The use of brood pheromones and feeding substitutes in the stimulation of honey bee *Apis mellifera* L. (Hymenoptera: Apidae)

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Abstract— The study was conducted at the Apiary site of the College of Agriculture University of Baghdad to evaluate the effect of brood pheromone (Superboost) and feeding substitutes on stimulation of colony growth brood and honey area, building wax foundation and quantity of pollen collected. Results showed that was a significant difference on the amount of change in brood area between treatments. The synthetic pollen patty was given the highest brood area of 234.142 inch², followed by treatment of superboost brood pheromone, synthetic pollen patty+ superboost brood pheromone, artificial mesquite patty+ superboost brood pheromone and mesquite patty treatment which reached 192.857, 179.571, 169 and 114.714 inch² respectively, compared with 77.285 inch² for the control treatment. Results of the speed of building wax foundation were showed no significant differences within 24 hour for all the treatments. Within 120 hour The highest average of 272 inch² was recorded on colonies treated with superboost pheromone only and synthetic pollen patty+ superboost brood pheromone, followed by 180, 150, 120 inch² for the treatment of artificial pollen patty, superboost + mesquite patty and mesquite patty. The highest pollen weight of 41.1 gm was observed on colonies treated with super boost only compared with 10.454 , 9.172 and 1.2 gm for other treatments of synthetic pollen patty+ superboost, pollen patty and control treatments. The results of using superboost brood pheromone with locally feeding substitutes for increasing the pollination activating and efficiency of honey bee colonies also discussed.

Keyword— Brood pheromones, honey bees, *Apis mellifera*, Superboost, feeding substitutes.

INTRODUCTION

Brood pheromone is produced in salivary glands of the honey bee (*Apis mellifera* L.) Larvae. It tells nurse bee workers that larvae are present and require food. Beekeepers may provide a pollen /protein supplement to honey bee colonies during the shortage of pollen resources to stimulate colony growth (Nabors, 2000, safari et al., 2004 and van der Steen, 2007). The protein content of pollen is considered as a feeding index and it has effect on performance and age of the honey bee colony (Nicolson , 2011) without pollen presence the quantity of royal jelly required for the production small larvae to feed the queen is reduced(Fujita et al., 2012).

Adding brood pheromone to honey bee colonies may increases the number of pollen foragers by 150% significantly increased the quantity of pollen collected and increase colony growth rate (Pankiw, 2004 and Pankiw et al., 1998). A commercial formulations such as superboost placed inside the colony will stimulate the presence of more brood, which stimulate the collecting of more pollen by foragers and visit more pollen resources (Best management practices for pollination in Ontariocrops. www.pollinator.ca/canpolin/pheromones.html.

Little or nothing studies has been reported on the use of brood pheromone in Iraq to stimulate honey bee colonies during the very hot summer and cold winter which honey bee worker spending only few hours during the day in foraging activities. Honey bee keepers also suffer from the shortage of resources available for their colonies. Here, we tested the effects of addition of Brood pheromone and some locally prepared feeding alternatives on the growth and stimulation of honey bee colonies.

II. Material and methods

2.1 Colony preparation

Eighteen local colonies of beehives with naturally mated queen in spring 2016 were used in this experiment. These colonies were transferred into a wooden hives and received the same required maintains till the beginning of the experiment in Feb 21, 2017. Each treatment was comprised of 3 colonies (3 replicates) and was randomly distributed around the apiary field. There were 6 treatments as follows:
1. Treatment 1 (T1): feeding with mesquite pie.
2. Treatment 2 (T2): feeding with synthetic pollen pie.
3. Treatment 3 (T3): feeding super brood pheromone twice during the season.
4. Treatment 4 (T4): feeding with mesquite pie and super brood pheromone.
5. Treatment 5 (T5): feeding with synthetic pollen pie and super brood pheromone.
6. Treatment 6 (T6): feeding naturally (control treatment).

## 2.2 Pie Preparation

The synthetic feed pies were prepared by grinding a 150 gm dried pollen in house mortar and mixed with 75 gm of grinded sugar. A 50% sugar solution was prepared and mixed with 25 gm of Peking powder, the mixing was kneaded. Fifty grams of these pie per/ colony were put in a special paper and fixed on bee hive frames. Feeding with these synthetic Feed bees was done weekly.

### 2.2.3: Mesquite Preparation

The mesquite cumin was collected from Abu-Graib region during June, and kept in the laboratory till get dried and then grinded and sieved. A 100gm sample was weighted, and added to 50 gm of grinded sugar. A 100%-sugar solution was prepared and mixed with 50gm of Peking powder and kneaded, after kneading completion, 50 gm of this mesquite were put on a special paper and fixed on the honey bee frame for every colony. Mesquite pie feeding was done regulary at 7 days intervals. Honey bees

### 2.2.4: Super Boost Pheromones

Super brood pheromones of the Canadian company were brought from the agent company in the north of Iraq and put in a freezer till it used in this experiment. One slide of the super boost pheromone for every colony replicate were used and replaced every 30 days according to the company instruction.

## 2.3 Effect of Super Boost Pheromone on Brood and Honey Area

Brood and honey area before and after the experiment was measured by using the standard fram count every 12 days from the first feeding to the end of the experiment.

## 2.4 Effect of Super Boost on Waxy Base Construction

Fram of the honey bee hive counted by putting the frame of honey bee hive on a standered honey bee hive and measuring its area according to the squares number (square inches) for both frame sides after and before the experiment every12 days from first feeding to end of the experiment.

## 3- Rate of Waxy Base Construction

Waxy base was put for each replicate and area of performance was measured every 24 hour by fram count for 5 days.

### 4- Use of Pollen Traps

Traps were put in hives door for each treatment of the experiment treatments for 24 hours per week, then the collected pollen by the honey bees were weighted out to stimulate the range of feeding and pheromone effect in increasing honey workers bees activity in collecting plant pollen.

### Statistical Analysis

Data were statistically analyzed using Sass program (SAS, 2012) the differences between treatments means according to CRD design compared using L.S.D \( P=0.05 \)

## III. RESULT AND DISCUSSION

### 1- Brood Area

The results of table (1) showed superiority of (T3) increasing the brood area to 234.142 inch², followed by (T5) (T4),(T3) and (T1) compared with(T6) treatments, the brood areas were 192.857 , 179.571, 169114.714 and 77.285 inch² respectively.

The time readings of broods area increase (as it is shown in table (1) was positively correlated with broods area, and the boost area average was 34.166 inch² in the beginning of the experiment (21/2/2017) and the highest increase average was 268.833 inch² at 3/5/2017. It may be concluded that addition of broods pheromone to honey bee hives in spring and summer encourage the nurse bee workers in scouting and transferring bee workers at early stages and this increased the number of pollen foragers and pollen load returned to the hive (Pankiw, 2007).

Increase of broods area in pollen pies treatment may be to the structure of the synthetic pollen pie, pecking powder an sugar which provided the nutrients requirement to broods built in their high proteins, sugars , fats , vitamins and minerals contents and these contents helped in pharyngeal glands development which play important role in broods feeding besides its high content of saccharides which is considered as feeding stimulant , as increase saccharides ratio in the nutrient materials may be more attractive to bees because bees don’t feed on food having less than 20% sugar . Al- Hjemey (2009) reported that shrush treatment which consists of 50% shrush and 50ml of 60% sugar solution increased the numbers of pollen foragers from 366 to 667 per day.
Table 1: Effect of superboost brood pheromone and feeding substitutes on bees colonies and the brood area.

| Sampling date | 1/4 | 5/3 | 3/17 | 3/29 | 4/10 | 4/21 | 5/3 | Means |
|---------------|-----|-----|------|------|------|------|-----|-------|
| T₁            | 43  | 63  | 90   | 120  | 142  | 155  | 190 | 114.714 |
| T₂            | 37  | 168 | 230  | 261  | 282  | 311  | 350 | 234.142 |
| T₃            | 35  | 82  | 136  | 157  | 249  | 254  | 270 | 169     |
| T₄            | 34  | 107 | 123  | 177  | 226  | 273  | 317 | 179.571 |
| T₅            | 31  | 90  | 129  | 197  | 221  | 318  | 364 | 192.857 |
| T₆            | 25  | 39  | 73   | 79   | 94   | 109  | 122 | 77.285  |
| Means         | 34.166 | 91.5 | 130.166 | 165.166 | 202.333 | 236.666 | 268.833 | ---         |

L.S.D Treatment : 14.74’ Duration: 16.46’ Interference: 28.53’

2- Honey area

Results in table 2 show significant differences in honey area between treatments. Values of honey area were 41 and 40.857 inch² for (T₅) and (T₄) respectively compared with 24.142 , 22 , 19.857 and 16.857 inch² for the treatments T₂, T₆, T₃ and T₁ respectively. The range of honey area at the beginning of the experiment were between (107-128) inch² and its began to decrease gradually may due the shortage of flowering plant pollen in the foraging environment, which negatively affect the amount of honey collected in all treatments (Pankiw et al. 2008). Daily changes in the environmental conditions such as temperature, relative humidity and speed of wind may affect the activity of the pollen foragers.

Table 2: Effect of superboost brood pheromone and feeding substitutes on bees colonies and the honey area

| Treatments | Sampling date |
|------------|---------------|
|            | 21/2 | 3/3 | 17/3 | 3/29 | 10/4 | 21/4 | 3/5 | Mean |
| T₁         | 107  | 9   | 2    | 0    | 0    | 0    | 0   | 16.857 |
| T₂         | 128  | 33  | 8    | 0    | 0    | 0    | 0   | 24.142 |
| T₃         | 118  | 18  | 3    | 0    | 0    | 0    | 0   | 19.857 |
| T₄         | 111  | 104 | 47   | 17   | 7    | 0    | 0   | 40.857 |
| T₅         | 112  | 122 | 39   | 14   | 0    | 0    | 0   | 41    |
| T₆         | 110  | 39  | 5    | 0    | 0    | 0    | 0   | 22    |
| Mean       | 114.333 | 54.166 | 17.333 | 5.166 | 1.66 | 0.00 | 0.00 | ---- |

L.S.D Treatment: 5.36’ Duration: 5.77’ Interference: 9.69’.

Wax base construction rate

The results in Fig. 1 indicated that there was no significant difference between the treatments after 24 hours. However, The treatments of the (T₅) and (T₂) show a superiority over all other treatments were wax base construction area were 272 inch² for each of them, followed by (T₆), (T₁), (T₃) and (T₄) in which the value of wax base were 180, 150, 120 and 74.5 respectively. These results indicated that the increase or decrease of the wax base area varies with season, bee hive activity, numbers of foragers in the hive and the availability of alternative pollen resource.
Fig.1: Effect of superboost brood pheromone and feeding substitutes on the speed of building wax foundation of honey bee colonies

Weight of collected Pollen

The weights of pollen collected by the scout bees in traps are presented in table 6. The highest pollen weight were in (T5) of 14.1 gm which significantly different from all other treatments, followed by 10.45, 9.17 for (T4) and (T2) respectively. The least pollen weight of 3.5 gm was collected in (T1) which is not significantly different from 2.1 gm in (T6). our result agree with that of pankiw (2004) who stated that the addition of brood pheromone to honey bee colonies in spring and summer change in the job of nurse bee to scout bee worker in early age and this encourage bee scouting to collect more pollen at time unite also, the pollen mass transferred in to the hives increased, this result increase of 150% in stocked pollen in the growth of bee hives.

Table (3) Average pollen weight collected by forager’s bees in pollen traps.

| Treatments | Sampling date | Mean |
|------------|---------------|------|
|            | 21/2 | 28/2 | 7/3 | 14/3 | 21/3 | 28/3 | 4/4 | 11/4 | 18/4 | 25/4 | 2/5 |
| T1         | 0    | 0.5  | 4.3 | 4.3  | 6    | 4.2  | 1.7 | 4.8  | 1.7  | 7.2  | 3.8  | 3.5 |
| T2         | 0    | 0.7  | 4.5 | 6.2  | 10.4 | 8    | 10.5| 18   | 12.6 | 5.8  | 24.2 | 9.172|
| T3         | 0    | 0.7  | 3.2 | 2    | 5    | 5.3  | 6.5 | 6.7  | 9.3  | 13.4 | 4.8  |
| T4         | 0    | 2.5  | 5   | 7    | 10   | 9    | 8.9 | 10.2 | 16.5 | 19.4 | 26.5 | 10.454|
| T5         | 0    | 2.2  | 3.5 | 4.6  | 8.6  | 10.4 | 16.7| 21   | 25.5 | 30.1 | 32.5 | 14.1 |
| T6         | 0    | 0.4  | 0.8 | 1.7  | 2    | 6    | 1.6 | 2.2  | 2    | 2.4  | 4    | 2.1  |
| Mean       | 0.00 | 1.166| 3.13| 4.5  | 6.5  | 7.1  | 7.45| 10.45| 10.833| 12.366| 17.4| ---  |

L.S.DTreatment: 2.67” Duration: 3.98”Interference: 2.67”
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