

Effect of some pollen grain supplemented foods on the honey bee (*Apis mellifera* L.) colonies and worker activities.

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Abstract

This study was conducted in march 2020 included Four seasons, spring, early summer, late summer and autumn respectively at the apiary of the agriculture office, Sulaymaniyah / Kurdistan Region - Iraq. Sixteen colonies with the almost same densities (4-5 bee frames) were randomly divided in to four groups, comprising four feeding treatments (Sugar solution as control, Wady Nektapol+Sugar solution, Varroa Nektapol+Sugar solution and Candy Shahang+Sugar solution).

The result showed that the highest numbers of workers prefer fed on Varroa nektapol plus Sugar solution was (627.500) worker inside the hives and (350.000) worker outside the hives The highest percentage of protein content in the workers body was (164.187%) which fed with Varroa nektapol +Sugar solution and the lowest was (39.5%) fed with sugar solution .The highest average of body weights of 5th larvae stage fed on Wady nektapol + Sugar solution was (116.250) mg in late summer season but the lowest was (63.750) mg fed on Sugar solution in autumn season. The highest body weight of 10-day old workers was (110.750) mg fed on Varroa nektapol in spring season and the lowest was (78.500) mg fed on Sugar solution in autumn season.

Introduction

Beekeepers in the Kurdistan region face many challenges around the year due to the changing weather condition. Honeybees have shown to be affected by environmental factors such as temperature, abundance of nutrients, floral patterns, consequently, any change in these factors will affect the honeybee colonies in terms of activity, brood rearing, honey making, and many other aspects in their colonies. One of these factors being nutrient availability, can be controlled by supplying the colonies with artificial food supplements.

Bees collect nectar from flowers and transport it to honeycombs, where it is processed into honey, a high-energy food source. Forager bees collect pollen from flowers and store it in the honeycomb, where it transformed into bee bread, which contains protein, fats, minerals and vitamins. (Morais, *et al.*,2013).

The nourishment (royal jelly) that worker bees feed young bee larvae and the queen provides them with protein food resources are substantially decreased during certain times of the year such as winter and dry season. (Saffari, *et al.*, 2010). Supplemental foods are supplied to honey bee colonies to

meet their nutritional needs in places where natural food supplies (pollen, nectar and honey) are insufficient or unavailable. The amount and quality of incoming pollen and nectar, as well as the food stores in the hive, will determine if the bees want additional food. (Sihag and Gupta, 2013).

Beekeepers can supplement colony demands with artificial foods to reduce the effects of dearth seasons. These supplements must have well-balanced formulae that suit the dietary requirements of the bees, assuring the colony's longevity and performance. (DeGrandi-Hoffman, *et al.*, 2016) A well-fed hive may endure difficult times, allowing the queen to continue laying eggs and producing healthy brood. When pollen becomes accessible again, the colony is ready to collect it, replenishing food reserves and allowing honey production.

The aim of the study is an attempt to find out:

1-To find out the complementary diets of honey bees which play an important role in the development and activation of honey bees.

2-To find out the best food that can be used during the food deficiency in order to design the scientific methods for development bee's.

3-To find the effect of supplemental food on the bioactivity honey production and honeybee development.

Materials and methods

This study was conducted in march 2020 at the apiary of the agriculture office, Sulaymaniyha / Kurdistan Region - Iraq. Sixteen colonies with the almost same densities (4-5 bee frames) were randomly divided in to four groups, comprising four feeding treatments (Sugar solution as control, Wady Nektapoll+Sugar solution,

Varroa Nektapoll+sugar solution and Candy Shahang+Sugar solution, in order to study the effect of some pollen grain supplemented foods on the honey bees (*Apis mellifera* L) workers activities and production of (brood, honey, wax, and pollen) during spring, early summer, late summer and autumn season under field condition. At the beginning of the research sixteen colonies of *Apis mellifera* were selected from apiary depending on the following characters.

To make two Langstroth frames used for measurement, were prepared, first changed in way by dividing each frame in two 17 inches (43.2cm) in length and 8 inch in width, holes were made to each frame by using frame fastening wires (silk) which was across of two wires equal in length and wide with on square inch in way that the total number of the inch were equal to 136 inch. (Shamdin, 2003 ; Targany, 2008)

This frame used for measured area of the brood, honey, pollen and wax in different seasons (spring, early summer, late summer and autumn of 2020 measured every 12 days until the end of the study. The measurement was taken using a typical frame with the method (Targany, 2008 ; Bas, 2013).

Sugar solution (control) was prepared by dissolving one kilo gram of crystal sugar in one liter of water after the water was boiled to 70°C, each colony was supplied with one liter of sugar solution at the different concentration weekly during different season (spring, early summer, late summer and autumn). The ratio of concentration was 1:1in spring and autumn 1:2 in early summer and late summer (Targany, 2008). foods prepared food by dissolving 80 gm of candy for each treatment add to 4 liter and mixed by using hand mixer until the candy was dissolve completely in the Sugar solution to give the colony except control (Sugar solution).

The prepared food was given to the bee by using two type of feeder, frame feeder plastic which was put inside the hive and top feeder put on the hive instead inner cover. The frame feeder was appropriate way to allow the food (Neupane and Thapa 2005).

Sample were taken from experimental colonies containing a considerable number of labeled workers with known ages on colony inspection (Bas, 2013).

The preference food and acceptance studying by the honey bees for all replicates and for a period of four continuous days at three different times (9-10, 12-1, 3-4) hourly of the day and calculated the number of honey bee workers coming to feed on the food inside and outside the hives in the each replicate for 45 minutes at the day (Targany, 2008).

Determination of honey bee longevity, numbers of honey bee workers caged for each treatment contained (60 workers) of different ages in small caged with small empty piece of comb used for water and fed on the same prepared food the caged workers bee with the food were put in the hive to determine the longevity monitoring calculating the dead bees and adding food to avoid bee death when needed food during shortage of food. (Abou-EL-Enain *et al.*, 2006)

The climatic data was taken information from Sulaymaniyha general directorate of metrology and seismology.

Statistical analysis:

The results were analyzed statistically using factorial RCBD design with four replicate and performed using XLSTAT program (2016). The leading data analysis and statistical solution for Microsoft Excel is a powerful yet flexible Excel data analysis add-on that allows users to analyze,

customize and share results within Microsoft Excel. Duncan's multiple range Test was used to determine the differences between means at $P=0.05$.

Results and discussion

Food preference by honey bee workers inside and outside the hives, Table (1) show that prefer feeding for the workers bee on Varroa nektapol + Sugar solution inside and outside the hive then, Wady nektapol + Sugar solution, Sugar solution and finally Candy Shahang +Sugar solution. The highest numbers of workers prefer fed on Varroa nektapol was (627.500) workers inside the hives but outside the hives was (350.000) workers. Statistical analysis showed significant difference at level 0.05 between treatments and prefer the foods where the much higher of Varroa nektapol plus Sugar solution in comparison with other treatments. Al-Ghamdi (2002) who observed that the Carniola bee colonies which were fed on pollen supplementary feeding during the experiment period indicated to the highest level of population density, brood rearing, pollen and honey storage compared with the control colonies which were only fed on natural food sources and the number of combs covered with bees in colonies supplement with patty increased gradually. Also this result agreed with Al-Sharhy and Al-Ghamdi (2006) who indicated that honey bee colonies fed on a mixture of yeast meals with gluten or yeast has a high palatability by bees similar to palatable natural commercial pollen, Al-Anssari (1998) stated that the beekeepers often feed their colonies by alternative food or complementary food during the summer to make up for the shortage of both pollen and nectar. Heyes (1984) who recommended the soybean flour and yeast added to candy as the food to be given to colonies in the spring.

Table (1): Food preference by honey bee workers inside and outside the hives during different times of day.2020.

Types of food	Time of day (9-10, 12-1, 3-4) hours		Means
	Inside	Outside	
Sugar solution	550.776 c	252.333 g	401.554 c
Wady nektapol	603.333 b	303.500 f	453.416 b
Varroa nektapol	627.500 a	350.000 e	488.750 a
Candy shahang	452.750 d	201.250 h	327.000 d
Means	558.589 a	276.770 b	

Means with the same letter are not significantly different at the level 0.05.

The result data in table (2) demonstrated that effect of some supplemental foods on the nitrogen

Content (total protein) in the body of honey bee workers recorded the highest ratio which

nitrogen content was 26.27% fed with Varroa nektapol + Sugar solution while the lowest

percentage of nitrogen content was 6.32% fed with Sugar solution also concerning the total

protein showed the highest number was 164.187% fed with Varroa Nektapol plus sugar solution

but the lowest ratio was 39,5% fed with Sugar solution in both circumstance there were

significantly at 0.05 ($P < 0.05$) effected on Nitrogen content (total protein) in the body workers

honey bee, this result was agreed with Haydak, (1970) the nitrogen content is more than (%93,

%76 and %37) in the head, thorax and abdomen respectively 5 days after feeding on the pollen,

also, confirme Szymas, (1996) feeding honey bees with material proteins from potatoes and fish

protein and fat content.

Table (2): Percentage of nitrogen (total protein) in the body of honey bee workers.

Foods	Nitrogen content %	Total protein %
Sugar solution	6.32	39.5
Wady nektapol	22.1	138.125
Varroa nektapol	26.27	164.187
Candy shahang	17.35	108.437

Table (3) shows some supplemental food effected the body weights of 5th larvae stage supplemented with Sugar solution, Wady nektapol, Varroa nektapol and Candy shahang in spring season were 92.250, 107.250, 97.000 and 89.500 mg respectively. The highest average was 107.250 mg fed on Wady nektapol + Sugar solution while the lowest average was 89.500 mg fed on Candy shahang. statistically significant difference at level 0.05 between treatments and body weight of mature larvae fed on Wady nektapol + Sugar solution where superior compared with other treatments.

The average of body weights of 5th larvae stage supplemented with Sugar solution, Wady nektapol, Varroa nektapol and Candy shahang in early summer season were 81.250, 96.750, 87.250 and 75.500 mg respectively. The highest average was 96.750 mg fed on Wady nektapol + Sugar solution while the lowest average was 75.500 mg fed on Candy shahang + Sugar solution. statistically shows significant difference at level 0.05 between treatments and body weight of mature larvae fed on Wady nektapol where superior compared with other treatments.

The average of body weight of 5th larva stage supplied with Sugar solution, Wady nektapol, Varroa Nektapol and Candy shahang in late summer season were 95.000, 116.250, 105.000 and 94.250 mg respectively. The highest average was 116.250 mg fed on Wady nektapol + Sugar solution while the lowest average was 94.250 mg fed on candy shahang. Statistically, there was significant difference at level 0.05 between treatments and body weight of mature larvae fed with Wady nektapol where superior compared with other treatments.

The average of body weights of 5th larvae stage supplemented with sugar solution,

Wady nektapol, Varroa nektapol and Candy shahang in autumn season were 74.000, 85.250, 84.750 and 63.750 mg respectively. The highest average was 85.250 mg fed on Wady nektapol + Sugar solution while the lowest average was 63.750 mg fed on Candy shahang + Sugar solution. Statistically, there was significant difference at level 0.05 between treatments and body weight of mature larvae fed with Wady nektapol where superior compared with other treatments. Statistically had significant difference at level 0.05 between treatments and body weight of 5th larvae stage in four seasons fed on Wady nektapol + Sugar solution were superiority in comparison with other treatment. Statistically the effect of supplemental foods there were a significant difference between the body weight and foods. Wady nektapol considered as a best food compared with other treatments as shown in table (6). This result agreed with Bas (2013) record highest rate of body weights (mg/larva) of mature larvae fed with geval protein were 121.50, 116.20 and 112.03 in spring, summer, autumn respectively.

Table (4,5,6, and7) shows some supplemental food effected on body weights of workers from four treatments with four ages in spring, early summer, late summer and autumn seasons. The comparison among the body weight in each ages showed significant differences at level 0.05 between treatments in all seasons.

Highest weight of one day old workers was 99.000 mg fed on Varroa nektapol + Sugar solution while the lowest weight was 74.000 mg fed on Sugar solution in autumn season. The highest mean of body weight was 93.875 mg fed on Varroa nektapol + Sugar solution and lowest mean was 77.937 mg fed on sugar solution. The result agree with Daly and Morse (1991) who mentioned that the amount foods of honey bee larvae had

effects on adult body weight, with Shamdin (2003) who found that the highest rate body weight of one day old was (109.15) mg and

(91.14) mg thirty day old workers when fed with soy bean.

Table (3): Effect of supplemental foods on the body weight of mature larva/mg

Foods	Weight of 5th larva stage/mg				
	Spring season	Early summer season	Late summer season	Autumn season	Means
Sugar solution	92.250 c-e	81.250 Gh	95.000 cd	74.000 I	85.625 c
Wady nektapol	107.250 b	96.750 c	116.250 a	85.250 fg	101.375 a
Varroa nektapol	97.000 c	87.250 e-g	105.000 b	84.750 Fg	93.500 b
Candy shahang	89.500 d-f	75.500 hi	94.250 cd	63.750 J	80.750 d
Means	96.500 b	85.187 c	102.625 a	76.937 d	

Means with the same letter are not significantly difference at the level 0.05.

Table (4): Effect of supplemental foods on body weight of one day old workers/mg

Foods	Weight of one day old worker/mg				
	Spring season	Early summer season	Late summer season	autumn	Means
Sugar solution	85.250 a-d	78.000 b-d	74.500 d	74.000 d	77.937 d
Wady nektapol	89.667 a-d	80.750 b-d	76.750 cd	83.250 a-d	82.604 c
Varroa nektapol	93.500 ab	88.750 a-d	94.250 ab	99.000 a	93.875 a
Candy shahang	78.000 b-d	84.250 a-d	92.000 a-c	90.000 a-d	86.062 b
Means	86.604 a	82.937 b	84.375 ab	86.562 ab	

Means with the same letter are not significantly difference at the level 0.05

The highest body weight of ten days old workers was 110.750 mg fed on Varroa nektapol + Sugar solution in spring season while the lowest body weight was 78.500 mg fed on Sugar solution in autumn season. The highest mean of body weight was 98.000 mg fed on Varroa nektapol + Sugar

solution and lowest mean was 80.312 mg fed on Sugar solution. This study agrees with result of Ayoub, (2011) who found that the weight in 10 days old workers higher than the body weight of newly emerged and forager workers.

Table (5): Effect of supplemental foods on body weight of ten days old workers/mg

Foods	Weight of ten days old worker/mg				
	Spring season	Early summer season	Late summer season	Autumn season	Means
Sugar solution	81.500 e	82.250 e	79.000 e	78.500 e	80.312 d
Wady nektapol	93.250 b-e	84.750 de	87.750 c-e	85.500 de	87.812 c
Varroa nektapol	110.750 a	91.000 b-e	99.250 a-d	91.000 b-e	98.000 a
Candy shahang	81.000 a	92.750 b-e	102.000 a-c	105.000 ab	95.187 B
Means	91.625 ab	87.687 b	92.000 a	90.000 ab	

Means with the same letter are not significantly difference at the level 0.05

The highest body weight of twenty days old workers was 90.250 mg fed on Varroa nektapol + Sugar solution in late summer season while the lowest body weight was 68.500 mg fed on Sugar solution in early summer season. Also the highest mean of body weight was 85.062 mg fed on Varroa nektapol and the lowest means was 69.812 mg fed on Sugar solution, Bas (2013) who recorded the highest rate of body weight of newly emerged workers in colonies fed with geval protein was 113.60 mg/workers in

autumn season and the highest body weight of seven day old workers in colonies fed with geval protein was 123.97 mg. in spring season, the highest body weight of twenty one day old workers in colonies fed with apricot juice was 97.33 mg in summer season and the highest body weight of twenty eight day old workers in colonies fed with geval protein was 96.73 mg in spring season, this study disagreed with Winston (1987) who stated the weight of honey bee workers ranged from 81-140 mg. .

Table (6): Effect of supplemental foods on body weight of twenty old workers/mg

Foods	Weight of twenty days old worker/mg				
	Spring season	Early summer season	Late summer season	Autumn season	Means
Sugar solution	73.000 c-e	68.500 e	69.000 e	68.750 e	69.812 c
Wady nektapol	85.000 a-c	74.750 b-e	70.500 e	82.750 a-d	78.250 b
Varroa nektapol	85.250 a-c	78.250 a-e	90.250 a	86.500 ab	85.062 a
Candy shahang	69.500 e	74.000 b-e	83.250 a-c	84.500 a-c	77.812 bc
Means	78.187	73.875	78.250	80.625	

Means with the same letter are not significantly difference at level 0.05

The highest body weight of thirty days old workers was 83.000 mg fed on Varroa nektapol plus Sugar solution in autumn season while the lowest body weight was 61.000 mg fed on sugar solution in late summer season and the highest mean of

body weight was 78.125 mg fed on Varroa nektapol and lowest mean was 61.812mg fed on Sugar solution. Shamdin (2003) who found that the highest rate of body weight of thirty day old was (91.14) mg when fed with soy bean.

Table (7): Effect of supplemental foods on body weight of thirty old workers/mg

Foods	Weight of thirty days workers old/mg				
	Spring season	Early summer season	Late summer season	autumn	Means
Sugar solution	61.250 de	61.750 de	61.000 de	63.250 de	61.812 d
Wady nektapol	80.500 a	65.750 de	61.500 de	79.500 ab	71.812 b
Varroa nektapol	82.500 a	70.250 b-d	76.750 a-c	83.000 a	78.125 a
Candy shahang	62.750 de	65.500 de	69.250 c-e	79.500 ab	69.250 c
Means	71.750 b	65.812 d	67.125 c	76.312 a	

Means with the same letter are not significantly difference at level 0.05.

Table (8) shows the effect of some supplemental food on length and width of body dimension of the 5th larvae stage fed on Sugar solution, Wady nektapol, Varroa nektapol and Candy shahang in different seasons.

In spring season, the lengths were 7.41, 7.67, 10.10 and 8.76 mm respectively. The widths were 2.42, 2.61, 3.35 and 2.22 mm respectively. Highest average length was 10.10mm fed with Varroa nektapol + Sugar solution while the lowest average length 7.41 mm fed with Sugar solution also the highest average width was 3.35 mm fed on Varroa nektapol while the lowest average width was 2.22 mm fed on Candy shahang + Sugar solution.

In early summer season the lengths were 7.31, 9.17, 10.90 and 8.72 mm respectively. The widths were 2.69, 3.29, 3.58 and 2.42 mm respectively. The highest average length

was 10.90 mm fed with Varroa nektapol + Sugar solution while the lowest average length was 7.31 mm fed on Sugar solution also the highest average width was 3.58 mm fed with Varroa nektapol + Sugar solution while the lowest average width was 2.42 mm fed on Candy shahang + Sugar solution.

In late summer season the lengths were 7.17, 9.05, 10.74 and 8.53 mm respectively. The widths were 2.55, 3.14, 3.47 and 2.57 mm respectively. The highest average length was 10.74 mm fed with Varroa nektapol + Sugar solution while the lowest average length was 7.17 mm fed on Sugar solution also the highest average width 3.47 mm fed on Varroa nektapol + Sugar solution while the lowest average width 2.55 mm fed with Sugar solution.

In autumn season the length were 7.15, 8.09, 10.69 and 8.67 mm respectively. The widths were 2.63, 3.09, 3.42 and 2.48 mm

respectively. The highest average length was 10.69 mm fed on Varroa nektapol + Sugar solution while the lowest average length was 7.15 mm fed on Sugar solution also the highest average width was 3.42 mm fed on Varroa nektapol + Sugar solution while the lowest average width was 2.48 mm fed with Candy shahang + Sugar solution.

Effect of foods on the body dimensions the highest average length of Varroa nektapol was 10.61 mm where superior in compare with the lowest average length by effect food was 7.26 mm in Sugar solution. The highest average width of Varroa nektapol was 3.45 mm where the superior in comparison with the lowest average width

was 2.42 mm in Candy shahang. Statistically there were significant difference between treatments and average length and width. the effect of season of body dimensions the highest average length and width in early summer season compare with another season. The result agrees with Shamidn (2003) who recorded that the highest rate of length was 12.16 mm and the highest width 4.27mm but without significant difference between treatments and body dimension. Bas (2013) he recorded highest rates of length of body dimensions of mature larvae were 9.93, 9.57 and 9.20 mm in colonies fed with apricot juice in spring, geval protein in summer and autumn seasons respectively.

Table (8): Effect of supplemental foods on body dimension of 5th larval stage.

Foods	Body dimensions of mature larvae/mm								Food effect	
	Spring season		Early summer season		Late summer season		Autumn season			
	length	width	length	width	length	Width	Lengt h	width	length	Width
Sugar solution	7.41 d	2.42 c	7.31 d	2.69 bc	7.17 d	2.55 c	7.15 d	2.63 c	7.26 a	2.57 b
Wady nektapol	7.67 cd	2.61 c	9.17 bc	3.29 a	9.05 bc	3.14 ab	8.09 cd	3.09 ab	8.49 a	3.03 a
Varroa nektapol	10.10 ab	3.35 a	10.90 a	3.58 a	10.74 a	3.47 a	10.69 a	3.42 a	10.61 a	3.45 a
Candy shahang	8.76 b-d	2.22 c	8.72 b-d	2.42 c	8.53 b-d	2.57 c	8.67 b-d	2.48 c	8.67 a	2.42 b
Season effect	8.48 b	2.65 a	9.02 a	2.99 a	8.87 b	2.93 a	8.65 b	2.90 a		

Mean with the same letter are not significantly different at level 0.05.

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