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EFFECT OF SOME SUPPLEMENTARY FEEDING ON PHYSIOLOGICAL CHARACTERS OF HONEYBEE WORKERS

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Abstract: The investigations were carried out under laboratory conditions on small groups of caged honeybee to evaluate the suitability of five proteinic diets for bee feeding. This experiments was designed with two trails; in the first trail, the food consumption and mortality of newly emerged bees under feeding with different diets were determined. The highest food consumption was recorded when bees fed on date palm pollen (diet 2) followed by bee bread (diet 1) and the lowest one was recorded for Haydak's diet (diet 4) feeding bees on bee bread gave the longest LT_{50}. On the other hand, the honeybee workers fed on date palm pollen and Haydak's diet were gave LT_{50}, 18.16 and 19.52 days, respectively. In the second trail, the development of hypopharyngeal glands and rectal contents were evaluated. The results indicated that the normal source of protein for honeybee workers as bee bread or date palm pollen were the best sources for hypopharyngeal gland development (3.83, 3.41 degrees) respectively. Meanwhile, the glands of worker feeding on a mixture of protenic materials (diet 5) was 2.86 degrees. This results indicated that, the addition of pollen to honeybee diet activated the hypopharyngeal glands development. The results of honeybee rectal contents weight reflected the suitability of diets which contain pollen grains for feeding honeybee colonies.

Key words: feeding, physiological, honeybee workers.

Introduction

The basic food for honeybee is represented by honey as energetic source and pollen grains that rich in protein, vitamins, enzymes, mineral salts, lipids, etc., which necessary for the growth, development and activity of honeybees. Shortage of pollen results in decreasing of brood rearing, developmental abnormalities, decreased length of workers life and poor honey production (Haydak, 1970; Kleinschmidt and Kondos, 1978; Doull, 1980 and Winston *et al.*, 1983).

Since pollen is often not present in adequate quantities in the field, the beekeepers used to feed bees with pollen supplements or substitutes to promote colony development and health. In Egypt, several beekeepers use to feed their bee colonies on sugar syrups during late autumn and early

spring to accelerate brood rearing. This feed stimulates the oviposition (Doull, 1975) but it has usually a limited effect due to the to lack of pollen. Pollen grains of palm date which collected by hand were used effectively for feeding of honeybee colonies in Assiut area (Hussein, 1981). Omar (1989) suggested that the probability of proteinious feeding during autumn is essential to increase the ability of brood rearing and to prepare the bees for overwintering. Moreover, obtaining high population of bees in the spring is of a particular interest to beekeepers.

The useful pollen substitute should stimulate colony growth and support aspects of worker quality, such as high brood survival and long adult life (Winston *et al.*, 1983). Meanwhile, the pollen substitute should be acceptable and has the necessary stimulation for bee to consume their food (Doull, 1973).

This investigation aimed to study the effect of some proteinic material by determing the food consumption, longevity, development of hypopharyngeal glands and intestinal contents. Such information obtained from these studies may be lead to develop a suitable substitute recommended to improve the development of honeybee colonies.

Materials and Methods

The experimental work was carried out in laboratory of Plant Protection Department, Faculty of Agriculture, Assiut University. First hybrid of Carniolan honeybee *Apis mellifera* L. was used in the present investigations.

Nutritive material used:

The materials used in feeding bees in the experiments were bee bread, date palm pollen and some selected materials.

– Bee bread was collected from combs during active season of honeybee colonies (representing the control) and stored under freezing condition until used.

– Date palm pollen (*Phoenix dactylifera* L.) collected directly by hand from male date palm trees in March and kept under freezing condition until used.

– Four materials that are available in local market, were chosen for investigation as pollen substitutes. These material were:

- Defatted soybean meal (*Glycine max* (L.) Merr.).
- Brewer's dried yeast.
- Skimed milk powder
- Corn flour (*Zea mays*).

Variant of proteinic diets tested:

Different five tested diets were compared together against the control as been explained in Table (1).

Table (1): Description of diets administrated to honeybee workers.

Variants	Bee bread	Date palm pollen	Corn flour	Soybean meal	Dried skimed milk	Brewers yeast	Sugar
Diet 1 (bee bread)	100	-	-	-	-	-	-
Diet 2 (date palm pollen)	-	50	-	-	-	-	50
Diet 3 (corn flour)	-	-	50	-	-	-	50
Diet 4 (Haydak diet)	-	-	-	30	10	10	50
Diet 5 (mixture diet)	-	10	10	10	10	10	50
Control	Feed with sugar syrup only						

The present study was carried out in experimental cages using newly emerged honeybee workers obtained from one colony to prevent any genetic variations as possible and sampled in summer period (June-July, 2004). Sealed brood combs, free from bee bread, were placed in an incubator at 32±1°C, 65±5 R.H in screen cages to obtain newly emerged bees (0-12 hours).

Experimental cages used:

Experimental wooden cage of 15x15x5 cm dimensions with a glass side and other was covered with black muslin. Every cage was provided with a vial of tap water and other vial of sugar solution 1:1 (w/v) and a pieces of wax comb were attached to the top of cage Four cages (each contains fifty workers) were used for every tested material.

The paste-like diets from each variant mentioned above were introduced for each cage, into a small plastic feeder (1 cm height and 3 cm diameter) covered with a small scheets of polyethylene which are pastorate to avoid water evaporation and any loss which could occurred at free access of honeybees to the whole quantity of food (sticking on the legs, wings, the small hairs on the body, etc.). Each feeder contains an average amount of 5 gm proteinic food. Stock diets are kept into the refrigerator at 4°C, until administration. The diets were changed in each cage every 3 days. All cages were held in the dark in an incubator at 32±1°C and 65+5% RH.

The experiments was carried out under laboratory conditions in two trials:

- **In the first trial, the following parameters were measured**:

- Daily food consumption of nurse bees.
- Honeybee workers longevity.

- **In the second trial, the following parameters were measured**:

- Development of hypopharyngeal glands.
- Rectal contents weight.

Measurements:

a. Food consumption:

Food consumption was calculated every 3 days and represented as mg/bee/3 days. The amount of food/cage was compared to the number of live bees existing in each *cage during the investigation period.*

b. Longevity:

Dead bees in each cage were counted and removed every 3 days interval until half of the initial number of honeybees were dead. The LT_{50} (the time required to reach 50% mortality) was estimated. Values in days of bees fed with different diets were determined by a computerized probit analysis program

c.Determination of the hypopharyngeal gland development:

The test was carried out on honeybee workers of 3, 6, 9, and 18 days-old to determine the development of the hypopharyngeal glands of each age. Twenty bees were used to assimilate each age from each treatment. The degree of development of hypopharyngeal glands was determined according to Maurizio (1954). An arbitary scale (I to IV) was used to determine the degree of development; grade 1, represented undeveloped gland and grade IV, represented complete development.

d. Weight of rectal content:

The same twenty bees which were selected for determining the development of hypopharyngeal glands were used to determine the weight of rectal content by extracted *the rectum with a fine forceps, put it on a cover glass, previously weighed,* and then reweighed on an analytical balance.

The F-test was used in order to calculate and test the significance of the administrated diets. The multiple scale of Duncan (1955) was used in order to compared the means.

Results and Discussion

The first trail:

The first trail was carried out to test the food consumption and mortality rate of honeybee workers.

Table (2) and Fig. (1) illustrated the food consumption (mg/bee/ 3day) of newly emerged bees from different four diets against the control which fed on bee bread. Honeybee workers started to consume a considerable amount at the first period after emergence (1-3 days) then increased their feeding amount to reach a peak of consumption at (4-6 days) which differed significantly over all the compared periods. The consumed diets decreased gradually from the third period. Similar results were also

obtained by Pain (1961) and Jaycox (1981) who reported that the consumption peak of pollen obtained at 3-5 days of worker age and then decreased to two level by time.

The total food consumed by the workers during the twelve days after emergence was calculated for each tested diet. The deviation in the total consumption of each diet from that of bee bread (control) was estimated (Table 2). It was found that the highest food consumption was recorded when the workers were fed on date palm pollen diet (40.39 mg/bee/ 12 days) followed by that consumed from the bee bread (38.89 mg/bee/12 days). The two mentioned treatments have a same significant level. The other compared diets were significantly different. The lowest consumption was recorded for Haydak's diet (diet 4) which differed significantly from any compared diet.

The results indicate that the type of basic material used in pollen substitute greatly affected the consumption of the foods. Kleinschmidt and Kondos (1977), Herbert and Shimanuki (1980), Mostafa (2000) stated that consumption of food is clearly stimulated by adding the pollen to the mixed diets for honeybee.

Table(2): Food consumption by honeybee workers from used proteinic diets.

Tested / Diets	Mean of consumption (mg/bee/3 day)				Total
	1-3 days	4-6 days	7-9 days	10-12 days	mg/bee/ 12 days
Diet 1 (bee bread)	11.63 b	12.88 b	9.38 de	5.00 h	38.89 A
Diet 2 (date palm pollen)	9.88 cd	16.88 a	7.63 fg	6.00 gh	40.39 A
Diet 3 (corn flour)	8.00 ef	11.13 bc	6.25 fgh	5.88 gh	31.26 B
Diet 4 (Haydak's diet)	7.13 fg	11.25 bc	3.13 i	1.75 i	23.26 C
Diet 5 (mixture diet)	7.25 fg	11.75 b	6.63 fgh	5.88 gh	31.51
Mean	8.78 (B)	12.78 (A)	6.60 (C)	4.90 (C)	

Means followed by the same letter do not differ significantly at the 5% level of probability.

It can be concluded that nurse workers prefer to fed on diet supplied with pollen.

The cumulative mortality percentages as well as the longevity of worker's at each treatment were

recorded. The half life was estimated LT_{50} (number of days required for 50% of the bee to die). The mortality percentages and LT_{50} were summarized in Table (3) and Figure (2). Data showing that feeding caged bees on sugar syrup only shortened the longevity of honeybee workers (LT_{50}, 15.73 days) in comparison with those fed on bee bread (LT_{50}, 20.79 days) while those fed on date palm pollen (diet 2) their LT_{50} extended to 18.16 days. The results also indicated that the life length of honeybee workers fed on Haydak's diet (diet 4) have the longest record for LT_{50} among the compared diets (19.52 days). Diet 3 which contains corn flour only gave a poor result as that of sugar syrup (15.66 day, 15.73 days, respectively).

Table (3): Cumulative mortality percentages of honeybee workers fed on some proteinic diets.

Proteinic diets / Age of Bees (days)	Diet 1 (bee bread)	Diet 2 (date palm pollen)	Diet 3 (corn flour)	Diet 4 (Haydak's diet)	Diet 5 (mixture diet)	Control (Sugar syrup)
3	0.0	6.0	3.0	7.5	7.0	2.5
6	7.5	10.0	8.5	14.0	13.5	9.0
9	11.0	14.5	14.5	23.0	20.5	16.0
12	25.5	22.5	29.5	30.0	29.0	32.0
15	36.5	37.5	54.5	35.0	38.0	49.0
18	44.5	48.5	70.0	41.0	52.0	64.5
21	52.5	59.5	79.5	50.5	64.5	76.5
24	62.0	76.5	86.5	61.5	79.5	86.5
27	68.5	82.5	91.5	70.5	91.0	95.0
30	74.5	90.5	95.0	82.0	97.0	-
33	83.5	98.0	-	90.5	-	-
36	91.5	-	-	95.0	-	-
39	99.5	-	-	-	-	-
LT_{50} days	20.79 A	18.16 BC	15.66 D	16.88 CD	19.52 AB	15.73 D

Means followed by the same letter do not differ significantly at the 5% level of probability.

The above results indicated that protein source played an important role influencing the longevity of honeybee workers.

The second trail:

The degree of the hypopharyngeal glands development in newly emerged workers which fed on proteinic diets as well as bee bread were presented in Table (4) and illustrated in Figure (2). First of all the results indicate that the age of bees in which the development of hypopharyngeal glands could be influenced occurred during the 12

Fig. (1): Variations of food consumption by honey bee workers fed on proteinic diets.

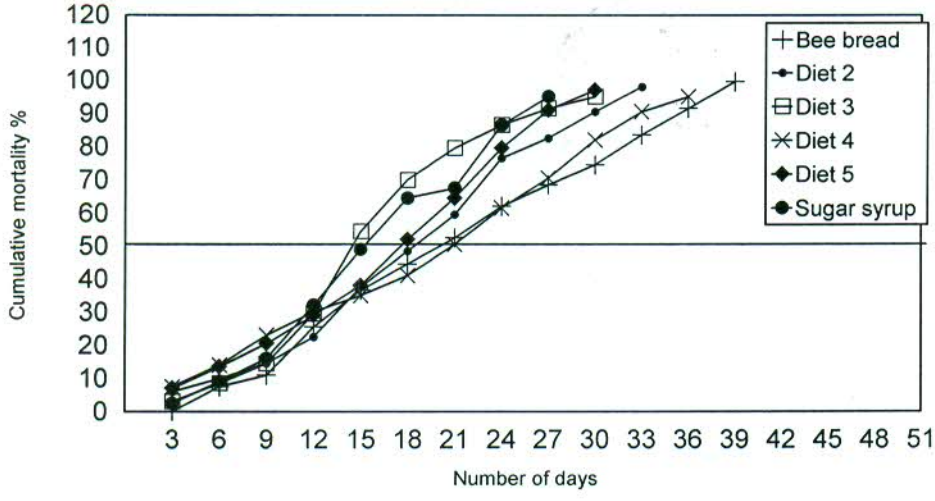

Fig.(2): Cumulative mortality percentage under feeding on proteinic dites.

days old. The considering increment in the gland development has been obtained during this period. This results were in agreement with those obtained by Khodairy (1990).

Table (4): Development of hypopharyngeal glands in honeybee workers fed on proteinic diets.

Tested / Diets	Degree of hypopharyngeal gland development					Mean
	3 days	6 days	9 days	12 days	15 days	
Diet 1 (bee bread)	3.66 abc	3.87 a	3.69 abc	4.00 a	4.00 a	3.83 A
Diet 2 (date palm pollen)	2.40 cdefg	3.26 abcd	3.65 abc	3.95 a	3.81 ab	3.41 A
Diet 3 (corn flour)	3.61 abc	2.74 abcde	2.91 abcde	2.35 cdefg	2.15 defg	2.75 B
Diet 4 (Haydak's diet)	2.45 bcdefg	2.82 abcde	1.29 fgh	1.20 gh	1.35 fgh	1.82 C
Diet 5 (mixture diet)	3.05 abcde	3.26 abcd	2.64 abcdef	3.26 abcd	2.10 defg	2.86 B
Control (sugar syrup)	1.77 efg	2.80 abcde	0.42 h	1.99 defg	2.12 defg	1.82 C
Mean	2.81 (AB)	3.13 (A)	2.43 (B)	2.79 (AB)	2.59 (B)	

Means followed by the same letter do not differ significantly at the 5% level of probability.

When honeybee workers fed on bee bread, the grade of hypopharyngeal gland was the highest degree of development among other compared diets. It followed by development of glands in bees fed on date palm pollen diet (3.41 degree) with insignificant difference with those fed on bee bread. The glands of honeybee workers fed on a mixture diet and corn flour diet had a 2.86 and 2.75 degrees with insignificant differences with those fed on bee bread and date palm pollen diet. The poorest result was obtained when bees fed on Haydak's diet (diet 4) or on sugar syrup only.

The present results are in agreement with those obtained by Maurizio (1950) and Standifer *et al.* (1960) who indicated that the development of the glands is related to/or promoted by high protein content of the diet. Many authors described that soybean flour, dried yeast and dried milk were two most suitable substance used for supplementary feeding of honeybee colonies after pollen of bee bread (Spencer-Booth,

1960; Standifer *et al.*, 1973 and Mostafa, 2000).

The rectal contents estimated in order to explain the food preference and digestability of consumed food by bees. Table (5) and Fig. (4) explain that for most of the tested diets. The rectal contents started with low weight on the first inspected age then grew up to reach the highest weight at the eldest age (15 days). The means of rectal contents weight regardless to the age of honeybee workers show that values of those fed on diet which contains date palm pollen was smaller to those fed on bee bread without any significant difference. The next descendent mean belong to the bees fed on corn meal with a significant difference from the mean of honeybee workers fed on bee bread followed by those fed on diet 5 (a mixture of protenic material). The lowest mean of rectal weight was reported for the bees fed on Haydak's diet that differs significantly less than any compared diet.

The pervious results reflect the suitability of diets contain pollen for feeding honeybee colonies during nursing period of honeybee workers.

Table (5): Weight of rectal content of honeybee workers fed on proteinic diets.

Tested / Diets	Weight of rectal contents (mg/bee) at different age					General mean
	3 days	6 days	9 days	12 days	15 days	
Diet 1 (bee bread)	6.35 k	8.56 fghijk	12.20 bcde	20.79 a	19.30 a	13.44 A
Diet 2 (date palm pollen)	8.97 efghijk	11.49 cdef	10.41 defghi	14.48 bc	18.85 a	12.84 A
Diet 3 (corn flour)	7.99 ghijk	9.73 defghij	11.76 bcdef	14.86 b	12.72 bcd	11.41 B
Diet 4 (Haydak's diet)	7.55 ijk	6.99 jk	6.80 jk	6.28 k	12.56 bcd	8.04 D
Diet 5 (mixture diet)	6.76 jk	10.40 defghi	11.26 cdefg	11.64 cdef	10.45 defghi	10.11 C
Control (sugar syrup)	7.68 hijk	6.69 jk	8.02 ghijk	10.24 defghi	10.94 defgh	8.71 D
General mean	7.55 (C)	8.98 (B)	10.08 (B)	13.05 (A)	14.14 (A)	

Means followed by the same letter do not differ significantly at the 5% level of probability.

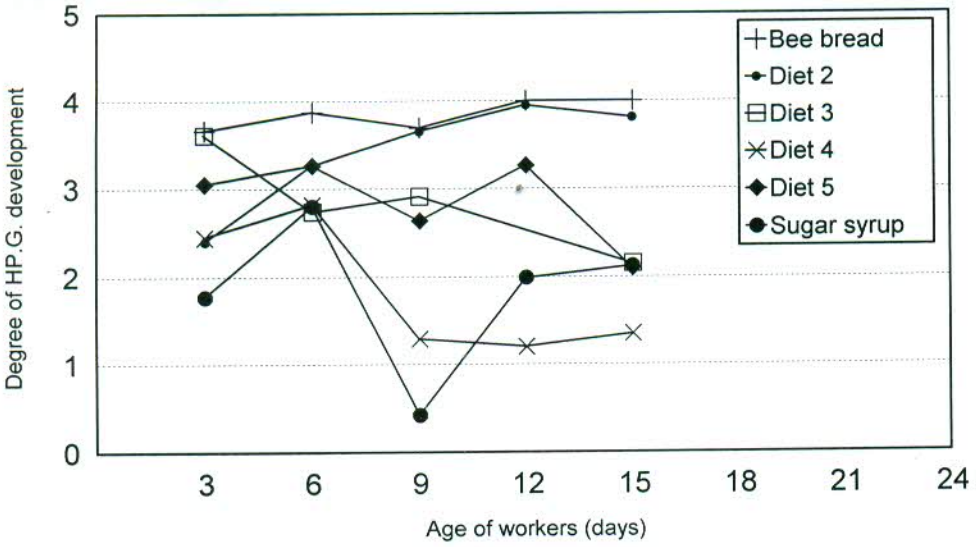

Fig.(3): Variations of hypopharyngeal gland development of honeybee workers fed on proteinic diets.

Fig.(4): Variations of rectal contents of honeybee workers fed on proteinic diets.

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تأثير بعض أنواع التغذية الاضافية على الخصائص الفسيولوجية لشغالات نحل العسل

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أجرى البحث تحت الظروف المعملية بقسم وقاية النبات بكلية الزراعة جامعة أسيوط- مصر وذلك على مجموعات صغيرة من شغالات نحل العسل داخل أقفاص وذلك لتقييم ملائمة خمسة أنواع من التغذية الإضافية البروتينية لتغذية نحل العسل .

وقد قسمت التجربة إلى قسمين :

- **القسم الأول** : وفيه تم تقييم الاستهلاك الغذائى وحساب طول فترة حياة الشغالات حديثة الخروج تحت ظروف التغذية بمختلف الأغذية البروتينية وقد سجل أعلى معدل للإستهلاك الغذائى لشغالات نحل العسل عند تغذيتا بحبوب لقاح النخيل تلاها تلك المغذاه بخبز النحل وكان أدناها فى الإستهلاك المغذاه بمخلوط هيداك . وقد سجل النحل المتغذى على خبز النحل أطول فترة معيشة حيث كان الوقت اللازم لموت ٥٠% من الأفراد ٢٠,٧٩ يوم بينما أستمر النحل المتغذى على حبوب لقاح النخيل ومخلوط هيداك ١٨,١٦ ، ١٩,٥٢ يوماً على التوالى وذلك لموت ٥٠% من الأفراد .

- **القسم الثانى** : وفيه تم تقييم نمو غدد الغذاء الملكى ووزن مكونات المستقيم وقد دلت النتائج على استخدام مصادر البروتين الطبيعية مثل خبز النحل وحبوب لقاح النخيل حيث كانت الأفضل لنمو غدد الغذاء الملكى حيث سجلت درجات النمو ٣,٨٤ ، ٣,٤١ درجة على التوالى بينما التى غذيت على مخلوط من المواد البروتينية كانت درجة نمو الغدد بها ٢,٨٦ درجة مما يوضح أن اضافة حبوب اللقاح إلى أغذية النحل تنشط نمو غدد الغذاء الملكى وفى نفس الوقت فإن أوزان مكونات المستقيم عكست مدى ملائمة الأغذية المحتوية على حبوب لقاح لتغذية شغلات نحل العسل .