

EFFECT OF POLLEN SUBSTITUTES ON SOME DIFFERENT ACTIVITIES IN HONEYBEE COLONIES

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ABSTRACT

The present work aimed to evaluate the efficiency of different low cost and availability of natural raw materials as a food for honey bees under laboratory and apiary conditions. The effects of food materials on some activities in honey bee colonies such as, brood rearing, pollen gathering, pollen substitutes consumption, development of hypopharyngeal gland and longevity of honey bees were used as screening parameters. After long time studying, it was very seen that the Wheat Germ is the best pollen substitute at all, as it increase the bee's activities, especially in the lack of pollen grains sources, followed by Dried Brewer's yeast, then Soybean flour and the Palm Date came the last in order.

INTRODUCTION

Natural food of honey bees is composed commonly of two components, pollens and bee honey or nectar. Pollens are the main source of proteins, amino acids, vitamins, fats and minerals; whereas honey and nectar represents the main sources of carbohydrates. Honey bees can be fed on various pollen substitutes to supplement inadequate supplies of pollen. In early spring before pollen and nectar are available or at other times of the year when these materials are in short supply (dearth periods). Pollen substitutes help the colony to survive or to make it more populous and productive (El-Banby and El-Sherif 1987a,b).

Practically, beekeepers feed their bees on Pollen substitutes in autumn and winter seasons to develop and maintain colonies with optimum populations for nectar flows, queens, package bee production and wintering. Proper pollen substitutes and sugar syrup as nutritional requirements for honeybee colonies should be offered to give the best wintering results as brood rearing, pollen gathering, longevity and sexual maturity (El-Sherif *et al.* 1994). Older worker bee use protein directly from pollen. Queen in ages, larvae queens and young larvae of both sexes receive protein from nurse bees supplied with pollen (Stanlely and Linskens 1974).

Present work aimed to evaluate the efficiency of different low cost and availability of natural raw materials as a food for honey bees under laboratory and apiary conditions. The effects of food materials on some activities in honey bee colonies such as, brood rearing, pollen gathering, pollen substitutes consumption, development of hypopharyngeal gland and longevity of honey bees were used as screening parameters.

MATERIALS AND METHODS

The apiary experiments were carried out at Mit Ghamr region, Dakahleya Governorate, on honey bee colonies picked from Apiculture Research Department, Dokki, Giza. While the laboratory experiments carried out in Faculty of Agriculture, Moshtohor, Benha University. This work was conducted during the two successive years 2004 and 2005.

1- Design of experiment:

Fifteen randomly selected honeybee colonies of first hybrid Carniolan bees, *Apis mellifera* L. Of about equal strength containing at least seven standard langstroth frames evenly covered with adult bees from both sides and headed by mated first hybrid Carniolan queens at the same age. These colonies have been divided into five groups, and each group has three colonies as replicates.

2- Preparation of diets:

This study demonstrated by choosing four diets and using them as a pollen substitutes which are not expensive and available.

These pollen substitutes are:

- a) **Soybean flour + Honey (2:1 v/v)**, the chemical compositions of Soybean flour determined by Hammad (2000) as follows: humidity 6.58% , protein 50.88% , fat 5.41% , fiber 6.54% and ash 6.73%.
- b) **Wheat Germ + Honey (2:1 v/v)**, the chemical compositions of Wheat Germ determined by Kent *et al.* (1967) as follows: humidity 9-13%, protein 22-32%, fat 6-11%, fiber 2.5-1.8%, ash 4-5% and carbohydrates 35-45%.
- c) **Dried Brewer's yeast + Honey (2:1 v/v)**, the chemical compositions of Dried Brewer's yeast determined by Atallah (1975) as follows: humidity 9.5%, crude protein 40.5%, other extracts 1.5% and ash 6.25% in addition to vitamin B1, B2 and nicotinic acid.
- d) **Palm Date + Powder Sugar (2:1 w/w)**, the chemical compositions of Palm Date determined by Atallah (1968) as follows: humidity 19.16%, protein 3.59%, crude fat 0.58%, crude fiber 3.1%, ash 1.62% and carbohydrates 71.95%.

The colonies have been treated with the above mentioned pollen substitutes in addition to feeding sugar solution (1:1w/w) in lateral feeder (Dummy) inside the hive, except "Control" treated with the sugar solution only. These pollen substitutes was offered to the bees in cake form and placed directly over the brood nest.

3- Estimation of brood rearing:

To estimate of brood rearing rate in the colonies, areas of sealed worker brood of the tested colonies were measured at 13 day intervals during two successive years using a typical Langstroth wire frame divided into square inches, this frame was laid against any comp in which it was desired to count the inches of sealed brood Omer (1998).

4- Estimation of stored pollen:

The same previous method of brood rearing counting was used at 13 days intervals to count the number of square inches of pollen grains stored by the field workers, the areas of stored pollen were measured during two successive years at the same time of brood measure. The amount of pollen

stored in honey bee combs was estimated in grams, five samples of stored pollen blocks were taken randomly from the combs in the experimental colonies. Each sample (containing 25 cells of pollen) was weighed and the average weight of stored pollen per cell was estimated. One stored pollen cell has about 0.16 grams and one square inch of stored pollen weights about 4 grams.

5- Estimation of pollen substitutes consumption:

Each colony received 300 g every 13 days of each of the tested pollen substitutes during the two successive years. The unconsumed pollen substitutes were collected and weighed to determine the rate of consumption of each pollen substitutes. The selected colonies including the control were fed with 500 g sugar as a sugar solution (1:1) at seven days intervals during the dearth periods of nectar.

6- Development of hypopharyngeal glands:

Five wooden cages were used. Their dimensions were 11×15×18 cm with two sides of metal net. Groups of newly emerged bees from each group of experimental colonies were inserted into each cage and fed with tested pollen substitutes in paste form. The cages bees were provided with sugar solution (2:1w/w) and water in little jars.

Twenty nurse bees were taken from each cage in each of the following ages 1, 3, 6, 9, 12, 15 and 18 day(s). Samples were quickly frozen until ready for dissection.

The hypopharyngeal gland was laid bare with a level cut through the head using a razor blade. The worker bee was put in a dissection plate and was flooded with distilled water as a dissection fluid. The gland was taken out from the front region of the head and mounted on a glass slide. Since the hypopharyngeal gland development was investigated under the stereo Binocular at 32x.

Degrees of development of the hypopharyngeal gland were evaluated by referring to the 4 stages (with 1 representing no gland development and 4 representing complete development), according to Maurizio (1954)

7- Longevity:

Fifteen wooden cages were used with the same dimensions before. These cages have been divided into five groups 3 cages each. Groups of newly emerged bees (each of 50 individuals) from each experimental colony were inserted into each cage and fed with tested pollen substitutes in paste form. The cages bees were provided with sugar solution (2:1w/w) and water in little jars.

Daily inspection carried out to count died bees in each cage till all bees died, and then calculating the average life length of worker bees for each cage at the end of the test.

8- Statistical analysis:

The F-test was applied for analysis of variance and the L.S.D with level of 0.05 was used to determine significance of differences between the means.

RESULTS AND DISCUSSION

1. Effect of pollen substitutes on brood rearing activity in honeybee colonies:

The obtained results in table (1) showed that the mean of sealed worker brood area measured from each treatment of Soybean flour, Wheat Germ, Dried Brewer's yeast, Palm Date and Control during the first year (2004) were 186.27, 244.2, 219.06, 148.26 and 154.74 inch²/ colony, respectively. While the mean of sealed worker brood area measured during the second year (2005) were 195.69, 251.47, 207.54, 130.31 and 149.19 inch²/ colony, respectively.

Statistical analysis from the data recorded in table (1) indicated that there were significant differences between Wheat Germ and all other treatments (Dried Brewer's yeast, Soybean flour, Control and Palm Date), respectively. While there were insignificant differences between Control and Palm Date.

Table (1): Effect of feeding pollen substitutes on brood rearing activities in square inches during experimental period.

years	Months	Treatments				
		Soybean flour	Wheat Germ	Dried Brewer's yeast	Palm Date	Control
2004	January	62.33	93.17	81.67	35.17	38.33
	February	164.78	222.56	174.56	93.33	85.22
	March	237.17	333.00	251.83	108.50	126.67
	April	229.83	327.17	305.17	179.83	188.33
	May	279.00	362.22	310.22	255.11	253.78
	June	277.33	366.00	335.17	285.67	293.17
	July	287.33	298.33	309.00	209.83	248.83
	August	175.33	255.78	243.33	141.44	188.44
	September	165.67	237.00	197.17	174.00	140.83
	October	206.83	240.67	236.00	188.00	180.00
	November	107.78	119.67	122.33	67.78	72.11
	December	41.83	74.83	62.33	40.50	41.17
	Total	2235.22	2930.39	2628.78	1779.17	1856.89
	Mean	186.27	244.20	219.06	148.26	154.74
2005	January	117.78	121.00	103.33	35.89	59.44
	February	202.83	260.00	237.33	66.17	75.33
	March	243.67	373.83	244.50	116.17	137.17
	April	253.50	346.50	291.50	144.67	187.00
	May	292.56	369.56	352.44	256.33	290.56
	June	240.00	338.83	241.50	228.00	237.33
	July	237.17	305.33	238.33	208.17	238.33
	August	233.67	231.78	245.78	153.33	209.22
	September	221.00	285.00	201.67	139.83	134.50
	October	158.50	183.50	148.33	109.50	96.33
	November	86.78	121.78	121.56	69.11	93.56
	December	60.83	80.50	64.17	36.50	31.50
	Total	2348.28	3017.81	2490.44	1563.67	1790.28
	Mean	195.69	251.47	207.54	130.31	149.19

L.S.D.for 2004 2005
at 0.05 11.69 20.53

The above results are supported by Mishref *et al.* (1995), El-Waseef (2002), Kalev *et al.* (2002) and Serag El-Dien And Eissa (2003) whose studies indicated that the honeybee colonies provided with pollen substitutes reared more worker brood than in case of unfed ones (Control colonies) during the observation period.

2. Effect of pollen substitutes on pollen gathering activity in honeybee colonies:-

The results in table (2) indicated that the mean amounts of stored pollen measured from each treatment of (Soybean flour, Wheat Germ, Dried Brewer's yeast, Palm Date and Control) during the first year (2004) were 177.37, 196.52, 191.7, 136.63 and 152.8 g/colony, respectively. While the mean amount of stored pollen measured during the second year (2005) were 208.58, 243.41, 215.44, 140.94 and 188.13 g/colony, respectively.

Statistical analysis of the data recorded in table (2) indicated that there were insignificant differences between Wheat Germ and Dried Brewer's yeast in the first year (2004), while there were significant differences between Wheat Germ and all other treatments (Dried Brewer's yeast, Soybean flour, Control and Palm Date) , respectively, in the second year (2005).

Table (2): Effect of feeding pollen substitutes on stored pollen (g/colony) during experimental period.

years	Months	Treatments				
		Soybean flour	Wheat Germ	Dried Brewer's yeast	Palm Date	Control
2004	January	59.33	60.67	68.67	38.00	34.67
	February	68.00	90.67	76.89	59.56	46.67
	March	116.67	106.67	94.00	98.67	90.67
	April	139.33	189.33	136.00	137.33	107.33
	May	329.78	390.67	311.11	235.56	270.67
	June	321.33	303.33	398.67	178.67	249.33
	July	240.00	351.33	293.33	210.67	268.67
	August	326.67	382.22	371.11	288.44	268.44
	September	216.67	176.67	241.33	136.00	238.67
	October	113.33	102.00	121.33	121.33	95.33
	November	105.33	118.67	114.67	94.67	105.78
	December	92.00	86.00	73.33	40.67	57.33
Total		2128.44	2358.22	2300.44	1639.56	1833.56
Mean		177.37	196.52	191.70	136.63	152.80
2005	January	92.00	74.22	118.67	37.78	50.67
	February	106.67	93.33	83.33	18.67	64.67
	March	114.67	125.33	91.33	73.33	70.67
	April	124.67	199.33	105.33	122.00	93.33
	May	352.00	368.89	354.67	200.89	325.78
	June	365.33	514.00	335.33	206.67	358.00
	July	194.07	408.07	253.27	210.00	214.00
	August	425.33	299.11	396.00	309.33	294.67
	September	379.33	420.00	440.67	240.00	438.67
	October	156.00	146.67	168.67	86.67	108.67
	November	128.89	190.67	186.67	144.00	155.11
	December	64.00	81.33	51.33	42.00	83.33
Total		2502.96	2920.96	2585.27	1691.33	2257.56
Mean		208.58	243.41	215.44	140.94	188.13

L.S.D.for
at 0.05

2004
15.8

2005
16.88

The above results are agreed with El-Sherif *et al.* (1994) who found that the ample pollen period extended from mid-March to mid-September (about 6 months). pollen dearth, (from September to December) monthly harvested pollen as well as drone and worker production gradually decreased and began to increase from January till the beginning of the ample pollen period in mid-March.

3. The consumption of pollen substitutes in honeybee colonies:-

In table (3) the results showed that the mean amounts of pollen substitutes consumed by bees from each of (Soybean flour, Wheat Germ, Dried Brewer's yeast and Palm Date) during the first year (2004) were 145.87, 166.08, 162.91 and 119.87 g/colony, respectively. While the mean amounts of pollen substitutes consumed by bees during the second year (2005) were 153.54, 181.63, 179.64 and 127.75 g/colony, respectively.

Table (3): The consumption of pollen substitutes during experimental period.

years	Months	Treatments			
		Soybean flour	Wheat Germ	Dried Brewer's yeast	Palm Date
2004	January	93.90	122.43	124.97	55.63
	February	123.61	154.97	157.20	86.31
	March	165.85	183.83	173.52	119.23
	April	165.02	219.47	203.55	147.50
	May	225.59	240.71	248.19	169.09
	June	230.85	246.98	240.73	210.72
	July	216.82	209.15	199.45	216.35
	August	152.32	145.11	146.43	108.73
	September	113.97	117.98	85.28	111.93
	October	106.90	121.43	111.90	93.67
	November	92.83	112.24	134.27	68.58
	December	62.82	118.62	129.42	50.67
Total		1750.47	1992.93	1954.91	1438.41
Mean		145.87	166.08	162.91	119.87
2005	January	128.11	162.79	163.11	84.40
	February	131.62	173.78	170.13	83.80
	March	156.27	187.45	185.58	118.98
	April	175.33	230.98	221.95	177.23
	May	208.22	245.74	245.61	202.99
	June	242.73	255.57	254.25	224.00
	July	200.88	199.58	186.53	170.15
	August	137.07	188.82	180.51	151.20
	September	170.28	162.03	157.58	116.88
	October	103.97	126.55	115.43	58.67
	November	84.51	113.70	130.96	84.08
	December	103.47	132.53	143.98	60.57
Total		1842.46	2179.54	2155.64	1532.95
Mean		153.54	181.63	179.64	127.75

L.S.D.for
at 0.05

2004
6.39

2005
8.49

Statistical analysis from the data recorded in table (3) indicated that there were insignificant differences between Wheat Germ and Dried Brewer's yeast, while there were significant differences between Wheat Germ and both of Soybean flour and Palm Date.

The above results are agreed with Mohanny (1999) who found that the consumption of pollen substitute (Wheat Germ) was higher than another diet during different seasons.

4. Effect of pollen substitutes on hypopharyngeal gland development:

The results obtained in Table (4) showed that, the mean of the hypopharyngeal gland development stage was 3.02 with Soy been flour, the highest average of the hypopharyngeal gland development stage in six days old was 3.55. The mean of the hypopharyngeal gland development stages was 2.95 with Wheat Germ, the highest average of the hypopharyngeal gland development stage in six days old was 3.45. The mean of the hypopharyngeal gland development stage was 2.77 with Dried Brewer's yeast, the highest average of the hypopharyngeal gland development stage in six days old was 3.25. The mean of the hypopharyngeal gland development stages was 2.13 with Palm Date, the highest average of the hypopharyngeal gland development stage in six days old was 2.55.

Statistical analysis from the data recorded in Table (4) indicated that there were insignificant differences between Soybean flour and Wheat Germ, but there were highly significant differences between Soybean flour and each of Dried Brewer's yeast, Palm Date and Control. Whereas there were significant differences between different ages of bees.

The above results are agreed with Darhous (1990), El-Dakhakhni and Metwally (1995) and Mohanny (1999).

Table (4): The effect of feeding different pollen substitutes on the development stages of the hepopharyngeal glands.

Age \ Treat.	Soybean flour	Wheat Germ	Dried Brewer's yeast	Palm Date	Control	Total	Mean
1day	2.60	2.55	2.50	2.15	1.90	11.70	2.34
3days	2.90	2.95	2.75	2.35	2.25	13.20	2.64
6days	3.55	3.45	3.25	2.55	2.60	15.40	3.08
9days	3.35	3.40	2.95	2.20	2.45	14.35	2.87
12day	3.30	3.15	3.05	1.95	2.05	13.50	2.70
15day	2.85	2.75	2.60	2.00	1.90	12.10	2.42
18day	2.60	2.40	2.30	1.70	1.55	10.55	2.11
Total	21.15	20.65	19.40	14.90	14.70	90.80	18.16
Mean	3.02	2.95	2.77	2.13	2.10	12.97	--
L.S.D.for	Treat	Age					
at 0.05	0.13	0.16					

5. Effect of pollen substitutes on longevity of honey bees:

The results tabulated in Table (5) showed that the mean of longevity in newly emerged caged bees during the experiment were 15.07, 28.94, 23.01,

15.13 and 16.31 day /worker for Soybean flour, Wheat Germ, Dried Brewer's yeast, Palm Date and Control, respectively.

These results indicated that (Wheat Germ) gave the longest longevity followed by (Dried Brewer's yeast, Control and Palm Date), respectively, while (Soybean flour) gave the shortest longevity during the experiment.

Statistical analysis from the data recorded in Table (5) indicated that there were highly significant differences between Wheat Germ and all other treatments, while there were insignificant differences between palm Date and Control. The results agreed with Werner (1987) and Mansour (2002).

Table (5): The effect of feeding different pollen substitutes on the longevity (day/worker) of caged bees taken from the tested colonies.

Treat. Rep.	Soybean flour	Wheat Germ	Dried Brewer's yeast	Palm Date	Control
1	16.08	29.38	22.60	15.46	17.84
2	13.56	27.84	23.40	16.06	16.10
3	15.58	29.60	23.04	13.88	14.98
Total	45.22	86.82	69.04	45.40	48.92
Mean	15.07	28.94	23.01	15.13	16.31
L.S.D. at 0.05 = 2.02					

In conclusion, the results obtained from biological study, may indicate the great effect of the carbohydrate and protein in pollen substitutes. The best and most effective results on wheat germ. This was probably due to the large amount of protein.

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تأثير بدائل حبوب اللقاح علي بعض الانشطة المختلفة في طوائف نحل العسل

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أجريت هذه الدراسة علي ١٥ طائفة نحل هجين اول كرنيولي متساوية القوة تقريبا، وقسمت هذه الطوائف الي ٥ مجموعات كل مجموعة ٣ طوائف ، وغذيت هذه الطوائف ببدايل حبوب اللقاح المتوفرة في الاسواق المحلية وهي دقيق فول الصويا، جنين القمح، الخميرة الطبية الجافة غير النشطة والعجوة، الي جانب التغذية بالمحلول السكري (١:١) فيما عدا طوائف المقارنة التي كانت تغذي بالمحلول السكري فقط. وذلك طيلة عامي الدراسة (٢٠٠٤-٢٠٠٥) في منطقتي ميت غمر ومشتهر.

وكانت نتائج الدراسة كما يلي :-

١- نشاط الطوائف في تربية الحضنة: أوضحت النتائج أن الطوائف المغذاة بجنين القمح أعطت أعلى مساحة حضنة خلال عامي الدراسة ثم الخميرة الطبية الجافة غير النشطة ثم دقيق فول الصويا ثم المقارنة بينما تأتي العجوة في المؤخرة حيث كان متوسط مساحة الحضنة المقللة في العام الأول ٢٤٤,٢ ، ٢١٩,٠٦ ، ١٨٦,٢٧ ، ١٥٤,٧٤ ، ١٤٨,٢٦ بوصة مربعة/ طائفة لكل من المعاملات السابقة على التوالي ، بينما كان متوسط مساحة الحضنة المقللة في العام الثاني ٢٥١,٤٧ ، ٢٠٧,٥٤ ، ١٩٥,٦٩ ، ١٤٩,١٩ ، ١٣٠,٣١ بوصة مربعة/ طائفة لكل من المعاملات السابقة على التوالي .

٢- نشاط الطوائف في تخزين حبوب اللقاح: أوضحت النتائج أن الطوائف المغذاة بجنين القمح أعطت أعلى مساحة حبوب لقاح مخزنة خلال عامي الدراسة ثم الخميرة الطبية الجافة غير النشطة ثم دقيق فول الصويا ثم المقارنة بينما تأتي العجوة في المؤخرة حيث كان متوسط مساحة حبوب اللقاح المخزنة مقدرة بالجرام في العام الأول ١٩٦,٥٢ ، ١٩١,٧ ، ١٧٧,٣٧ ، ١٥٢,٨ ، ١٣٦,٦٣ جرام/ طائفة لكل من المعاملات السابقة على التوالي ، بينما كان متوسط مساحة حبوب اللقاح المخزنة مقدرة بالجرام في العام الثاني ٢٤٣,٤١ ، ٢١٥,٤٤ ، ٢٠٨,٥٨ ، ١٨٨,١٣ ، ١٤٠,٩٤ جرام/ طائفة لكل من المعاملات السابقة على التوالي .

٣- معدلات إستهلاك الطوائف للبدائل: أوضحت النتائج أن الطوائف المغذاة بجنين القمح أعطت أعلى معدل إستهلاك للبدائل بينما الطوائف المغذاة بالعجوة أعطت أقل معدل إستهلاك للبدائل خلال عامي الدراسة ، حيث كانت الكمية المستهلكة من البدائل في العام الأول ١٤٥,٨٧ ، ١٦٦,٠٨ ، ١٦٢,٩١ ، ١١٩,٨٧ جرام/ طائفة لكل من دقيق فول الصويا ، جنين القمح ، الخميرة الطبية الجافة غير النشطة ثم العجوة على التوالي ، بينما كانت الكمية المستهلكة من البدائل في العام الثاني ١٥٣,٥٤ ، ١٨١,٦٣ ، ١٧٩,٦٤ ، ١٢٧,٧٥ جرام/ طائفة لكل من المعاملات السابقة على التوالي .

٤- تطور غدد الغذاء الملكي: أوضحت النتائج أن النحل المتغذي على دقيق فول الصويا أعطي أعلى معدل في تطور غدة الغذاء الملكي بينما نحل المقارنة أعطي أقل معدل، حيث كان متوسط درجة تطور غدة الغذاء الملكي ٣,٠٢ ، ٢,٩٥ ، ٢,٧٧ ، ٢,١٣ ، ٢,١٠ لكل من دقيق فول الصويا ، جنين القمح ، الخميرة الطبية الجافة غير النشطة ، العجوة ثم المقارنة على التوالي . وكانت أعلى معدلات تطور الغدة في الأعمار من ٦ الي ١٢ يوم من وقت خروج الشغالة من العين السادسة .

٥- طول عمر النحل: أوضحت النتائج أن النحل الذي تم تغذيته على جنين القمح أعطي أطول متوسط عمر في حين أن النحل المتغذي على دقيق فول الصويا أعطي أقصر متوسط عمر ، وكان متوسط عمر النحل في المعاملات المختلفة ١٥,٠٧ ، ٢٨,٩٤ ، ٢٣,٠١ ، ١٥,١٣ ، ١٦,٣١ يوم/ شغالة لكل من دقيق فول الصويا، جنين القمح ، الخميرة الطبية الجافة غير النشطة ، العجوة ثم المقارنة على التوالي .

وتوصي النتائج بتغذية طوائف نحل العسل على بدائل حبوب اللقاح المتوفرة في السوق المحلية مثل (جنين القمح، الخميرة الطبية الجافة غير النشطة، دقيق فول الصويا المنزوع الدسم والعجوة) بجانب المحلول السكري والتي أظهرت نشاطاً ملحوظاً في تربية الحضنة وتطور غدد الغذاء الملكي الي جانب نشاط الطوائف في جمع حبوب اللقاح وتخزينها في الأقراص هذا بالإضافة الي إطالة عمر الشغالات الناتجة من الطوائف المغذاة بهذه البدائل عن الطوائف غير المغذاة.