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Influence of Different Factors on Graft Acceptance in *Apis mellifera* Linnaeus colonies during Autumn in Punjab

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ABSTRACT: Three factors *viz.* bee strength/ crowdiness, queenliness of the colony and priming status of queen cell cups were evaluated on graft acceptance. The combined effect of the different levels of bee strength, queenliness and priming status of cell cups was non-significant on the mean number of accepted grafts (out of 90) (5.33-43.67 i.e. 5.92-48.52%).

Key words: *Apis mellifera*, graft acceptance, queen cell cups, royal jelly

INTRODUCTION

After achieving a breakthrough in honey production in the Punjab, currently the emphasis is on promoting and adoption of diversification in apiculture itself. According to an estimate, while migratory beekeeping ensures a gross profit of Rs 4,000 per annum, a well conceived diversification plan (including queen bee rearing and royal jelly production) would pay a profit of at least Rs. 7,000 (Makkar *et al*., 2010). Royal jelly has a high nutritional and pharmaceutical value and its demand is continually increasing in the world market. It is rich source of organic fatty acids, amino acids, minerals and vitamins (Krell, 1996). Some research has been conducted in abroad and also in India to evolve / standardize basic technology for royal jelly production from *Apis mellifera* Linnaeus colonies (Rana, 1996; Singh, 1997). Even the effect of various pollen substitutes on various colony parameters was studied and found helpful in hypopharyngeal glands development and this would be useful for royal jelly production (Chhuneja *et al*., 1993)

A scan through literature has revealed that the most of the work on royal jelly production pertained to standardizing only the basic parameters / requirements *e.g.* age of worker larvae for grafting, number of grafts to be used, position of grafts in queen rearing frame, priming of queen cell cups, royal jelly extraction time following larval grafting, *etc.* (Rana, 1996; Singh, 1997). The present studies were conducted to observe the impact of bee strength of colony/ crowdiness, queenliness (queenless v/s queen-right) and priming status of the queen cell cups on the absolute number of accepted grafts and per cent graft acceptance during the autumn season.

MATERIALS AND METHODS

Treatments: Three

The different levels of various treatments, evaluated for royal jelly production during the experiment have been given below:

T1: Bee strength and crowdiness in cell builder colonies

1. 10 bee-frames on 10 combs
2. 15 bee-frames on 15 combs
3. 20 bee-frames on 20 combs
4. 10 bee-frames on 8 combs
5. 15 bee-frames on 12 combs
6. 20 bee-frames on 16 combs

T2: Queenliness of cell builder colonies

1. Queenless
2. Queen-right

T3: Priming status of queen cell cups

1. Primed
2. Not primed

Modified Doolittle method as standardized by Singh (1997) was basically followed for the study. The above experiment was conducted using 90 grafts in plastic gyne cell cups without any artificial feeding to the experimental colonies.

Imported brown plastic queen cell cups were purchased from the local market for grafting.

Cell builder colony preparation

The comb arrangement in cell builder colony was H S S E Y C P E S where

H: Honey comb,
S: Sealed brood comb,
E: Comb with sealed brood near adult emergence,
Y: Brood comb with > 3 day old larvae,
P: Pollen comb, and
C: Grafted cell cups queen rearing frame

In the case of queen-right 15 and 20 bee-frame strength colonies, the queen cells raising frame(s) was/ were given in brood /lower chamber and the existing queen bee was restricted in the upper chamber with the use of horizontal queen excluder in between the two chambers.

Priming of queen cell cups

Dry grafting (without priming the cell cups with royal jelly) or wet grafting (after priming queen cell cups with speck of royal jelly) of larvae were evaluated.

Larval grafting

Young worker bee larvae of < 24 h age were transferred into queen cell cups.

Royal jelly extraction

Royal jelly was extracted 72 h after larval grafting.

Statistical analysis

Data were statistically analysed using Factorial Completely Randomized Design for determining the significance of differences of various levels of the treatment means and the means of combinations (interactions) among the various levels of different treatments.

RESULTS AND DISCUSSION

Absolute number of accepted grafts: Bee strength of 15/15 in the colonies resulted in the highest mean number of grafts accepted (23.92) cell cups and it was on par with all the other bee strengths except 8/10 bee-frame strength (11.00). Other bee strengths showed acceptance of 23.50 (10/10 bee-frames), 22.08 (16/20 bee frames), 20.58 (12/15 bee-frames) and 20.17 (20/20 bee frames) grafted cell cups (Table 1). The effect of queenliness on the number of the accepted grafts was significant. Queen-right colonies showed higher number of accepted grafts (23.42) than queenless colonies (16.50). The mean number of accepted cells in colonies provided primed cell cups was significantly higher (26.00) than those provided unprimed cell cups (14.42).

The interaction between bee strength and queenliness of the cell builder colonies proved to be non significant, with the mean number of accepted grafts ranging between 8.50-27.50. The various combination interactions between queenliness and priming status of cell cups, however proved to be non-significant (11.89-30.89 grafts).

Bee strength of 16/20 frames coupled with priming of cell cups resulted into significantly higher graft acceptance (33.00) which was followed by 12/15 bee-frame strength under primed status of cell cups (30.17). The other combinations resulted in 10.33 (8/10 bee-frame colonies with primed cell cups) to 28.83 (10/10 bee-frame colonies provided with primed cell cups) accepted grafts. The interaction among the different levels of three treatments proved to be non significant (5.33-43.67 grafts).

Per cent graft acceptance: Bee strength of 15/15 frames resulted in the highest per cent acceptance (26.57%) which was on par with 10/10 bee-frame strength (26.11%), 16/20 bee-frame strength (24.53 %), 20/20 bee-frame strength (24.40 %) and 12/15 bee-frame strength (22.87%) colonies. The least per cent graft acceptance was recorded in 8/10 bee-frame strength colonies (11.39%) (Table 2).

The queenliness of cell-builder colonies showed significant effect on the per cent acceptance of grafted cell cups. Between the two conditions (queenless and queen-right), it was higher in queen-right colonies (26.57%) than in queenless colonies (18.33%). Priming of cell cups proved to be better (28.89%) than no priming (16.01%) in this respect. Interaction among the combinations of bee strength and queenliness of cell builder colonies was non-significant w.r.t. per cent graft acceptance (9.44-30.56). The combined effect of the given levels of queenliness and priming status of cell cups was non-significant on the per cent acceptance of larval grafts (13.21-34.32). The combined effect of different levels of bee strength and priming status of cell cups was significant with maximum percent acceptance in 16/20 bee-frame strength colonies with primed cell cups (36.66%) which was on par with colonies with primed cell cups with bee-strength of 12/15 bee-frame (33.52%), 10/10 bee-frame (32.03%), 15/15 bee-frame (30.92%), 20/20 bee-frame (28.70%). This was followed by 15/15 bee-frame strength colonies with non-primed cell cups (22.22%) which was on par with other colonies with non-primed cell cups and bee strength of 10/10 bee-frame (20.18%), 20/20 bee-frame (16.11%), 16/20 bee-frame (12.41%), 8/10 bee-frame (12.96%), 12/15 bee-frame (12.22%) and least acceptance was observed in 8/10 bee-frame strength colonies with primed cell cups (11.48%). Interaction among all the three combinations was non-significant in this respect (5.92- 48.52%).

The above results *w.r.t.* the effect of bee strengths on graft acceptance for royal jelly production are in conformity with Aulakh *et al* (2002) who have also reported that graft acceptance by using 24 h old larvae in 15-20 bee frame strength *A. mellifera* colonies was significantly higher than 10 bee-frame strength colonies. Considering the effect of queenliness, queen-right condition proved to be better than queenless condition and our results are in conformity with Adam (1975) who reported 80 per cent acceptance of grafts using queen-right cell finisher colonies. Szabo (1987) recommended the use of queenless starter colonies and queen-right cell finisher colonies using 15-16 beeswax cell cups per bar on three bar queen cell raising frame (total 45-48 cell cups/ colony) under Californian conditions. Webster (1988) also recommended the use of queen-right cell builder colonies by restricting the queen under queen excluder and keeping the grafts above the excluder until the 10^{th} day of grafting when the sealed queen cells were transplanted in the mating nuclei. The results of the effect of priming status of cell builder colonies are in conformity with those of Macicka (1985) who reported that the mean acceptance of larvae grafted with cell cup priming was 75.6 per cent in comparison with 41.3 per cent without priming with royal jelly. The similar kinds of observations were recorded by Pickard and Kitner (1983) and Morton (1992).

Table 1. Effect of bee strength and queenliness of cell builder *A. mellifera* colony on the number of grafts accepted under primed vs dry grafting condition during autumn.

Bee strength (No. of combs / No. of bee-frames)	Mean number of accepted grafts per colony*								
	Queen-right colony			Queenless colony			Mean		Grand mean
	Primed cell cups	No priming	Mean	Primed cell cups	No priming	Mean	Primed cell cups	No priming	
8/10	14.33	12.67	**13.50**	6.33	10.67	**8.50**	10.33	11.67	**11.00**
	(3.90)	(3.69)	**(3.79)**	(2.58)	(2.98)	**(3.33)**	(3.24)	(2.78)	**(3.29)**
10/10	31.67	18.33	**25.00**	26.00	18.00	**22.00**	28.83	18.17	**23.50**
	(5.70)	(4.39)	**(5.04)**	(5.13)	(4.14)	**(4.26)**	(5.41)	(4.63)	**(4.84)**
12/15	43.67	11.33	**27.50**	16.67	10.67	**13.67**	30.17	11.00	**20.58**
	(6.66)	(3.46)	**(5.06)**	(4.18)	(3.33)	**(3.39)**	(5.42)	(3.75)	**(4.41)**
15/15	32.33	21.33	**26.83**	23.33	18.67	**21.00**	27.83	20.00	**23.92**
	(5.72)	(4.65)	**(5.18)**	(4.92)	(4.42)	**(4.53)**	(5.32)	(4.67)	**(4.93)**
16/20	34.00	17.00	**25.50**	32.00	5.33	**18.67**	33.00	11.17	**22.08**
	(5.90)	(3.76)	**(4.83)**	(5.74)	(2.28)	**(3.02)**	(5.82)	(4.01)	**(4.42)**
20/20	29.33	21.00	**25.17**	22.33	8.00	**15.17**	25.83	14.50	**20.17**
	(5.50)	(4.66)	**(5.08)**	(4.66)	(2.72)	**(3.69)**	(5.08)	(3.69)	**(4.39)**
Mean	**30.89**	**16.94**	**23.92**	**21.11**	**11.89**	**16.50**	**26.00**	**14.42**	**20.21**
	(5.56)	**(4.10)**	**(4.83)**	**(4.53)**	**(3.31)**	**(3.92)**	**(5.05)**	**(3.71)**	**(4.38)**

* Figures in parentheses are the means of √n+1 transformations

LSD (p = 0.05) for : Bee strength (A) = (0.88), Queenliness (B) = (0.51), Priming vs no priming (C) = (0.51), A x B = (NS), B x C = (NS), C x A = (1.25)
A x B x C = (NS)

Table 2. Effect of bee strength and queenliness of cell builder *A. mellifera* colony on graft acceptance under primed vs dry grafting condition during autumn.

Bee strength (No. of combs / No. of bee-frames)	Mean per cent graft acceptance*								
	Queen-right colony			Queenless colony			Mean		Grand mea
	Primed cell cups	No priming	Mean	Primed cell cups	No priming	Mean	Primed cell cups	No priming	
8/10	15.92	14.07	**14.50**	7.03	11.85	**9.44**	11.48	12.96	**12.22**
	(23.43)	(21.96)	**(22.69)**	(14.28)	(15.99)	**(15.13)**	(18.85)	(18.97)	**(18.91)**
10/10	35.18	20.37	**27.77**	28.88	20.00	**24.44**	32.03	20.18	**26.11**
	(36.29)	(26.77)	**(31.53)**	(32.18)	(25.22)	**(28.70)**	(34.24)	(26.00)	**(30.12)**
12/15	48.52	12.59	**30.56**	18.52	11.85	**15.18**	33.52	12.22	**22.87**
	(44.11)	(20.45)	**(32.28)**	(25.36)	(19.52)	**(22.44)**	(34.73)	(19.99)	**(27.36)**
15/15	35.92	23.70	**29.81**	35.92	20.74	**23.30**	30.92	22.22	**26.57**
	(36.59)	(28.71)	**(32.65)**	(30.54)	(27.00)	**(28.77)**	(33.56)	(27.85)	**(30.71)**
16/20	37.77	18.89	**28.33**	35.55	5.92	**20.74**	36.66	12.41	**24.53**
	(37.85)	(21.41)	**(29.63)**	(36.57)	(11.19)	**(23.88)**	(37.21)	(16.30)	**(26.75)**
20/20	32.59	23.33	**27.96**	24.81	8.89	**16.85**	28.70	16.11	**22.40**
	(34.76)	(28.73)	**(31.75)**	(28.97)	(14.17)	**(21.57)**	(31.86)	(21.45)	**(26.66)**
Mean	**34.32**	**18.82**	**26.57**	**23.45**	**13.21**	**18.33**	**28.89**	**16.01**	**22.45**
	(35.50)	**(24.67)**	**(30.09)**	**(27.98)**	**(18.85)**	**(23.41)**	**(31.74)**	**(21.76)**	**(26.75)**

* Figures in parentheses are the means of arc sine √ percentage transformations

LSD (p = 0.05) for: Bee strength (A) = (7.30), Queenliness (B) = (4.22), Priming vs no priming (C) = (4.22), A x B = (NS), B x C = (NS), C x A = (10.32), A x B x C = (NS)

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