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EFFECT OF GRAFTING SUBSTRATES USED IN WET GRAFTING METHOD ON THE CHARACTERISTICS OF PRODUCED VIRGIN QUEENS

BY

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ABSTRACT

This study was carried out during 2000 & 2001 to compare between the importance of grafting substrates, which are used in wet grafting and its effect on the characteristics of produced virgin queens. This work considers the consequence of transplantation with pure royal jelly; royal jelly + 10% honey; royal jelly + 10% stored pollen, and royal jelly + 5% honey + 5% stored pollen to deter mine the best technique used for queen production. The results revealed that the acceptance and the different characters of virgin queens emergence were highest by using royal jelly as grafting substrate. The lowest acceptance and different characters of virgin queens emergence were observed, when using royal jelly + 10% stored pollen as grafting substrate. The statistical analysis showed that there were significant difference between the used grafting substrates.

INTRODUCTION

Honey production, is known to be affected by various important factors among them queen weight, amount of protein in ovaries of virgin queens, diameter of spermatheca and length and width of 3rd and 4th of tergites.

Queens vary greatly in size and weight and the greater weight gives more ovarioles and more eggs. Quality of queens is not only hereditary controlled, but also depends on the conditions in which it grows as larvae. The size and vigor of a colony of honey bee are a direct reflection of the genotype of the queen, as her body weight increases (Hoopingamer and Farrar, 1959) queen producers strive to provide optimum condition as larvae develop. However, they did not know accurately the proper procedures, which could enhance the required queen qualities (Delaplame and Harbo, 1988). It was found that artificial queen rearing could be used to control the queen qualities. This was carried out by using different wet grafting methods.

Some authors have stated that Royal Jelly priming is not an essential requirement for successful transplantation but may serve to facilitate the process, when other conditions are less than optimal (York, 1975; Laidlaw, 1979; Morse,

1979) some authors have reported that priming result in larval rejection and stunfed growth (Johansson & Joliansson, 1978), whilst others have found it to be particularly beneficial (Vuillaunee, 1959; Bodolanova, 1974; Bobrzecki & Prabucki, 1975). Ebadiacary (1980) obtained 86.2% acceptance for cells primed with 10 {SYMBOL 109 \f "Symbol"}l of Royal Jelly and 10% honey. The present study considers the consequence of transplantation with a pure royal jelly, Royal Jelly + 10% honey, royal Jelly + 10% stored pollen and Royal Jelly + 5% stored pollen + 5% honey (R.S. Pickard and G.Y. Krrhb, 1983).

Volsevich (1954) concluded that the quality of the queen was determined by the number of ovarioles, length of ovary, diameter of spermatheca, length and width of both 3rd and 4th tergites and area of the forewing. Woyke (1971) found that number of spermatozoa in the spermatheca was directly related to the queen body weight. He concluded that the process of virgin queens weight could be help in their selection. Eid *et al.* (1978) concluded that the size of the queens at emergence could be used as a criterion for determining their prolificness. In the present work the weight of virgin, queens diameter of spermatheca, length and width of 3rd and 4th tergum, area of the forewing were determined aiming to study the effect of different wet grafting methods on the characteristics of produced virgin queens to find out the most suitable way and the best condition for queen rearing.

MATERIALS AND METHODS

This study was performed in apiary of Faculty of Agriculture Al-Azhar University at Mustored during 2000, 2001, to determine the best way for queen rearing and to establish the importance of grafting substrates, which are used in wet grafting for commercial queen production.

Grafting techniques :

Experiments were conducted on rearing honey bee queens during nectarflow to evaluate the effect of different ways of wet grafting on the characteristics of produced virgin queens. These methods were :

a. Pure royal jelly diet.
b. Royal Jelly + 10% honey.
c. Royal Jelly + 10% stored pollen.
d. Royal Jelly + 5% stored pollen + 5% honey.

Royal jelly was collected 1 day before starting the experiment from queen cell containing larvae 72 h old; it was kept at 5{SYMBOL 176 \f "Symbol"}C during storage. Bee bread (stored pollen in worker cells covered with a thin layer of honey by bees) from various floral sources was collected with a small spatula from different combs of several colonies and mixed one frame of 30 cells was placed in each cell-builder colony. The location of each treatment replication on each bar was random.

The queen rearing technique was carried out by transplanting (grafting) one day old worker larvae of carniolan race into queen cell cups which were given to the queen rearing colonies (Doolittle 1909).

Larvae age about 24 hours were usually grafting into wax cup by using different substrate of wet grafting.

The larvae were transferred from their cells to different artificial queen cells containing different substrates.

As soon as all queen cell cups were grafted the frame holding then was insorted in the prepared cell building units.

All the queen rearing units were fed on sugar syrup.

Nine days after larval grafted the seald cells were carefully removed from bars, each queen cell was placed in a screened cage incubated in strange colony until emergence.

The criteria used throughout this work of virgin queen produced from different wet grafting were the weight of virigin queen, the area of forewing, length and width of 3rd and 4th tergites and the diameter of spermatheca.

RESULTS AND DISCUSSION

1. The successful number of queen cell cups produced by different wet grafting experiments :

These experiments were carried out during nectar flow of season, in order to study the effect of different wet grafting substrates on the production of queen cells and percentage of acceptance.

Four different wet grafting experiments were used in this work, the first, pure royal jelly, second, royal jelly + 10% honey, third, royal jelly + 5% honey + 5% stored pollen and the fourth royal jelly + 10% stored pollen. The queen rearing method described before were used. On day larvae were used in all these experiments. Generally, in this work 45 queen cell cups were attached in three wooden stick bars prepared for grafting. 450 queen cell cups were grafted for each wet grafting trial. The number of successful queen cell cups produced form each wet grafting trial during all experiments were calculated. Data in Table (1) showed that the average number of the successful queen cell cups produced from different wet grafting experiments were 34.7, 30.2, 24.23 and 20.1 queen cell cups respectively. It constituted 77.11%, 67.11%, 54.0% and 44.66% to the total cups added for different wet grafting trials respectively. From the previous data it is clear that the best wet grafting was when using the pure royal jelly, after that cane, the grafting with royal jelly +10% honeys the last number of the successful queen cell cups was obtained by using royal jelly +10% stored pollen. Statistical analysis showed that there was a significant difference between the numbers of successful queen cells produced by the different wet grafting experiments.

This result agree with the findings obtained by Askew (1957).

In this respect Mohanna (1969) reported that the number of successful queen cell cups differs from one month to another.

Mohamed (1999) mentioned that most suitable time for obtaining highest number of the successful queen cell cups was late summer and during spring. Ahmed (2004) found that the Italian bees was more active in queen cell production than the carniolan race during late summer and spring under Mustored environmental conditions. It could be suggested that when stored pollen was including in the priming substrate, the acceptance of cells was reduced greatly (Table 1). Stored pollen apparently changes the feeding behaviour of nurse bees, perhaps by inhibiting cell provisioning with royal jelly. In our opinion in this work, cell acceptance and rejection are perhaps controlled by the same underlying mechanisms, and they are clearly the result of many variables acting in concern. Organization of these variables to enhance acceptance may well influence the quality of queen cell provisioning throughout larval development, and the quality of queen produced. It could be suggested that many factors, such as foreign odours or queen pheromones on or in the wax cells may stimulate rejection. Most of the mechanisms regulating acceptance and provisioning remain to be determined.

Finally, the condition of colony could be improved for the cell building by the addition of more bees from other colonies or by the addition of emerging brood and bees. Continuous feeding of sugar syrup, pollen and dilute honey was essential to assure optimum condition for queen cells production.

Acceptance of grafted larvae is the good indication of colony condition. The obtained results showed clearly that, the most suitable wet grafting method for queen cell production was using fresh royal jelly that could bee advised for bee keepers.

2. Effect of various substrates in wet grafting on the characters of produced virgin queens :

Data presented in Table (2), showed clearly that there were significant differences in weight of virgin queens, diameter of spermetheca and length and width of both 3rd and 4th tergites produced by different wet grafting methods. Also, statistical analysis of the data revealed significant difference for area of forewing of virgin queens produced by different wet grafting methods.

From these results, it is clear that the different characters of emergence of virgin queens were highest by using fresh royal jelly as grafting substrate. The lowest different characters of emergence of virgin queens produced by using royal jelly +10% stored pollen as grafting substrate. Statistical analysis showed also significant difference between the different grafting substrate, used in wet grafting.

Table (1) : The successful number of queen cell cups produced by different wet grafting substrates.

No. of Bacles	No. of cups added	Grafting methods							
		Pure royal jelly		Royal jelly +10% honey		Royal jelly +5% honey +5% stored pollan		Royal jelly 10% stored pollan	
		Accepted	Rejected	Accepted	Rejected	Accepted	Rejected	Accepted	Rejected
1	45	33	12	30	15	22	23	19	26
2	45	35	10	29	16	28	17	22	23
3	45	41	4	28	17	28	17	21	24
4	45	36	9	24	21	32	13	25	20
5	45	34	11	31	14	25	20	20	25
6	45	37	8	29	16	26	19	19	26
7	45	30	15	26	19	14	31	16	29
8	45	32	13	35	10	27	18	18	27
9	45	29	16	32	13	23	22	20	25
10	45	40	5	35	10	26	19	21	24
Total	450	347	103	302	148	243	207	201	249
Mean		34.7		30.2		24.3		20.1	
%		77.11		67.11		54.0		44.66	

L.S.D. = 9.853 AT 5%.

Table (2) : Effect of various substrates in wet grafting on the external and internal characters of produced virgin queen.

Character of virgin queens	Methods of different wet grafting				L.S.D.
	Pure royal jelly	Royal jelly +10% honey	Royal jelly +5% honey +5% stored pollan	Royal jelly +10% stored pollan	5%
Area of forewing	16.457	16.243	15.652	15.414	0.1152
L. of 3rd tergite	3.346	3.152	2.346	2.110	0.205
W. of 3rd tergite	9.305	8.860	7.120	5.930	0.0680
L. of 4th tergite	3.336	3.000	2.244	2.070	0.004
W. of 4th tergite	10.448	10.085	9.190	8.530	0.053
Diamter of spermatheca	1235.5	1122.0	1025.5	829.0	203.63
Weight of virgin queens	196.5	179.95	165.2	159.5	19.58

Significant.

In summary, the most suitable substrate for rearing virgin queens is by using royal jelly as grafting substrate. Szabo (1973) found that the weight of newly emerged virgin queens was correlated with the capacity of egg laying and the weights of queens at emergence seems to be a reliable index for the beekeepers in selecting them (Szabo, 1974).

From the obtained data clear it is that the morphological variation can be useful for separating at least some population within Apis mellifera, however, it could be said that virgin queens vary greatly in size and weight. The length and width of both 3rd and 4th tergites, area of forewing can estimate the quality of virgin queens. This agree with both Volesvish (1954) and Savin (1956) they found that the quality of queens was estimated by the number of ovariols, diameter of spermatheca, length and width of 3rd and 4th tergites and length and width of forewing.

From the foregoing results, it could be said that variation of different characters of virgin queens produced by different wet grafting method are influenced by different environmental factors such as using different wet grafting methods (pure royal jelly technique) which lead to the improvement of different characters of resulted virgin queens and could bee advised for use in the commercial apiaries.

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تأثير المواد المستخدمة فى التطعيم المبتل على صفات الملكات العذارى الناتجة

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فى هذه الدراسة تم استخدم أربعة انواع من المواد المستخدمة فى التطعيم المبتل فى تطعيم الكئوس الشمعية وهى -:

١- استخدام الغذاء الملكى النقى.
٢- استخدام غذاء ملكى +١٠% عسل.
٣- استخدام غذاء ملكى +٥٠% عسل +٥٠% حبوب اللقاح.
٤- استخدام غذاء ملكى + ١٠% حبوب لقاح.

وتم دراسة تأثير استخدام هذه المواد المختلفة فى التطعيم المبتل على الصفات المورفولوجية والفسيولوجية للملكات العذارى وكانت الصفات التى درست كما يلى -:

١- مساحة الجناح الأمامى.
٢- طول وعرض الترجة الثالثة والرابعة.
٣- وزن العذارى.
٤- قطر القابلة المنوية.

ولقد أوضحت النتائج وجود فروق معنوية بين الصفات المختلفة للعذارى الناتجة لكل نوع من أنواع التطعيم .وتبين ان استخدام الغذاء الملكى النقى Pure royal Jelly قد أعطى أعلى النتائج، بينما اعطى استخادم الغذاء الملكى مع + ١٠% حبوب لقاح أقل النتائج من حيث الصفات المورفولوجية والفسيولوجية.

واوضحت النتائج كذلك ان استخدام الغذاء الملكى النقى Pure royal jellyكان له أكبر الأثر فى تحسين صفات الملكات العذارى الناتجة.