفقس الملكات و كذلك وزن الملكات العذارى حديثة الفقس المرباة في طوائف بيتيمة ذات تعداد مختلف من الشغالات (وزن الشغالات 1 كيلوجرام، 1.5 كيلوجرام) بمنطقة المنزلة المعزولة. تراوحت النسبة المئوية لقبول اليرقات المطعومه بين 52.4 الى 100 % وكان هناك فرق معنوي بين الطوائف مرتفعة و منخفضة التعداد. اعلى قيمة لنسبة القبول سجلت في 14 مايو 2016 بينما اقل قيمه سجلت في 16 فبراير 2016. كذلك كان هناك فروق معنوية بين نسب فقس الملكات بين طوائف التربية في التجربة وكانت النسب تتراوح بين 84.1 الى 100%. متوسط وزن الملكات للطوائف منخفضة التعداد (159.63 ملجم) و للطوائف مرتفعة التعداد (160.58 ملجم) مع عدم وجود فروق معنوية بالنسبة للوزان بين طوائف التجربة. كان هناك تأثير ايجابي لدرجات الحرارة المختلفة (صغرى، قصوى، متوسطة) على الطوائف ذات التعداد المنخفض من الشغالات. ولذلك ينصح باستخدام طوائف تربية تحتوى على 1.5 كيلوجرام نحل فما فوق لانتاج ملكات ذات جودة عالية.