IMPACT OF USING POLLEN SUBSTITUTES ON PERFORMANCE OF HONEY BEE (*Apis mellifera* L.) COLONIES UNDER HARSH ENVIRONMENTAL CONDITIONS

AMRO, Abdulraouf Mohamed ¹; OMAR, Mohamed Omar ²; AL-GHAMDI, Ahmed Alkhazim ³.

Received: 07/11/2020 *Accepted:* 28/12/2020

¹Bee Research Department, Plant Protection Research Institute, Agricultural Research Center, Dokki, Giza, Egypt; ²Department of Plant Protection, College of Agriculture, Assiut University, Assiut, Egypt; ³Department of Plant Protection, College of Food and Agriculture Sciences, King Saud University, Kingdom of Saudi Arabia.

ABSTRACT

ollen grains are the main source of protein for honey bees. In Central region of Saudi Arabia, the lack of this natural diet during dry seasons in summer is a primary concern for beekeeping industry. Effect of using alternative protein feeding on performance, feed consumption, brood rearing activity and storing bee bread of honey bee colonies was tested during late summer under Riyadh region condition. Soybean flour, mesquite pods flour, date paste, corn gluten and Feedbee[®] diets were the main tested material in the present study. Preference was determined by using multiple choice tests (MCT). The obtained results revealed that honey bees preferred firstly the commercial product Feedbee[®], followed by date paste and mesquite diets. Also, the highest consumption was recorded in honey bee colonies fed Feedbee[®] and was equal to approximately 1.2, 1.3 and 1.8 fold of amounts consumed from date paste, mesquite and corn gluten diets, respectively. Effect of tested diets on brood rearing activity of honey bee colonies during two successive years was studied. Results showed that the superior response had been obtained using Feedbee[®] followed by mesquite and date paste diets. Due to the fact that Feedbee[®] is expensive and not available in the local markets of Saudi Arabia, beekeepers are advised to use mesquite pods flour and date paste as pollen substitute in the dearth periods to enhanced brood rearing of honey bee colonies during late summer.

Keywords: Honey bees. Substitute diets. Feedbee[®]. Mesquite pods flour. Bee bread.

INTRODUCTION

For many years, the development of a pollen substitute for honey bees has long been an area of interest to the beekeeping industry. The suitability of different protein feeds in the nutrition of honey bees has been tested all over the world (AL-GHAMDI et al., 2011; AMRO et al., 2016, 2020; CRAILSHEIM; STOLBERG, 1989; ZERBO et al., 2001). Stimulating effect of the protein feeds on the development of bee colonies, oviposition and the condition of young queens have been demonstrated. The practical value of a feed is critically dependent on its acceptance by bees. Thus, providing a proteinaceous feed to stimulate colony strength would assist in the maximization of colony population, honey production and crops pollination.

The large variety between materials used as pollen substitutes has encouraged several investigators to study their impacts on honey bees (AL-GHAMDI, 2002; AMRO et al., 2020; DEGRANDI-HOFFMAN et al., 2010; DE JONG et al., 2009; MATTILA; OTIS, 2006; PĂTRUICĂ et al., 2013; SZYMAS; MALISZEWSKA, 1999). Results of Abbasian and Ebadi (2002) showed that soybean flour, soybean meal, wheat gluten and yeast can be used as a basic material for preparing pollen supplement and substitute cakes. Also, the results of Amro et al. (2016) suggested the possibility of using mesquite pods powder and date paste to make a mixed diet for feeding honey bee colonies during the shortage of pollen sources.

For improving nutritional value of pollen substitutes and the intake of complex protein and sugars by bees, the individual components have to be affective for the enzymatic system for honey bees and reduce their oxidative stress (TAWFIK et al., 2020). Szymas and Maliszewska (1999) improved a pollen substitute formula by adding methionine and lysine to enhance pollen substitutes and ensure their balance was brought to the level found in natural pollen. Moreover, Rogala and Szymas (2004) demonstrated that supplementing the pollen substitute with amino acids, which are absent, brings them up to the pollen level. Bringing up the pollen level has improved the nutritional value of the protein in the surrogate to equal that of pollen.

Recently, some commercial pollen substitutes as Feedbee[®] and Bee-pro[®] were offered for application. The bees fed Feedbee[®] had the highest level of body protein, only slightly higher than for those fed Bee-Pro[®], but about 5% higher than those fed pollen and about 3 times

than those fed only sugar syrup (DE JONG et al., 2009; SAFFARI et al., 2004). Also, Amro et al. (2016) highlighted the impacts of pollen substitutes on honey bee colonies performance under the fully absence of natural protein source (pollen) and recorded that Feedbee[®] was the best one, which encouraged bees for rearing brood.

Under the environmental conditions of central region in Saudi Arabia, where dearth periods are long and the raining and flowering periods are short, the protein source feed is a critical factor. Therefore, during these periods, special care should be taken in management of bees. One of the solutions is migration beekeeping, however it causes lots of discomfort and time and money wasting. The other solution is to feed honey bee colonies with proteinaceous diets, so that brood rearing activity can be enhanced and strength colonies. In this regard, Alqarni (1995) and Al-Ghamdi (2002) reported that the stored pollen areas in the comb were less than 10 inch²/colony during the summer season and they recommended supplementary feeding during dearth period.

Alqarni and Alatawi (2008) investigated the attractiveness effects of adding five different volatile oils to the pollen substitute. They found that coriander oil, fennel and spearmint are important agents in attracting honey bees to the supplementary feeds. Al-Sharhi (2008) evaluated different pollen substitutes and supplements during different periods of the year and measured the effects on the brood rearing, honey production and pollen collection of honey bee colonies. He found that a mixture of yeast and corn gluten (1:1) had high acceptance and better performances of colonies.

Despite the presence of some local preliminary works, the studies were not exhaustive and were mostly focused on laboratory and honey bee preference tests (AL-GHAMDI et al., 2011). In the present study, an attempt has been done to compare the impact of proteinaceous diet formulations fed to bee colonies during late summer on some parameters as brood rearing and stored bee bread areas, so that suitable pollen substitute can be developed to improve beekeeping practice in Riyadh region during summer season.

MATERIAL AND METHODS

The experiments were conducted in the apiary of Ministry of Environment, Water and Agriculture, Riyadh (24° 34′ 27″ N 46° 41′ 18 ″ E) during late summer of 2014 and 2015 season.

Proteinaceous materials used as substitutes

Seven materials that are rich in protein and are available in local area were selected for testing as pollen substitute. They are described in Table 1. Total protein of these raw materials was determined by Kjeldahl method (KIRK, 1950). Five proteinaceous mixtures were prepared from the raw materials (Table 2). Feedbee[®] is a commercial substitute tested by Saffari et al. (2004). Centrum[®] multivitamins and minerals (from A to Zink[®] 221 g) produced by Pfizer (formerly Wyeth).

| Fable 1 - Total | protein percentage of | of raw materials used for | pollen substitutes. |
|-----------------|-----------------------|---------------------------|---------------------|
| | | | |

| Raw materials | % | | |
|---------------------------------------|-------------------|--|--|
| Soybean meal (Glycine max (L.) Merr.) | 39.88 c* ± 0.13** | | |
| Brewer's dried yeast | 40.57 b ± 0.19 | | |
| Skimmed milk powder | 29.87 d ± 0.13 | | |
| Corn gluten | 55.94 a ± 0.07 | | |
| Dates paste (Phoenix dactylifera) | 24.55 f ± 0.26 | | |
| Flour of Mesquite pods | 16 E9 g ± 0.27 | | |
| (Prosopis juliflora (Sw.) DC.) | 10.56 g ± 0.27 | | |
| Feedbee® | 28.82 e ± 0.19 | | |

*Means (± SD**) followed by the same letter do not differ significantly at the 5% level of probability.

| Matorials | Composition of the diets (Kg) | | | | | |
|--|-------------------------------|--------|--------|--------|--------|--|
| Iviaterials | Diet 1 | Diet 2 | Diet 3 | Diet 4 | Diet 5 | |
| Soybean meal | 252 | | | | | |
| (Glycine max) | 252 | | | | | |
| Mesquite pods powder | | 250 | | | | |
| (Prosopis juliflora) | | 256 | | | | |
| Date paste | | | 274 | | | |
| (Phoenix dactylifera) | | | | | | |
| Feedbee®* | | | | 379 | | |
| Corn gluten | | | | | 424 | |
| Dried skim milk | 84 | 86 | 91 | | | |
| Brewer's yeast | 84 | 86 | 91 | | | |
| Sugar powder | 378 | 385 | 411 | | 381 | |
| Fresh mixed pollen pellets | 42 | 43 | 46 | | 42 | |
| Multivitamins and minerals (Centrum ^{®**}) | | 4 | 5 | | | |
| Coriander oil (ml) | | 9 | 9 | | | |
| Sucrose solution (2:1) (ml) | | | | 606 | | |
| Honey (ml) | 17 | 17 | 18 | 15 | 17 | |
| Water (ml) | 143 | 112 | 55 | | 136 | |
| Total | 1000 | 1000 | 1000 | 1000 | 1000 | |
| Total protein % in diet | 16.89 | 10.87 | 7.17 | 28.82 | 45.24 | |

 Table 2 - Description of mixed proteinaceous diets administrated to honey bee workers.

*Commercial substitute tested by Saffari et al. (2004).

** Multivitamin from A to Zink[®], produced by Pfizer (formerly Wyeth).

Diet preferences

To determine the honey bee preference for the tested diets, the diets were offered together in each honey bee colony using free choices and replicated in four colonies in the same strength. From each diet 70 g were poured in lid of Petri-dish (100x15 mm) and placed together over the brood nest of every examined colony. The preference was measured as amount consumed from every diet in the same colony and calculated the differences in the weight of every diet before and after feeding period (g/colony/week).

Colony performance evaluation

The experiment started on August 14 in two successive years (2014 and 2015), when the stored pollen area (square inche²/colony) inside honey bee colony was very low as described

with Al-Ghamdi (2002). Twenty-five honey bee colonies headed with sister queens and nearly of similar strength and stored food were selected for the first season in 2014, and the experiments were repeated in another colonies in the second season of 2015. The colonies were randomly divided into five groups. The diets were offered in cake form at the rate of 250 g/colony/6 days interval during the experimental period as described by Amro et al. (2016) to ensure a sufficient amount of food for bees and access to food for all mature and immature stages. The cakes were poured in the lid of Petri-dishes (100x15 mm) and covered with perforated polyethylene bags to reduce the water evaporation and placed over the brood nest. The treated colonies were inspected to determine several parameters such food consumption, sealed brood area and bee bread area.

Measurements

Diet consumption

The diet consumptions were recorded by calculating the difference between the weight of diet before and after feeding (g/colony). This procedure was applied every 6 days interval for every variant during the period of study.

Determination of sealed brood and bee bread areas

Areas of sealed brood and bee bread were recorded from mid-August until mid-October of the two studied seasons (2014 and 2015) at 12-days intervals by using graduated grid divided into square inches after the honey bees had been shaken from the combs as described by Jeffree (1958).

Statistical analysis

The experiment was set up according to a randomized complete block design (RCBD). Three replicates (colonies) were used per treatment (5 treatments or diets) and control group. Means ± standard deviation (SD) were calculated for studied parameters. The data presented no deviation from normality accordingly with Shapiro-Wilk's W test (SHAPIRO; WILK, 1965). For the aforementioned parameters, ANOVA was performed. The means were compared using Duncan's multiple range test (DUNCAN, 1955) at a significance level of 0.05. Simple

correlations between some factors were calculated using a Pearson correlation coefficient (r). The statistical analysis was performed with a SAS 9.1.3 program (SAS INSTITUTE, 2004).

RESULTS and DISCUSSION

Preference of the tested proteinaceous diets

The consumed amounts of tested pollen substitutes based on honey bee preference are recorded in Table 3. The means of the two applications insured that the four tested diets differed significantly to the control (p<0.05). The highest consumption amount was recorded when honey bee colonies fed with Feedbee[®] (48.43 g) followed by date paste, mesquite, soybean and corn gluten, which consumed 41.30, 40.20, 29.85 and 14.74 g/colony/6 days, respectively. The obtained results showed that honey bees were attracted and prefer to consume Feedbee[®] and date paste diets in high rates and very fast regardless of their protein content. These findings were harmonious with Amro et al. (2016). However, they recorded that date past was more consumed than Feedbee[®]. This may be due to the fact that their honey bee colonies were under isolation condition and in this study the honey bee colonies were in free flight condition. Also, Hanna and Schmidt (2004) attributed the preference of honey bee workers to diets with high sucrose content, which serve as phagostimulants. Also, the addition of 5% pollen grain (AMRO et al., 2016) and coriander oil in small amount (0.3%) increased palatability of the diets and the consumption rate (ALQARNI; ALATAWI, 2008).

| | Means (± S.D.**) of consumed amount | | | | | |
|--------------------------------|-------------------------------------|---------------|--------------|--------|--|--|
| Diets | (g/colony/6 days) | | | | | |
| | 2014 | 2015 | Average | R.O*** | | |
| Diet 1 (soybean meal | 31.35+3.98 d* | 28.35+2.84 c | 29.85+1.25 d | 4 | | |
| control) | 01.00_0.00 0 | 20100-210 - 0 | 23100_1120 G | | | |
| Diet 2 (mesquite) | 46.60±1.50 b | 33.80±1.80 b | 40.20±1.47 c | 3 | | |
| Diet 3 (date paste) | 39.30±6.77 c | 43.30±4.49 a | 41.30±4.84 b | 2 | | |
| Diet 4 (corn gluten) | 13.60±4.69 e | 15.88±3.49 d | 14.74±1.78 e | 5 | | |
| Diet 5 (Feedbee [®]) | 56.30±4.21 a | 40.55±4.74 a | 48.43±4.37 a | 1 | | |

| able 3 - Preference of pollen sul | stitute diets administrated to honey | bee colonies in 2014 and 2015 in Riyadh. |
|-----------------------------------|--------------------------------------|--|
|-----------------------------------|--------------------------------------|--|

*Means followed by the same letter within the column are not significantly different at 0.05.

**S. D= standard deviation.

*** Ro. = Ranking order.

Consumption of proteinaceous diets

Consumption of each tested diet fluctuated during the period of feeding in late summer. Data obtained during two successive years (2014 and 2015) were illustrated in Figure 1. The obtained data at first season 2014 revealed that commercial pollen substitute Feedbee[®] (diet 5) and date paste (diet 3) were consumed more than mesquite (diets 2) and corn gluten (diet 4), respectively. The treatments could be arranged descending according to their consumption means as follows: diet 5 (157.92 g), diet 3 (129.74 g), diet 2 (116.85 g) and diet 4 (71.44 g). Significant differences in feed consumption were achieved among all tested diets (p<0.05).

Consumption rates of 2015 showed a similar trend. The highest consumed diet was diet 5 (148.33 g). The other tested diets were consumed in lower quantities with significant differences among them (p<0.05). The highest consumption was recorded in honey bee colonies fed with Feedbee[®] and it was equal to 1.2, 1.3 and 1.8 fold of amount consumed from date paste, mesquite and corn gluten diets, respectively. In this approach, Hanna and Schmidt (2004) stated that phagostimulants consist of matching set of diverse components that additively or synergistically act to exceed a threshold level of stimulation necessary for

feeding. The addition of 5% pollen grain to pollen substitute increased the past consumption significantly (AMRO et al., 2016). However, the same previous authors, reported that honey bee colonies encouraged to consume pollen substitutes in fully absence condition of natural protein course (pollen). On the other hand, Alqarni and Alatawi (2008) in Saudi Arabia reported that the addition of 0.03% coriander oil to the artificial diet promote the bees to consume more amount of introduced pollen substitutes.

The present results were matches with Saffari et al. (2010) who stated that Feedbee[®] showed highly palatable and greater consumption than the other feedings introduced to honey bees, especially during the shortage or absence of pollen sources. In the present study, corn gluten consumption rates were recorded as the last ranking order all over the study period. In the same line, Amro et al. (2016) recorded the lowest consumption rate for corn gluten diet when compared with another pollen substitute's diet. Additionally, Irandoust and Ebadi (2017) found that honey bees prefer to consume higher amounts of pollen cakes, while bread yeast and soybean cakes were consumed more than soybean meal and wheat gluten.



Figure 1 - Consumption of pollen substitute diets by honey bee colonies during 2014 and 2015 seasons in Riyadh. The letters indicate the significant differences between means according to Duncan's multiple range test 0.05.

Impact of proteinaceous diets on sealed brood areas

Data of sealed brood areas of two successive seasons (2014 and 2015) are depicted in Figure 2. The results of first year (2014) indicated that seasonal fluctuations of sealed brood areas were similar. It started with low record at mid-August in all colonies groups. Mean of sealed brood area of control colonies decreased from 73.8 sq. inch²/colony to reach the lowest level (61.4 sq. inch²/colony) in mid-October, 2014. The same trend was observed in 2015. The resurgence in the brood rearing activity was noticed during the feeding period by recording both the highest measurements at mid-October during study seasons (2014 and 2015).

When the tested diets were offered to honey bee colonies, the sealed brood areas started to increase in all treatments and responded to the kind of tested diet. The statistical analysis showed significant difference (p<0.05) among the compared treatments at the inspected dates of October, 2014. The superior response had been obtained when diet 5 (Feedbee[®]) was used for feeding the colonies (77.15 sq. inch²/colony) followed by diet 2 (mesquite mixture, 69.72 sq. inch²/colony) and diet 3 (date past, 63.08 sq. inch²/colony), with significant difference (p<0.05) among all the arrangement of the compared diets followed the same respective order of previous studies.

In the present study, the treatments were applied in late summer of 2014 and 2015, when the natural pollen sources were rather scarce in the study region as detected before by Al-Ghamdi (2002). When the data of both seasons were taken into consideration, it is obvious to record the lowest values of sealed brood areas for all the compared colonies during the first two readings of August. This status was recorded at starting of proteinaceous diets administration, when the natural feeding sources diminished from the field and the reverse weather factors affect the honey bee activity as described previously by Al-Ghamdi (2002).

In the present investigation, brood area was gradually increased after supplementary feeding in all tested honey bee colonies except the control (un-fed) throughout the trial period. These results are confirmed by the findings of Srivastava et al. (2004), Dastouri et al. (2007) and Pande et al. (2011), which reported that the sealed brood area increased after honey bee supplementary feeding comparing to un-fed colonies. However, Mohebodini et al. (2013)

showed that scarcity of pollen source in beekeeping areas lead to decrease of brood areas in honey bee colonies. According to Zerbo et al. (2001) and Hoover et al. (2006), protein is a structural and functional constituent of tissues, thus playing an important role in brood rearing and in the satisfactory development of adult bees. Therefore, honey bee (*Apis mellifera* L.) colonies required 29.5-34.0% dietary protein content before mixing with sucrose and water for maximum population growth and maximum worker quality (HERBERT JR. et al., 1977). Contrastingly, the previous authors recorded that extra protein content (more than 50%) decreased brood rearing because it may inhibit the absorption of other nutrients.

In the present investigation, Feedbee[®] and mesquite diet highly and effectively stimulated bees to rear more brood and increase sealed brood area. Also, Saffari et al. (2010) and Amro et al. (2016) recorded the highest brood areas in honey bee colonies fed with Feedbee[®] in comparison with the rest of pollen substitutes. In addition, the mesquite pods flour diet come in the second ranking order after Feedbee[®] in this study. This might be due relatively to the high protein content in mesquite (AMRO et al., 2016) and phagostimulative effect of coriander oil (ALQARNI; ALATAWI, 2008), which induced high royal jelly production, resulting in increasing the ability of nursing workers to feed and rear more brood.

In the present investigation, date past (diet 3) had a good impact on sealed brood areas in tested colonies in compare with control (un-feed). Also, Amro et al. (2016) found that date paste diet was significantly better than corn gluten diet or control colonies (un-feed) in its impact on brood rearing activity under isolation condition. However, corn gluten diet recorded the lowest sealed brood areas in this study. Similarly, Irandoust and Ebadi (2017) mentioned that dietary inclusion of wheat gluten supplements caused the lowest bee mass reduction, and respective colonies had the greatest laying area at the end of winter.



Figure 2 - Average of sealed brood areas in honey bee colonies fed with different pollen substitute diets during 2014 and 2015 in Riyadh. The letters indicate the significant differences between means according to Duncan's multiple range test 0.05.

Impact of proteinaceous diets on storing bee bread

A remarkable increase of stored bee bread area was recorded under feeding conditions for all tested diets during 2014 and 2015 (Figure 3). In general, a significant increment (p<0.05) was recorded for diet 5 followed by diet 2 and 3, while diet 4 presented the lowest increment. The un-fed colonies (control) were arranged in the last rank at the same inspecting dates.

During the second year of study (2015), data revealed that the bee bread areas were also low at the beginning of the season, and gradually increased under supplementary feeding until the end of the season. The highest stored bee bread areas were recorded during mid-October. Feedbee[®] and mesquite diets took the first and second ranking orders, respectively. The remaining diets revealed the same trend of those obtained during 2014 season. The general mean of bee bread areas showed that hives fed with Feedbee[®] produced the highest square area, with an average of 44.87 sq. inch²/colony. The rest of the tested diets enabled honey bee to collect the following stored pollen areas: diet 2 (mesquite), 40.63; diet 3 (date paste), 34.70; and diet 4 (corn gluten), 31.03 sq. inch²/colony, respectively. So, both diets 5 and 2 were superiors in the effect and/or enhancement on stored pollen areas during late summer of 2014 and 2015. However, diet 4 failed to induce such enhancement. In the present investigation, diet 5 (Feedbee[®]) and diet 2 (mesquite) were superior in their effect in enhancement of pollen collecting and storing, and control colonies (un-fed) were the lowest one. The present results are in accordance with data of Srivastava et al. (2004), Sihag and Gupta (2011), and Pătruică et al. (2013). They reported that after feeding of pollen supplement, pollen hoarding capacity of bee colonies was better than the control. These findings suggest that the colonies continuously produce and rear more brood, and forage for more pollen and nectar. Also, Mattila and Otis (2006) recorded that colonies that supplemented with pollen or a pollen substitute in the spring started to collect pollen and rear brood earlier than control colonies (un-fed) and produced the most workers by late April or early May, which were able to collect pollen.



Figure 3 - Average of stored bee bread areas in honey bee colonies fed with pollen substitute diets during 2014 and 2015 in Riyadh. The letters indicate the significant differences between means according to Duncan's multiple range test 0.05.

Correlation coefficient between honey bee colonies performance and proteinaceous diet consumption rate

Correlation coefficient (r) values between honey bee colonies performance parameters and proteinaceous diet consumption rates during 2014 and 2015 seasons were calculated and presented in Table 4.

| consumption during 2014 and 2015 seasons in Riyadh. | | | | |
|---|------------|-----------|--|--|
| Comparisons | (r) values | | | |
| | 2014 | 2015 | | |
| Consumption rate x brood area | + 0.543** | + 0.783** | | |
| Consumption rate x bee bread | + 0.845** | + 0.760** | | |
| Brood area x bee bread area | + 0.457* | + 0.896** | | |

| Table 4 - Correlation coefficient values between honey bee colonies performance parameters and die |
|--|
| consumption during 2014 and 2015 seasons in Riyadh. |

* Significant ** Highly significant (p<0.05).

This data shows a highly positive significant correlation between consumption rate x sealed brood area during 2014 and 2015 seasons. Also, highly positive significant correlation between consumption rates from the tested diets and storing bee bread area was recorded in both seasons of study. Correlation coefficient (r) between sealed brood and bee bread areas showed significant and highly significant values during 2014 and 2015 seasons, respectively. Dependent of the bee bread in the colony as a responsible factor of brood rearing, logically reflects the number and activity of honey bee workers inside the tested colonies. This finding insure the knowledge that colony growth is dependent upon some factors dealing within the colony such as brood rearing ability and other factors resulting from the external conditions, which affect the foraging activity and nectar secretion (CRAILSHEIM; STOLBERG, 1989).

As a result, feeding honey bee colonies with mesquite pods powder (diet 2) and date paste (diet 3) were positivity correlated with enhancement of honey bee activity. These diets constitute from cheap local materials and are widely used by several investigators and recorded as rich protein materials (GOLUBOV et al., 2001), increasing the performance of honey bee colonies (AMRO et al., 2016) and enhancing the physiological condition of workers (AMRO et al., 2020). Data presented in Table 5 show the prices of used materials. The main constituent of mesquite (diet 2) is the mesquite pods present in the wild area surrounding the study location. In addition, the mesquite pods can be taken from the beekeeping market as a

powder for direct use in a price not more than 4 USD/kg. Also, the main constituent of date paste is easy to obtain from the local marketing with not more than 2.67 USD/kg.

The additional materials, as dried skim milk and brewer's yeast that had high nutritional values for honey bee physiology (AMRO et al., 2020), as well as high protein constituents (AMRO et al., 2016), are presented also in the local marketing with a cost of 2.67-6.66 USD/kg each one, respectively. Also, fresh mixed pollen pellets added to honey bee diets were collected during active season using pollen traps. Although, the price of the used vitamins (Centerum[®]) is not expensive (0.59 USD/kg) when used in low quantities. Honey, powdering sugar and coriander oil also can be used as attractant agents for honey bee, as mentioned before by Alqarni and Alatawi (2008).

| Treatment | Required amount of raw material (g) for ca 1 kg. of pollen substitute | | Cost of raw material | Cost of raw material used in 1 kg of | Total cost |
|------------------------|---|-----------------|-------------------------|--|-----------------|
| incutinent | Material required | Quantity (g) | (USD/kg) | pollen substitute (USD) | (USD / 1 kg) |
| | Flour | 258 | 4 | 1.03 | |
| | Dried skim milk | 86 | 6.66 | 0.57 | |
| | Brewer's yeast | 86 | 2.67 | 0.23 | |
| Mesquite | Sugar powder | 386 | 1.47 | 0.56 | |
| (Prosopis | Pollen | 43 | 8 | 0.35 | 3.75 |
| juliflora) | Centrum®* | 4 | 143.96 | 0.59 | |
| | Coriander oil | 9 | 31.99 | 0.29 | |
| | Honey | 17 | 8 | 0.13 | |
| | Water | 112 | - | - | |
| | Date past | 274 | 2.67 | 0.72 | |
| | Dried skim milk | 91 | 6.66 | 0.61 | |
| Date paste | Brewer's yeast | 91 | 2.67 | 0.24 | |
| | Sugar powder | 411 | 1.47 | 0.61 | |
| (Phoenix | Pollen | 46 | 8 | 0.37 | 3.69 |
| dactylifera) | Centrum® | 5 | 143.96 | 0.72 | |
| ,,,,, | Coriander oil | 9 | 31.99 | 0.29 | |
| | Honey | 18 | 8 | 0.13 | |
| | Water | 55 | - | - | |
| | Flour | 379 | 39.99 | 15.17 | |
| Feedbee ^{®**} | Sucrose solution (2:1 w:w) | 606 | 1.33 | 0.80 | 16.10 |
| | Honey | 15 | 8 | 0.13 | |

Table 5 - Cost of pollen substitute patties

* Multivitamin from A to Zink[®], produced by Pfizer (formerly Wyeth).

**Commercial substitute tested by Saffari et al. (2004).

CONCLUSION

Feeding honey bee colonies with mesquite pods powder (*Prosopis juliflora*), date paste (*Phoenix dactylifera*) and commercial pollen substitute (Feedbee[®]) was found to enhance the activity of honey bee workers and promote colonies to rear brood, collect pollen and stored more bee bread areas at late summer and early autumn season. The activity of brood rearing would be resulted in much more honey bee population and then allowed to get strength

colony at the following spring. The high price of the imported commercial diet (Feedbee[®]), as well as the unavailability in the local marketing, makes the mesquite and date paste diets more robust and the better choice for beekeepers, especially because their components are available in the local marketing with a cheap price.

IMPACTO DO USO DE CERTOS SUBSTITUTOS DO PÓLEN NO DESEMPENHO DE COLÔNIAS DE ABELHAS (*Apis mellifera* L.) SOB CONDIÇÕES AMBIENTAIS ADVERSAS

RESUMO

s grãos de pólen são a principal fonte de proteína para as abelhas. Na região central da Arábia Saudita, a falta desta dieta natural durante as estações secas no verão é a principal preocupação para a indústria apícola. O efeito do uso de certas proteínas, como alternativa alimentar, no desempenho, quantidade de consumo, atividade de criação de ninhadas e armazenamento de pão de abelha, de colônias de abelhas, foi testado durante o final do verão nas condições da região de Riade. Farinha de soja, farinha de vagem de algaroba, pasta de tâmara, glúten de milho e as dietas Feedbee® foram os principais produtos testados no presente estudo. A preferência foi determinada por meio de testes de múltipla escolha (TMC). Os resultados obtidos revelaram que as abelhas preferiram primeiramente o produto comercial Feedbee[®] seguido das dietas com pasta de tâmara e algaroba. Além disso, a maior quantidade de consumo foi registrada em colônias de abelhas alimentadas com Feedbee[®] e foi aproximadamente igual a 1,2, 1,3 e 1,8 vezes as quantidades consumidas de dietas de pasta de tâmara, algaroba e glúten de milho, respectivamente. Foi estudado o efeito das dietas testadas na atividade de criação de ninhadas de colônias de abelhas durante dois anos sucessivos. Os resultados mostraram que a melhor resposta foi obtida usando Feedbee® seguido por dietas de algaroba e pasta de tâmara. Porém, o Feedbee[®] tem custo elevado e não está disponível nos mercados locais da Arábia Saudita. Portanto, os apicultores são aconselhados a usar farinha de vagens de algaroba e pasta de tâmaras como substituto do pólen nos períodos de escassez, para melhorar a criação de ninhadas de colônias de abelhas durante o final do verão.

Palavras-chave: Mel de abelhas. Dietas substitutas. Feedbee[®]. Farinha de vagens de algaroba. Pão de abelha.

IMPACTO DEL USO DE CIERTOS SUSTITUTOS DEL POLEN EN EL RENDIMIENTO DE COLONIAS DE ABEJAS MELÍFERAS (*Apis mellifera* L.) BAJO CONDICIONES AMBIENTALES ADVERSAS

RESUMEN

os granos de polen son la principal fuente de proteínas para las abejas melíferas. En la región central de Arabia Saudita, la falta de esta dieta natural durante las estaciones ■secas en verano es una preocupación principal para la industria apícola. Se probó el efecto del uso de ciertas proteínas alternativas sobre el rendimiento, la cantidad de consumo, la actividad de cría y el almacenamiento de pan de abeja de las colonias de abejas melíferas a fines del verano en las condiciones de la región de Riad. La harina de soja, la harina de vainas de mezquite, la pasta de dátiles, el gluten de maíz y las dietas Feedbee® fueron el principal material probado en el presente estudio. La preferencia se determinó mediante pruebas de opción múltiple (POM). Los resultados obtenidos revelaron que las abejas melíferas prefirieron en primer lugar el producto comercial Feedbee[®] seguido de la pasta de dátiles y las dietas de mezquite. Además, el mayor consumo se registró en las colonias de abejas alimentadas con Feedbee[®] y aproximadamente fue igual a 1,2, 1,3 y 1,8 veces las cantidades consumidas de las dietas de pasta de dátiles, mezquite y gluten de maíz, respectivamente. Se estudió el efecto de las dietas probadas sobre la actividad de cría de las colonias de abejas melíferas durante dos años sucesivos. Los resultados mostraron que la mejor respuesta se había obtenido utilizando Feedbee[®] seguido de dietas de pasta de mezquite y dátiles. Pero Feedbee[®] es caro y no está disponible en los mercados locales de Arabia Saudita. Por lo tanto, se aconseja a los apicultores que utilicen harina de vainas de mezquite y pasta de dátiles como sustituto del polen en los períodos de escasez para mejorar la cría de las colonias de abejas melíferas durante el final del verano.

Palabras clave: Miel de abejas. Dietas sustitutivas. Feedbee[®]. Harina de vainas de mezquite. Pan de abeja.

REFERENCES

ABBASIAN, A. R.; EBADI, R. Nutritional effect of some protein sources on longevity, protein and fat body of bee workers (*Apis mellifera* L.). Journal of Crop Production and Processing. v. 6, n. 2, p. 149-158, 2002.

AL-GHAMDI, A. A. The effect of pollen supplementary feeding on some activates of honey bee colonies during summer season in Riyadh, Saudi Arabia. **Saudi Journal of Biological Sciences**, v. 9, n. 2, p. 85-94, 2002.

AL-GHAMDI, A. A.; AL-KHAIBARI, A. M.; OMAR, M. O. M. Effect of honey bee race and worker age on development and histological structure of hypopharyngeal glands of honey bee. **Saudi Journal of Biological Science**, v. 18, n. 2, p. 113-116, 2011.

ALQARNI, A. S. **Morphometrical and biological studies of the native honeybee race**, *Apis mellifera* L.; the Carniolan, *Apis mellifera carnica* Pollmann, and their F1 hybrid. Riyadh: KSU, 1995. Thesis, College of Food Science and Agriculture, King Saud University, 1995.

ALQARNI, A. S.; ALATAWI, F. J. Use of some volatile oils as attractive agents to increase food consumption in honey bee colonies. **Arab Universities Journal of Agricultural Sciences**, v. 16, n. 1, p. 203-210, 2008.

AL-SHARHI, M. M. M. **Effect of feeding types and its seasonal timing on activities of honey bee colonies (***Apis mellifera carnica* **hybrid)**. Riyadh: KSU, 2008. Thesis, College of Food Science and Agriculture, King Saud University, 2008.

AMRO, A.; OMAR, M.; AL-GHAMDI, A. Influence of different proteinaceous diets on consumption, brood rearing, and honey bee quality parameters under isolation conditions. **Turkish Journal Veterinary of Animal Sciences**, v. 40, n. 3, p. 468-475, 2016.

AMRO, A.; YOUNIS, M.; GHANIA, A. Physiological effects of some pollen substitutes diets on caged honey bee workers (*Apis mellifera* L.). **International Journal of Environment**, v. 9, n. 1, p. 87-99, 2020.

CRAILSHEIM, K.; STOLBERG, E. Influence of diet, age and colony condition upon intestinal proteolytic activity and size of the hypopharyngeal glands in the honey bee (*Apis mellifera* L.). **Journal of Insect Physiology**, v. 35, n. 8, p. 595-602, 1989.

DASTOURI, M. R.; MAHERI-SIS, N.; AGHAJANZADEH-GOLSHANI, A.; EBRAHIM-NEZHAD, Y. The effect of replacement feeding of some protein sources with pollen on honey bee population and colony performance. **Journal of Animal and Veterinary Advances**, v. 6, n. 11, p. 1258-1261, 2007.

DEGRANDI-HOFFMAN, G.; CHEN, Y.; HUANG, E.; HUANG, M. H. The effect of diet on protein concentration, hypopharyngeal gland development and virus load in worker honey bees (*Apis mellifera* L.). **Journal of Insect Physiology**, v. 56, n. 9, p. 1184-1191, 2010.

DE JONG, D.; SILVA, E. J.; KEVAN, P. G.; ATKINSON, J. L. Pollen substitutes increase honey bee haemolymph protein levels as much as or more than does pollen. **Journal of Apiculture Research**, v. 48, n. 1, p. 34-40, 2009.

DUNCAN, D. B. Multiple range and multiple F tests. Biometrics, v. 11, n. 1, p. 1-42, 1955.

GOLUBOV, J.; MANDUJANO, M. C.; EGUIARTE, L. E. The paradox of mesquites (*Prosopis* spp.): invading species or biodiversity enhancers? **Boletín de la Sociedad Botánica de México**, v. 69, p. 23-30, 2001.

HANNA, A.; SCHMIDT, J. O. Effect of phagostimulants in artificial diets on honey bee feeding behavior. **Southwestern Entomologist**, v. 29, n. 4, p. 253-261, 2004.

HERBERT JR., E. W.; SHIMANUKI, H.; CARON, D. Optimum protein levels required by honey bees (Hymenoptera, Apidae) to initiate and maintain brood rearing. **Apidologie**, v. 8, n. 2, p. 141-146, 1977.

HOOVER, S. E. R.; HIGO, H. A.; WINSTON, M. L. Worker honey bee ovarian development: seasonal variation and the influence of larval and adult nutrition. **Journal of Comparative Physiology**, v. 176, n. 1, p. 55-63, 2006.

IRANDOUST, H.; EBADI, R. Nutritional effects of high protein feeds on growth, development, performance and overwintering of honey bee (*Apis mellifera* L.). **International Journal of Advanced Biological and Biomedical Research**, v. 5, n. 4, p. 178-184, 2017.

JEFFREE, E. P. A shaped wire grid for estimating quantities of brood and pollen in combs. **Bee World**, v. 39, n. 5, p. 115-118, 1958.

KIRK, P. L. Kjeldahl method for total nitrogen. **Analytical Chemistry**, v. 22, n. 2, p. 354-358, 1950.

MATTILA, H. R.; OTIS, G. W. The effects of pollen availability during larval development on the behaviour and physiology of spring-reared honey bee workers. **Apidologie**, v. 37, n. 5, p. 533-546, 2006.

MOHEBODINI, H.; TAHMASBI, G.; DASTAR, B.; AHANGARI, Y. J.; ZEREHDARAN, S. Effect of dietary thiamine on growth of the Iranian honey bee colonies (*Apis mellifera meda*) in different seasons. **Agriculture & Forestry**, v. 59, n. 3, p. 119-126, 2013.

PANDE, R.; FIRAKE, D. M.; KARNATAK, A. K. Development of pollen substitutes for dearth period management of honey bee (*Apis mellifera* L.) colonies in foothills of Shivalik range of Himalayas. **Indian Journal of Agricultural Sciences**, v. 81, n. 9, p. 861–866, 2011.

PÅTRUICÅ, S.; POPOVICI, D.; COLIBAR, O. Researches on the influence of some apicol stimulators use in the supplemental feeding of honey bee colonies. **Scientific Papers: Animal Science and Biotechnologies**, v. 46, n. 1, p. 277-280, 2013.

ROGALA, R.; SZYMAS, B. Nutritional value for bees of pollen substitute enriched with synthetic amino acids. Part I. Chemical methods. **Journal of Apicultural Science**, v. 48, n. 1, p. 19-27, 2004.

SAFFARI, A. M.; KEVAN, P. G.; ATKINSON, J. L. A promising pollen substitute for honey bees. **American Bee Journal**, v. 144, n. 3, p. 230-231, 2004.

SAFFARI, A. M.; KEVAN, P. G.; ATKINSON, J. L. Palatability and consumption of pattyformulated pollen and pollen substitutes and their effects on honey bee colony performance. **Journal of Apicultural Science**, v. 54, n. 2, p. 63-71, 2010.

SAS INSTITUTE. The SAS System Version 9.1.3. SAS Institute, Cary, NC, 2004.

SHAPIRO, S. S.; WILK, M. B. An analysis of variance test for normality (complete samples). **Biometrika**, v. 52, n. 3-4, p. 591–611, 1965.

SIHAG, R. C.; GUPTA, M. Development of an artificial pollen substitute/supplement diet to help tide the colonies of honey bee (*Apis mellifera* L.) over the dearth season. **Journal of Apicultural Science**, v. 55, n. 2, p. 15-29, 2011.

SRIVASTAVA, B. G.; TIWARI, A.; MEENAKSHI, M. Development of pollen supplements for *Apis cerana indica* Fabricius. **Indian Journal of Entomology**, v. 66, n. 2, p. 121-123, 2004.

SZYMAS, B.; MALISZEWSKA, R. The condition of honey bee feed pollen substitute enriched with methionine and lysine amino acids. **Scientific Papers Series D. Animal Science**, v. 1, p. 47-53, 1999.

TAWFIK, A. I.; AHMED, Z. H.; ABDEL-RAHMAN, M. F.; MOUSTAFA, A. M. Influence of winter feeding on colony development and the antioxidant system of the honey bee, *Apis mellifera* L. **Journal of Apicultural Research**. v. 59, n. 5, p. 752-763, 2020.

ZERBO, A. C.; LÚCIA, R.; MORAES, M. S.; BROCHETTO-BRAGA, M. R. Protein requirements in larvae and adults of *Scaptotrigona postica* (Hymenoptera: Apidia, Meliponinae): midgut proteolytic activity and pollen digestion. **Comparative Biochemistry and Physiology Part B: Biochemistry and Molecular Biology**, v. 129, n. 1, p. 139-147, 2001.

> Corresponding author: Abdulraouf Mohamed Amro. Agricultural Research Center, Dokki, Giza, 12619, Egypt. raoof_amro@yahoo.com