FEEDING PREFERENCE FOR SEVEN TYPES OF BEE BREAD BY HONEY BEES, Apis mellifera L. UNDER LABORATORY CONDITIONS.

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Abstract: In the first hybrid of Carniolan bees, feeding preference on different types of bee bread from monofloral sources was studied under laboratory condition at Faculty of Agriculture, Assiut University, testing the feeding preference, newly emerged bee workers were given a choice among seven types of bee bread from maize, coriander, canola, caper, broad bean, Egyptian clover and fennel plants. During the period of 1-3 days, bees consumed the highest percentage of total pollens (51.36%) followed by the period of 4-6 days (36.25%), then the pollen consumed decreased sharply at the period of 7-9 days (9.22%). Significant differences in pollen consumption by bee workers among all pollen types were recorded. The total consumption from maize, coriander, canola, caper, broad bean, Egyptian clover and fennel pollens were 16.9, 14,4, 11,2, 9.7, 8.0, 5.4 and 0.6 mg/bee/18 days, respectively. The

results indicated that honey bee workers exhibit a preference for pollen types more than the others. After the present results, the pollen types were divided into three groups dependant on preference level of pollen feeding by bee workers. The first group (more preferred), pollen consumption was more than 20% from total pollen consumed, included maize (25.53%) and coriander pollen (21.75%). The second (considerably preferred), the pollen consumption ranged from 10-20%, included canola (16.92%), caper (14.65%) and broad bean pollen (12.08%). Whereas, the third (slightly preferred), the consumption percentage was less than 10%, included Egyptian clover (8.16%) and fennel pollen (0.91%). It can be summarized that the maize and coriander pollens were the most favourable and best attractant pollens than other tested pollens. While fennel pollen was less favourable and lowest attractant.

Key words: honey bee, pollen, bee bread, consumption, feeding preference.

Introduction

Pollen is very important for bee colonies, it is the only protein source

naturally available to honey bee colonies. Pollen supplies dietary requirements including, lipids,

vitamins and minerals necessary for normal growth and development of bee colonies (Hydak, 1970; Winston 1987 and Roulston and Cane, 2000). The quantity and the quality of pollen collected by honey bees affect reproduction, brood rearing and longevity, thus ultimately the of the productivity colony (Kleinschmidt and Kondos, 1978). The proportions of these nutrients can vary widely among pollens of different plant species (Stanley and Linskens, 1974). So, the survival of honey bee colony is vitally linked to its ability to collect sufficient quantities of pollen to rear brood and maintain adults. Bees collect pollen from a wide variety of floral have and distinct sources preferences for some pollen types over others, as demonstrated in natural setting (Jay and Jay, 1984 and Free, 1993). Although it is clear that bees posses the ability to discriminate among pollen types, the way in which they utilize pollenbased cues are poorly understood.

Honey bees depend on visual and olfactory stimuli to locate flowers and their rewards (Backhaus, 1993 and Menzel et al., 1997). Foragers have innate abilities for discriminating colour and retain certain colour cues more effectively than others (Frisch, 1967 and Menzel, 1990). Also important as visual stimuli are floral shape or form (Free, 1970 and Lamb and Wells, 1995), pigmentation patterns

(Petrikin and Wells, 1995) and floral symmetry (West and Laverty, 1998). Although colour is the main stimulus used by bees to locate flowers at a distance, odour is also used in flower selection (Beker et al., 1989 and Kirchner and Grasser, 1998). The use of odour is most important during close-range orientation when bees inspect flowers both before and after alighting (Dobson, 1991), floral odour is the result of compounds produced from several structures including the petals, sepals, gynoecium, anthers and pollen (Dobson et al., 1990). Such observation are further substantiated by qualitative differences in the profiles of volatiles produced by whole-flowers and pollen (Dobson et al., 1990 and 1996), within pollen, odour-producing compounds are associated with the pollenkitt layer surrounding each grain (Dobson, 1988). Honey bees have the ability to discriminate between the odour of pollen and that of other floral volatiles, and can be trained to collect pollen based its odour alone, even in the absence of supplementary dance information (Aufsess, 1960). Furthermore, the selection of food substances that have little or no nutritional value by bees may stimulated by the addition of pollen lipid odour components (Hohmann, 1970 and Starrat and Boch, 1971). There are other factors affect the foraging as, the size of pollen grains (Harder, 1998), the

amount of pollen kitt surrounding grains (Stanley and Linskens, 1974), the external morphological features of pollen grains (Vaissiere and Vinson, 1994) and their associated electrostatic charges (Erickson and Buchmann, 1983 and Chaloner, 1986). Factors such as the pH or age of pollen may also be criteria for its acceptability to bees (Schmidt, 1982 and Schmidt and Johnson, 1984).

The aim of the present study is to investigate the feeding preference of honey bee workers for bee bread (pollen stored in comb cells) from seven monofloral sources under laboratory condition.

Materials and Methods

The experiments were carried out at Faculty of Agriculture, Assiut University under laboratory conditions during 2006. The first hybrid of Carniolan bees, *Apis mellifera* L. worker were used.

Bee bread extraction:

In the present study, bee bread was used because bees do not consume fresh pollen. Bees collect pollen directly from the flowers and store it in the comb cells inside the hive. During collection and storage the pollen composition is changed through the addition of mainly nectar and also glandular secretions. Then, it undergoes a fermentative process (Winston, 1987 and Roulston, 2005).

During the different times of 2005 season, bee bread was extracted from the first hybrid of Carniolan bee colonies, as following protocol:

Numbers of bee colonies were placed in seven farms in Assiut area containing following monofloral species: Egyptian clover (Trifolium alexandrinum), broad bean (Vicia faba), canola (Brassica napus), (Zea maize mays), fennel (Faeniculum vulgare), coriander (Coriandrum sativum), caper (Brassica kaber). All these sources were the common cultivated plants in Assiut Governorate. At the end of each flowering period, the bee bread was extracted from the different bee colonies for each of monofloral farm. Bee bread from each source was stored under freezing condition until using. For sure, identification of bee-stored pollen types was done microscopically in comparison with standard pollen grains collected from the anther flowers.

Preparation of bee cages and bioassay protocol:

Sealed brood combs, containing hatching brood, were taken from queenright colony, then incubated at 32°C±1 and 60% RH. The brood were observed until adults emergence. Four hundred workers, less than 12-hour-old, were placed inside four wooden cages (12x12x5 cm), one hundreds per cage. The cages were provided with a vial of

tap water and a vial of sucrose solution (1:1 w/v), and a piece of bee wax comb (7x5 cm) attached to the top of the cage as a clustering plat form for the bees. The cages were continuously supplied with water and sugar solution. Each cage was provided with equal amounts (3 g) of bee bread from the seven sources under experiment. Four cages as replicates were used for each treatment. The cages were held in a dark incubator at 32°C±1 and 60% RH.

Determination of bee bread preference:

To investigate the pollen preference by honey bees, the tested pollens were offered as a group in

multiple choice to each cage. Equal amounts of each tested pollen were weighed into separate clean plastic feeders and placed at random into the cage. Three days later the weight of the remaining pollens in the feeders was recorded and fresh bee bread were provided to replace residual diets. At this time the positions of the test pollen feeders were reversed. This procedure was repeated 5 times at 3 day intervals until pollen consumption stopped. The preference was measured as relative consumption for tested pollen type to total consumption of all pollen types. The consumption of each test pollen was calculated according to the following equation:

% Pollen consumption (preference) = $\frac{\text{Weight pollen consumed for each type}}{\text{Weight pollen consumed for all types}} \times 100$

Statistical analysis:

For the purpose of statistical analysis, data obtained were statistically analysed. Means were compared according to Duncan's multiple range test (SAS Institute, 1990).

Results and Discussions

During study, in feeding preference tests, newly emerged bee workers were given a choice among seven types of pollen. The bees showed a preference for some pollen types than others.

During the period of 1-3 days, the bee workers consumed the greatest amount of total pollen consumed. The highest percentage of pollen consumed was 51.36%, followed by the period of 4-6 days, was 36.25%. Then, the pollen consumed, decreased sharply at the periods of 7-9 days (9.22%) and 10-12 days (2.41%). Whereas, the lowest pollen consumed was 0.76% during the period of 13-15 days. At the period of 16-18 days, the bee workers stopped for consumption more pollens (Table 1).

Table(1): Bee bread consumption by honey bee workers in laboratory condition at different periods.

	Bee bread consumption (mg/bee) & (%)							Total	Grand
Periods (days)	Maize	Coriander	Canola	Caper	Broad bean	Egyptian clover	fennel	consumption mg/period & (%)	Mean ±SE (%)
1-3	12.0	6.3	3.9	5,3	3.7	2.7	0.1	34.0	4.86a ±0.63
	(35.29)	(18.53)	(11,47)	(15.59)	(10.89)	(7.94)	(0.29)	(100)	(51.36)
4-6	4.3	6.2	4.9	3.6	3.1	1.6	0.3	24.0	3,43b ±0.88
	(17.92)	(25.83)	(20.41)	(15.00)	(12.92)	(6.67)	(1.25)	(100)	(36.25)
7-9	0.4	1.5	2.1	0.7	0.8	0.5	0.1	6.1	0.87c ±0.26
	(6.56)	(24.58)	(34.43)	(11.48)	(13.11)	(8.20)	(1.64)	(100)	(9.22)
10-12	0.2	0.3	0.2	0.1	0.3	0.4	0.1	1.6	0.23c ±0.04
	(12.50)	(18.75)	(12.50)	(6.25)	(18,75)	(25.00)	(6.25)	(100)	(2.41)
13-15	0.0	0.1	0.1	0.0	0.1	0.2	0.0	0.5	0.07c ±0.03
	(0.00)	(20.00)	(20.00)	(0.00)	(20.00)	(40.00)	(0.00)	(100)	(0.76)
16-18	0.0	0.0	0.0	0,0	0.0	0.0	0,0	0.0	0.0
Total consumption mg/bee/18 days & (%)	16.9	14.4	11.2	9.7	8.0	5.4	0.6	66.2	(100)
	(25.53)	(21.75)	(16.92)	(14.65)	(12.08)	(8.16)	(0.91)	(100)	(100)
Grand Mean ±SE	4.23	2.88A	2.24B	2.43C	1.6C	1.08CD	0.15E		
	±0.10	±0.05	±0.5	±0.85	±0.3	±0.71	±0.03		{

In general, there were significant differences in the pollen consumption by bee workers fed on pollen from seven plant sources under experiment. The calculated amounts of pollen consumed per period indicated different pref-

erence. The total consumption of pollen was 16.9, 14.4, 11.2, 9.7, 8.0, 5.4 and 0.6 mg/bee/18 days for bees fed on maize, coriander, canola, caper, broad bean, clover and fennel pollens, respectively (Table 1 and Fig. 1).

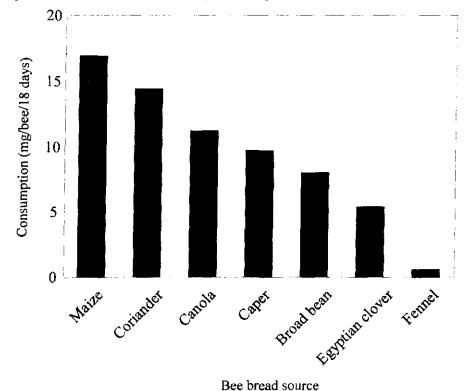


Fig.(1): Total consumption of bee bread for honey bee workers which fed on seven types during the experimental period.

Throughout the present results, could suggested to divide the pollen consumed into three groups, dependant on pollen feeding preference of bee workers as follows: The first group (more

preferred), the percentage of pollen consumption was more than 20% from total pollen consumed. This group included maize (25.53%) and coriander pollen (21.75%) (Fig. 2).

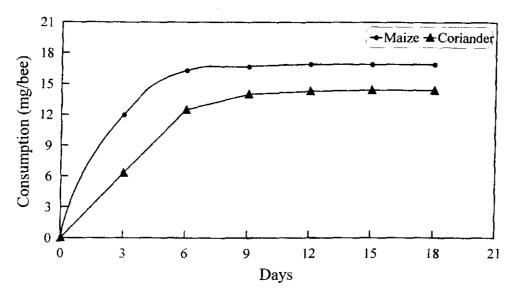


Fig.(2): Cumulative consumption of bee bread by bees fed on maize and coriander pollen, (more preferred group).

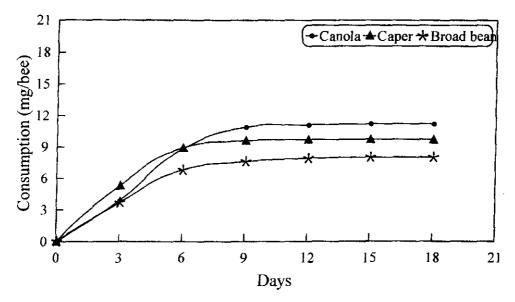


Fig.(3): Cumulative consumption of bee bread by bees fed on canola, caper and broad bean pollen (Considerably preferred group).

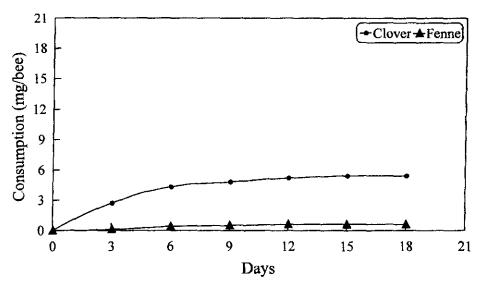


Fig.(4): Cumulative consumption of bee bread by bees fed on Egyptian clover and fennel pollen (Slightly preferred group).

In the second group (considerably preferred), the consumption percentages ranged from 10 to 20%, included canola (16.92%), caper (14.65%) and broad bean pollen (12.08%) (Fig. 3). Whereas, in the third group (slightly preferred), the percentage was less than 10%, included clover (8.16%) and fennel pollen (0.91%) (Fig. 4). So, the most preferred of tested pollen was maize and coriander pollen, but the lowest preferred was fennel pollen. These results indicate that the honey bees exhibit a preference for pollen types more than others. It can be summarized that maize and coriander pollen were the most favourable and the best

attractant pollen than other tested pollen. Whereas, the less favourable and lowest attractant was fennel pollen. These findings confirmed by Campana and Moeller (1977), who showed that when pollens from different sources were offered simultaneously within the hive, the bees ate more of some pollens than of others. Although honey bees are extremely polylectic and use an enormous variety of pollen sources in their diets (O'Neal and Waller, 1984), when given a choice, they are eclectic in their preferences. Honey bees are known to exhibit preferences in pollen selection. but the basis for preferences is not vet clearly

understood. Pollen phagostimulants consist not of a single or a few specific compounds but rather are a suite of diverse components that additively or synergistically serve to exceed a threshold level of stimulation necessary for feeding (Schmidt and Anita, 2006).

Honey bees appear to be capable of using a combination of sensory abilities to enhance their pollen collection. Some researchers have found that pollen odour (Levin and Bohart, 1955) and colour (Boch, 1982) are important factors in pollen attractiveness. The use of pollen odour as a primary cue for honey bees to evaluate whether to engage in pollen-collecting behaviours is supported by its ability to discern olfactory cues better than other types of stimuli. Odour is more important in conditioning foraging preferences than colour, form or time of day (Frisch, 1967; Koltermann, 1969). This seems to be especially promising given that pollen odour plays such a pivotal role in pollen attractiveness. Pernal and Currie (2002) determined that honey bees are most sensitive to the odour of pollen. Bees are appear unable to evaluate the protein content of the resource they are collecting. presence of pollen odour is dominant enough to override cooccurring factors that decease pollen collection behaviours. Their findings also suggest that honey bees do not discriminate among food

sources based on differences in quality, but instead evaluate factors that may increase their efficiency of collection and recruitment to such a food resource.

All plant pollens contain lipids. The lipid concentration differs markedly in pollens as well as in its fatty acid composition. According to a study by Singh et al. (1999) bees preferred pollens with highest amount of lipids. In addition to variation in lipid content, pollen also varies in the relative proportions of fatty acids as well as in their diversity (Manning, 2001 and Markowicz Bastos et al., 2004). The role of lipids as phagostimulants (attractants), appears to have merit when examples of pollen with nutrient qualities low in protein but high in fat content are far more attractive to foraging honey bees. This example like maize and coriander pollens here in the present study, it recorded the highest attractant pollens than others. Maize pollen has a low level of crude protein (14-15%) (Stace, 1996), but has a high in fat content (6%), whereas broad bean pollen resulted less attractant may be due to low in fat content (about 2%), resulted by Somerville (1995). Also, canola pollen recorded more attractant in the present results, due to its higher fat levels (10.7%) (Roulston and Cane, 2000). Somerville (2001), would support this view, as canola pollen has a high fat levels, appear

to be associated with attractiveness of the pollen to foraging bees. Also, Stace and Hayter (1994) suggested that a fat content of 6-10% in protein supplements could increase consumption.

Fatty acids are important in the reproduction, development, nutrition of hone bees (Farag et al., 1978 and Manning, 2001). Fatty acids as a percentage of total lipids are important for honey bee nutrition. A number of the plant species that have evolved with this honey bee have a higher percentage of the nutritionally important fatty acids such as oleic and palmitic acids. Palmitic acid was 26% in broad bean, 19% in clover and 15% in caper pollen in Egypt, recorded by Farag et al. (1978) and Shawer et al. (1987). Whereas the oleic acid was 15, 10 and 4% for broad bean, clover and caper pollen, in Egypt by the same authors. Palmitic and oleic acids were 33 and 7-42% in maize pollen (Battaglini and Bosi, 1968 and Shawer et al., 1987), whereas were 16 and 5% in canola pollen (Evans et al., 1991). Some types of pollens have a high level of oleic and palmitic acids probably have a greater role in honey bee nutrition (Manning, 2001).

On the other hand, there are some factors play role in the attractiveness of pollens to foraging honey bees. Pollen collecting bees are attracted by the form and size of the pollen

grains (Ohe, 1987 and Pernal and Currie, 2002). Instead, floral constancy may be influenced more by cues involved in perceptual conditioning, odour, colour and size (Wilson and Stine, 1996), which were carefully standardized among treatments. Schmidt and Johnson (1984) reported that honey bee tended to prefer less acidic pollen. Schmidt (1984) indicated that bees exhibits preference for polyphagy pollen rather than monophagy.

The effect of an amino acid, glycine on feeding preferences in the honey bee was studied by Kim and Smith (2000). They found that the bees preferred to feed on a sucrose stimulus that contained glycine, and the highest relative preference was recorded for the highest concentration of glycine. So, the modulate glycine can feeding preferences in honey bees. From the study not all the more attractive pollens have a high nutritional value for bees, but sometimes are poor and need to support with rich materials such as, maize pollen has more favourable and best attractant, but it is poor in protein content. According study and previous information, it can be concluded that the fat content of pollen, especially palmitic and oleic acids play an important and main role in the attractiveness of pollen to honey bee workers, together with other important factors, such as the form and size of the pollen grains, odour,

colour, pollen acidity and amino acid, glycine.

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التفضيل الغذائى لسبعة أنواع من خبز النحل بواسطة نحل العسل تحت ظروف المعمل

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أجريت هذه الدراسة بكلية الزراعة جامعة أسيوط في موسم ٢٠٠٦م بغرض دراسة الاستهلاك الغذائسي ودرجة التفضيل بواسطة تغذية شغالات النحل حديثة المخروج على سبعة أنواع من خبز الــنحل من مصادر نباتية رئيسية في منطقة أسيوط وهي النرة الشامية والكزبرة والكانولا والكبر والفــول الــبلدي والبرســيم المصري والشمر . أوضحت النتائج أنه توجد اختلافات معنوية بين الأنواع المختلفة من حيث التفضيل والاستهلاك الغذائي بصفة عامة سُجلت أعلى نسبة للاستهلاك الغذائي (٥١,٣٦%) من إجمالي الاستهلاك خلال التجربة في الفترة من ١-٣ يوم من عمر النحل تليها الفترة من ٤-٦ يوم (٣٦,٢٥%) ثم قل الاستهلاك الغذائي بصورة حادة بعد ذلك. وأوضحت الدراسة أن اجمالي الاستهلاك الغذائي لخبز النحل كان ١٦.٩ ، ١٤,٤ ، ١١,٢ ، ٩,٧ ، ٨,٠ ، ٥.٤ ، ٠,٦ ملجرام / نحلة / ١٨ يوم للشغالات التي تغنت على خبز النحل لكل من الذرة الشامية والكزبسرة والكانولا والكبر والفول البلدي والبرسيم المصري والشمر على التوالي . ومن خلال النـــتائج أمكــن تقسيم أنواع خبز النحل إلى ثلاث مجموعات بناءا على درجة تفضيل واستهلاك السنحل لهسا وهسى: المجمُّوعة الأولى (أكثر تفضيلًا) وتكون نسبة استهلاك خبر النحل في هذه المجموعــة أكـــثر مــن ٢٠% من الاستهلاك الكلى لجميع أنواع خبز النحل المختبرة وتضم هذه المجموعة خبز نحل كل من الذرة الشامية (٢٥,٥٣%) والكزبرة (٢١,٧٥%) والمجموعة الثانية (تفضيل معقول) وتتراوح نسبة الاستهلاك لخبز النحل من ١٠-٢٠% من إجمالي الاستهلاك وتضم خــبز النحل لكل من الكانولا (١٦,٩٢%) والكبر (٤,٦٥%) والفول البلدي (١٢,٠٨%) ، بينما المجموعـــة الثالثة (أقل تفضيلا) نسبة استهلاك خبز النحل أقل من ١٠% من إجمالي الاستهلاك وتضم خبز النحل لكل من البرسيم المصرى (٨٨,١٦) والشمر (١٩١٠) . من خلال ذلك كان أعلمي استهلاك وأكثر تفضيلا في التغذية على خبز النحل للذرة الشامية والكزبرة بينما كان أقل استهلاك وأدنس تفضيل خبز نحل الشمر . وفي الحقيقة ليس من الضرورة أن يكون كل غذاء جانب أن يكون له مستوى غذائي عالى مثل خبز نحل الذرة الشامية أكثر تفضيلا وأقل غذائيا فيجب أن يُوصى مربى النحل أن يقدم دعما غذائيا عالى القيمة في الفترات التي تنتشر فيها هذه المصمادر الفقسيرة . كمما يُوصى بإضافة خيز النحل الجاذب في حالات استخدام تغنية صناعية بدبلة.