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Nutritional Effects of Supplementary Diets on Colony Build up , Brood Development and Honey Production of *Apis mellifera* L.

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ABSTRACT

The study was conduct to determine the effect of artificial diet on colony build when enough floral resources were not available. Six diets were prepared which included Black gram (20)+ Yeast (20g) + Pollen (20g) + Honey (20g) + Glucose (20g), Soybean flour (60g) + Honey (35g) + Yeast (4g) + Vitamins (1 g/kg) HAU Diet, Defatted soya flour (20g) + skimmed milk powder (25g) + sugar (5g) + pollen (5g)+glucose (10g) + h, Soybean flour (25 g) + Yeast (10 g) + Pollen (15 g) + Skimmed milk powder (5 g) + Honey (22.5 g) + Sugar (22.5 g) GBPUA and T diet, Brewer's yeast (42 g) + Gram (4g) + Skimmed Milk Power (4g) + Sugar (50g) + Pollen (10 g) PAU Diet and Control (sugar feeding). The studies revealed that *Apis mellifera* bees exhibited differential consumption pattern during different weekly intervals throughout the season as per the needs of the colony. The preference of different diets was in the order 3 >4>2>5>1>6. Diet 3 was most preferred followed by 4, 2, 5, 1 and 6 respectively. In case of diet 1 maximum brood area was observed on 6th July 2017 (1802.8 cm^2) which decreased thereafter (1018.6 cm^2) on 20th July 2017 and minimum brood was observed on 14th September 2017 (107.3 cm^2). On average overall brood reared on this diet during the entire season was (526.80 cm^2). There was significant impact of effect of feeding artificial diets on the number of frames covered by bees. The feeding of artificial diet was helpful to maintain the strength of bee colonies. It was very interesting to find that all the diets were superior over control.

Keywords *Apis mellifera,* Protein diet, Bee flora, Beekeeping.

INTRODUCTION

Honeybees besides sweet honey peoduce a number of products such as beeswax, royal jelly bee venom and propolis. They are important pollinators of a wide variet of crops increasing productivity of agricultural crops through cross pollination (McGregor 1976, Sihag 1986, Free 1993). Besides, increasing the productivity of ctros, they are known for significantly improving the quality and quantity of crops. It has been recognized as one of the essential inputs in agriculture (Sihag 2001). However, the success of beekeeping depends upon the prevailing weather conditions, availability of bee flora and management practices. Proper manipulative and management prac-

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tices are required for honey production and successful pollination in an area.

The rainy period from July to August is the Most difficult time for bees as floral dearth coupled with attack of diseases and enemies require extra management practices like providing ollen substitutes or articicial diets. Several investigators have attempted different types of pollen substitutes/ diets for honeybee colonies during floral dearth periods such as pollen substitutes with skimmed milk whey and wheat, soy products, brewer's yeast, fish meal and meat scraps. (Haydak 1967, Zaytoon *et al.* 1988 Rana *et al.* 1996, Herbert, 1979 Herbert and Shimanuki 1979, Haydak 1967, Standifer *et al.* 1978, Kulincervic *et al.* 1982, Abdelatif *et al.* 1971, Herbert and Shimanuki 1979, Chalmers 1980).

In northern part of our country, summer season is little longer and harsh as compared to other parts. The natural bee flora starts disappearing in the month of may causing dearth of food (pollen and nectar) for bees. The dearth periods result into low nutrional reserves which seroiu; y affects the egg laying and brood rearing activity. Also due to poor strength, colonies may be attacked by various bee enemies like black ants, bee eating birds and / Varroa mite. All these factors lead to quick dwindling and sometimes even perishing of bee colonies. To avoid this situation, colonies are either physically moved to a bee flora rich area or provision of feeding artificial diet in form of pollen substitutes and supplements should be made.

The necessity of artificial diets to honey bees has been long-standing interest to the beekeeping industry (Haydak 1935, 1936, Standifer *et al.* 1960). Haydak (1967) observed a variety of pollen substitutes and reported mixture containing soy flour, brewer's yeast and skimmed milk powder as best formulation. Different type of artificial diets have been formulated and their effect on various colony parameters were observed by several foreign (Standifer *et al.* 1960, Doull 1968, Stranger and Gripp 1972, Herbert and Shimanuki 1978, Saffari *et al.* 2010a,b, De Grandi-Hoffman G. *et al.* 2008) and Indian (Chhuneja *et al.* 1993, Srivastava 1996, Sihag and Gupta 2011, Kumar *et al.* 2012) researchers It was reported that in some instances pollen substitute diets were even better than pollen/pollen supplement with regards to acceptability and nutritional value for honey bees. Some commercial pollen substitutes are available in international market, but they are formulated and tested in different eco-climatic conditions and their efficacy may not be satisfactory.

The artificial feeding or supplementing pollen substitutes help to sustain colonies for hobey production and pollination. Strong colonies produce more honey and serve as better pollinators. This is possible if sufficient brood stores, nectar pollen and bee population is available. Frurthermore, provision of artificial diets is especially important in areas where stationery beekeeping is practiced (Adbellatif *et al.* 1971, Herbert and Shimanuki 1980, 1983, Herbert 2000, Nabors 2000, Van der Steen 2007, De Grandii-Hoffman *et al.* 2008, De Jong *et al.* 2009,Saffari *et al.* 2010a, Sihag and Gupta 2011).

The productivity and effective nees of bees in an area depends on the bee breed and bee forage, whereas the foraging index is the key factor, which determines the performance of a colony. Honey bees increase yields of cross pollinated crops, thus increasing farm incomes and provide employment to the landless and unemployed rural people (Goyal 1989). Honey production and pollination potential deends upon the foraging efficiency of bees. The foraging pattern determines the strength of colonies, honey stores, pollen stores, number of bees. Brood rearing depends upon the availability of pollen and nectar, as also on climate factors prevalent in the locality. The nectar and pollen stores and supplementary feeding influences the colony foraging activity of colonies. Besides these, the foraging activity of colony is also influenced by the abiotic factors. The colony strength has also bearing on the incidence of various disease and natural enemies.

The state is suitable place for beekeeping owing to its topography, temperate climate and conducive environment for bees. Honey bees depend on pollen and nectar for their food but these provisions are not available to the bees right through the year, which causes the depletion of the on hand honey stores in the colony. There may also be a shortage of pollen, bees then reduce the amount of brood that they rear

resulting in quick dwindling and perishing of honey bee colonies. The major problem in beekeeping is to maintain good strength of honeybee colonies during dearth periods (summer and winter) as enough bee flora is not available during this period. The condition is more severe in hilly areas where dearth periods are little longer as compared to other parts sometimes resulting in the dearth of bee colonies. Therefore during these periods, special care should be taken in management of bees in terms of feeding artificial diets (Haydak 1935, 1936). Different diet formulated have been tested by several investigators. In India, work on the artificial diet formulation has been carried and various substitutes have been suggested with different compositions has been suggested (Chhuneja *et al.* 1992, Srivastava 1996, Sihag *et al.* 2011). The studies are therefore proposed with an attempt to compare the effect of various diet formulations fed to bee colonies during dearth periods on colony parameters and diseases and enemies incidence so that suitable pollen substitute can be developed to improve beekeeping practice in the state.

A number of attempts on formulation of suitable diet for honeybees during dearth period have been made by various workers, majority of them are from abroad and very few from India, but a standard and well accepted formulation is still not available in our country. During the current study, efforts were made to develop a highly palatable, nutritionally balanced and economically viable artificial diet for honeybees (*Apis mellifera*) in north Indian conditions. To assess the relative acceptance and consumption of stimulative feeding during dearth period, two artificial diets were formulated.

MATERIALS AND METHODS

The study was conduct to determine the effect of artificial diet on colony build, effect of different artificial diets on the incidence of diseases and enemies and qualitative and quantitative analysis of pollen loads to determine the major floral sources of *A.mellifera* in the university apiary located at experimental farm of Sher-e-Kashmir university of Agricultural Sciences and Technology Jammu at Chatha during the period 2017- 2018.

Experimental site

The experimental site is located in the sub- tropical zone at 32.73°N and longitude of 74.87°E. It has an average elevation of 327m from the mean sea level. Most of the rains are received during July to October (about 70%). The annual mean maximum and minimum temperature are 29.60 and 16.70°C, respectively. Summer months are hot with temperature and humidity ranging from 23.50 to 35.50°C and 53.0 to 73.50%, respectively. The winter months experience mild to severe cold conditions with average temperature ranging from 6.50 to 21.70°C. December is the coldest month, when minimum temperature touches 4°C.The highest temperature is recorded in the month of June (4°C).The daily maximum and minimum temperature and evaporation rate rise from March onwards. The detailed material sand methods employed for these studies are reported in this chapter objective wise.

Details of experiments

Effect of artificial diet on colony build up

The experiment shall be conducted in the Apiary, Division of Entomology. The following pollen substitute treatments on weight basis shall be administered to three colonies in each treatment, which served as replicates.

Bee strength of experimental colonies:

- 6 bee-frame
- 8 bee-frame
- 10 bee frame

No. of colonies to be examined =
24 (8 each in 6, 8 and 10 bee-frame bee strength)

Diet -I Defatted soya flour 20g + skimmed milk powder 25g + sugar 5g + pollen (5g)+ glucose 10g + honey 35g

Diet –II Black gram (20)+ Yeast (20g) + Pollen (20g) + Honey (20g) + glucose (20g)

Diet –III Brewer's yeast (42 g) + Gram (4g) + Skimmed Milk Power (4g) + Sugar (50g) + Pollen (10 g) PAU Diet

Diet –IV Soybean flour (60 g) + Honey (35 g) + Yeast (5g) + Vitamins (1 g/kg) HAU Diet

Diet-V Soybean flour (25 g) + Yeast (10 g) + Pollen (15 g) + Skimmed milk powder (5 g)

+ Honey (22.5 g) + Sugar (22.5 g) GBPUA and T diet
Diet -VI Control (sugar feeding)

Days to consume the diet

Provision of feeding the diet formulations to honeybees shall be made in the form of patties for a feeding period of 14 days. Weighed amount of patties shall be fed to *Apis mellifera* colonies by top bar method of feeding. The number of days taken by bees to consume the diet fully shall be recorded. Observations shall be recorded on daily basis to find out which diet is consumed fully. The least number of days taken to complete the diet can be an indication of its palatability and feeding preference to bees. In order to estimate the effect of diet formulations, the overall performance of colonies shall be judged by observing and comparing all the parameters viz, sealed and unsealed brood, egg laying, honey stores, and bee activity with control colonies. For studying the economics of feeding the pollen substitutes and supplements, the prices of the feedstuffs used in the various formulations shall be collected from the market and cost per kg of each of the pollen substitute and supplement shall be calculated.

Effect of artificial diet on seasonal incidence of diseases and enemies

The seasonal incidence of natural enemies shall be observed weekly throughout the study period. The observations on the incidence of enemies and diseases shall be recorded at weekly interval. Each sample shall consist of minimum of 20 larvae, pupae and adults from each colony and 20g debris from each colony. The collected samples shall be analyzed for mites and associated pathogens if any. The samples of the honeybee, *A. mellifera* shall be examined for the presence of mites. Each sample consisted of 100 suspect bees along with a part of the comb containing immature stages of bees and stored food materials. Samples shall be collected randomly from each hive at fort nightly intervals and examined directly under a stereo binocular for the presence of mites. Besides, hive debris shall also be collected and examined for the presence of mites. The collected mites shall be preserved in 70% alcohol and permanent slides prepared using Hoyer's medium. The mites shall be identified with the help of available literature and the identifications later got confirmed from the mite taxonomists.

The combs of *A. mellifera* shall be examined for the presence of eggs, larvae, pupae and adults of wax moths. If any of the stage shall be found in debris of combs and /or on combs, the hive shall be considered to be infested. Based on this the seasonal incidence pattern shall be worked out. The debris combs containing eggs/larvae shall be maintained in glass jars covered with muslin cloth and supplemented with the wax from old combs as food for developing larvae. The bee's population in the apiary shall be recorded by visual counts while walking from one corner of the apiary to the other. Weekly observations recorded thrice a day (morning, noon and evening) and diurnal fluctuation shall also be recorded.

RESULTS

Impact of artificial diets on colony build up, brood development add honey stores

The data presented in Table 1 revealed that *Apis mellifera* bees exhibited differential consumption pattern during different weekly intervals throughout the season as per the needs of the colony. The preference of different diets was in the order 3 >4>2>5>1>6. Diet 3 was most preferred followed by 4, 2, 5, 1 and 6 respectively. The results revealed that net consumption of diet 3 was (67.85) followed by 4 (61.06 g). The mean consumption of other diets 2, 5 and 1 was 52.44, 47.86 and 45.72 g per colony, respectively. All these diets were superior ove control (25.74). Analysis of data revealed that differences were statistically significant both for diets as well as time periods. The data further revealed that the consumption of diets was minimum on 6th July which increased gradually attaining peak on 31st August and consumption decreased thereafter and was minimum on 2nd November.

There was seasonal impact of diet on brood development (Table 2). In case of diet 1 maximum brood area was observed on 6th July 2017 (1802.8 cm^2) which decreased thereafter (1018.6 cm^2) on 20th July 2017 and minimum brood was observed on 14th September 2017 (107.3 cm^2). On average overall

Table 1. Amount of artificial diets consumed by *Apis mellifera* during 2017-18.

Feeding interval	Qt. Given	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Mean
6th Jul 2017	200	28.1	26.4	33.8	35.0	21.8	9.0	25.68
20 Jul	200	42.0	53.8	55.6	42.2	42.4	12.3	41.38
3rd Aug	200	39.6	51.2	69.2	58.8	48.2	18.2	47.53
17th Aug	200	97.2	104.6	124.5	123.4	94.4	67.8	101.98
31 Aug	200	93.6	103.3	138.3	118.6	96.8	66.8	102.90
14 Sep	200	80.0	73.2	88.4	73.6	70.0	43.0	71.36
28 Sep	200	38.9	49.3	59.8	56.8	40.8	20.6	44.36
5 Oct	200	21.0	34.3	48.9	45.4	33.9	13.5	32.83
19 Oct	200	12.9	16.3	36.6	35.2	19.6	6.2	21.13
2 Nov	200	3.9	12.0	23.4	21.6	10.7	0.0	11.93
Mean		45.72	52.44	67.85	61.06	47.86	25.74	
CD at 0.05	T (Treatment)		0.09					
	I (Period)		0.12					
	T X I (Treatment X Period)		0.29	``				

brood reared on this diet during the entire season was (526.80 cm^2). In case of diet 2 also maximum brood was observed on 6th and 20th July 2017 and decreased thereafter with minimum during 17th August 2017. On an average brood reared on this diet was 715.47 cm^2. Overall brood development was much higher in case of diet 4 (790.70 cm^2). The brood development was in the order 4>3>2>6>1. The minimum brood was observed in diet 5.The brood development was significantly different in all the treatments.

The results show the significant impact of effect of feeding artificial diets on the number of frames covered by bees (Table 3). The colonies fed with diet 3 and 2 had maximum strength of 5.8 frame of bees. The lowest bee strength (5.1) was observed in colonies fed with diet number 1. There was significant variation in strength of bee colonies with minimum strength during September and October. The feeding of artificial diet was helpful to maintain the strength of bee colonies. It was very interesting to find that all the diets were superior over control. The data presented in Table 4 revealed that artificial diets had significant impact on honey stores. The diet 3 resulted in production of highest honey stores (541.31 cm^2) and diet 5 minimum (295.00 cm^2), the other diets were in between the two. In general the production of honey stores was in the order: 3> 4 >2> 6> 1>5. The studies revealed that all artificial diets

Table 2. Effect of supplementary feeding on amount of sealed brood (cm^2) in *Apis mellifera* colonies 2017-18.

Treatment period	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Mean
6th Jul 2017	1802.8	1884.8	1263.2	1413.0	1393.5	1450.3	1534.60
20 Jul	1018.6	1573.5	830.5	1070.0	849.7	1137.0	1079.88
3rd Aug	327.0	588.0	399.6	462.0	263.3	318.0	392.98
17th Aug	120.3	161.0	396.7	288.3	28.0	167.3	193.60
31 Aug	118.7	178.7	396.0	452.0	48.3	176.0	228.28
14 Sep	107.3	288.0	477.0	488.0	140.4	398.0	316.45
28 Sep	231.3	472.0	788.6	785.3	176.0	487.2	490.06
5 Oct	488.4	577.8	1272.0	1367.0	288.0	588.0	763.53
Mean	526.80	715.47	727.95	790.70	398.40	590.22	
CD at 0.05	T (Treatment)		0.12				
	I (Brood cycle)		0.14				
	T X I (Treatment X Brood cycle)		0.36				

Table 3. Effect of supplementary feeding on number of frames covered by bees 2017-18**.**

Treatment period	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Mean
6th Jul 2017	6.0	6.0	6.0	6.0	6.0	6.0	6.0
20 Jul	5.7	6.0	5.6	5.9	5.8	5.9	5.8
3rd Aug	5.5	5.7	5.8	5.8	5.6	5.7	5.6
17th Aug	5.2	5.3	5.7	5.7	5.0	5.2	5.3
31 Aug	4.8	4.9	5.8	5.7	4.6	4.8	5.1
14 Sep	4.7	4.7	5.5	5.8	4.5	4.7	4.9
28 Sep	4.7	5.0	5.8	5.9	4.5	4.7	5.1
5 Oct	4.7	4.6	6.3	6.0	4.6	4.8	5.1
Mean	5.1	5.2	5.8	5.8	5.0	5.2	
CD 0.05	T (Treatment)		0.3				
	I (Brood cycle)		0.2				
	T X I (Treatment X Brood cycle)		0.8				

were superior over control.

DISCUSSION

The need for artificial diets has been must for sustainable beekeeping (Haydak 1935, 1936). In India, work on the artificial diet formulation has been carried and various substitutes have been suggested with different compositions has been suggested (Chhuneja *et al*. 1992, Srivastava 1996). Kumar *et al.* (2013) studied the preference of six protein rich artificial to *Apis mellifera* colonies in the form of patties and found that containing defatted soy four, brewer's yeast and soy protein hydrolysate powder proved to be most effective to produce higher sealed brood area, total bee strength in terms of frames covered and 9138.6 bee population. Tomar and Singh (2014) fed artificial diet (Sugar solution and pollen substances) to honeybee colonies and found 7.8, 6.2,4.4 and 3.3 frame bee strength after passing dearth period. The colonies whom artificial diet was not provided timely, could not survive till last week of June. Kumar and Agrawal (2014) developed highly palatable, nutritionally balanced and economically viable pollen supplement or substitute for *Apis mellifera*. Diet composed of soy flour-1part, brewer's yeast-1part, soy protein hydrolysate-1part, sugar-1part, glucose-1part proved to be best for honeybees in terms of biochemical composition, net consumption, positive influence on colony parameters and input cost involved. Abd El-Wahab *et al.* (2016) evaluated pollen supplement diet, the main components of orange juice, mint oil,

Table 4. Effect of supplementary feeding on honey stores in *Apis mellifera* colonies 2017-18**.**

Treatment period	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Mean
6th Jul 2017	645.3	815.3	752.7	805.3	795.6	771.80	764.33
20 Jul	562.4	594.0	628.0	698.3	554.2	630.10	611.16
3rd Aug	368.0	293.0	481.7	383.3	277.0	334.49	356.24
17th Aug	206.0	108.0	213.4	276.6	98.4	160.12	177.08
31 Aug	64.0	99.7	141.7	128.3	4.0	90.50	88.03
14 Sep	95.3	182.0	287.0	291.0	96.3	155.70	184.55
28 Sep	500.6	621.5	637.0	610.2	242.2	445.00	509.41
5 Oct	995.0	1220.0	1189.0	999.3	292.3	890.20	930.96
Mean	429.57	491.68	541.31	524.03	295.00	434.73	
CD 0.05	T (Treatment)		0.18				
	I (Period)		0.19				
	T X I (Treatment X Period)		0.50				

Turmeric and Fenugreek powders and vitamins (diet E) traditional pollen supplement diets, A (powdered sugar + sugar syrup), B (diet A+ date pollen grains), C (diet A+ bee honey) and D (date pollen grains + powdered sugar + sugar syrup). The diet E was found to be most preferred one resulting in higher brood production and population of bees.

Abdulraouf *et al.* (2015) tested the efficacy of 5 proteinaceous diets soybean meal, mesquite pod powder, date paste, Feedbee and corn gluten, on honey bees along with control (bees fed naturally on pollens). They found that date paste was most prefeerred followed by Feedbee, mesquite and corn gluten, respectively. Brodschneider and Crailsheim (2010) emphasized on nutritional demands of honey bee workers that strongly affected by shortages of this nutrient. Muhammad *et al*. (2016) reared artificially *Apis mellifera* L. (Hymenoptera: Apidae) at varying diets including royal jelly and reported that survival rates of the larvae and queen bee were higher in 8-g food as compared to 4 and 6g.

Sihag and Gupta (2011) used four pulses viz, soybean (*Glycine max* L. Merr.), mungbean (*Vigna radiata* L. Wilczek), chick pea (*Cicer arietinum* L.) and pigeon pea (*Cajanus cajan* L. Mill SP) for the preparation of artificial diets to feed colonies of honeybee (*Apis mellifera* L.) during the dearth period. Of all these pulses, soyabean was the most preferred with higher brood area and oulation of bees.

Soybean have been recommended as highly palatable proteinaceous diets (Sihag and Gupta 2011). They have been shown to play a substantial role in enhancing activities of colonies (De Groot 1953 and De Grandi Hoffman *et al*. 2008).

Artificial diets played a predominant role in enhanced brood development (Chhuneja *et al.* 1993, Abd El-Wahab *et al.* 2016). Shoreit and Hussein (1993) also reported higher honey production rates in colonies fed with proteinaceous diets as compared to those fed on sugar alone.

Sometimes pollen depression in colonies may be responsible for reduced rates of honey collection (Peixin and BaoHua 2010). Kumar and Agrawal (2013) found a direct correlation between developments of colonies with artificial diets. Mahfouz (2016) reported that colonies fed with artificial diets showed improved rates of honey and pollen collection.

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