

**REARING EFFICIENCY, FORAGING BEHAVIOR AND FOOD
MANAGEMENT OF HONEY BEE (*Apis mellifera* L.)
DURING DEARTH PERIOD**

B. M. ROKONUZZAMAN



**DEPARTMENT OF ENTOMOLOGY
SHER-E-BANGLA AGRICULTURAL UNIVERSITY
DHAKA-1207**

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BY

**B. M. ROKONUZZAMAN
REGISTRATION NO. 14-06334**

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Approved by:

Dr. Mohammed Ali
Professor
Department of Entomology
Sher-e-Bangla Agricultural University
Supervisor

Dr. Mohammed Sakhawat Hossain
Associate Professor
Department of Entomology
Sher-e-Bangla Agricultural University
Co-Supervisor

Dr. Mohammed Ali
Professor
Examination Committee
Department of Entomology
Sher-e-Bangla Agricultural University
Chairman



DEPARTMENT OF ENTOMOLOGY
Sher-e-Bangla Agricultural University
Sher-e-Bangla Nagar, Dhaka-1207
Bangladesh

PABX: +88029144270-9
Ext. 309 (Off.)
Fax: +88029112649
e-mail: bioc_sau@ymail.com

Ref:

Date: 20.10.2016

CERTIFICATE

This is to certify that the thesis entitled, “**REARING EFFICIENCY, FORAGING BEHAVIOR AND FOOD MANAGEMENT OF HONEY BEE (*Apis mellifera* L.) DURING DEARTH PERIOD**” submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE in ENTOMOLOGY**, embodies the result of a piece of *bona fide* research work carried out by **B. M. Rokonzaman**, Registration No.: 14-06334, under my supervision and guidance. No part of this thesis has been submitted for any other degree or diploma.

I further certify that any help or sources of information has been availed of during the course of this investigation has duly been acknowledged.

Dated: 20.10.2016
Dhaka, Bangladesh

(Dr. Mohammed Ali)
Professor
Department of Entomology
Sher-e-Bangla Agricultural University
Supervisor

Dedicated

To

*Almighty to bless me ever with the best of all
the choices*

&

*My loving parents
and teachers
who laid the foundation of my success*

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The Author

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ABSTRACT

A study on rearing efficiency, foraging behavior and food management of honey bee (*Apis mellifera* L.) during dearth period was conducted during dearth period (August 2015 to December 2015) in an apiary located at Sher-e-Bangla Agricultural University (SAU) campus. Study was also undertaken to evaluate the effectiveness of artificial nectar and pollen substitutes at four different concentrations supplied on the honey bee colonies on its population development as a preparation for migratory beekeeping. Study on the foraging behavior of honey bee during dearth period was also included. Treatments consisted of the substitute of nectar in the form sugar: water, and pollen in the form of mungbean flour applied in the ratios of 2:1, 1.5:1 and 1:1 and replicated 3 times. Treatments applied in four bee colonies in the apiary. In the month of August, maximum space (cm²) of the colony was covered by larva, pupae having 221.6 and 310, respectively. The highest number of worker per 4 cm² area were 3.238 and drone per frame was 1.913 in the colony treated by nectar substitute with sugar:water (2:1) and pollen alternative with mungbean flour. In the month of September, October, November and December the colonies treated with nectar and pollen substitutes with that higher ratio (2:1) showed the highest spaces of 145.33, 284, 300 and 397 cm² covered by eggs in four bee colonies, respectively. With similar substitute the highest space (cm²) covered by larvae and pupae were 300, 286, 298 and 306 cm² and 321, 288, 315 and 400 cm² and highest number of worker per 4 cm² was 3.79, 3.895, 4.566 and 5.083 and maximum number of drone per frame was 2.026, 2.40, 2.52 and 3.26 in the month of September, October, November and December, respectively. In this study foraging behavior of honey bee during dearth period was also conducted. Number of worker bee ingress per minute into the colony with pollen was counted. Highest number of worker bee was counted as 3.344, 10.40, 20.494, 20.056 and 29.944 in the month of August, September, October, November and December, respectively in the colonies treated by nectar substitute with sugar and water in the ratio of 2:1 and pollen alternative with mungbean flour.

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CHAPTER I

INTRODUCTION

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INTRODUCTION

Honey bees are of great economic importance because they not only produce honey and bees wax but also act as primary pollinating agents of many agricultural and horticultural crops due to pollination crop yield increases, quality of seed and fruit improves and heterosis can be exploited. Beekeeping can play a vital role in sustainable agricultural development as it increases resource without changing environmental balance. As a cottage industry, it is a source of income of the rural people. Beekeeping is one of the important components of integrated rural development programmes (Verma, 1990).

Bees are social insects of the order Hymenoptera which feed on pollen and nectar. There are many species of honey bee, but four species are common and useful honey producer and good pollinators. These are little honey bee known as *Apis florea*, giant honey bee known as *Apis dorsata*, Asian honey bee known as *Apis cerana* and European honey bee known as *Apis mellifera*. Due to domestic nature, *Apis mellifera* is the most popular worldwide and can be easily reared and safely migrated from one place to other for pollination and honey production. European honey bee (*Apis mellifera*) is the marvelous social insects known to the mankind. The peculiarities of this agro based industry for honey production are that it does not require any raw material like other industries. The raw material is the nectar and pollen from flowers which is freely available in nature for honey bee (Abrol, 2006).

A honey bee colony consists of a queen, several thousand workers and a few hundred drone at a certain season of the year. Among the members of the colony there is a division of labor and specialization in the performance of biological functions (Winston, 1987). As an inherited behavioral characteristic, all honey bee colonies tend

to store a certain quantity of honey and pollen as their food reserve. The quantity of food stored depends upon several factors including the availability of natural nectar and pollen, the worker population in the colony, its rate of reproduction etc. The success of beekeeping depends on strong, vigorous colonies. Colonies can only develop large populations when the queen maintains a high egg-laying rate. She can only do so when there are adequate stores of pollen and honey. When these forage resources are not available, they must be provided artificially. Supplemental feeding enables the colony to optimize its potential as a production unit and increase its ability to produce more honey and perform successful pollination (Anonymous, 2001).

Honey bees have no unusual nutritional requirements. Nectar and honeydew are the chief sources of supply for carbohydrates in the diet of bees and pollen furnishes all the other indispensable constituents. Adult bees can survive on carbohydrates (*i.e.*, honey or sucrose) and water; however, proteins, lipids or fats, minerals and vitamins are necessary for growth and development of young bees and rearing larvae (Standifer *et al.*, 1977; Hyser, 1980).

Honey bees (*A. mellifera*) are dependent on the supply of floral pollen and nectar. The activity of the honey bee is controlled largely by ambient conditions. In some habitats where the weather fluctuates annually, as in the “sub-tropical region”, the warm season coincides with the lack of flowers and is considered as the “Dearth period”. In the cold season, when the flowers appear, the bees are at full activity, collecting and storing food, along with fulfilling their reproductive duties. Although bees are physiologically capable of being active in hot deserts, they suffer from the lack of food sources and water, the latter being used for cooling hives in addition to its physiological function. Honey bees are thus restricted to areas where blooming occurs at least for part of the year (Echazarreta and Paxton, 1997).

Proper colony management should ensure adequate honey reserves but sometimes carbohydrate supplement feeding also become necessary. Whenever colonies have little honey reserves, they should be fed with artificial foods. Carbohydrate foods have some value for stimulating queens to begin laying eggs, but no carbohydrate will support sustained egg laying or brood rearing in the absence of pollen or a protein supplemental food (Shimanuki, 1971).

A. mellifera carries heavier pollen, less aggressive and produce more honey than the native bee *A. cerana*. It is less prone to swarming for beekeepers who naturally hope to lose their colonies as rarely as possible. Like other honey bee species *A. mellifera* has a high flight range for foraging. A worker of this species may fly maximum 2-3 km away from its colony (Abrol, 2006). There is a general agreement that introduction of the exotic *A. mellifera*, in Northern India, Bangladesh, Pakistan and Thailand is now the basis of flourishing apiculture industries. This exotic bee species produces three times more honey than the native, *A. cerana* and is more suited to modern bee management technology (Verma, 1990).

All types of flowers are not suitable in yielding similar quality or quantity of nectar and pollen. Some are rich in nectar rather than pollen and vice versa. Furthermore, all types of nectar or pollen are not similarly accepted to the worker honey bees. When flowers enriched by nectar and abundant in the nature and the pollen quantity or quality is poor, alternative pollen i.e. pollen substitute should be supplied artificially to maintain the population of the colony. Similarly, nectar substitute should be provided to the colonies if good quality natural nectar is rarely available. For successful management and rearing of honey bees in dearth period, it is imperative to adapt beekeeping measures for colony development. The annual cycle of colony development of European honey bee (*A. mellifera*) is described in detail in many independent studies in temperate climates from

North America (Farrar, 1937; Avitabile, 1978). Limited brood rearing may be initiated during winter months and brood rearing leading to colony expansion is often initiated before nectar and pollen become available. Furthermore, queen rearing is essential for improving existing stock, but has not been practiced successfully with *A. mellifera* in spite of many attempts. This species is very new in Bangladesh and the information in this country regarding this species are also scanty.

Sugar syrup (1:1) is one of the most popular nectar substitutes whereas soybean flower, mungbean flour, corn flour, mixed flour etc. are different pollen substitutes for honey bee used during dearth period (Anonymous, 2005). The reports on efficiency of such nectar and pollen substitutes at an economic dose in the maintenance of honey bee colonies are scanty.

Beekeeping is a valuable and profitable venture to farmers' supplement income. At present beekeeping activity is practiced on a part-time basis in Bangladesh (Anonymous, 2005). Bangladesh government and other non-government organizations, including Bangladesh Institute of Apiculture (BIA), Proshika and Mouchak Unnayan Sangstha (MUS) etc. have taken various schemes to provide technological support for training on beekeeping, developing marketing facilities and supplying necessary equipment for the economic production of honey in the country. Beekeeping may be very important for increasing family income if the women could be particularly engaged in beekeeping. In Bangladesh, generally women remain in home and have very limited opportunity for earning cash incomes outside their home. Beekeeping may be one of the ways to have women involved in the income generating process.

Honey bee colonies are a rich and stable habitat for parasites. Host bees, honey, pollen and wax are present year round and bees regulate temperature, humidity and carbon dioxide levels within narrow limits (Winston, 1987). As a consequence, honey bee

colonies are exploited by an array of parasites, pathogens and associated organisms (Bailey and Ball, 1991). They play as important selective force on the evolution of colony structure and function (Sherman *et al.*, 1988). Over 40 enemy species are known to be associated with honey bee colonies, several of which are parasitic, cause severe mortality of bee colony and worldwide losses in the beekeeping industry (Bailey and Ball, 1991). So some management approaches utilizing fumigation of tobacco leaf and sulphur dust to protect the colony from insect pests and diseases for successful migration of the hives after the dearth period are necessary. Therefore, the present study was undertaken as a preparation for migratory beekeeping with the following objectives:

1. to assess the rearing efficiency of *Apis mellifera* during dearth period,
2. to study the foraging behavior during dearth period of *Apis mellifera*,
3. to evaluate the effect of artificial nectar and pollen supplements with different concentrations in the colonies on honey bee population during the dearth period and
4. to evaluate different management approaches applied in the bee colonies of an apiary.

CHAPTER II

REVIEW OF LITERATURE

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REVIEW OF LITERATURE

Apis mellifera L. considered as European honey bee, is the most important among all the species as a pollinator, honey and bee wax producer. It makes parallel comb. In the hives it has prolific queen having the feature like medium sized, less stinging reflex, swarming habit, nearly no absconding, good honey gatherer and can guard its nest against enemies except wasps and mites (Verma, 1990) and gentle tempered. So it can be easily domesticated. *A. mellifera* is good for migratory beekeeping and produces more honey and other hive products such as propolis and royal jelly. A considerable number of papers have been published on *A. mellifera* which are more or less relevant to the present study. Briefing of some of the results of those research works are presented below:

2.1 The effect of nectar and pollen supplements on *A. mellifera* during dearth period

Pandey *et al.* (2014) conducted a study on Germinated pulses as a pollen substitute for dearth period management of honey bee colonies. Diet containing germinated (ger) pulses flour was evaluated to develop efficient and cheap pollen substitute for dearth period management of honey bee colonies. Bee colonies were provided by the four different pollen substitutes viz., ger horse gram, ger chick pea, ger greengram/mungbean, ger pea and compared with the control (no feeding) to determine their impact on bee colonies. Results indicated that the per cent palatability of all the prepared diets were more than 65 per cent. A gradual increase in brood area, honey and pollen store and foraging activity were observed after feeding in all the diet combination

viz., ger chick pea, ger greengram and ger horse gram. All the desirable parameters were found to be least in ger pea. However, all the diets were found significantly superior over control. Cost and shelf life of these patties were also determined, in support of the adoption of these patties. Present work revealed that among the evaluated ger pulses, ger chickpea was the best substitute for bees during the dearth period.

Standifer *et al.* (1977) showed that wheat, soybean flour and several brewer's yeast products singly or in combination are palatable to bees. They contain the quality and quantity of proteins and amino acids, lipids, vitamins and minerals required for growth and development of individuals and reproduction of the colony. The yeast products and soybean flour formulation can be fed as a dry mix or moist cake inside the hive or as a dry mix in open feeder outside the hive. Bees are unable to collect wheat in its original dry state because of its large particle size; therefore, it must be fed as moist cake inside the hive. Garcia *et al.* (1986) carried out an experiment providing the honey bee colonies with a diet of corn and soybean meal containing (A) 40% crude protein or (B) 30% crude protein + 0.29% methionine or (C) B + 0.77% lysine. Colonies fed with diet C consumed more than those on diet B; consumption was lowest in case of diet A, but in these colonies egg viability was higher. The feeding of amino acid (diets B and C) did not increase brood rearing or storage of honey or pollen and did not reduce the *Varroa* infestation.

Pesante *et al.* (1992) showed that in an apiary, European honey bee (*A. mellifera*) colonies at 2 locations were fed twice a week with 1 liter or 3 liters of 50% sugar syrup with protein and pollen twice a week gained significantly more weight than Africanized colonies. However, this was not found at other apiary where the nectar flow was poorer. Colonies fed with one liter feeds of syrup gained more weight than those fed with 3

liters; the higher rate of feeding resulted in slower brood nest expansion and thus restricted colony development.

Chang *et al.* (1993) in an experiment on honey bee (*A. mellifera*) used pollen supplements composed of sugar, dried yeast, lactalbumin, fermented soybean powder, egg powder, natural pollen, alfalfa meal, chlorella, rapeseed oil, lecithin, wheat bran, vitamin C and water. Zero, 0.43% or 0.86% of wheat bran and 0.00, 0.008% or 0.016% of vitamin C was mixed in the diet by 3x3 factorial design. Each of the 9 supplements was fed to 6 colonies of honey bees (*A. mellifera*) at a rate of 7 g a day and the consumption of each was determined. The mean relative consumption of the diet containing 0.86% wheat bran was higher than those of zero and 0.43% wheat bran diets. However, the different vitamin C levels did not cause any significant difference in palatability. The concentration of vitamin C in the experimental diet was lower when compared to 3000 mg per kg in natural pollen; e.g., it was 105 mg per kg of vitamin C in the supplement containing 0.43% wheat bran and 0.008% vitamin C. They opined that further research is needed to determine the supplementary level of vitamin C needed. Same year Szymas *et al.* (1993) provided protein diet based on milk powder, casein, feed yeast, extruded maize and vitamins, supplemented with blood meal + rapeseed oil meal (diet 1) or with fish meal + soybean oil meal (diet 2), with or without pollen addition to honey bee (*A. mellifera*). Hypo pharyngeal gland and fat body development in bees as well as the change in middle intestinal epithelium were examined in a laboratory experiment, while the development in bee families (brood surface increase) and feed palatability were studied in a hive experiment. Significantly better results were obtained in both experiments with feeding diet 2 than diet 1. Addition of pollen significantly increased palatability of both diets.

Augustin (1994) showed that in honey bee (*A. mellifera*) colonies of an apiary, the period October flight activity declined to nearly zero from a maximum of 28000 bees per day at the beginning of August. Activity is increased temporarily during sugar feeding. Mortality increased dramatically after the onset of feeding, perhaps because of the increased activity demanded by younger bees, which processed the sugar solution. During feeding, flight activity started later in the day and the proportion of short flights increased. In the same year, Moritz *et al.* (1994) reported that the diet of honey bees and their sociality; control of food procurement of the colony; nutrition and caste determination; prophylaxis. Honey bees are the most complex social insects; it is difficult to analyze the role of nourishment in the social evolution of honey bees. However, the overall impression is that the nourishment specialization of honey bees seems to be sophisticated adaptations to the social organization, rather than the proximate cause for social behavior.

Abbas *et al.* (1995) conducted a study on artificial feeding to *A. mellifera* reared in eight honey bee colonies, each with 4 frames of bees, were fed during the rainy season. The four colonies were fed with a pollen substitute containing 55% soya bean flour (also 25% sugar, 5% yeast, 5% milk powder, 10% water); another 4 colonies were fed with a diet consisting of blackgram (*Phaseolus mungo*) flour. After 3 months, the first group contained an average of 4.75 frame per colony and produced 7.12 kg honey per box, whereas the another group of colonies on an average had 5.75 frames and produced 8.62 kg honey per box. Four colonies which receive no pollen substitute had only 2.5 frame per colony having 1.87 kg of honey per box only. And Tahir *et al.* (1995) reported that the flour of blackgram (*Phaseolus mungo*) @ 550 g per kg diet was used in the diet of honey bees (*A. mellifera*) colonies fed with pollen substitute containing soybean meal

during summer rainy season produced a higher number of frames, resulting in higher production of honey compared to those fed on meal containing black gram flour.

Goodwin (1997) conducted trials over 4 seasons in 7 orchard using 379 *A. mellifera* colonies to test the effects of time and frequency of feeding, volume, concentration and grade of sugar and type of feeder. Syrup (but not dry sugar) feeding resulted in a mean increase of up to 7.9 folds in pollen collection over the flowering season, with up to 43.6 fold daily increases. Feeding of colonies with one liter of syrup per day between 08:00 hour and 10.00 hour consistently resulted in the greatest amount of pollen collected; grade sugar and syrup concentration (1 or 2 M) and type of feeder (division board or top feeder) had no effects. Three liters of syrup every 3rd day (to reduce labor costs) increased pollen collection by almost the same amount. Syrup feeding increased pollen collection even when there were significantly levels of competition from other flowers around the orchard and did not affect the colonies floral sex constancy.

Standifer *et al.* (1977) conducted a study to assess the general food requirements of honey bees, *A. mellifera* and presents formulas for supplementary diets and methods of feeding such foods to bee colonies. In early spring, before pollen and nectar are available or at other times of the year when these materials are not available for bees in the field or in the hive, supplementary feeding may help the colony survive or sustain brood rearing and colony development. None of the protein supplemental foods fed to honey bees is a complete replacement for natural pollen; however, several brewer's yeast products, wheat and soybean flour, fed singly or in combination, can be used to improve the nutrition of colonies when natural pollen is scarce. Cane or beet sugar and isomerized corn syrup can be used to supplement the bees' diet of nectar or honey.

The relationship between food consumption and royal jelly production was investigated at “Padre Assis” apiary, Brazil, from December 1996 to January 1997. Twenty Langstroth bee hives with nests separated by an excluding screen for queen bees were used. Treatments were applied randomly to all the hives. The effects of treatments were evaluated through the performance of beehives in the production of royal jelly. Treatment 1, comprising a powder based feed and refined sugar, resulted in an average production of 7.9 g of royal jelly per hive at each harvest. In treatment 2, comprising soybean meal and honey, the production was 4.32 g per hive. It was shown that the use of apiaries in the central region of Rio Grande do Sul for royal jelly production in December and January is successful when supplemental feeding constituting 3 parts of sugar and 1-part milk powder meal was used (Perlin, 1999).

Jhajj *et al.* (1992) found that in north Indian plains the colony increases are obtained from November through April but the queens reared and mated during February-March. In southern peninsula the best colony multiplication season is November-December and in north-eastern hills the colony multiplication is appropriate in spring. Haemolymph protein measurement methods were used to determine the efficiency of protein diet substitute of *A. mellifera*. Groups of 120 newly emerged worker bees were kept in small cases in the laboratory and feed on bee bread or unprocessed pollen (Natural protein diet), soybean or yeast or maize meal (alternative protein diet) or a sucrose solution (non-protein diet), from adult emergence until 6 d later, the protein content in haemolymph was determined in these bees at 0, 2, 4, 6 d of adult life. Additionally, vitellogenin (a major protein in young adult worker bees) titrate was measured through rocket immunoelectrophoresis of the haemolymph of 6-d-old bees. A significant and progressive rise in protein titrate was observed from 0 to the 6th day of adult life in the haemolymph of bees fed on bee bread, soybean or yeast or pollen. However, a

significant reduction was recorded when fed on maize or sucrose only (Cremonez, 2000). Newly emerged worker bees (*A. mellifera*) were fed honey, 22 and 40% pollen in honey and 22 and 40% royal jelly in honey for 14 days. Workers fed on royal jelly, pollen and honey had large, medium and small ovaries, respectively. Royal jelly had higher nutritive value for workers' ovarian development compared to pollen, possibly because royal jelly is predigested by nurse bees and easily used by adult and larval bees. Their results suggested that nurse bees could mediate workers' ovarian development in colonies via trophallactic exchange of royal jelly. Six level of royal jelly in honey i.e., 0, 20, 40, 60, 80 and 100% were tested for their effects on workers' ovarian development and mortality for 10 days. These findings suggested that nurse bees functioning as a unit which digest pollen and produce royal jelly and may feed some potentially egg-laying workers in a brood chamber with royal jelly when a queen is lost in a colony. Feeding workers on diet of 50% royal jelly in honey incubated at 34°C for 10 days is recommended for test of ovarian development (Lin-HuaRong and Winston, 1996).

An experiment was conducted by Chhuneja *et al.* (1996) to find out the efficiency of supplementary food of *A. mellifera*. Compared with 7 other diets, two types of patty containing soybean resulted in significantly higher mortality of unsealed and sealed brood. Brood mortality was lowest in colonies fed with pollen or with a diet of brewer's yeast + sugar meal; mean population in these colonies were 11650 and 9700 bees, respectively (compared with less than 7670 bees in colonies on other diets) and mean the weight of individual's nurse bees and foragers were significantly higher. Average wax production from colonies fed on a diet of brewer's yeast + sugar meal (B) for 6.5 months was 1.325 kg per colony and from pollen-fed (P) colonies 2.455 kg per colony. Colonies fed on other diets yielded significantly less wax. Honey yields were also

higher in colonies fed with B or P. Feeding pollen substitute resulted in a significant decrease in the average weight of pollen loads, including a fall in pollen foraging (Chhuneja *et al.*, 1993). Moeller *et al.* (1978) undertook an experiment to find out the necessity of carbohydrate substitute in the management of honey bee colonies. They reported that, proper colony management should ensure adequate honey reserves or stores in the hive at all times, but feeding sugar may sometimes be necessary. Whenever the honey supply in the colony is low and nectar in the field is in short supply due to adverse weather, the colonies should be fed sugar supplement. Brood rearing requires a large quantity of honey and pollen. Cane or sugar beet, isomerized corn syrup and type-50 sugar syrup are satisfactory substitutes for honey in the natural diet of honey bees. The last two are supplied only as a liquid for bees.

2.2 Effect of artificial nectar and pollen supplement and different management practices on mite infestation in the colony

Sukarsi (1993) reported that sulphur naphthalane trial in honey bee (*A. mellifera*) to control *Tropilaelaps clareae* in relation to life cycle of the bee. Two methods of applications of a mixture of sulphur and naphthalene for controlling *T. clareae* in *A. mellifera* were tested. A total of 15 colonies of *A. mellifera* was divided into 3 groups: A, treated every day at the first 10 days and then treated every 10 days for 6 treatments; B, treated every 10 days for 7 treatments and C, untreated control. The degree of mite infestation of brood was identified by removing 500 capped workers and drone brood cells from 3 combs for each colony and the presence and absence of mite were recorded. The degree of infestation of group A after the 16th day of treatment was 0.08%, significantly less than in group B and C. In group B, 11.36% of the brood was infested

and in group C, 24.88%. After 70th day of treatment in group A and B, similar results were obtained, in which infestation was undetectable.

Anderson (1994) observed the non-reproduction of *Varroa jacobsoni* in *A. mellifera* colonies in Papua New Guinea and Indonesia. Mite incidence and reproduction were determined in colonies of *A. cerana* and *A. mellifera* in Papua New Guinea, Arian Jaya and Java. At each locality and in colonies of each bee species, adult female mite was present in capped brood cells, with proportionally more drone than worker brood cells infested. In the *A. cerana*, female mites reproduced only in capped drone brood cells. In *A. mellifera* colonies there was no evidence of successful mite reproduction on either worker or drone brood. Although not reproducing in *A. mellifera* colonies, adult female mites were nevertheless feeding and surviving, they must have spread from nearby *A. cerana* colonies. There was no evidence that the mites inability to reproduce in *A. mellifera* colonies resulted from extremely slow reproduction, inter specific competition between *V. jacobsonia* and *T. clareae*, resistant bee population or climatic condition. These results have implications for findings and developing novel means of controlling *V. jacobsoni* in localities where the mite has become a serious pest of *A. mellifera*. And Boot *et al.* (1994) observed the behavior of *Varroa* mites invading honey bee brood cells. The invasion behavior of *V. jacobsoni* into brood cells of *A. mellifera* was studied using an observation hive. The mites were carried close to a suitable brood cell of the bees. Subsequently, the mites moved from the bees to the rim of the cells, walked quickly inside, crawled between the larvae and the cell wall and moved on to the bottom of the cell. *V. jacobsoni* mites were never seen walking across the comb and entering in the living brood cells as has been described for *T. clareae*. Differences in invasion strategies between *V. jacobsoni* and *T. clareae* are discussed.

Kumar *et al.* (1993) studied the development biology of *T. clareae* Delfinad and Baker (Acarina: Lepidoptera) vis the threshold stage in the life cycle of *A. mellifera* (Hymenoptera: Apidae). The biology of *T. clareae* was studied to identify the time of invasion of the mite into brood of *A. mellifera* and the threshold stage in the life cycle of the host. Honey bee brood was sequentially sampled on 0, 4, 8, 12, 16 and 20 days of development. Adult mites infested the 8 day-old larva shortly before the cell was capped. The larvae, protonymphs, deutonymphs and adults of *T. clareae* were all found parasitizing bright red eyed pupae during day 16 of brood development. This was identified as the most parasitized stage in the life cycle of the host. The mite developed from egg to adult in about 8 days. Another scientist Woyke (1993) found practical control method of the parasitic bee mite *T. clareae*. A biological method of controlling *T. clareae* in honey bee *A. mellifera* colonies involves caging the queen for 21 days or alternatively removing all brood from the colony. However, some beekeepers have reported that either procedure results in a decrease in colony strength. It is now recommended that if a queen is removed, this should be done during the final honey flow of the season. If brood is removed from a colony, any mites on emerging bees die within 3 days and therefore 3 days after emergence of the last bees the nucleus can be reunited with the original colony (after destroying queen cells).

Jain (1992) reported that efficacy of sulphur, sulphur smoke and formaldehyde against *T. clareae* Delfinado and Baker. Twelve colonies of *A. mellifera* infested with *T. clareae* were treated with fine sulphur dust (0.5 g per frame), sulphur smoke (produced by burning 2.0 g of sulfur powder in a smoker) or formaldehyde (10 ml of 15% commercial formaldehyde solution applied on a filter paper strip). The formaldehyde treatment caused maximum mortality of the mite, while both the sulfur treatments were relatively ineffective.

Sharma *et al.* (1994) reported the control of ectoparasitic mite, *Tropilaelaps clareae* Delfinado and Baker infestation using formic acid and sulfur. Mite infested *A. mellifera* colonies were treated daily with 85% formic acid (evaporation of 5 ml); another group of *A. mellifera* colonies were treated at 4-day intervals with sulphur dust (450 mg per frame). Daily counts of fallen mites showed that both treatments were quite effective when applied either during the monsoon season (July) or after it (September) and eliminated mites in 23-26 days. No adverse effects on brood or on adult workers or queens were observed. The next year Gatoria *et al.* (1995) reported the seasonal incidence of ectoparasitic mite, *T. clareae* and its control in *A. mellifera* colonies. The seasonal incidence of *T. clareae* and the use of sulfur for control were studied in the Punjab during 1984-89. The ectoparasite was observed throughout the year in colonies of *A. mellifera*, with peaks of infestation during March-April and October-November.

Arun *et al.* (2001) made a study to determine the infestation phenology of *T. clareae* and *A. woodi* (Rennie) on *A. mellifera* and *A. cerana*. Infestation of brood *T. clareae*, as well as the mite mortality, attained two peaks during June-July and October-November in *A. mellifera*. *A. woodi* infestation remained considerably low during March to May and high during October to November on *A. mellifera* and as well as in *A. cerana*. Strong correlation appeared between acarine infested bees and highest number of mites per tracheae in both the species of honey bees. Confinement of bees in the colony either due to rain or cold or the condition unfavorable to colony development may enhance mite infestation in the colonies.

Mahavir *et al.* (1999) showed that infestation of *A. mellifera* colonies with ectoparasitic mites. Two hundred samples of bees were taken from a colony of 5,000 *A. mellifera* in Hisar, Harayana, India. They were examined for ectoparasites each month from September 1994 to December 1996. *A. clareae* was the most abundant ectoparasite on

A. mellifera. Greatest infestation occurred in February, March and April and a decline from May to August. Poor management of bee colonies, hive microclimate, strength of the colony, inadequate food supply during winter and age of the colony increased the prevalence of ectoparasites in bee colonies. And Shah and Shah (1988) controlled *Varroa* infestation without any chemical. They dusted 10-15 g of wheat flour on to the bees on combs. Four dustings at 10 day's intervals were given. This gave complete control of *varroa*. This is attributed to the facts that dust on the bees adheres to the tarsal pads of the *Varroa* mite which makes it difficult for the mite to attach to the bee's surface.

2.3 Foraging activity and pollination of *A. mellifera*

Bhatia (2006) carried out a study about the foraging behavior of honey bee, *A. mellifera*. Foraging behavior is one of the distinctive behaviors of honey bees, *A. mellifera*. This behavior is the link between the honey bee colony and the ambient environment. Therefore, various in-colony and out-colony factors have an impact on this behavior and many studies have been employed to investigate these factors. Foraging behavior is not advantageous only for the colony and for plant pollination but also has other benefits. In contrast, some disadvantages have also been discovered to be linked with foraging activity. Practically speaking, the control over this behavior is very important to maximize colony products as well as to increase other agricultural benefits. This paper presents a review on foraging activity including; the regulation of foraging tasks, factors impacting this behavior, foraging preference, variations between subspecies, monitoring methods as well as the possible methods for controlling this behavior. As concluded from this review, more work needs to be performed in order to elucidate certain aspects of foraging behavior. Four *A. mellifera* colonies were

introduced to a litchi orchard in Himachal Pradesh, India, at the start of flowering (Badiyala and Grag, 1990). They observed that honey bees visited flowers frequently, especially in the morning (9.30-11.30 am) and less actively from 3 pm to 5 pm. A few *Vespa basalis* (wasps) were seen and hoverflies were also observed. Fruit set in inflorescences accessible to bees and other pollinators was 2 to 3 times greater than in bagged inflorescences; fruits/inflorescence and fruit weight were also greater. Results varied among different litchi cultivars. They strongly recommended the introduction of *A. mellifera* colonies to litchi orchards.

Experiments were done by Pandey and Yadava (2006) in orchards near water canals, in the foothills and in a submontane area. The quantity of fruit set in inflorescences open to insect visitors was ranged from 0.71 to 11.25% and in inflorescences caged against insects was 0.026 to 0.105% which indicated that litchi is highly self-sterile and that insect pollination is necessary for a fruit setting. Counts of insect visitors to the flowers showed that Apoidea (*Apis* spp. And *Melipona* spp.) Comprised 98-99% of the total. In the same year, a study was carried out by Bhatia *et al.* (2006) at each in 1992-93 to study the relative abundance of insect visitors on flowers of mango (*Mangifera indica*), litchi (*Litchi chinensis*) and *Citrus* spp. and their effects on fruit set. Of 34 insect species recorded on the flowers of loose-skinned mandarin (*Citrus reliculata*), Malta (*C. sinensis*), kinnow mandarin (*C. nobilis* X, *C. deliciosa*), mango and litchi, 15 were Diptera, 13 Hymenoptera, 4 Coleoptera and 1 each of Lepidoptera and Hemiptera. Honey bees (*A. mellifera*) were the main pollinators of *Citrus* spp., *A. florea* and hoverflies (*Episyrphus halteatus*) on litchi and flies belonging to the family Calliphoridae on mango. Fruit set on bagged flowers was zero on mango, 0.88% and 0.98% on litchi and 20-60% on *Citrus* spp. For unbagged flowers, corresponding fruit sets were 4.32%, 2.14% and 2.48% and 60-76%, respectively.

Phadke and Nairn (1976) observed higher honey bee (*A. mellifera*) visit to the litchi (*Nephelium litchi*) blossoms at Pusa (Bihar, India) compared to other flowers. Jay (1974) observed that more pollen was collected in the morning than in the afternoon. Bees collected nectar from male flowers in the afternoon. Jenkinson and Gkynne (1953) found that rape plant (*Brassica napus*) kept in the relatively still air of glasshouse produced only one third or half seed compared to open condition. The study revealed that the wind and insect particularly bee pollination is also needed to obtain a high seed set. Honey bees gather nectar and pollen from flowers from rape plant (*B. napus*) as their food. Pollen is the raw materials of the beekeeping industry. Every minute the bees remove pollen from their bodies with pollen brushes and collect surplus pollen in their baskets. Pollen collection in a colony generally influences the quantity of honey in the comb. Jeffree and Allen (1956) stated that the quantity of stored pollen in healthy colonies showed a well, marked peak during June, July and August while from September or August the pollen stores remained at a constant low level. They observed that the queenlessness in late summer frequently resulted in large stores of pollen in autumn and winter. The next year Rashad (1957) reported on litchi orchard that very little information is available about the effect of humidity on the flight or foraging range of honey bees except in the springs when high relative humidity decreased pollen collection.

Bisht and Pant (1968) observed on litchi flowers that the number of pollen gathering bees was the highest during the months of January, February and March. They observed lesser activity of the bee during April, August, September and October. The number of pollen gathering bee reduced further during November and December. Pollen gathering activity is dependent upon the availability of pollen yielding flowers and the environmental conditions like sunrise, sunset and day temperature etc. Pollen gathering

activity is, therefore, the interaction of available flora and these interactions presumably determine the pattern of pollen gathering activity. They also remarked that there was a negative correlation between the time when the bees started pollen gathering trips and the maximum temperature of the day. There was a positive correlation between relative humidity of the day and the periods when the bee started pollen gathering. And Free and Williams (1968) studied the behaviors and frequency of honey bee (*A. mellifera*) works in summer rape (*Brassica napus* var. *Nilla*). All honey bee (HB) collected nectar but none pollen only. During a single flower visit, most HB visited both inner nectarines. They touch the stamens of 76.2% flowers visited (32 flowers) and spent 4.1 (second) per flower. Some HB packed the pollen into their corbicule whereas others discarded it.

Adlakha and Sharma (1975) reported that at Nagrota, *A. mellifera* was active within 21.28°C range, inactive below 12.88°C; *A. cerana* was active even at lower temperatures. Foraging range of *A. mellifera* was 3-4 times higher than *A. cerana*. Naim and Phadke (1976) stated that January to March as the peak period of activity, March to April as honey storing activity, May, June, August, November and December as the period of mild activity and July, September and October as the period of dull activity in litchi (*Nephelium litchi*) at Pusa in Bihar.

Dhaliwal and Atwal (1978) says that for getting adequate pollination and honey production by *A. mellifera* bees, the bar seem fields for seed production be grown within 1 km of the apiary or the bee hives be kept within this range during its flowering period. The honey bee is more active at the time of 10-11 am. Jhajj and Goyal (1979) worked on the comparative behavior as pollen forager of *A. mellifera* and *A. cerana* in rape plant (*B. napus*). They observed that number of bees of either species collected mixed pollen of on all foraging trips on the same day or one each of the 5 days observed.

Pollen availability decreased from morning to afternoon and some pollen foragers of both species then changed to nectar collection next morning. The two species showed similar behavior in pollen foraging with minor differences.

Kubisova *et al.* (1980) observed that the bees visiting *B. napus* flowers worked systematically (visiting 7-10 flowers/min) collecting nectar and pollen simultaneously. The number of flowers visited by 1 bee increased from 7 to 10 per minute. Acceptable levels of pollination were achieved only when 4 bee colonies/ha were brought to the crop. They also reported that the individual flower of *B. napus* secreted nectar for 2 days which is equivalent to 0.68-1.09 mg of sugars. Each flower secreted nectar for 2 days, giving a total of 2.28-2.55 mg of nectar, with an average sugar content of 33-40%. Total nectar production was estimated 199-230 kg/ha and total sugar 62-89 kg/ha.

Murrell and Nash (1981) stated that *A. florea* appeared on the floret of *B. napus* in a small number than *A. cerana* and spent more time on one floret. *A. dorsata* was intermediate in foraging speed. They remarked that the most bees of all species collected both nectar and pollen but pollen collection by *A. cerana* was observed only in the morning. The highest proportion of *A. cerana* forager carrying pollen loads followed shortly after anthesis. They observed that maximum number of foragers was found at 10:30 am for *A. cerana* and at 1:30 pm for *A. florea*. Zuberi and Sarker (1982) found inadequate pollen transfer in rapeseed under natural open-pollination. Even if adequate and effective pollination had taken place, presence of large number of self-pollen on the stigma could result in poor fertilization and ovule development.

Verma and Dulta (1986) studied the comparative foraging behavior of *A. cerana* and *A. mellifera* on apple bloom and the results on these investigations reviewed were as follows: workerbees of *A. cerana* started their foraging activities earlier in the morning

(mean time 06:03 hours) than *A. mellifera* (mean time 06:27 hours). In the evening *A. mellifera* ceased its foraging activity (mean time 18:55 hours) than *A. cerana* (Mean time 19:13 hours). Thus the average duration of foraging activity in *A. cerana* was 13.10 hours and for *A. mellifera*, it was 12.28 hours. The mean duration of a foraging trip by *A. cerana* and *A. mellifera* was 11.85 and 17.92 minutes, respectively. Thus the duration of a foraging trip was more significant for *A. mellifera* than for *A. cerana*. Then Pinzauti *et al.* (1987) observed that the foraging behavior of marked workers of *A. mellifera* from 4 hives in the Pisa area and in the Lucia area of Italy. On dense plots of *Vicia foba* nectar robbing was observed but on *Hedysarum coronarium* the bees effected pollination. On *Brassica napus V. oleifera* the percentage of foragers collecting nectar was 75%, collecting pollen 15% and collecting nectar + pollen 10%, but on *Helianthus annuus* it was 39%, 52% and 9%, respectively.

Oh and Woo (1990) found that the flight activity of *A. mellifera* increased sharply after sunrise, decreasing gradually through the day and ceasing just before sunset. The total period of foraging increased as the duration of sunshine increased, from 09.00-20.00 h in April to 06.00-21.00 h in May. Pollen foraging usually started 1-2 h before flying ceased, with a maximum level of activity at 13:00 h.

Jain (1992) conducted an experiment on pollen collection activity of honey bee in sunflower seed crops in semi-tropical region. He reported that *A. florea* and *A. dorsata* visited alfalfa (*Medicago saliva*) mainly for nectar. *A. mellifera* colonies placed in alfalfa crops showed a decline in honey and pollen reserves and in brood area. On sunflower crops *A. mellifera* was much more active than *A. florea* and *A. dorsata*, foraging for nectar. Adeagas and Nogueira-Couto (1992) revealed that honey bees collected nectar from the flower of rape cultivars, CTC-4 throughout the day and a bee spent on an average, 3.2-3.5 s on a flower, honey bees (*A. cerana*) also collected pollen

from 07.00 to 11.00 h, spending 20 s on each flower. And Ghoniemy and Abu-Zeid (1992) observed that more bees (*A. mellifera*) foraged for nectar than for pollen in 2 cultivars of swede rape (*B. napus*). The average number of flowers visited per minute for nectar collection reached its peak (10.14) at midday, whereas for pollen collection the peak (2.55) was at 9:00-10:00 h. Same year Hossain (1992) stated that foraging activity of honey bees (*A. mellifera*) was greatest in January, October and November, with maximum pollen collection in January, February and November. Maximum values for pollen collection was recorded during the monsoon (June-August) in litchi flower. Rahman and Rahman (1993) studies pollen gathering activity of *A. cerana* and *A. mellifera* and compared them in *B. napus*. In *A. cerana* colonies, the maximum area of stored pollen was recorded in February and the minimum in October, in *A. mellifera* colonies the maximum and minimum quantities were recorded in March and October, respectively. Throughout the year, the average of pollen collecting bees/day was significantly higher in *A. cerana* colonies than in *A. mellifera*.

Alam (1988) revealed that the in absence of self-pollination, up to 81% cross fertilization can be achieved in *B. napus* with honey bee under caged conditions. Estimates of self-and cross fertilization can be made considering open pollination as 100% by inclusion or elimination of different pollinating agents. Wind contributed 12 and 20% cross-and self-pollination, respectively. Bhat *et al.* (1996) observed the pollinator visits for 5 m at 5 times from 08:30 to 16:30 h for 3 days on 5 panicles from 3 trees of *Peltophorum pterocarpum* (DC.) (Fam: Leguminosae). Four pollinators were found: *A. dorsata*, *A. cerana*, *Xylocarpa* spp. and an unidentified syrphid. Honey bee visits were most at 08:30 h (for pollen foraging) with other peaks at 12:30 h (*A. dorsata*) and 14:30 h (*A. cerana*) for nectar collection. *Xylocarpa* (carpenter bee) activity (nectar collection) increased through the day. The syrphid was observed from 10:30 h onwards

and was possibly involved in residual pollen collecting activity. Mamood *et al.* (1996) observed honey bees foraging on the *B. napus* flowers most actively between 10.00 and 13.00 h, with a peak at 11.00 hours.

Pernal and Currie (1998) examined the nectar sugar composition and temporal patterns of nectar sugar production in oilseed summer rape (*B. napus* sub spp. *olifera*) from 6 open pollinated, 8 cytoplasm male-sterile (CMS) hybrid and 7 dominant self-incompatible (SI) hybrid cultivar at 3 field plot sites in Manitoba. Hybrid and open pollinated cultivar flowers had similar sugar content. However, for all cultivars, total nectar sugar content per flower was lower during the 8.00 and 11.00 h sampling periods and increased to maximum levels during the 14.00 and 16.00 h sampling periods. Significant differences in nectar sugar content were also found in relation to the flowering phenology of cultivars. Selecting for higher total sugar content may produce nectars more attractive to foraging honey bee (*A. mellifera*), thereby ensuring adequate pollination of hybrid parental lined and F1 hybrid plants. Selecting for lower nectar glucose will produce honeys with more desirable granulation characteristics. Overall, the production and quality of nectar sugar in oilseed rape hybrids are similar to those of open-pollinated cultivars and are not likely to adversely affect the pollinating activities of honey bees or their potential for honey production.

Sihag *et al.* (1999) revealed that both foraging by honey bees and anthesis in flowers seemed to be regulated by the ambient temperature. They studied the foraging pattern of *A. dorsata*, *A. mellifera* and *A. florea* on eight cultivars of oilseed crops viz., brown sarson (*Brassica campestris* var. *brown sarson* cv. BSH-1), yellow sarson (*Brassica campestris* var. *yellow sarson* cv. YSPB-1), toria (*Brassica campestris* var. *toria* TH-68), raya (*B. juncea* cvs. RH-8812, RH-819 and RH-30), taramira (*Eruca saliva* [E. vesicaria] cv. T-27) and sunflower (*Helianthus annuus*), during their entire blooming

period. Honey bee visitation frequency was low at the time of initiation of flowering, then increased gradually and reached a peak during the peak flowering time. This peak continued for over two weeks and then declined suddenly with the decline in flowering on these crops. This pattern of honey bee visitation provided useful information for selecting the time of deployment of honey bee colonies on these crops for their pollination. In this study also the three honey bee species were found to show clear preferences for the various crops and seemed to partition the food resources. Arshad *et al.* (2000) observed that the maximum average number of visits of worker honey bees was done during 10:00-11:00 hours (25.21 flower, 10 minutes, 1 followed by the rest of the individuals in a day).

2.4 Honey Production of *A. mellifera*

Atwal and Sharma (1970) reported that honey yields of *A. mellifera* was four times more than that of *A. cerana*. Anonymous (2001) reported that average honey production per *A. mellifera* colony was 60 kg/year. On the other hand, the average production of *A. cerana* colony was 10 kg/year. The honey production by European honey bee, *A. mellifera* varies in different countries due to weather condition. Average honey yield of 14.5 kgs per *A. mellifera* colony and a maximum of 51.5 kg per colony have already been obtained in Punjab.

Koltai (1988) reported that approximately 30,000 beekeepers and more than 6,00,000 colonies of *A. mellifera* produces annual honey amounting to more than 10,000 metric tons in Hungary. Zhen-Ming *et al.* (1990) found that the European honey bee, *A. mellifera* was the most important species in Chinese agriculture. In European honey bees, the honey output of each colony is 30 kg/year and the highest can be as much as 150-200 kg. Verma (1990) reported that the, *A. mellifera* introduced by Friz Maurer in

1980 in Bhutan & he (Maurer) observed the average honey yield of 30 kg/colony/annum.

Anonymous (1993) carried out a research work on the estimate of number of beekeepers and number of colonies, production and value of honey and wax in Canada. He stated that in Canada during 1992, 13100 beekeepers kept 501420 colonies and produced 30339 tons of honey. Portch (1994) recorded annual honey yields for the period 1982-1992 for 100 honey bees (*A. mellifera*) colonies kept at 10 apiaries in Wiltshire, England. Total annual yields. Presented graphically, ranged from 1200 lb (545 kg) in 1986 to 6500 lb (2950 kg) in 1989. Over 10 year's period, the average yield per colony was 36 lb (16 kg) with an annual maximum in 1989 of 65 lb (29.5 kg). The pattern of results was compared with variations in local weather conditions, e.g. annual average rainfall, hours of sunshine. Allsopp (1994) obtained the honey production in South Africa to be 29.3 kg per colony per year. He also found that deep supers with worker foundation are most suited for increased honey production. Chaudhary (1997) reported that the Italian honey bee, *A. mellifera* was introduced to Tamil Nadu, India from Maharashtra, to overcome an epidemic of Thai sac brood virus (TSBV) disease in the area. He extracted 80 kg of honey from 10 colonies during the first 15 days of the honey flow season. Chaudhary (2000) conducted an experiment on performance and prospects of *A. mellifera* in southwestern Haryana, India and he extracted an average of 18.5 kg honey per colony during February-March from rape seeds. Anonymous (2000) reported that the average honey production per *A. mellifera* colony was 60 kg/year.

2.5 Environmental factors influencing foraging behavior of four honey bee species

Honey bee species *A. dorsata*; *A. mellifera*; *A. cerana* and *A. florea* were the most important and efficient pollinators of litchi flowers (*Litchi chinensis*). They constituted more than 65% of the total pollinating insects. The ecological threshold for commencement and cessation of flight activity of each honey bee species varied from one another. In general, 15.5-18.5°C temperature, 600-1700 lx light intensity and 9-20 m W/square meter solar radiation appeared to be the minimum ecological conditions for commencement of flight activity in *Apis* species. Cessation of activities in all the honey bee species was controlled mainly by decline in values of light intensity and solar radiation irrespective of other factors. Between commencement and cessation, the foraging activity of all honey bee species followed the same general pattern as temperature, light intensity, solar radiation and nectar sugar concentration and inversely with relative humidity. Path analysis revealed that all honey bee species differed in their responses to temperature, light intensity and solar radiation, the three most important factors in foraging behavior (Abrol, 2006).

2.6 Space (cm²) covered by *A. mellifera* eggs, larvae and pupae in the comb

Standifer *et al.* (1977) showed that wheat, soybean flour and several brewer's yeast products singly or in combination are palatable to bees. They contain the quality and quantity of proteins and amino acids, lipids, vitamins and minerals required for growth and development of individuals and reproduction of the colony. The yeast products and soybean flour formulations can be fed as a dry mix or moist cake inside the hive or as a dry mix in open feeders outside the hive. Bees are unable to collect wheat in its original dry state because of its large particle size; therefore, it must be fed as moist

cake inside the hive. Chhuneja *et al.* (1993) revealed that diets containing soybean resulted in significantly lower mortality of unsealed and sealed brood. Brood mortality was lowest in colonies fed with pollen or with a diet of brewer's yeast + sugar meal; mean populations in these colonies were 11650 and 9700 bees, respectively (compared with less than 7670 bees in colonies on other diets) and mean weights of individual nurse bees and foragers were significantly higher.

2.7 Food substitutes with other effects

Allsopp *et al.* (1994) stated that the pentose sugar xylose has recently been reported as a major sugar in the nectar of *Protea* and *Faurea* (Proteaceae). Honey bees are potentially important pollinators of both *Protea* and *Faurea*, the authors investigated the responses of Cape honey bees to xylose solutions. He observed that when bees were fed sucrose, glucose, fructose and xylose (all 30% w/w) and water only, survival on xylose was as poor as on water. With different glucose/xylose mixtures, survival time was inversely related to the proportion of xylose in the diet and each 5% increment in xylose causing an additional increase in mortality. It was concluded that the xylose in *Protea* and *Faurea* nectar is not there for the benefit of honey bees. Bitondi and Simoes (1996) showed that the relationship between the amount of pollen consumed and vitellogenin (Vg) and juvenile hormone (JH) titers in the blood of Africanized worker honey bees (*A. mellifera*). Analysis of blood from young workers fed on diets differing in pollen content revealed that Vg, but not JH titers revealed that is dependent on pollen consumption. Workers fed on a 50% pollen diet had higher Vg levels than workers fed on a 15% pollen diet. A 0% pollen diet severely impaired the increase in Vg titer normally observed during the first days of adult life. However, the quantity of pollen in the diet was not reflected in the measured JH titer, which was not significantly different

in bees fed on the various diets. The results are discussed in terms of regulation of Vg synthesis.

Pernal (1998) showed that the development of hypopharyngeal glands and ovaries varied with diet and, collectively, proved to be sensitive measures of protein utilization and pollen quality. For workers fed 1-year-old *Phacelia* pollen, protein was utilized in a differential fashion, promoting the development of ovaries over that of hypopharyngeal glands. Development of glands and ovaries was strongly correlated with the amount of protein consumed by workers from pollen diets and to a lesser extent, the crude protein content of diets. Storing pollen for 1 year by freezing did not affect gland or ovary development. Perlin (1999) compared the relationship between food consumption and royal jelly production. Comprising powdered milk and refined sugar, resulted in an average production of 7.9 g of royal jelly/bee hive for each harvest. Again in mother diet comprising soybean meal and honey, the production was 4.32 gm. It was shown that, the use of apiaries for the production of royal jelly when supplemental feeding constituting 3 parts of sugar and 1-part milk powder meal was used. Cremonez *et al.* (2000) observed a significant and progressive rise in protein titers from 0 to the 6th day of adult life in the haemolymph of *A. mellifera* bees fed on beebread, soybean/yeast or pollen. He recorded a significant protein reduction was recorded in bees fed on maize meal or sucrose only. The protein titers of 6-d-old bees gave the best discrimination between diets. Protein titers were highly correlated with vitellogenin levels in 6-d-old bees, when the different diets were compared. The protein values reflected the quantity and usability of the protein in the diets and not the consumption, which was similar for all protein diets used. Both total protein measurement and vitellogenin level determination proved to be objective methods for comparing the effectiveness of protein diets. Evans *et al.* (2000) found that queen

production is the result of interaction between environmental factors, such as diet and genetic factors. Foraging, hygienic and aggressive behaviors appear to have genetic components, which are mediated by the environment.

Siddiqui (1970) revealed that honey and nectars contain traces of toxic sugars such as raffinose, mannose and galactose. Sub lethal levels of these sugars in pollen, honey or nectar could modify effects of sugars in supplementary diets. Doull (1975) observed that sugar syrups produced by the hydrolysis of wheat starch were detrimental to bees in confinement. He suspected undigested polysaccharides, particularly starch, to be harmful. He obtained better results sucrose than with his invert syrups. Bailey (1991) found that semi-refined cane sugar was harmless but that semi-refined beet sugar decreased the life of bees. So, impurities in his unrefined beet sugar must be toxic. Nectar is the major source of carbohydrate in the natural diet of honey bees. It may contain 5 to 75 percent soluble solids (sugars) although most nectars are in the 25 to 40 percent range. The primary sugars are sucrose, glucose and fructose. As nectar is manipulated and finally stored as honey, much of the sucrose is inverted to approximately equal parts of glucose and fructose. A normalized honey bee colony may use the nectar equivalent of 300 to 500 pounds a year.

CHAPTER III

MATERIALS AND METHODS

CHAPTER III

MATERIALS AND METHODS

The study was undertaken utilizing honey bee (*Apis mellifera*) reared on artificial diet at dearth period at an apiary of the Department of Entomology, SAU during August to December 2015 utilizing nectar and pollen substitutes at different concentrations on honey bee population and applying different management approaches in the bee colonies. The foraging behavior of *A. mellifera* in colonies was also studied. Data collection regarding the predetermined parameters and the analysis of data was performed to measure the effects of food substitutes and management practices on the colony development as a preparation for migratory beekeeping. The detailed methodology and other related procedures followed in the studies are discussed below:

3.1 Design and layout of the Experiment:

The experiment was set up in a Randomized Complete Block Design (RCBD). Food substitute of three different concentrations were considered as the three different treatments viz., application of Nectar (sugar:water=1:1), Nectar (sugar:water=1.5:1) and Nectar (sugar:water=2:1). With all of three different concentration of nectar, same quantity of pollen supplement is also used. The treatments were randomly allotted for three replications. Therefore, four treatments may be designated as follows:

T₁=Nectar (sugar:water=1:1) and mungbean flour

T₂=Nectar (sugar:water=1.5:1) and mungbean flour

T₃=Nectar (sugar:water=2:1) and mungbean flour

Untreated Control

3.2 Rearing technique of honey bee

To study the foraging behavior and management practices of *A. mellifera*, the stock of bees was reared on artificial nectar and pollen substitutes in the apiary during the dearth period.

3.3 Preparation of Nectar substitute

For preparation of nectar substitute at three different concentrations of sugar and water are mixed with the desired ratio. In a bowl 1 kg of sugar was taken and then the 1 L of water was poured on it (1:1). For preparing the nectar substitute at 1.5:1 and 2:1 concentration 1.5 kg and 2 kg of sugar was dissolved in 1 liter of water, respectively by following the same procedures. Then the solution was poured in sugar syrup feeding pot in the hive (Plate 1).



Plate 1. Sugar syrup given on feeding pot

3.4 Preparation of Pollen substitute

For preparation of pollen substitute mungbean flour and icing sugar was mixed properly. The pollen substitute was then supplied in the hive.

3.5 Application of Nectar substitute (Sugar syrup)

1 liter of sugar syrup of aforesaid concentrations was applied in the feeding pots of colonies. This was done in the morning 7:00 am to 8:00 am at 7 days' interval and continued for five months (Plate 1).

3.6 Application of Pollen substitute (Mungbean flour)

The mixture of mungbean flour and icing sugar was provided as pollen substitute. For honey bee colonies, 100 g pollen substitute was given in a petridish. The pollen substitute was applied in the morning at 10 days' interval and continued for five months.

3.7 Prophylactic measures taken against ant and other intruders

For this purpose, the hives were fumigated using coconut leaf powder. Coconut leaf was placed on coconut husk and kept in smoker and fired for getting smoke.

Smoker was puffed 10 times instantaneously. A delivery tube of the smoker was extended to the entrance of hive and puffed 5-10 times to deliver and circulate sufficient smoke in the hive. Smoking was done at 20 days' interval.

The entrance of the hive was cleared regularly for uninterrupted foraging and movement of the bees (Plate 2). For sanitation, cleaning of upper surface of the frame was done before each application of pollen substitutes. Cells were cleaned by using small camel hair brush. Plastic cover was provided to avoid sudden rain that can harm

the apiary. Water tray was provided at the base of stand of a hive to avoid ant movement. Sevin powder was also used under the hive to prevent ant (Plate 3).



Plate 2. Clear entrance of the hive



Plate 3. Maintenance of the hives

3.8 Measuring space occupied by eggs, larvae, pupae, number of workers, queen cells, drone and to check the pollen in the hive

In the dearth period from August to December 2015, the relative space covered by eggs, larvae, pupae and number of workers in the hives (4 hives) of the apiary was measured

by a 4 cm² hollow scale with a handle made up of drawing sheet (Plate 4). The hollow scale was placed on the space covered by eggs, larvae, pupae and number of worker within the frame and total space covered by them was calculated.

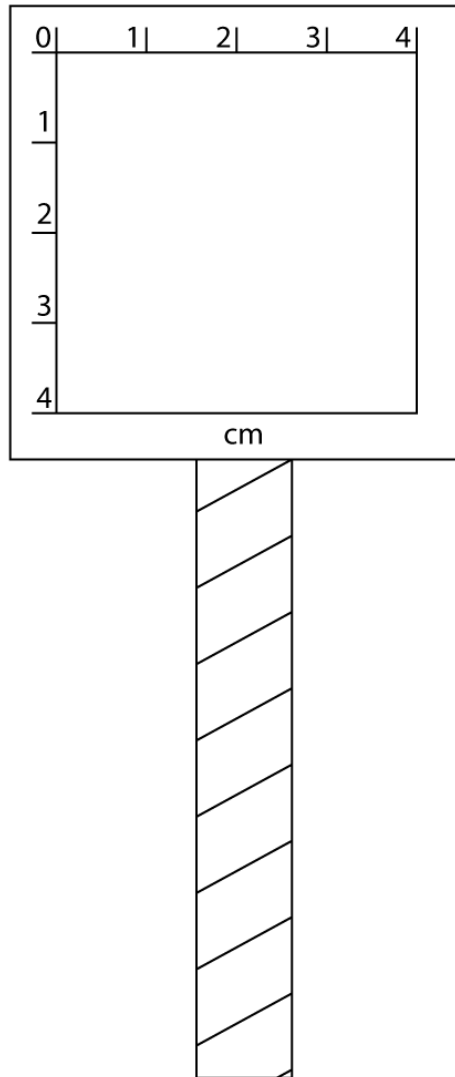


Plate 4. Hollow scale made up of drawing sheet with 4 cm² space within

3.9 Collection of data

Five frames of the experimental colonies were selected before data collection to maintain the homogeneity of honey bee population. The data were collected at 7 days' interval for 5 months on the following parameters:

3.9.1 Space (cm²) covered by eggs in the comb

The total space (cm²) of the comb covered by eggs was recorded for each treatment from all the selected colonies and the mean was calculated in the month of August through December.

3.9.2 Space (cm²) covered by larvae in the comb

The total space (cm²) of the comb covered by larvae was recorded for each treatment from all the selected colonies and the mean was calculated.

3.9.3 Space (cm²) covered by pupae in the comb

In this case the total area (cm²) of the comb covered by pupae was measured for each treatment from each of the selected colony and the mean was calculated.

3.9.4 Number of worker per 4 cm² area

Total number of workers covering within a 4 cm² space of the frame were counted from each of the frame and the mean was recorded.

3.9.5 Number of drone per frame

Number of drone per frame was counted for each of the colony and the mean was calculated.

3.9.6 Number of worker bees entering the colony with pollen and nectar

The foraging activities of *A. mellifera* were studied for a period of August to December 2015 i.e., at the time (season) of honey flow. The number of worker bees entering the colony with pollen and nectar were recorded separately on weekly basis. The numbers

of bees carrying pollen or nectar during 7:00 to 8:00 am were recorded once in a week. Pollen collectors were identified by the presence of pollen load on their hind legs. Nectar collectors do not bear such pollen load on their hind legs. Data was taken by observing worker bees carrying pollen or nectar from flowers and landing on the colony entrance. Worker bees entering the colony throughout the day starting from pollen or nectar collection in morning 6:00 am and continued till the bees stopped foraging activity in the afternoon 6:00 pm.

3.10 Statistical analysis

The data of tested parameters were analyzed statistically to find out the variations among the treatments. The analysis for variance of different parameters was done following the standard procedure of MSTAT-C software. The means were separated by Least Significant Difference (LSD).

CHAPTER IV

RESULTS AND DISCUSSION

CHAPTER IV

RESULTS AND DISCUSSION

4.1 Foraging of honey bee at the experimental site during dearth period

During the study period a survey was conducted to identify the flowers of plant species which act as the natural pollen and nectar source for honey bee (*A. mellifera*). Among all of the plant present in SAU campus some species were identified which provide nectar and pollen during dearth period. The list of honey and pollen supplying flowering plants recorded in the area are present in Table 1.

Table 1. List of flowering plant species that are the sources of nectar and/or pollen for *A. mellifera* in SAU* campus during dearth period (August to December, 2015)

Common Name	Scientific Name	Status of the observed plants	Resource of Nectar, Pollen
Allamanda	<i>Allamanda cathartica</i>	Planted	N, P
Asoka tree	<i>Saraca indica</i>	Feral, ornamental	N, P
Bougainvillea	<i>Bougainvillea</i>	Planted	p
China Rose	<i>Hibiscus rosa sinensis</i>	Planted	N, P
Coconut	<i>Cocos nucifera</i>	Planted	N, P
Duranta	<i>Duranta repens</i>	Planted, ornamental	P
Gardenia	<i>Gardenia jasminoides</i>	Planted	N
Guava	<i>Psidium guajava</i>	Planted	N, P
Ixora	<i>Ixora sp.</i>	Planted	P
Indian lilac	<i>Lagerstroemia indica</i>	Planted	N, P
Indian gum tree	<i>Acacia arabica</i>	Feral	N
Jasmine	<i>Jasminum auriculatum</i>	Planted	N
Karambola	<i>Averrhoa karambola</i>	Planted	P
Madhabilata	<i>Hiptage madablota</i>	Planted	N, P
Oleander	<i>Nerium oleander</i>	Planted	P
Passion fruit	<i>Passiflora edulis</i>	Feral	P
Salvia	<i>Salvia splendens</i>	Planted	N, P
Touch me not	<i>Mimosa pudica</i>	Feral	N, P
Mussaenda	<i>Mussaenda erythrophylla</i>	Planted	P
White siris	<i>Albizia procera</i>	Planted, feral	N, P

* Sher-e-Bangla Agricultural University

N=Flowering plants providing nectar

P=Flowering plants providing pollen

4.2 Effect of artificial food on population density of honey bee

During dearth period when supplementary diet was added, the population densities at different life stages of honey bee *A. mellifera* fed on nectar and pollen substitutes provided at four different concentrations were found different. The total population density was determined by measuring the space covered by eggs, larvae, pupae, number of workers per 4 cm² area and number of drone per frame. Result indicated that, food substitute at different concentration influencing the population density of *A. mellifera* (Table 2).

4.2.1 Space (cm²) covered by eggs, larvae, pupae, number of workers and drone per frame in August, 2015

The effect of supplementary diet at different concentration on queen's ability to lay eggs and ultimate production of larvae, pupae, workers and drone were evaluated during August, 2015 (Table 2). No egg was found in the hive during August, 2015.

The highest space (120.6 cm²) covered by egg was found in the hive during August, 2015 in the colonies supplied with nectar substitute (sugar:water) at 2:1 ratios + 100 g mungbean flour as pollen substitute which was statistically similar (111.00 cm²) to that of nectar and pollen substitute provided at the ratio of 1.5:1 but differed significantly when nectar and pollen supplemented at the lowest concentration of 1:1 and the untreated control. The space (90.00 cm²) covered by egg was obtained in the colony treated with nectar and pollen substitute at a ratio of 1:1 which was found statistically similar to that of untreated control (82.3).

The highest space (221.6 cm²) covered by live larvae available before the dearth period was observed in the colonies supplemented with nectar (sugar:water) at 2:1 ratios plus mungbean flour as pollen substitute but this was statistically similar to that where nectar

substitute was provided at the ratio of 1.5:1 (207.67 ab) + 100 g mungbean flour as pollen but differed significantly where nectar was supplemented at lowest concentration of 1:1 + 100 g mungbean flour as pollen. The space (190.67 cm²) covered by larvae was obtained in the colony treated with nectar at a ratio of 1:1 + 100 g mungbean flour as pollen which was found statistically similar to those provided with nectar in the ratios of 1.5:1 + 100 g mungbean flour as pollen. All were statistically different from lowest space (115 cm²) covered by larva in the untreated control (Table 2).

Table 2. Effect of food supplements in the bee colony in August, 2015 on the growth and development of honey bee, *A. mellifera*

Food supplements	Parameter				
	Space covered by egg (cm ²)	Space covered by larvae (cm ²)	Space covered by pupae (cm ²)	No. of worker per 4 (cm ²) area	No. of drone per frame
T ₁ =Nectar (sugar:water=1:1) and mungbean flour	90 b	190.67 b	267.33 b	2.443 b	1.12 c
T ₂ =Nectar (sugar:water=1.5:1) and mungbean flour	111 a	207.6 ab	290 a	2.795 b	1.36 b
T ₃ =Nectar (sugar:water=2:1) and mungbean flour	120.6 a	221.6 a	310 a	3.238 a	1.913 a
Untreated Control	82.3 b	115 c	136 c	1.936 c	0.946 d
CV	5.11%	5.66%	4.09 %	7.58 %	5.95%

Mean followed by uncommon letters in a column differed significantly ($p < 0.05$) from each other by LSD.

In case of pupae of *A. mellifera* the highest space covered (310 cm²) by pupae was found in the colony treated with nectar substitute sugar:water at 2:1 plus pollen (mungbean flour) which was statistically similar to that of the treatment where nectar

(sugar:water) at 1.5:1 and pollen mungbean flour were used. Both are significantly different from (267.33 cm²) that of the treatment where nectar substitute (sugar:water) at 1:1 plus mungbean flour were supplied. Lowest space (136 cm²) was obtained from the untreated control colony (Table 2).

The number of workers per 4 cm² area was found highest (3.238) in the colony treated with T₃ composed of nectar supplements, sugar:water=2:1 and pollen substitute, Mungbean flour, which was significantly different (2.795) from the colony treated with T₂ comprising nectar supplements, sugar:water=1.5:1 and pollen substitute, (mungbean flour). The second lowest number of workers (2.443) was found in the colony treated with T₁ having lowest concentration (1:1) of sugar:water and mungbean flour as pollen substitute. There is no significant difference between the last two food supplements (T₁ & T₂). The lowest number of workers (1.936) was found in untreated control colony which was significantly different from all others treatment (Table 2).

The colony treated with T₃ provided the highest number of drone (1.913) per frame (4 cm²) that differs significantly with the colonies treated with T₂ (1.36) and T₁ (1.12) and T₂ and T₁ treatments were also significantly different from each other. The lowest number of drone (0.946) was counted in the untreated control colony that also differed significantly from all other treatments (Table 2).

From this result it is evident that, having nectar supplements at higher concentration plus pollen substitute (mungbean flour) provided better colony development than that of other food substitutes. It is also clear that the colonies treated with T₃ having the highest nectar concentration plus pollen substitute (mungbean flour) resulted strong colonies with higher number of population. This result was similar to those of Standifer *et al.* (1977) who reported that the presence of pollen and nectar supplement in adequate quantity initiate better colony development.

4.2.2 Space (cm²) covered by eggs, larvae, pupae, number of workers and drone per frame in September, 2015

The effect of nectar and pollens substitute at different concentration plus pollen supplement on laying eggs by queen and ultimate production of larvae, pupae, workers and drone in the month of September 2015 was shown (Table 3).

In September maximum space covered by eggs (145.33 cm²) was recorded in the colony treated with T₃ (sugar:water=2:1 and mungbean flour) which was statistically similar to that of colony (136.67 cm²) treated with T₂ (sugar:water=1.5:1 and mungbean flour) but significantly different from the colony (128.33 cm²) treated with T₁ (sugar:water=1:1 and mungbean flour). Food substitute of T₂ and T₁ were statistically similar. The lowest space (94 cm²) was covered in the control colony which was significantly different from all others treatment (Table 3).

The maximum space (300 cm²) covered by larvae was found in the colony treated with the treatment T₃ (sugar:water=2:1 and mungbean flour) followed by (242.3 cm²) and (164.3 cm²) when colonies are treated by T₂ (sugar:water=1.5:1 and mungbean flour) and T₁ (sugar:water=1:1 and mungbean flour), respectively and it varied significantly each other. The lowest space (110 cm²) covered by larva was obtained in untreated control which is significantly different from all other treatments (Table 3).

The highest space (321 cm²) covered by pupae was also found in the colony treated with treatment T₃ (sugar:water=2:1 and mungbean flour) followed by (261 cm²) and (216 cm²) when colonies are treated by T₂ (sugar:water=1.5:1 and mungbean flour) and T₁ (sugar:water=1:1 and mungbean flour), respectively and they differed significantly from each other. The lowest space (155 cm²) covered by pupae was obtained in untreated control colony which is significantly different from all other treatments (Table 3).

The maximum number of worker per 4 cm² area (3.79) was obtained in the colony treated with T₃ (sugar:water=2:1 and mungbean flour) which is significantly different from that of colony (3.105) treated with T₂ (sugar:water=1.5:1 and mungbean flour) and colony (2.703) treated with T₁ (sugar:water=1:1 and mungbean flour). But T₂ and T₁ were statistically similar to each other. The lowest number of worker (1.33) was found in the untreated control colony which was significantly different from all others treatment (Table 3).

Table 3. Effect of food supplements in the bee colony in September, 2015 on the growth and development of honey bee, *A. mellifera*

Food supplements	Parameter				
	Space covered by egg (cm ²)	Space covered by larvae (cm ²)	Space covered by pupae (cm ²)	No. of worker per 4 (cm ²) area	No. of drone per frame
T ₁ =Nectar (sugar:water=1:1) and mungbean flour	128.33 b	164.3 c	216.67 c	2.703 b	1.242 c
T ₂ =Nectar (sugar:water=1.5:1) and mungbean flour	136.67 ab	242.3 b	261.67 b	3.105 b	1.57 b
T ₃ =Nectar (sugar:water=2:1) and mungbean flour	145.33 a	300 a	321.67 a	3.79 a	2.026 a
Untreated Control	94.33 c	110 d	155 d	1.33 c	1.026 d
CV	3.92%	4.88 %	5.41 %	8.32 %	6.75 %

Mean followed by uncommon letters in a column differed significantly ($p < 0.05$) from each other by LSD.

The colony treated with T₃ provided the highest number (2.026) of drone per frame that differs significantly with the colonies treated with T₂ (1.57) and T₁ (1.242) treatments. But here drone frame⁻¹ in T₂ and T₁ were significantly varied from each other.

Significantly the lowest number of drone (1.026) was obtained in the colony of untreated control (Table 3).

The result revealed that in September T₃ having nectar and pollen supplement with higher concentration plus pollen substitute (mungbean flour) provided better colony development than those of other treatments. Like those of August untreated colony having the lowest number of population. Beside this, T₁ treated colonies having second lowest concentration of nectar alternative and pollen substitute resulted weak colonies with lower number of population. The present result was almost similar to that of Abbas *et al.* (1995) who reported that adequate pollen and nectar supplement with higher concentration initiate healthy colony development.

4.2.3 Space (cm²) covered by eggs, larvae, pupae, number of workers and drone per frame in October, 2015

The effect of nectar and pollens substitute applied at different concentration on the queen's ovipositional activity and subsequently emergence of larvae, pupae, workers and drone per frame in October, 2015 were shown in Table 4.

Significantly the highest space (284 cm²) covered by eggs was obtained in the colony treated with T₃ food supplements which was followed by T₂ (258 cm²) and T₃ (235 cm²) treated colonies. Numerically the lowest space covered by eggs was found in the colony of untreated control (183 cm²) which was statistically different from all other treated colonies.

In October the maximum space (286 cm²) covered by larvae was observed in the colony provided with T₃ treated control which was followed by T₂ (257 cm²) and T₁ (237 cm²) ones and their level of significance varied from each other. Significantly the lowest

space covered by eggs was found in the colony of untreated control (192 cm²) which was statistically different from all other treated colonies (Table 4).

In this month the highest space covered by pupae (288 cm²) was obtained in the colony treated with T₃ (sugar:water=2:1 and mungbean flour) which is significantly different from that of the colony (263 cm²) treated with T₂ (sugar:water=1.5:1 and mungbean flour) and (244 cm²) treated with T₁ (sugar:water=1:1 and mungbean flour). But space covered by T₂ and T₁ treated colonies were statistically similar to each other. The lowest space covered by pupae (1.87 cm²) was found in the untreated control colony was significantly different from all others food substitutes.

Table 4. Effect of food supplements in the bee colony in October, 2015 on the growth and development of honey bee, *A. mellifera*

Food supplements	Parameter				
	Space covered by egg (cm ²)	Space covered by larvae (cm ²)	Space covered by pupae (cm ²)	No. of worker per 4 (cm ²) area	No. of drone per frame
T ₁ =Nectar (sugar:water=1:1) and mungbean flour	235 c	237 c	244 b	2.988 b	1.515 c
T ₂ =Nectar (sugar:water=1.5:1) and mungbean flour	258 b	257 b	263 b	3.42 ab	1.9307 b
T ₃ =Nectar (sugar:water=2:1) and mungbean flour	284 a	286 a	288 a	3..895 a	2.40 a
Untreated Control	183 d	192 d	187 c	1.595 c	1.004 d
CV	3.51 %	2.82 %	3.87 %	13.34 %	9.28 %

Mean followed by uncommon letters in a column differed significantly ($p < 0.05$) from each other by LSD.

The number of workers per 4 cm² area was found highest (3.895) in the colony treated with T₃ treatment which was statistically similar to that of the colony treated with T₂ (3.42) treatment but differed significantly with those of the colony treated with T₁. Later treatment (T₁) accommodate 2.988 workers per 4 cm² frame which was statistically similar with those of the colony had T₂ treatment (Table 4). The lowest number of worker found in the untreated control colony (1.595) which was significantly lower than any other treated colony.

The colony provided with T₃ treatment gave rise the highest number (2.40) of drone per frame that differs significantly with those of the colonies treated with T₂ (1.93) and T_a (1.515). But the number of drone frame⁻¹ in T₂ and T₁ also significantly varied from each other. As usually the lowest number of drone (1.004) was obtained in the colony of untreated control (Table 4).

The result reflected that T₃ having the nectar substitute at higher concentration plus pollen supplement ensured better colony development compared to other treatments with lower concentration of nectar alternative. The colonies with nectar and pollen substitute having the lowest concentration plus the pollen supplement (T₁) resulted reduced population of various developmental stages of the bees. Almost comparable finding was reported by Pesante *et al.* (1992) who found that the presence of pollen and nectar supplement in sufficient quantity ensured better colony development.

4.2.4 Space (cm²) covered by eggs, larvae, pupae, number of workers and drone per frame in November, 2015

The impact of nectar and pollens substitute at different concentration on egg deposition ability of queen and subsequent development of larvae, pupae, workers and drone in the month of November, 2015 was shown in Table 5.

Significantly the highest space covered by eggs (300 cm^2) was obtained in the colony treated with T_3 followed by spaces 282.0 cm^2 and 248.8 cm^2 when hives were treated with T_2 and T_1 , respectively in November and it varied significantly from each other. Significantly the lowest space (182 cm^2) covered by eggs was observed in the colony of untreated control (Table 5).

Similar trend was found for space covered by larvae in this month. Significantly the highest space covered by larvae (298 cm^2) was found in the colony treated with T_3 , followed by the space 261 cm^2 and 221 cm^2 when the colonies were treated with T_2 and T_1 treatments, respectively and they are significantly different from each other. Like the previous months significantly the lowest space (209.7 cm^2) was covered by larvae in the colony of untreated control (Table 5).

Like the space covered by egg and larvae, the highest space (315 cm^2) covered by pupae was found in the colony treated with treatment T_3 (sugar:water=2:1 and mungbean flour) which was followed by (288 cm^2) and (273 cm^2) when colonies are treated by T_2 (sugar:water=1.5:1 and mungbean flour) and T_1 (sugar:water=1:1 and mungbean flour), respectively and T_3 was significantly different from those of T_2 and T_1 but the area covered by T_2 and T_1 was statistically similar. The lowest space (188 cm^2) covered by pupae was obtained in untreated control colony which was significantly different from all other treatments.

The number of workers per 4 cm^2 area was the highest (4.566) in the colony treated with the treatment T_3 followed by the area (4.105 cm^2) and (3.24 cm^2) in colonies treated with by T_2 and T_1 treatment, respectively and they were varied significantly from each other. The lowest number of workers (1.9367) was obtained in the untreated control colony which was statistically different from all other colony treated with food supplements (Table 5).

Table 5. Effect of food supplements in the bee colony in November, 2015 on the growth and development of honey bee, *A. mellifera*

Food supplements	Parameter				
	Space covered by egg (cm ²)	Space covered by larvae (cm ²)	Space covered by pupae (cm ²)	No. of worker per 4 (cm ²) area	No. of drone per frame
T ₁ =Nectar (sugar:water=1:1) and mungbean flour	248 c	221 c	273 b	3.24 c	1.84 b
T ₂ =Nectar (sugar:water=1.5:1) and mungbean flour	282 b	261 b	288 b	4.105 b	2.133 b
T ₃ =Nectar (sugar:water=2:1) and mungbean flour	300 a	298 a	315 a	4.566 a	2.52 a
Untreated Control	183 d	163 d	188 c	1.936 d	1.03 c
CV	3.24 %	4.76 %	4.82 %	6.01%	7.81 %

Mean followed by uncommon letters in a column differed significantly ($p < 0.05$) from each other by LSD.

The highest number of drone per frame (2.52) was found in the colony treated with T₃, followed by T₂ (2.133) and T₁ (1.84), respectively and it significantly different from the later two treatments. But drone frame⁻¹ of T₂ and T₁ was statistically similar. The lowest number of drone per frame (1.03) was obtained in the untreated control colony and it was significantly lower than any other treated colony.

The present promising results were due to use of nectar and pollen substitutes which might encouraged queen to lay healthy egg and subsequently increased the production of larvae, pupae, workers, drone. This finding may be explained particularly with the findings of the other author. Augustin (1994) found increased number of bee flight (28000 bees per day) due to addition of supplementing sugar solution in the month of August. Flight activity may drop to zero if sugar feeding is stopped. Tahir *et al.* (1995)

reported that the flour of black gram at 550g per kg added in the diet of *A. mellifera* colonies as pollen substitute during summer rainy season produced a higher number of frames, resulting in higher production of honey compared to pollen substitute using soybean meal.

4.2.5 Space (cm²) covered by eggs, larvae, pupae, number of workers and drone per frame in December, 2015

Positive impact of nectar and pollen substitute on the production of eggs and subsequently on the growth and development of larvae, pupae, workers and drone in December, 2015 was shown in Table 6.

The highest space covered by eggs (397 cm²) was found in the colony treated with T₃ followed by space 363 cm² and 285 cm² occupied by eggs in the colony treated with T₂ and T₁, respectively and they varied significantly from each other. The lowest space occupied by eggs (195 cm²) was found in the untreated control colony (Table 6).

Similarly, the maximum space covered by larvae (306.0 cm²) was observed in the colony treated with T₃ followed by space 280 cm² and 256 cm² occupied by larvae in the colonies treated with T₂ and T₁, respectively and they were statistically different from each other. Like other months the lowest space (181 cm²) was covered by larvae was the untreated control colony (Table 6).

In the month of December, 2015 again the highest space covered by pupae (420 cm²) was measured in the colony treated with T₃ treatment followed by space 361 cm² and 281 cm² when the colonies were treated with T₂ and T₁ treatment, respectively and these were significantly different from each other. Similarly, the lowest space (200 cm²) was covered by pupae in the untreated control colony which was significantly different from all other treatments (Table 6).

Table 6. Effect of food supplements in the bee colony in December, 2015 on the growth and development of honey bee, *A. mellifera*

Food supplements	Parameter				
	Space covered by egg (cm ²)	Space covered by larvae (cm ²)	Space covered by pupae (cm ²)	No. of worker per 4 (cm ²) area	No. of drone per frame
T ₁ =Nectar (sugar:water=1:1) and mungbean flour	285 c	256 c	281 c	3.773 c	1.932 c
T ₂ =Nectar (sugar:water=1.5:1) and mungbean flour	363 b	280 b	361 b	4.296 b	2.746 b
T ₃ =Nectar (sugar:water=2:1) and mungbean flour	397 a	306 a	420 a	5.083 a	3.26 a
Untreated Control	195 d	181 d	200 d	2.048 d	1.073 d
CV	3.35 %	4.33 %	5.38 %	5.57 %	5.16 %

Mean followed by uncommon letters in a column differed significantly ($p < 0.05$) from each other by LSD.

The highest number of worker per 4 cm² area (5.083) was found in the colony treated with T₃, treatment consisting of the higher concentration of nectar substitute plus pollen supplement. This was followed by 4.296 and 3.773 numbers when colonies were treated with T₂ and T₁ treatment, respectively. Significantly the lowest number of workers (2.048) was obtained in the colony treated with untreated control colony (Table 6).

The colony treated with T₃ produced highest number (3.26) of drone per frame followed by those of the colonies treated with T₂ (2.746) and T₁ (1.932) treatments and they were significantly different from each other significantly. The lowest number of drone per frame (1.073) was counted in the untreated control colony. (Table 6).

The results in the month of December suggested that the nectar supplement at higher concentration plus pollen substitute contribute in better colony development compared

to food substitute at lower concentration. Lower concentration of nectar and pollen substitutes unable to contribute positively in the growth and development of different stages of bees. Similarly, Moeller *et al.* (1978) found necessity of carbohydrate substitute for the sound management of honey bee colonies. Because proper colony management should ensure adequate honey reserves in the hive at all times but feeding sugar may sometimes be necessary. Whenever the honey supply in the colony is low and nectar in field is short supply due to adverse weather, the colonies should be fed sugar supplement. Brood rearing requires a large quantity of honey and pollen. Cane or sugar beet, isomerized corn syrup and type-50 sugar syrup are satisfactory substitute for honey in the natural diet of honey bees (Moeller *et al.*, 1978).

4.2.6 Comparative effect of with highest concentration (2:1) nectar substitute plus pollen supplement in August through December, 2015 on the growth and development of bees

In the diet with the highest concentration (2:1) of nectar plus pollen substitutes was found to be most effective compared to medium (1.5:1) and lower (1:1) concentration of nectar supplements. The effects of the nectar substitutes at highest concentration on the growth and development of the bees was varied in different months from August to December, 2015 (Figure 1 and Figure 2).

The effect of nectar substitutes at highest concentration plus pollen supplement on egg deposition ability of queen and subsequent development of larvae and pupae in the months of August through December 2015 was shown in Figure 1.

In first month, August, 2015, no eggs were laid by the queen but in the next month (September) space covered by eggs was 145.33 cm² and the increasing trends of laying

eggs increased in October (284 cm²), November (300.0 cm²) and in December (397 cm²) (Figure 1).

Space (cm²) covered by larvae was varied in different months and the curves showing (Figure 1) gradual raising pattern during August through December, 2015. The highest space (306 cm²) covered by larvae was observed in December, 2015 and the lowest (221 cm²) was in August, 2015.

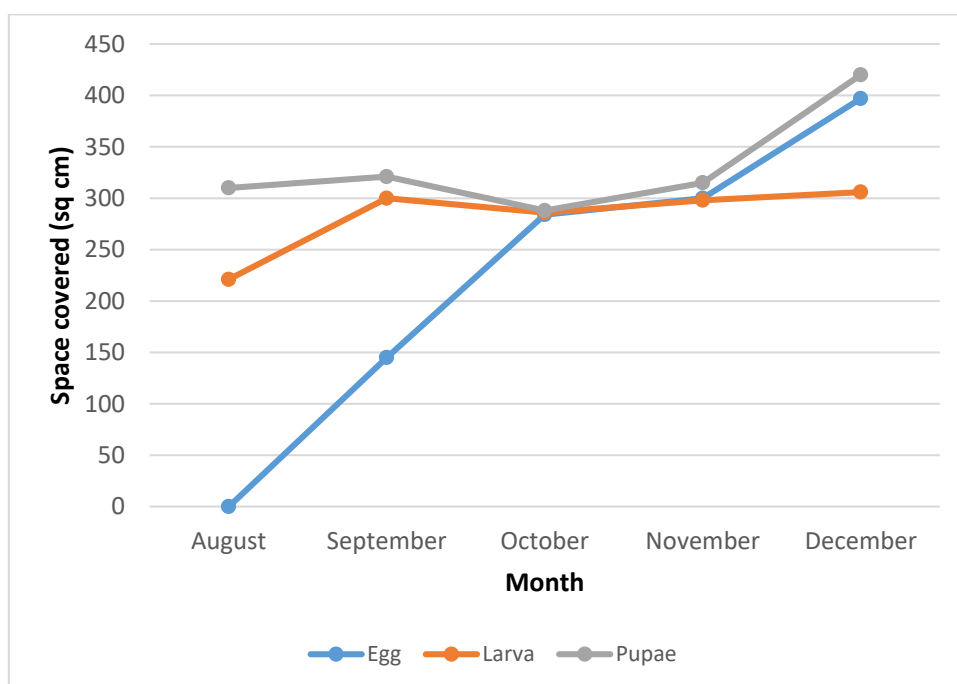


Figure 1. Space (cm²) covered by egg, larvae and pupae oh honey bee in August through December 2015 in the colonies treated with highest concentration of nectar substitute plus pollen supplement.

During August through December 2015, space occupied by pupae providing the dissimilar patterns of curve (Figure 1). In the month of September and October 2015 space covered by pupae was 321 cm² and 288 cm², respectively the curves showing decreasing patterns of pupal growth and development. In the month of October pupal population was lowest (288 cm²) and it might be due to adverse climatic condition. In

the next month (November) space covered by pupae was increased to 315 cm² and the trends remained the same up to December when the space occupied by pupae was the maximum (400 cm²).

Due to use of artificial food supplements at highest concentration encouraged queen fecundity and encouraged to lay healthy egg and subsequently increased the production of larvae and pupae in the colony. The results were found to be almost similar with the findings of Chhuneja *et al.* (1996) who reported that adequate food supplements prevent early brood mortality and initiate healthy colony multiplication and development through the dearth period when there is scarcity of natural food supplements.

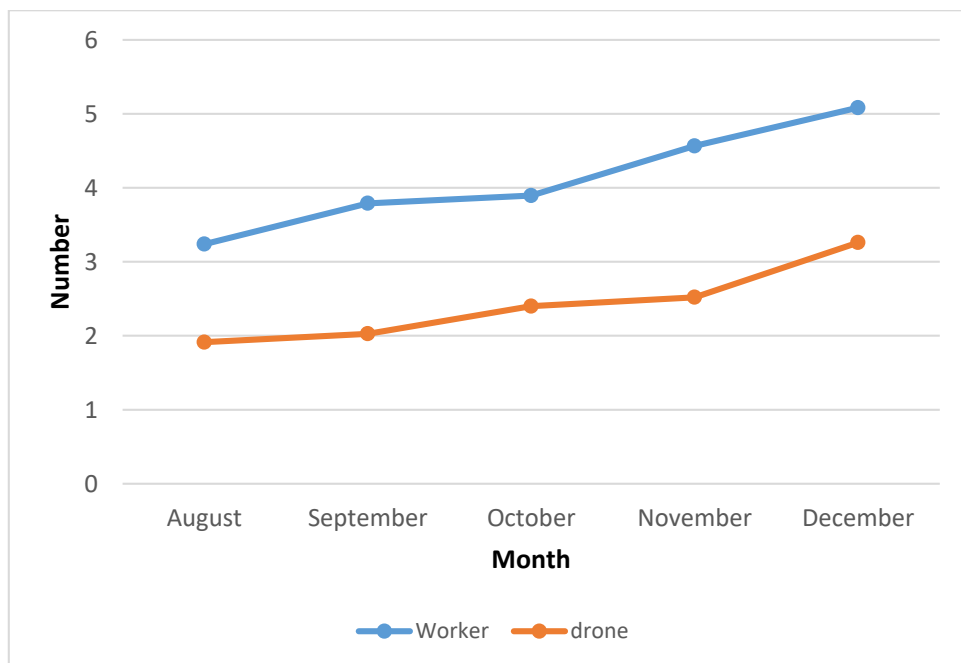


Figure 2. Contribution of highest concentration of nectar substitute plus pollen supplement on number of worker and drone population and queen cells development during August through December, 2015.

Artificial nectar substitutes at higher concentration (2:1) on worker population and drone per frame during August through December, 2015 was presented in Figure 2.

Number of workers per 4 cm² area from August to December, 2015 providing gradual increasing trend. The lowest (3.238) number of workers per 4 cm² area was observed in the month of August, 2015 and the highest (5.083) number of workers was in the month of December, 2015 (Figure 2).

Drone number per frame showed positive trends of increase during August through December, 2015 (Figure 2). The highest number of drone (3.26) per frame were observed in the month of December. The minimum number of drone (1.913) was found in the month of August, 2015.

4.3 Foraging behavior: Number of worker bees ingressed in the hive per minute

In a hive at SAU apiary, Substantial variation was observed in number of worker bees ingressed in the hive per minute during 07:00 to 8:00 am of a day in different weeks of a month. And the number of bees ingressed in the hive varied in different months throughout the dearth period (Table 7).

In first month, August 2015 significantly the highest number of worker bees ingressed (3.344) min⁻¹ in the colony of treatment T₃ during 07:00 to 8:00 am followed by 2.75 min⁻¹ and 2.00 min⁻¹ in colony treated with T₂ and T₁, respectively. The lowest number of worker ingressed (1.455 egress) was from the untreated control colony (Table 7).

In 2nd month, September 2015, significantly the highest number of worker bees ingressed was 10.40 min⁻¹ from the colony treated with treatment T₃ followed by 8.889 min⁻¹ and 7.416 min⁻¹ in the colony treated with treatment T₂ and T₁, respectively. The lowest number of worker ingressed (4.633 min⁻¹) from the untreated control colony (Table 7).

In October 2015, significantly the highest number of worker bees ingressed was 20.494 min^{-1} from the colony which was treated with treatment T₃ followed by 16.139 min^{-1} and 12.506 min^{-1} in the colony treated with treatment T₂ and T₁, respectively. Same as previous month the lowest number of worker ingressed (6.528 min^{-1}) from the colony of untreated control (Table 7).

Table 7. Number of worker bees (*A. mellifera*) ingressed per minute in a hive from 7:00 to 8:00 am of the day for five months in an apiary at SAU during the month of August to December 2015.

Food supplements	No. of worker bee ingressed per min				
	August	September	October	November	December
T ₁ =Nectar (sugar:water=1:1) and mungbean flour	2.00 c	7.417 b	12.506 c	17.71 c	19.389 c
T ₂ =Nectar (sugar:water=1.5:1) and mungbean flour	2.75 b	8.889 ab	16.139 b	21.017 b	23.528 b
T ₃ =Nectar (sugar:water=2:1) and mungbean flour	3.344 a	10.40 a	20.494 a	28.056 a	29.944 a
Untreated Control	1.455 d	4.633 c	6.528 d	9.294 d	10.139 d
CV	8.69 %	13.36 %	7.01 %	8.35 %	6.44 %

Mean followed by uncommon letters in a column differed significantly ($p < 0.05$) from each other by LSD.

In November 2015, the highest number of worker bees ingressed was 28.056 min^{-1} from the colony treated with treatment T₃ followed by 21.017 min^{-1} and 17.71 min^{-1} in the colony treated with treatment T₂ and T₁, respectively and they significantly varied from each other. Same as previous month the lowest number of worker ingressed (9.294 min^{-1}) from the untreated control colony (Table 7).

In December 2015, the maximum number of worker bees ingressed was 29.944 min^{-1} from the colony which was treated with treatment T₃ followed by 23.528 min^{-1} and

19.389 min⁻¹ in the colony treated with treatment T₂ and T₁, respectively and all were significantly different from each other. As usually the lowest number of worker ingressed (10.139 min⁻¹) in the untreated control colony (Table 7).

CHAPTER V

SUMMARY AND CONCLUSION

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SUMMARY

The present study on the "rearing efficiency, foraging behavior and dearth period management of honey bee (*A. mellifera*)" was undertaken in the SAU campus during August to December 2015 to determine the effect of artificial nectar and pollen supplement at different concentration in the dearth period. Four treatments consisted of the substitute of nectar in the form sugar:water applied in the ratios of 1:1, 1.5:1 and 2:1 plus mungbean flour as pollen supplement. The study was replicated 3 times following RCBD, from each of the colonies six frames were selected to maintain the homogeneity of honey bee population. Several management practices like sanitation of the colony, fumigation of the colony by tobacco leaf and Sevin powder were sprayed to reduce the ant infestation in the colony. Data were collected at seven days' intervals on space (cm² covered by eggs, larvae and pupae of the comb, number of workers/4 cm² area, number of drone per frame in the treated and untreated colonies from August to December 2015.

In the present study by the artificial nectar and pollen supplements at different concentration during August to December 2015 the population of honey bee of different parameters viz., eggs, larvae, pupae, workers, drone etc. were affected. During the month of August, no eggs were found. The highest space covered by eggs (397 cm²) was found in the colony provided with T₃ (using highest concentration of nectar substitute 2:1 plus mungbean flour as pollen supplements) during the month of December and the lowest space was found (94.33 cm²) in the untreated control colonies. And among the treatment T₃ (using lowest concentration of nectar substitute 1:1 plus

mungbean flour as pollen supplements) treated colonies gave the lowest space (145.33 cm²) in month of September which is significantly different from that of untreated control. The maximum space covered by larva (306 cm²) was found in the colonies provided with T₃ treatment during the month December and lowest space covered larva (110 cm²) in September in untreated control which varied significantly from second lowest (164 cm²) in same month in case of T₁ treatment. The maximum space covered by pupae (420 cm²) was found in the colonies provided with T₃ treatment during the month December and lowest space covered larva (155 cm²) in September in untreated control which varied significantly from second lowest (216 cm²) in the same month in T₁ treatment. Highest number of worker (5.083) and drone (3.26) were recorded in the month of December in the colonies treated with T₃ food substitutes. The minimum number of worker and drone were in untreated control hive which was significantly different from the second lowest number of workers (2.443) and drone (1.12) in the month of August.

Number of honey bee ingressed in the hive during dearth period from 07.00-08.00 am in five months was observed and found that, number of worker bee ingressed per minute was increased per month in all treatment. Highest number of worker bee ingressed from hive (29.944) was in the month of December in the colony treated with treatment T₃. Lowest number of worker bee ingressed in the hive in the month of August, was 1.455 in the untreated control colony and it was 2.00 in the T₃ treated colony and they were varied significantly.

CONCLUSION

The overall results of the present study indicated that for the management of *Apis mellifera* during dearth period from August to December 2015 artificial nectar substitute at various ratios (1:1, 1.5:1, 2:1) plus mungbean flour as pollen supplements influence the eggs, larvae, pupae, number of workers and drone, fecundity of the bee colony. Space (cm²) covered by eggs, larvae, pupae, number of workers per 4 cm² area and drone per frame (4 cm²) was statistically higher in the colonies fed with sugar:water=2:1 as nectar plus mungbean flour as pollen supplements as compared to those colonies treated with nectar substitute at the ratios of 1:1 and 1.5:1 plus mungbean flour as pollen supplements.

The worker bees ingressed into the hive in response to the availability of pollen and nectar yielding plants. Number of ingress of worker bee increased with progress of month and highest number of worker bees ingress in the colony which treated with sugar:water=2:1 as nectar plus mungbean flour as pollen supplements as compared to those colonies treated with nectar substitute at the ratios of 1:1 and 1.5:1 plus mungbean flour as pollen supplements.

RECOMMENDATIONS

Based on the above results following recommendations may be made:

The higher nectar substitute concentration (sugar:water at 2:1) plus pollen supplements significantly increase bee population during dearth period which ensured maximum honey production following this period due to higher foraging tending of these honey bees.

Therefore, the migratory beekeepers of Bangladesh may be suggested to follow this practice during the dearth period to get good harvest of honey in the rest following season.

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