EFFECT OF DIFFERENT FACTORS ON THE EFFICIENCY OF HONEY BEE QUEEN REARING

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S u m m a r y

The objective of the study was to investigate the effect of selected environmental factors on the efficiency with which honey bee queens are reared. The research material collected over the years 1990 - 2001 comprised nearly 31 thousand grafted larvae and over 14 thousand isolated queen cells reared in 623 replications. A substantial variation in queen rearing success was recorded as measured by the percentage of isolated queen cells vs. grafted larvae over the study years caused mainly by different time of queen rearing. The highest efficiency rate occurred at the beginning of the season in the second decade of May (55.8%) and then declined until the second decade of July (25.8%) to rise again in the first decade of August (44.6%). Rearing success rate was influenced slightly but significantly by air temperature, solar radation and relative humidity on the day of larva grafting. The site of larva introduction and the multiple use of a nurse colony had no impact. Queen rearing was best stimulated by moderate nectar flow whereas there was an unequivocal negative impact of both the very high flow (24.4%) and of the absence of flow (36.1%). Over the successive years of the study from 33.8 to 45.4% of the grafted larvae emerged as queens. Out of 311 non-emerged queens 32.8% died at the light pupa stage and 20.2% at the imago stage. Sporadically, the failure to emerge was caused by the presence of two individuals in a cell, each of them having died at a different development stage (2.6%).

Keywords: queen rearing, time of season, flow conditions, death of queen cells, weather conditions.

INTRODUCTION

A regular replacement of queens in bee colonies is a prerequisite for a successful beekeeping venture. A prevalent opinion is that a queen should not remain in the colony for more than two years. It means that every year the number of queens to be reared is equal to half the number of colonies in the apiary. Quality queens are reared in specialized apiaries that do the rearing on a large scale and during the whole season.

Rearing efficiency is to a large extent dependent upon ambient conditions but it is largely influenced by the choice and the preparation of the nurse colony and by the rearing conditions. It is necessary to start with a nurse colony that is healthy and strong and has brood of different age (Biłasz 1962, Taranow 1968, Skowronek and Skubida 1988). Rearing can be done in the presence of the queen but rearing performance is better in queenless colonies (Fell and Morse 1984, Chuda-Mickiewicz and Prabucki 1995) and in the absence of emergency queen cells (Free et al. 1984).

Cell cups in which the larvae are grafted can be made of plastic but beeswax cups perform better (Chang 1977). The diameter of 9 mm is regarded as optimal (Weiss 1967, Skowronek, Skubida 1988). A moot point is the necessity to introduce empty cups in advance to precondition the

Journal of Apicultural Science

colony. According to some authors the preconditioning positively influences the acceptance of grafted larvae (Kither and Pickard 1983, Delaplane and Harbo 1988). According to other investigators the treatment serves no purpose (Skowronek and Skubida 1988).

When grafted in cell cups the larvae normally are not placed on the bare bottom but on diverse substrates that can serve as the food for the larvae or they just help maintain the right humidity. A small drop of royal jelly or of royal jelly diluted in plain water are the best substrates (Bobrzecki, Prabucki 1975, Ebadi and Gary 1980, Pickard and Kither 1983, Skowronek, Skubida 1988). The best quality of reared queens is secured by a double transfer that consists in the grafting of a larva to replace another one that has been removed from an already existing queen cell (Roberts 1965, Weiss 1974).

Acceptance of larvae is also affected by the number of grafted larvae. Smaller numbers are preferable (Pusca 1970) and they should not exceed 50 (Konopacka 1971). The site of grafting can also be of some importance but no significant relationships have been demonstrated (Fell and Morse 1984, Visscher 1986).

The aim of the study was to analyze the effect of different factors on efficiency of queen rearing that was conducted in the same apiary operation for more than 10 years using an unchanged rearing method.

MATERIAL AND METHODS

The observations were taken in the apiary of the Institute of Pomology and Floriculture in the years 1990 - 2001. In 1991 no observations were taken. The location of the apiary is characterized by moderately high nectar flow and by a high number of bee colonies. Fruit trees, dandelion, locust, lime and goldenrod are the major honey plants of the location.

The rearing was done in a total of 145 nurse colonies to which nearly 31,000 larvae were introduced in 623 replications also referred to as rearing series. The rearing was done in colonies with Carniolan and Caucasian bees or with the hybrids thereof.

Larvae aged up to 24 hrs were used for rearing. Their age was estimated by size. The larvae were grafted from comb cells to beeswax queen cups 9 mm in diameter using a conventional grafting needle. Prior to grafting a drop of royal jelly diluted with boiled water was put on the cup's bottom. Attached to cork stoppers the larva-containing cups numbering from 45 to 90 were put on one or two frames and the frames were inserted in a nurse colony.

The rearing was done in queenless nurse colonies derived from healthy colonies in a swarming mood or showing the first symptoms of swarming. The nurse colonies contained at least four brood combs mostly at capped stage. The nurse colonies were strong and usually with a lot of young bees. The colony nest was packed very tightly so that the excess of bees overflowed and clustered behind the division board. The nurse colonies were supplied every week with two combs of brood of different age but mostly of capped brood. The nurse colonies were used repeatedly with new batches of larvae being grafted at intervals of several days. In a single nurse colony the rearing was done in 1 to 13 replications. The number of replications depended on the acceptance of successive grafts.

After 2 - 3 days following grafting the colonies were inspected for larva acceptance. Once the queen cells were capped they were placed in styrofoam blocks with holes 13 mm in diameters and kept in incubators at 34°C and at relative humidity of ca. 80%. One or two days before emergence the queen cells were transferred to a queen cages. At the bottom of the cage there was an attached beeswax cup containing a drop

of honey. The queens emerged following 7 days of incubation. Non-emerged queens were incubated for 2 more days thus giving an opportunity to emerge to queens with a slightly longer development period.

The data were analyzed using the multiple regression analysis. The calculation were made using Statgraphics Plus software program. Studente's t-test was used to test for a significant differences between means. Two-factor ANOVA was used to compare the variables of the decade rearing efficiency against sets of different meteorological data. The coefficients of correlation between analyzed data were calculated.

RESULTS AND DISCUSSION

A total of 30907 larvae in 623 series (replications) grafted in queen cells were experimentally reared in the years 1900--2001. It translates to an average of 49.6 larvae per replication. Of that number 14,272 individuals were accepted by the rearing colonies and brought to the pupa stage in capped queen cells. The rearing success rate as measured by the percentage of capped queen cells vs. the number of grafted larvae varied substantially over the years with the lowest rate of 32.9% in 1990 to the highest of 58.1% in 1999 (Table 1). The year to year differences were proved to be statistically significant. The causes of that variation are difficult to explain but the impact of the timing of rearing was obvious. The highest success rate was obtained in the years in which rearing was terminated by the beginning of July (1992, 1998, 1999). It is confirmed by the results of two-factor ANOVA (year and decade) which showed very high significance of decade-to-decade differences within the season while at the same time year-to year differences being non-significant.

The rearing success within a beekeeping season was analyzed. The acceptance rate by the rearing colonies was highest at the beginning of the season in May (Table 2). The success rate in May averaged across 11 years of the experiment was 52.1%. In the

Table 1

Queen rearing efficiency over successive seasons of 1990 to 2001.

Year	Rearing period	Duration of rearing period	Number of replications	Number of grafted larvae	Isolated queen cells	
					No.	%
1990	16.05-31.07	76	69	3105	1022	32.9 a
1992	20.05-6.07	47	43	1980	1101	55.6 d
1993	20.05-22.08	94	61	2745	1194	43.5 bc
1994	23.05-19.07	57	73	3600	1772	49.2 cd
1995	25.05-26.07	62	17	1020	577	56.6 d
1996	23.05-7.08	76	50	2964	1298	43.8 bc
1997	16.05-20.08	96	50	3030	1311	43.3 bc
1998	8.05-10.07	63	49	2337	1205	51.6 d
1999	6.05-10.07	65	72	3180	1846	58.1 d
2000	7.05-15.07	69	57	2941	1067	36.3 ab
2001	8.05-1.07	54	82	4005	1879	46.9 cd
	Total	69.0	623	30907	14272	46.2

Means followed by different letters are significantly different $P<0.05$

Table 2

Queen rearing efficiency in successive months averaged over the years 1990 - 2001.

Month	Number of replications	Number of grafted larvae	Isolated queen cells	
			Number	%
May	180	9283	4841	52.1 b
June	362	17262	7820	45.3 ab
July	72	3792	1372	36.2 a
August	9	570	239	41.9 ab
Total	623	30907	14272	46.2

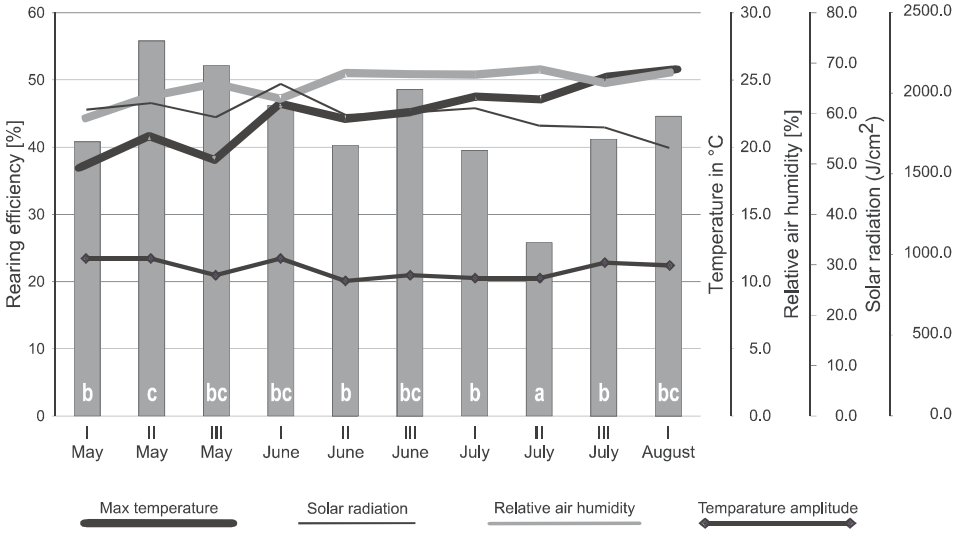

Fig. 1. Queen rearing efficiency over the years 1990 - 2001 against the variables of decade-based average maximum temperature, temparature amplitude, solar radiation and relative air humidity.

successive months the success rate declined to 36.2% in the month of July. In August the rearing success increased again. The differences between the extreme rates were found to be significant.

The assessment of the seasonal efficiency of queen rearing made over the decades (Fig. 1) showed that at the beginning of May the acceptance of larvae grafted in cell cups was poor. In the second decade of May there was an abrupt increase in the acceptance rate and it was also the top decade of the season for raising queens by the colonies. From the third decade of May onwards there was a gradual decline in the efficiency of queen rearing and in mid-July the efficiency dropped to half of that in mid-May. That decline in rearing efficiency was checked in the third decade of June with the percentage of capped queen cells being higher than in the first decade of June. The decade to decade differences were found to be signifi cant in several cases.

In order to find the cause of that variability rearing efficiency vs. temperature, solar

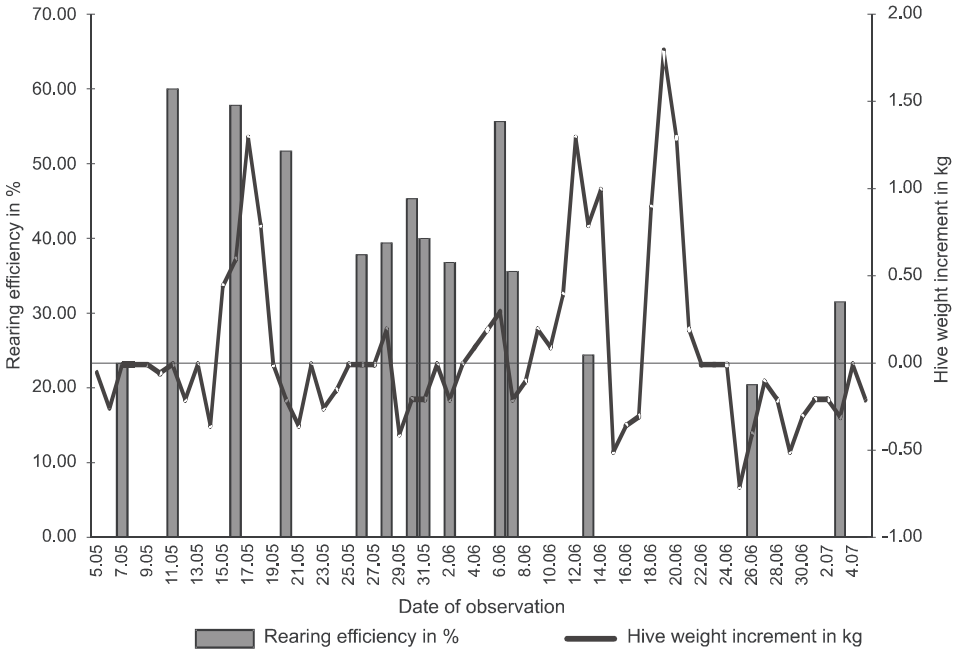

Fig. 2 Queen rearing efficiency in colony vs. hive weight increment in 2000

radiation, and relative air humidity were compared. The plotted curves do not point to any relationship between rearing efficiency over decades and the meteorological factors under investigation. Neither do the calculated correlations point to such a relationship. However, the correlation coefficient for those factors proved to be low but none-the-less higly significant $P<0.01$ and reached the following values:

- rearing efficiency vs. average diurnal relative humidity $r= 0.114$
- rearing efficiency vs. solar radiation J/cm^2 $r = 0.102$
- rearing efficiency vs. maximum temperature $r = 0.106$
- rearing efficiency vs. temperature amplitude $r = 0.146$

It was expected that the rearing efficiency could be affected by the magnitude of nectar flow. For that purpose the data from the individual rearing series of 2000 were compared against daily weight increments of the reference hive on the grafting day. The data curves shown on Fig. 2 do not point to such a simple relationship. The acceptance of larvae grafted on the days with a small weight decrement was high (20.05) and it was quite poor when grafts coincides with daily weight increments of more than 1 kg (13.06). A more detailed comparison of those data was given in Table 3. It can bee seen that the acceptance rate was highest on days with moderate weight increment (52.7%) and lower with the daily balance of food supply vs. consumption being zero. Grafts were unwillingly accepted on the days of marked weight decrease but the acceptance was poorest (24.2%) when grafting coincided with 0.8 kg of daily hive weight increment recognized as tantamount to high flow rate for the purpose of our study. It should be added that all nurse colonies were supplemented with sugar syrup that may have mitigated the adverse effect of natural flow. The statistical analysis showed that the variability was significant and the rearing effi-

Table 3

Queen rearing efficiency in colony vs. hive weight increment

Hive weight increment [kg]	Number of replications	Number of grafted larvae	Isolated queen cells	
			Number	%
Hive weight loss from -0,10 do -0,40 kg	54	2513	908	36.1 b
Unchanged (net increment = 0.0 kg)	13	795	340	42.8 bc
Moderate hive weight increment (+0.10 do +0.70 kg)	39	2091	1101	52.7 c
Large hive weight increment ≥ +0.80 kg	12	583	142	24.4 a
Total	118	5982	2491	41.6

Means followed by different letters are significantly different P<0.05

Table 4

Queen rearing efficiency in nurse colonies under the multiple grafting scheme in the years 1990 - 2001

Rearing series	Number of replications	Number of grafted larvae	Isolated queen cells	
			Number	%
1	139	7239	3358	46.4 a
2	119	5736	2630	45.9 a
3	104	5209	2471	47.4 a
4	76	5209	2471	45.7 a
5	52	2483	1245	50.1 a
6	44	2083	923	44.3 a
7	36	1582	685	43.3 a
8	25	1166	515	44.2 a
9	15	765	362	47.3 a
10	7	358	158	44.1 a
11	3	174	66	37.9 a
12	2	120	44	36.7 a
13	1	90	32	35.6 a
Total	623	30907	14272	46.2

Means followed by different letters are significantly different P<0.05

cacy averaged over the periods of good nectar flow was significantly lower than that for periods of slight weight increments or even for periods of marked food deficit. The regression equation for that relationship can be represented as a parabole and the formula for the expected value is as follows:

$$Y = 44.5667 - 22.7995 \times increment^2$$

Table 5

Queen rearing efficiency as affected by the positioning of grafted larvae in 2001

Graft positioning	Number of replications	Number of grafted larvae	Isolated queen cells	
			Number	%
Upper bar	56	841	432	51.4 a
Mid-position bar	56	886	461	52.0 a
Lower bar	53	828	385	46.5 a
Total	165	2555	1278	50.0

Means followed by different letters are significantly different $P<0.05$

Table 6

Percent loss on the preceding stage of queen larvae reared from grafting in a cell cup to queen emergence

Year	Grafted larvae	Accepted larvae		Capped queen cells		Emerged queen cells	
		Number	%	Number	%	Number	%
2000	1600	712	44.5	662	41.4	541	81.7
2001	1057	572	54.1	543	51.4	480	88.4
Total	2657	1284	48.3	2760	45.6	2388	86.5

Means followed by different letters are significantly different $P<0.05$

The nurse colonies were used repeatedly sometimes as many as 13 times. Table 4 shows how the repeated grafts affected rearing efficiency. High efficiency that came within 45-50% was maintained during the first five grafting series. Over the subsequent grafts it usually did not exceed 45% and, starting with the 10^{th} graft, it fell below 40%. However, the differences could not be validated statistically. Interestingly, the number of replications decreased with advancing rearing series. In occasional nurse colonies rearing was confined to one series only, the majority involved 4 - 5 series. The decision about the continued use of a nurse colony was based on rearing performance.

Another characteristic to be analyzed was rearing efficiency vs. the placement of grafted larvae on the grafting frame. Based on the years 2000-2001 observations it was found that the acceptance of grafted larvae varied only slightly with the site of attachment on the grafting frame (Table 5). The efficiency was highest for grafted larvae placed on the central bar lower for the upper bar and the lowest for the lower bar. As in the previous studies (Fell and Morse 1984, Visscher 1986) the differences were not significant.

In this study the rearing efficiency was assessed as the number of capped queen cells vs. the number of grafted larvae. An important issue to be resolved was the assessment of losses over the successive rearing stages. Observations to that effect were taken in some rearing the years 2000-2001. They showed that the majority of larvae (50%) perished at juvenile stage upon rejection of introduced cell cups (Table 6). Ultimately, an average of 38.4% of grafted larvae emerged as queens.

An attempt was made to evaluate the causes of death of larvae and pupae in capped queen cells. Of 3048 queen cells 311 died which accounts for 10.2% (Table 7). Most individuals died at the beginning of the pupa stage (items 5,6, and

Table 7

Death stage or cause of death of individuals in capped queen cells

No.	Status of dead queen cell	Number of non-emerged queen cells	Percentage of queen cells	
			Non-emerged n-311	Capped n-3048
1	empty queen cell (!!!)	2	0.6	0.1
2	spinning larva	5	1.6	0.2
3	stretched larva	50	16.1	1.6
4	pre-pupa	40	12.9	1.3
5	light-eyed light pupa	94	30.2	3.1
6	dark-eyed light pupa	8	2.6	0.3
7	light dark bodied pupa	15	4.8	0.5
8	dark pupa	27	8.7	0.9
9	pupa + greater wax moth (!!!)	1	0.3	0.1
10	pupae - 2 individuals in one queen cell (!!!)	3	1.0	0.1
11	winged queen	43	13.8	1.4
12	wingless queen	13	4.2	0.4
13	queen - wings with liquid !!!	1	0.3	0.1
14	queen positioned head uppermost (!!!)	1	0.3	0.1
15	queen + maternal pupa (!!!)	4	1.3	0.1
16	imago queen + imago worker bee (!!!)	1	0.3	0.1
17	unidentified stage	3	1.0	0.1
	total non-emerged queen cells	311	100.0	10.2
	total isolated queen cells	3048		100.0

7 of the table) accounting for one third of the total losses. Next in the order of importance came stretched out larvae, fully developed queens and dark pupae. Generally, dying events were most frequent immediately upon the successive transformations. More rare causes of death involved 8 cases of two individuals being present in one queen cell (2.6%) usually these were two queens at different stages of development but there was one case of a queen and a worker. Occasionally, death of the queen cell was caused by the positioning of the queen with the head uppermost or by the presence of a greater wax moth larva in a capped queen cell.

CONCLUSIONS

The efficiency of queen rearing varies over the years the time of rearing influencing that variability to the greatest extent.

Within the season queens are most readily reared in the second and in the third decade of May. As the season advances the readiness to rear queens declines and is at its lowest in mid-July. It rebounds again towards the end of July and in the beginning of August.

There was a slight beneficial effect on rearing success of maximum air temperature, temperature amplitude, and solar radiation expressed as J/cm^2. There was a slight adverse effect of high relative air humidity.

There is a well marked relationship between queen rearing efficiency and the nectar flow availability. Rearing efficiency

is most beneficially affected by moderate food availability, the absence of flow has a negative effect, the impact of abundant flow is the least beneficial.

Neither the site at which the larva is grafted in the hive nor the repeated use of a colony have any clear impact on rearing efficiency.

Multiple raising of queens in cell cups resulted in 50% of successful grafts and nearly 40% of emerged queens. After cell capping the greatest losses were at the beginning of the pupa stage.

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Journal of Apicultural Science

WPŁYW RÓŻNYCH CZYNNIKÓW NA EFEKTYWNOŚĆ WYCHOWU MATEK PSZCZELICH

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S t r e s z c z e n i e

Celem przeprowadzonych obserwacji było zbadanie wpływu wybranych czynników środowiskowych na efektywność wychowu matek pszczelich. Materiał badawczy z lat 1990-2001 obejmował prawie 31 tysięcy poddanych larw i ponad 14 tysięcy zaizolowanych mateczników, wychowanych w 623 powtórzeniach. Stwierdzono bardzo dużą zmienność w efektywności wychowu, mierzoną procentem mateczników zaizolowanych w stosunku do poddanych larw w kolejnych latach powodowaną głównie różną porą prowadzenia wychowu. Najwyższa efektywność była na początku sezonu w II dekadzie maja (55,8%) i wykazywała tendencję malejącą aż do II dekady lipca (25,8%), a następnie wzrastała w I dekadzie sierpnia (44,6%). Na efektywność wychowu ma niewielki ale istotny wpływ temperatura powietrza, promieniowanie słoneczne i wilgotność względna powietrza panująca w dniu poddawania larw. Nie ma wpływu miejsce poddania larw oraz wielokrotne wykorzystanie rodziny wychowującej. Najkorzystniej stymulował wychów umiarkowany wziątek (52,7%), natomiast zdecydowanie niekorzystnie bardzo dobry pożytek (24,4%) oraz okres bezpożytkowy (36,1%). W poszczególnych latach uzyskano od 33,8% do 45,4% matek wygryzionych w stosunku do liczby poddanych larw. Spośród nie wygryzionych 311 mateczników najwięcej zamarło w stadium jasnej poczwarki (32,8%) oraz stadium imago (20,2%). Sporadyczną przyczyną zamierania była natomiast obecność w jednym mateczniku 2 osobników zamarłych w różnych stadiach rozwojowych (2,6%).

Słowa kluczowe: wychów matek, pora sezonu, warunki pożytkowe, zamieranie mateczników, warunki atmosferyczne.