

**STUDIES ON BROOD SURVIVAL RATE AND
FORAGING BEHAVIOUR OF APIS MELLIFERA L.
ON MUSTARD**

BY

SAIKAT GHOSH



**MASTER OF SCIENCE IN AGRICULTURE
(ENTOMOLOGY)**

DEPARTMENT OF ENTOMOLOGY

**DR. RAJENDRA PRASAD CENTRAL AGRICULTURAL UNIVERSITY
PUSA (SAMASTIPUR) – 848 125, BIHAR**

2021

Regd. No. - M/ENTO/331/2019-20

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A THESIS SUBMITTED TO THE DR. RAJENDRA PRASAD
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PUSA (SAMASTIPUR) - 848 125, BIHAR, INDIA**

2021

Regd. No. - M/ENTO/331/2019-20



*Dedicated to my
beloved parents and my
chairman*

Dr. Nagendra Kumar



DR. RAJENDRA PRASAD CENTRAL AGRICULTURAL UNIVERSITY
PUSA (SAMASTIPUR) – 848 125, BIHAR

Dr. Nagendra Kumar
Assistant Professor
Dr. Rajendra Prasad Central
Agricultural University



Department of Entomology

Dated..15..1.5.7.. /2021

Certificate

This is to certify that the thesis entitled “Studies on brood survival rate and foraging behaviour of *Apis mellifera* L. on mustard” submitted in partial fulfilment of the requirements for the award of the degree of MASTER OF SCIENCE IN AGRICULTURE (ENTOMOLOGY) of the Post Graduate College of Agriculture, Dr. Rajendra Prasad Central Agricultural University, Pusa (Samastipur), Bihar, is genuine record of bonafide research work carried out by Mr. SAIKAT GHOSH, under my guidance and supervision. No part of the thesis has been submitted for any other degree or diploma.

It is further certified that such help or information received during the course of this investigation and preparation of the thesis have been duly acknowledged.

Nagendra Kumar
19/07/2021

(Dr. NAGENDRA KUMAR)
Chairman
(Advisory Committee)

ENDORSED

A. K. Misra
19/07/2021
Prof. & Head
P.G Deptt. of Entomology
RPCAU, Pusa (Samastipur)

(Dr. A. K. MISRA)


Professor and Head

Department of Entomology

Dr. Rajendra Prasad Central Agricultural University
Pusa (Samastipur), Bihar


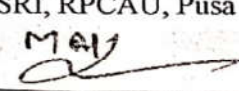

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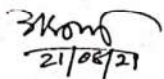
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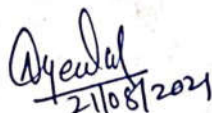
1. 
19/07/2021
(Dr. ANIL KUMAR)
Assistant Professor
Entomology
SRI, RPCAU, Pusa
2. 
(Dr. Md. MINNATULLAH)
Assistant Professor
Plant Pathology
SRI, RPCAU, Pusa
3. 
19.7.2021
(Dr. SANGEETA DEO)
Professor and Head
Department of Textile and
Apparel Designing
Director of Education Nominee
RPCAU, Pusa

Certificate

This is to certify that the thesis entitled "Studies on brood survival rate and foraging behaviour of *Apis mellifera* L. on mustard" submitted in partial fulfilment of the requirements for the award of the degree of MASTER OF SCIENCE IN AGRICULTURE (ENTOMOLOGY) of the Post Graduate College of Agriculture, Dr. Rajendra Prasad Central Agricultural University, Pusa, Samastipur, Bihar was examined and approved on 21/08/2021.

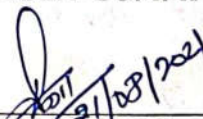

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EXTERNAL EXAMINER


21/08/2021

(Dr. NAGENDRA KUMAR)
Chairman
Advisory Committee

MEMBERS OF THE ADVISORY COMMITTEE


21/08/2021

(Dr. ANIL KUMAR)
Assistant Professor
Department of Entomology
SRI, RPCAU, PUSA


21/08/2021

(Dr. Md. MINNATULLAH)
Assistant Professor
Department of Plant pathology
SRI, RPCAU, PUSA

ENDORSED


21.8.2021

(Dr. A. K. Rai)
Professor and Head
Department of Entomology
Dr. Rajendra Prasad Central Agricultural
University,
Pusa (Samastipur), Bihar.


21/08/2021

(Dr. SANGEETA DEO)
Professor and head
Department of Textile and Apparel
Designing
Director of Education Nominee
RPCAU, PUSA

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Place: RPEAU, PUSA

Date: 19.07.2021

*Saikat Ghosh
(SAIKAT GHOSH)*



**DR. RAJENDRA PRASAD CENTRAL AGRICULTURAL UNIVERSITY
PUSA (SAMASTIPUR), BIHAR- 848 125**

Post Graduate degree programme : M.Sc. (Ag.)
Department : Entomology
Name of student : Saikat Ghosh
Major subject : Entomology
Minor Subject : Plant Pathology
Registration number : M/ENTO/331/2019-20
Major advisor : Dr. Nagendra Kumar
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ABSTRACT

The experiment on “Studies on brood survival rate and foraging behaviour of *Apis mellifera* L. on mustard” were carried out at University Apiary, RPCAU, Pusa during the *rabi* 2020-21. Objectives of this experiment were to determine the brood survival rate in the existing *A. mellifera* population, study the impact of brood survival rate on behavioural, economic and resistant traits of *A. mellifera* and to study the foraging behaviour of *A. mellifera* L. on mustard.

The obtained results showed that out of 10 colonies used for research 1 colony showed low brood survival rate, 3 colonies witnessed medium brood survival rate and 6 colonies showed high brood survival rate. Excellent, medium and poor brood pattern was found in 7, 2 and 1 colonies respectively in the experiment on scattered brood pattern.

The obtained data on bee activity showed that colony no. 9 having lowest number of outgoing bees with average 9 bees / minute. Highest outgoing bees were recorded in colony no. 3 with average 12.11 bees / minute. In case of incoming bees colony no. 5 having the highest value with average 11.78 incoming bee and colony number 9 having the less value with average 8.56 of bees came back to the hive.

Total honey content was maximum in colony no. 5 with 394.94 g and lowest in colony no. 9 having only 249.86 g of honey. For pollen content the same trend was

followed as colony no. 5 having maximum area of pollen of 36.13 cm² and lowest in colony no. 9 with total area of 25.16 cm² pollen.

The obtained results of hygienic behaviour showed that colony no. 3 having 100 per cent efficiency while colony no. 9 having lowest efficiency of 76%.

The duration of foraging of *Apis mellifera* was experimented out and it was noticed that the mean time of initiation time of foraging was at 7:28 am and mean cessation time of foraging time was 16.45 pm. The mean duration of foraging was 9.17 hours for all 10 colonies. It was noticed that duration of foraging kept increasing as the blooming period progressed. The foraging rate of *A. mellifera* was also calculated during different day hours and the studies shows that mean rate of foraging was highest during 15:00 hours of the day which was 19 flowers/ min while the lowest foraging rate was 0.24 flowers/ min during 07:00 hours. Foraging speed of *A. mellifera* was maximum during 09:00 hours of the day which was 2.94 sec/ flower and on the other side the lowest value was 1.13 sec/ flower obtained during 07:00 hours of the day.

Population dynamics of *A. mellifera* on mustard bloom was recorded during the whole flowering period and it was correlated with average minimum temperature, average maximum temperature, average temperature, average relative humidity (RH) at morning hours, average RH at evening hour, average RH and with total rainfall, the obtained results showed that average maximum temperature(°C) and average temperature(°C) showed positive and strong correlation with significant effect while average minimum temperature(°C) showed positive correlation with significant effect on bee population($r = 0.93^{**}$, 0.94^{**} and 0.76^*) On the other side average morning relative humidity(%) and average relative humidity(%) showed negative correlation with significant effect while average evening humidity showed negative but non-significant effect on bee population on mustard bloom($r = -0.70^*$, -0.64^* and -0.18). There was no rainfall was recorded during the whole experiment so no correlation was found between total rainfall (mm) and bee population. However all the weather parameters influenced 86 % bee population ($R^2 = 0.86$).



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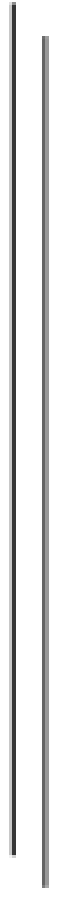
ABBREVIATIONS USED

%	: Per cent
/	: Per
@	: At the rate of
±	: Increase or decrease
⁰ C	: Degree Celsius
m	: Meter
et al.	: (et alii) other people
Fig.	: Figure
g	: Gram
kg	: Kilo gram
ha	: Hectare
hrs.	: Hours
h	: Hour
i.e.	: That is
Viz.	: Namely
No.	: Number
Avg.	: Average
Temp.	: Temperature
RH.	: Relative humidity
sp.	: Species
Max.	: Maximum
Min.	: Minimum
Etc.	: Etcetera
CV	: Coefficient of variance
CD	: Critical difference
S Em.	: Standard error of mean





CHAPTER - I



INTRODUCTION



INTRODUCTION

Honey bees are highly advanced and familiar groups of social insects. Bees are capable of increasing to the yield of cross pollinated and to some extent in case of self-pollinated crops through their pollination activities. It has been found that presently there are equal or more than 25,000 species of honey bees exist in the world and this holds for about 65 per cent pollination activities of different flowering plants. They are working as important pollinating agent for more than 80% of all flowering plants including the majority of agricultural and horticultural crops. Directly or indirectly this group of the insect order hymenoptera holds important role in maintaining the health of human beings and as well as for the ecosystem. Honey bee delivers us vast majority of essential products like honey, venom, pollen, bee wax *etc.* and thereby they directly and indirectly beneficial to the human beings. Rearing of honey bees for their economic products and benefits of human being is popularly called as apiculture or beekeeping.

Within a honey bee colony of a standard size, there are generally 20,000 to 30,000 bees. Their population consist of normally of a single queen and a few hundred drones or males and the remaining population is of workers, which making up over 90% of the population. The queen is the only functional and fertile female of the colony. She is larger in size than the worker. Queen has no wax plates and pollen baskets in her body. She has a well-developed ovipositor which functions also as a sting. The only function she performs is the activity related with reproduction and maintaining the colony strength. She gets mated by drones during the nuptial flight. After successful mating she is confined to her hive and lay eggs through entire her life. A queen met with several drones for successful conception and to full fill her spermatheca.

The worker is the imperfectly developed female or sterile female member of the colony and is the smallest of three in respect to body size. The workers work selflessly for the colony and devoting all their time and attention ever since their time of emergence as adults to the service of the colony as a whole. Young workers performs all the vital domestic or in colony duties like secretion and feeding the immature ones with royal jelly, repairing or building the combs with help of wax secreted by them, attending on the queen, keeping the hive warm by regulation of

temperature, maintain hygiene of the colony and defending the colony from enemies. Many of the experienced and matured workers go out on foraging for collection of pollen, nectar and propolis. They show various structural adaptations to do these varied duties. During both early part of their life when they are confined within the hive and later part of life when they go out on foraging they show certain division of labour, attending to specific duties depending upon their age.

The drone is the functional male and is larger and darker than the worker. Compound eyes are larger than those of a worker and they meet at the top of head. It has no sting, wax glands or pollen baskets. In a colony only a few hundred or sometimes no drones may be found. Its main duty is to take part in the nuptial flight of the queen and to mate her. After the successful mating the male dies as his copulating organ damaged during the mating. Other function of a male in a hive is to maintain the hive temperature and maintaining air circulation in to the hive. Otherwise it remains enjoying the warm sun and the food gathered by the workers.

The queen is capable of laying both fertilized and unfertilized eggs at the same time. From the fertilized eggs females are produced which means the queen and the workers and from the unfertilized eggs males are born. The queen is reared in a special cell which is situated at the lower portion of the comb and that particular cell is much larger than the others. The egg of honey bee generally hatches in three to four days and the larva is fed with a nutritious food rich in protein known as royal jelly. The composition of royal jelly is 15-18% proteins, 18% carbohydrates, 2-6.1% lipids and ash content is 0.7 to 1.2%. The larval stage commonly lasts for 5 to 6 days and the pupal stage lasts for seven days. It is found that the ectodermal secretion of the queen known as 'queen substance' which contains ecto-hormone plays the vital role in the production of a new queen. When the pheromone is in good quantity, it is taken by worker bee and this stops the workers from producing new queen and then only workers are produced. When the queen substance is relatively low, then new queen cells being constructed by the workers and new queens are produced. The only function of the queen is to participate in nuptial flight or is concerned with controlling the colony strength. Once the queen starts egg-laying she does not participate in any more nuptial flights. She can lay up to 500 eggs a single day but on an average during favourable seasons her egg laying ability extends. Queen bee lacks the basic motherly instincts of rearing the immature ones, as these are done by the nurse bees. When a

queen becomes old and become incapable of laying fertilized eggs, another queen replaces her and the old one is discarded from the colony.

There are major five species of honey bees common in India viz., Indian bee or *Apis cerana indica*, rock bee or *Apis dorsata*, European or Italian honey bee or *Apis mellifera*, little bee or *Apis florea* and stingless bee or *Melipona irridipennis* out of which four species are domesticated except *Apis dorsata* and mostly used species in apiculture is *A. mellifera*. Two more species also exist in India which is known as Himalayan bee or *Apis laboriosa* and the black dwarf bee or *Apis andreniformis*. Among them *A. mellifera* and *A. cerana indica* are generally used for domestication and commercialization as their nature, morphology and physiology is more or less same and are suitable for rearing, while the largest honey bee species is *A. dorsata* which is totally unsuitable for domestication as they are more prone to swarming and ferocious in their nature.

According to the records of beekeeping, apiculture seems to have been well known and practised in ancient India. Though, there is very little material as evidence of apiculture during the mediaeval period is available there. Now days, it has witnessed a great comeback as an industry and in some other parts of our country apiculture is increasing slowly and gradually. It got the boost after the successful introduction of high yielding moveable European or Italian honey bee *A. mellifera* L. from Europe to India. *A. mellifera* was introduced from Europe in to our country during 1962 with the major contribution from Dr. A. S. Atwal. Immediately after its introduction, it became popular in states like Haryana, Jammu and Kashmir, Punjab Himachal Pradesh and Uttar Pradesh. After that gradually culturing of *A. mellifera* bee spread in other states like Kerala, West Bengal, Bihar and Karnataka and even in some North-Eastern states.

In Europe and America *A. mellifera* is extensively reared. It is closely similar to *A. cerana indica* in respect to structure and habits but on the other hands it has a prolific queen, having less swarming tendency and has good honey collecting qualities. Annual yielding potential of *A. mellifera* is about 45-180 kg. Fertile and best performing queens of this species had been imported and introduced into Indian colonies in the 1960s. This special importation was mainly done in some parts of our country specially Punjab, Jammu and Kashmir and Maharashtra with the main target

of increasing the yield of honey. It has now well established in the country and slowly replacing *A. cerena indica* hives used for domestication and commercial purpose.

Honey bees are most common and important pollinators of several agricultural or field crops and mustard (*Brassica juncea*) is one of them. Mustard belongs to cruciferae family and generally cultivated as *rabi* crops in India. In Bihar also it is cultivated in winter season. It is one of the popular and mostly cultivated oilseed crops next to sunflower. Mustard is generally called as rai. There are total six cultivated species under the genus Brassica and among them more than 80% of total cultivated area holds by Indian mustard or scientifically known as *Brassica juncea* (Chandrashekhar *et al.*, 2013). Mustard seed are rich in energy content, total oil content in them is around 28-32 % with relatively good amount of protein (28- 36%), while the amino acid presents in mustard proteins are very finely balanced, it is much more rich in essential amino acid required by human beings, it contain 20-28% oleic acid, linoleic acid in 10-20% and lastly 30-40% erucic acid present in mustard protein (sonvance and Pathak 2016). Mustard is highly cross pollinated crops and heavily depends upon pollinators like honey bees for successful fruiting. Being the main pollination service provider, the honey bees adds quite well in increasing the overall production and productivity of these kind of cross pollinated crops with their efficient pollination in an obscure and silent way (Singh *et al.*, 2005 and Mohapatra *et al.*, 2010). If we take into consideration of Indian mustard then it is mainly grown in the states like UP, Rajasthan, Haryana, Madhya Pradesh and Gujarat also found in good numbers in some areas of south India like Telengana, Karnataka, Andhra Pradesh and Tamil Nadu. Yellow sarson is generally taken as *rabi* crop or cultivated during winter season in the states like Bihar, Orissa, Assam and West Bengal where in case of north Indian states like UP, Haryana, Punjab and Himachal Pradesh it is cultivated as catch crop (a quick growing crop or short duration crop grown in between the span of two main crops). Currently rapeseed mustard total area under sowing in India is 5.96 m ha, with a total annual production of 8.32 million tonnes, while the average productivity of it is 1397 kg /ha (Anonymous, 2018).

For successful pollination different traits or characters of visiting agents are directly responsible. *A. mellifera* is one of the pollinating agents having most of the foraging characters in great amount like duration of foraging (the gap between initiation time of foraging and cessation time of foraging), foraging rate (number of

flowers visited by *A. mellifera* per minute), foraging speed (time spent on a single flower by *A. mellifera* in seconds) *etc.* Others parameters also directly and indirectly associated with the performance of *A. mellifera* and thereby involved in foraging behaviour, these are brood survival rate, hygienic behaviour, nest cleaning ability, scattered brood pattern *etc.*

Apart from that, behaviour of bee is very much important in managing honey bee colonies for pollination requirements of crops and production of honey. Bees are attracted to the flowering plants and identify them by their odour, shape, floral rewards and colour. Abiotic factors like humidity, wind speed, air temperature, day length, sun shine hour, *etc.* While the biotic factors includes quality of flora, availability of flower (presence or absence) pollen, extend of nectar, *etc.* also decides bees foraging efficiency which ultimately determines the overall honey production and efficiency in pollination. Mustard is one of the entomophilous crops that require many insect pollinators specially and they provide rich source of pollen and nectar flow throughout the whole flowering period. Among different insect pollinators *A. mellifera* is one of the major and most dominant species, so there contribution should be studied thoroughly in respect to pollinating mustard to understand the gaps to be fulfilled in future. Keeping all these aspects in mind the experiment had been chalked out with the following three objectives:

1. Determination of brood survival rate in the existing *Apis mellifera* L. population.
2. Impact of brood survival rate on behavioural, economic and resistant traits of *Apis mellifera* L.
3. Studies on foraging behaviour of *Apis mellifera* L. on mustard.





CHAPTER - II



REVIEW OF LITERATURE



RIVEW OF LITERATURE

1. Determination of brood survival rate in the existing *Apis mellifera* L. population.

1.1) Studies on Brood survival rate:-

Fukuda and Sakagami (1968) conducted their research on worker brood survival in honeybees to construct life table. They have found that the brood survival rate is high in the centre portion of the hive while it was very less in the peripheral area or outside region of the hive. They performed the experiment with 100.0 eggs, 86.4 sealed brood, 94.2 unsealed brood and 85.1 with adults. The total duration in case of immature stages was 20 days in 87.1%, while for 21 days in 8.3% and then for 19 days or more than 22 days in the residual fraction of successfully workers that were emerging out.

Woyke (1963 and 1972) recorded the rate of survival of broods was less those were produced with instrumentally mated queen bees. This was done in the apiary specially made for mating with the drones originating from queen bees those were closely related and informed that there was increase in the number of sex alleles in their population and it was also increased the average brood's survival rate for the colony.

Woyke (1980 and 1981) discovered that during spring and autumn the three different groups of colonies that having various queens producing brood with survival rate of 100, 75 and 50 per cent respectively witnessed more or less similar brood areas. While in summer, the case is different he witnessed that colonies that having brood survival rate of 75 and 50 per cent capable of producing about 82 and 68 per cent respectively when compared to the brood area of some normal colonies. He also informed that colonies brood survival rate within 75 and 50 per cent had 88 and 79 per cent respectively when compared to the normal spring population of worker bee. While during summer these colonies had 93 and 35 per cent when they consider the population of normal worker bee.

Page and Metcalf (1982) also reported the effect of natural mating on queen bees producing brood that having various survival rates. They also noted that increase

in the successful mating will also increase the number of classes that queen bees producing brood having different survival rates.

Woyke (1984) had observed during spring that colonies those were producing brood having survival rate with 100, 75 and 50 per cent they had only 7, 10 and 20 per cent of the total brood cells vacant. While in summer the values showed that about 0, 14 and 42 per cent of the total brood cells were vacant and in case of autumn season, it was about 10, 13 and 16 per cent were vacant.

Ruttner (1988) observed that huge curtailment of overall productivity took place with the degree of inbreeding and it was when there was more than 25 per cent vacant cells were found inside the hive in sealed brood.

Farhenholz *et al.* (1989), Yi and Jung (2010) and Jeong *et al.* (2016) showed that brood area temperature was narrowly maintained near to be 35 °C for the best development of honey bee larvae. They also observed that the inner temperature of bee hive and inside the cluster of bee ball during winter season was around 21– 24 °C.

Mishra and Kumar (1995) seen that 4.83 to 7.73 per cent vacant brood cells in the bee hives at Hisar (Haryana) and that was much more lower than the hazardous level. After few days Punjab reported that the per cent mortality of immature stages or specifically the larval stage of *A. mellifera* was between 5.76 to 16.94 during other months (Anon, 1996).

Al-Lawati and Bienefeld (2009) observed that correlation between two factors that is in vitro survival rates and parental colony shows that genetically effects or maternal effects can perform a vital role in the brood survival of honey bee.

Ashley and Ellis (2018) conducted a research and showed that the 11-day brood survival percentage may be used when selection will be done for finding suitable colonies to use them as source colonies for testing in vitro-rearing risk assessments. As per the results, colonies having brood survival percentages more than or equal to 80% are very much suitable and from that the source of larvae for in vitro-rearing for risk assessments should be used.

1.2) Studies on Scattered brood pattern:

Kepeña (1976) showed that colonies developed with inbreeding were developed slowly because of the scatterings of the brood. This was just due to the homozygosity of the locus of those mated eggs.

When the eggs are eaten by bees the empty cells are observed and therefore scattered brood is observed and it became more frequent in older queens, poor quality of queen and failing of old queen, (Sammataro and Avitabile, 1998; Jabde, 2005; Head 2010, Koeniger *et al.*, 2014). The supply of sperm in the spermatheca decreases with the queen's age (Akyol *et al.*, 2008; Gregorc *et al.*, 2008; Al-Lawati *et al.*, 2009), which turns in to irregularities in deposition of eggs (Woyke, 1976) and emergence of scattered brood observed.

Borsuk *et al.* (2018) noticed that those eggs fertilised by sperm cells involved with DNA fragmentation (SDF) cannot develop normally and are generally eaten by worker bees and empty brood area produced likewise the same thing happens with eggs contained diploid drones (Woyke 1963).

2) Impact of Brood survival rate on behavioural, economical and resistant traits of *Apis mellifera* L.

Plass (1953) noticed that inbreed honey bee colonies had deficiencies in brood-rearing and a declination was observed in nest cleaning behaviour also.

Rothenbuhler (1964) experimented out that the behaviour of nest-cleaning in honey bees is directly related to the brood diseases. He proposed that the variations in the behaviour of nest cleaning were mainly due to recessive genes presents at one or more loci. Bruckner (1978) noticed that non-inbred worker bee recruited much more than the inbred workers.

Islam *et al.* (2015) carried out a research and demonstrated that European or Italian honeybee (*Apis mellifera* L.) was reared in bee box or hives and placed around Bangladesh Agricultural University (BAU) campus in Mymensingh mainly to study their behavioural activity, life cycle, pollen gathering, production of honey and ultimately its effect on overall yield of mustard. They observed that there having no relationship between sunrises, sunset, first out from the box and last entrance into the

box. But positive relationship was found with day temperature to first out and last entrance. The highest pollen collections by bees were recorded in the 3rd week of March. 12.00 to 1.00 pm was the time range when maximum pollen gathering activities was recorded. The highest amount of honey production was 4.00 kg per box. Per cent pollen gathering activity and honey production showed positive correlation. The highest number of queen cell was recorded in the month of March.

3) Studies on foraging behaviour of *Apis mellifera* L. on mustard.

Eckert (1933) stated that the best method to capture bees is to use the pan traps, this are also known as 'bee bowls.' These are made of simple plastic, this plastic cups are painted in various colour like white, fluorescent yellow and fluorescent blue which holds soapy water. Honey bees generally get attracted to those colours, and then they fall into the soapy water and drown and died. They also clarified that the area of foraging for bees is around a bee colony extends for near about two miles (3.2 km), though honey bees have been observed to forage around twice and three times from the distance of their hive. Their experiment had shown that bee colony or bee hives within the range of 4 miles from a food source will obtain weight, but beyond that distance their total energy expended in that process is much more than that their gained during the flight for foraging.

Butler (1940) observed that water foragers were seen to obtain continuous or same water sources compared to the stable ones. They also noted that large water containers were mostly preferred than tiny ones. They further added that forager bees love to collect water from some of the unusual and unique sources such as cow dung compared to clean water.

Kraai (1962) showed that when honey bees went out for searching a comparatively larger area for food resources during flowering, bees generally forage during the day hours or in daylight and are unlikely to sustain pollen viable enough to cause fertilisation by 12:00 hours.

Weaver (1965) showed that the detected differences or variation in foraging behaviour of honey bee on hairy vetch (*Vicia villosa* Roth) flowers. He also observed that some bees used the base of the flower while others use the mouth of the flower.

Kapil and Brar (1971) found that little bee *Apis florea* foraging activity was much high on 21-35⁰C temperature and favourable RH was 50-57 % in toria.

Szabo and Smith (1972) informed that leaf cutter bee or *Megachile rotundata* had maximum foraging activity on relatively higher temperature (30°C).

Kapil and Kumar (1974) showed that the minimum threshold temperature was 15-18°C of honey bees for the foraging activity and informed that temperature had profound effect on pollination services of honey bees.

Nuenz (1977) stated that *A. mellifera* activities in the morning totally depend on flow of nectar of the crop and activities during the afternoon depends on photoperiod.

Ewies and EL- Sahhar (1977) recorded that European honey bee *Apis mellifera* foraging activity hits peak at the day hours of 11:00-14:00 h of the day. Oh and Woo (1990) recorded that insect pollinators populations increased after sunrise while decreased sharply just before sunset.

Thakur *et al.* (1982) and Rana *et al.* (1997) both noticed that there was the peak in foraging activity at 12:00 hrs. for both the honey bee species *Apis mellifera* and *Apis cerena indica* compared to at 09:00 hrs.

According to a study it was recorded that *Apis mellifera* generally carries 11.19 ± 0.11 mg of pollen at 1200 hrs. of the day, while Verma and Dutta (1986) have observed that this was 12.22 mg in apple, Rana (1989) has noticed that this as 15.50 mg in plum and Negi and Joshi (2006) has observed that this weight becomes 11.20 mg in Indian mustard or *Brassica juncea*. The general behavior of an effective pollinator of apple should be that the insect touches the stigma of the inflorescence in many of their foraging trips and should be able to carry much germinable and viable pollen grain of the respected cultivars on its body surface.

Burril and Dietz (1981) showed that bees foraging activity was totally depends upon increasing air temperature and not related with relative humidity (RH) and atmospheric pressure.

Thakur *et al.* (1982) demonstrated that the activity of *A. cerena indica* and *A. mellifera* on mustard flower obtained peak value at 12:00h. There were less peaks at 14:00 followed by at 15:00 h. although, they found that there was a marginal increase between 15:00 to 15:30h, followed by a slow and gradual decrease.

Pham-Delegue *et al.* (1984) observed the recruiting behaviour in case of *Apis mellifera* in colonies. He noticed that the number of honey bees making a trip to artificial flowers which containing a solution of sugar scented with the main component geraniol, increased slowly to a fixed rate of 10 visits per 5 minute period. It was also recorded that the recruitment was much more rapid in case of hybrid colonies.

Osgood (1974) and Sihag (1984) showed that temperature plays vital or key role in initiation of foraging activity of leaf cutter bee *Megachile rotundata* and light intensity has profound effect on cessation of its activity. Gary (1975) informed that for initiation of *Apis mellifera* was 13⁰C and it is the minimum threshold temperature for it.

Free *et al.* (1985) observed that there are several factors that has direct impact on foraging activity of bees. These factors may be classified into two great groups first one is in-colony factors and the other one is out-colony factors. The first group consists of presence of queen and case of the queen bee (virgin or mated). Colony headed by virgin queens witnessed higher foraging activity with less pollen collection than colonies those were headed by mated queens witnessed lower foraging activity and collection of pollen were found in colonies that are queen less when compared to colonies with a virgin queen or mated one.

Sarviva (1985) informed that daily activities of insect pollinators rely on different metrological parameters like humidity, temperature etc.

Goyal *et al.* (1989) noticed that the optimum activity of foraging of house fly, syrphid fly and honeybee was in morning time at 10:00– 11:00 hours of the day on the concerned crop carrot.

Plowright *et al.* (1993) stated that the visitation time of foraging insects, commonly occur in the early morning, related to their plant material, and specially depends upon pollen and nectar availability. The most probable reason is that, both availability of nectar and pollen are much higher during early morning compared to afternoon and evening time. Most of the honey bees (Hymenoptera) collect nectar to meets their energy requirements and pollen to supplies their protein requirement especially for larval growth.

Rana *et al.* (1993) and Chand *et al.* (1994) also observed that the activity of foraging of Indian bee (*Apis cerana indica*) started to increase at 10:00 hr of the day and obtained its peak value at 11:00 hrs. on mustard flower. Also on the other side, the foraging activity of rock bee (*Apis dorsata*) started to increase from 14:00 hr and obtained the peak digit at 16:00 hrs.

Chand and Singh (1994) observed that the foraging activity of Indian bee *i.e.* *A. cerana indica* increased at 10:00h and achieved peak value at 11:00h on mustard bloom.

Kirk *et al.* (1995) found in the United Kingdom that foraging behaviour of honey bee may also be controlled by the presence of natural enemies of them. They observed that the *Meligethes aeneus* (pollen beetle) has an important influence on the foraging behaviour of bees on the crop known as oilseed rape flowers. They concluded that bees like to forage on the flowers that are fully opened and no beetles on those flowers.

Picard-Nizou *et al.* (1995) observed that no effects was there when introduction was done of genetically modified gene in oilseed crop rape (*Brassica napus* L.) a chitinase gene was imported to increase the disease resistance character and the foraging behaviour of *Apis mellifera* L.

Page *et al.* (1995) showed that the inheritance for the trait of high pollen-hoarding, is a recessive trait, but in case of honey storing behaviour, it shows a great dominant pattern of inheritance.

Collins *et al.* (1997) observed that several other environmental factors may have a significant role on foraging activity. They had found that there was important role of solar ultraviolet-B (UV-B) directly on honey bee and their foraging activity on specially two species of mustard crop, *Brassica nigra* and *B. rapa* grown when the crop was raised under controlled conditions or in green house condition.

Pierre *et al.* (1999) observed that in *Brassica napus*, maximum nectar secretion was found at the starting of flowering when the mean temperature was about 12.1⁰ C and the minimum secretion of nectar occurred in the day after when the mean temperature reached 9.1⁰ C, the second lowest secretion of nectar took place during end of flowering, despite a good warm weather was present at that time (19.9⁰ C).

Cresswell (1999), Ramsay *et al.* (1999) and Pierre (2001) showed that honey bees can travel many kilometres just with the purpose of foraging. They also concluded that generally the best of the foragers tries to concentrate within the range of 0.8 km from their hive when there is a favourable food resource is available. Many other studies have depicted that a quite good proportion almost up to 80 per cent of honey bee flights are generally less than 1 m in distance. While in case of pollen collection the bulk amount of pollen being moved by bees less than 5 m. Rarely honey bees may fly much further and several studies have showed that the calculated flight of honey bees may reach up to the distances of 1 - 2 km (Eckert 1933), while the maximum distance was recorded to be of 4 km Ramsay *et al.* (1999), Thompson *et al.* (1999).

Fulop and Menzel (2000) stated that take the out-colony factors like the main criteria is the amount of availability of preferable plant resources has a good influence on foraging activity, and at the same time it was recorded that forager bees have some preference for particular resources compared to others. The volume of reward (*e.g.* sucrose solution or nectar) that has an important influence on foraging activity of bees and those bees may recognize the quantity of total reward from those feeding sources.

Atwal (2001). Showed that when there are abundant flowers present in cultivated fields then individual honey bee tends to forage or tends to collect pollen and nectar from those flowers present immediately to each other or from adjacent plants. With the respect to collection of nectar, pollen or both there are different honey bee species. It was found that 58, 25 and 17 per cent of the total honeybees found on mustard crop were pollen collectors, nectar collectors or both pollen and nectar collectors, respectively.

Sihag (2001) observed that pollination services by insects pollinators is unavoidable for *Brassica sp* as they are commonly incompatible for self-pollination and the nature of pollen is sticky and much heavier, for this their pollen is which is not suitable for wind borne. Although, honey bees are indicated as excellently evolved agents for pollen transfer in *Brassicac sp*.

Ahmed and Rehman (2002) plotted their research and showed that the number of siliqua per mustard plant was significantly higher in open pollinated (OP) (189.60-

190.24) while for PWI (pollination without insects) the value is less and varies from (120.93-120.69).

Pankiw *et al.* (2002) experimented that the genotypic character of different strains of honey bee (*e.g.* low and high pollen-hoarding bees) heavily controlled the foraging behaviour when nectar or for pollen amount is concerned.

Schulz *et al.* (2002) observed that an earlier initiation was meant of foraging in younger honey bees in hives treating them with octopamine. In an addition, the pollination helped by the European honey bees *A. mellifera* may be developed by the presence of many other honey bee species in the orchards as observed by Brittain *et al.* (2013) in California, in some almond orchards.

Harrison and Fewell (2002) informed after their successful experiment that genetic factors and environmental factors influenced the activities of *Apis mellifera* for successful field crop pollination.

Mohapatra *et al.* (2003) observed that there are more than 50 per cent of the existing plant species are directly rely upon insects for proper pollination and seed set.

Steffan and Kuhn (2003) and Beekman *et al.* observed that several other factors can also alter the foraging behaviour. For example, distance of foraging was found to be changed during different time of year.

Chandel *et al.* (2004) informed that *Apis dorsata* was the most important and major pollinator of *Allium cepa* (onion) and it was during peak time of their foraging activity (12:00-14:00 h) and the period of foraging was (06:30-18:55 h) followed by syrphids and *Apis cerana* with foraging period ranging from 06:35-18:30 h and 07:25-18:20 h respectively.

Kalmath and Sattigi (2005) informed that the activity of different pollinators were much more during 10:00-12:00 h. Yucel and Duman (2005) recorded that the positive effects of foraging activity of Italian or European honey bees (*Apis mellifera* L.) on onion and they reported that foraging on onion plant started from 8.15 to 16.30 h while peak foraging time was between 11:00 and 12:00 hr.

Mostajeran *et al.* (2006) conducted their research and observed that the bees having large and broad wings were capable of flying high while on the other hand the

bees having small and lesser wing area reported to fly less and they cannot fly very high and therefore it impact in their foraging activities.

Young *et al.* (2007) stated that beside of the primary role of foraging activity for honey bee colonies like collecting nectar, pollen, water *etc* there are many evidences of its significant role in pollination of plant especially in those plant species in which honey bees are the main and only pollinator.

Fohouo *et al.* (2008) observed that more number of forager bees or worker bees was on *Syzygium guineense* var. *guineense* and on the other hand the lowest number of worker bees was recorded on *Psorospermum febrifugum*.

Yang *et al.* (2008) observed that insecticides also play a vital role in influencing the foraging behaviour of honey bees. They showed that the effects of imidacloprid when used in sub lethal doses resulted in delay in honey bees in their visit to their respective feeding site. The delay observed in that case was directly related to the concentration of the imidacloprid.

Amdam *et al.* (2009) and Abou-Shaara *et al.* (2013) observed that foraging activity of *Apis mellifera* was impacted by the strength of the colony and the activity of brood rearing and also depend upon the degree or amount of pollen need Weidenmuller and Tautz (2002). Type of beehive has also an important impact on the activity of foraging of *A. mellifera* bees Abou-Shaara *et al.* (2013).

Sharma and Kumar (2010) observed that there was a strong negative effect of an electromotive field with foraging behaviour of honey bees. At the same time surprisingly, it was found that the diesel exhaust can reduce the efficiency of foraging of worker honey bee. The main mechanism behind that was reduction in the ability of worker bees to locate the proper floral odours (Girling *et al.*, 2013).

Abrol (2011) showed that *Apis mellifera* activity was much higher at 16⁰C temperature, favourable relative humidity was recorded 75% and lux light intensity favoured around 800 and 10 mW/cm² solar radiations.

Higginson *et al.* (2011) observed that the bees having damaged wings were capable of less number of foraging trips compared to the healthy and normal ones. They also noticed that bees with damaged wings tried to fly closer or near to their hives than healthy bees.

Hagler *et al.* (2011) observed after their successful experiment that the foraging bees used many marking devices. Those self-marking devices were used mainly to study the range of foraging of honey bees on an alfalfa crop used for seed production.

Jung and Burgett (2011) and Jung *et al.* (2011) showed after their successful research that there is a huge role of synthetic brood pheromone which if presents in more quantity on colony then that colony shows higher activity related foraging and pollen collection. They also suggested that the number of active foraging bees in a hive was a vital factor.

Abou-Shaara (2012) showed that forager bees prefer to collect food resources like water, nectar, resin or pollen from the same place or resources over others.

Schneider *et al.* (2012) noted a remarkable deduction in foraging activity and also in longer foraging flights at two different doses of new molecular insecticides that is neonicotinoid insecticides; 0.5 mg/honey bee or more deduction was recorded for clothianidin and 1.5 mg/ honey bee and it was recorded more in case of imidacloprid mainly during the first 3 h after the particular treatment. In contrast, on rapeseed mustard and corn crop the presence of residues in the nectar and as well as in pollen was found due to seed treatment done by using thiamethoxam and it was reported to cause a low risk to bees (Pilling *et al.*,2013).

Leonhardt and Bluthgen (2012) suggested that all insect foragers with respect to their food collection are not same. They also noticed that the main differences presents between the honey bee and the bumble bees is the foraging pattern. They showed that bumblebees generally visits and collect pollens from at least double as many species of plants compared to what honey bees generally do.

Tan *et al.* (2012) observed that environmental factors that has profound influence on foraging activity of *A. mellifera* bees were noted that to initiate their foraging activity at favourable temperatures range with a mean value of the temperature was 26.57°C while on the other hand in other studies showed that this value was around to be 16°C (Joshi and Joshi 2010). At ambient or favourable temperatures that is about 20 °C, and in this temperature the highest activity was recorded (Tan *et al.*, 2012) while in case of lowest foraging activity was concerned

the temperature was at 43°C (Blazyte-Cereskiene *et al.*, 2010) and the in lower temperature range it was at or below 10°C (Joshi and Joshi, 2010).

Abou-Shaara *et al.* (2013) observed that the foraging activity of Yemeni honey bees was higher than Carniolan honey bees when tested under desert conditions. On the other side, no obvious impact of bee race was established in case of ARS Russian or Italian honey bee species mainly with special reference to the percentages of flight activity or pollen foragers (Danka *et al.*, 2006).

Brittain *et al.* (2013) reported that presence of other honey bee species in an area also influences the foraging behaviour of *Apis mellifera*. They also informed that alterations was recorded in foraging behaviour of honey bee in California almond orchards and that was mainly due to the presence of several other bee species or other bee communities.

Pearce *et al.* (2013) observed that there were no remarkable effects on foraging activity of honey bees when moving beehives from their original location to another and new location was done as far as 26 km.

Sushil *et al.* (2013) observed that foragers loves to spent less time in a flower under open conditions compared to that same flower cultivated in controlled or net house conditions.

Tan *et al.* (2013) observed that the foraging activities of honey bee may also be impacted by the presence or absence of predators. Presence of predator species like hornets and its presence caused a reduction in the foraging activity or visits of honey bees by 55–79% and on the other hand residence times by 17–33%. Also in case of the presence of bee-eaters affected indirectly or passively on foraging activity of honey bees (Ali and Taha 2012).

Abou-Shaara (2014) stated that foraging behaviour of *Apis mellifera* is one of the peculiar behaviours. This character is directly related between the honey bee colony and the immanent atmosphere. For that, various colonies related activities have an effect on this behaviour. Several scientists during many times had conducted many studies regarding this and employed to look in to these factors.

Goswami and Khan (2014) experimented at Pantnagar on the diversity and abundance of different insect pollinators on mustard (*Brassica juncea*). Total 19 insect pollinators maximum belonging to order Hymenoptera (15) and Diptera (4)

were found in the blossoms at Pantnagar. The total population or abundance (per cent of insect/m²/2 min.) of hymenopterans visitors were the maximum followed by the dipterans and others. In hymenopterans, bees (*Apis* bees) were recorded maximum followed by non *Apis* bees and the scolitud wasp. It was found that insect visitors increased total number of pods and per cent of pod set.

Goswami and Khan (2014) noticed that few flower of Indian mustard (*Brassica juncea* L.: Cruciferae) at Pantnagar started opening during evening time of a day before anthesis (first opening of flower) and most of flowers were opened during morning, mainly the time between 06:00 hrs. and 09:00 hrs. They also noticed that opening of new flowers was usually completed by 10:00 hours of the day.

Badade (2014) experimented and informed that the intensity of little bee, Indian bee and Stingless bee were reached at its foraging activity at peak between 12:00 – 14:00 h, while rock bee and European bee and other pollinator reached their peak between 14:00 – 16:00 h.

Narendra (2014) reported that foraging activity of *A. dorsata* and *A. cerana* obtained its peak value at 10.00 hrs. and for little bee or *A. florea* reached its highest value between 12.00 to 14.00 hrs.

Sanas *et al.* (2014) observed that *Apis mellifera* enhance the total number of seed per pod or per siliqua (23.27%) in mustard crop in Maharashtra under Konkan region.

Maity *et al.* (2014) and Reddy *et al.* (2015) observed that several climatic factors holds a vital role in controlling the activity related foraging and general behavior of social insects especially for honeybees.

Amin *et al.* (2015) showed that the peak foraging activity of the major pollinators found in an agroforestry mainly mango-based at 11:00 hour of the day at that particular time the mango flowers generally opened.

Tribhuvan (2016) stated that both European honey bee and Indian honey bee reached optimum foraging activity between day hours of 10:00 to 12:00.

Patidar *et al.* (2017) plotted out their research and showed that there were good amount of differences for all the quantitative parameters that were investigated among all the pollination methods. The mean data was found highest obtained from

open pollination, then plants caged with European honey bee or *A. mellifera* and plants caged without any pollinators (PE). The mean value of number of siliqua/pod per plant was recorded maximum (186.44) in case of plots having free access to all the pollinators (OP) followed by 154.82 in BP. The lowest value was found in PE (133.38).

Rinku and Chaudhary (2018) recorded that *Apis mellifera* was mostly abundant during 0800 and 1000 hrs. of the day (2.259 and 2.185) and started to decrease by 12:00 hours of the day (1.667) and their population minimum by 18:00 and 14:00 hours of the day (0.852 and 0.963 bees). They also observed that *Apis cerana indica* had foraged this crop with their maximum population at early and late hours of the day (0.70 and 0.82 bees). While *Apis florea* showed their peak abundance at 08:00 h (0.333), then started down considerably later on and was almost absent at 12:00 and 14:00 hour the day.

In a very recent study by Klein *et al.* (2019). Suggested that only about 19% of the total active foraging bees involved in performing near about 50% of the colony's total foraging activities and foraging trips. Thus, these factors may be the possible reasons for the varied pollen amounts and difference in foraging pollen amounts of the different honey bee colonies.

3.1) Duration of foraging

Jain and Kapil (1980) stated that the air temperature has profound effect on initiation of foraging activity while cessation was completely independent of air temperature but somewhat depended on light intensity of evening time.

Kapil and Brar (1986) conducted a research and showed that rock bee (*A. dosata*), little bee (*A. florea*) and Indian bee (*A. cerena*) began foraging at 07:00 to 10:00h on rapeseed mustard. They recorded that peak value of foraging rate was recorded in between 12:30 to 14:00 and ceased by 17:00h.

Kapoor and Dhaliwal (1989) showed that *Apis cerana indica* initiated their foraging activities one hour earlier compared to *Apis mellifera* on cauliflower as day temperature and light intensity was lower and relative humidity was high. While the cessation time of foraging activities was one hour later in case of *Apis cerana indica* compared to *Apis mellifera*.

Rao and Suryanarayana (1989) recorded that *Apis cerana* and *Apis florea* initiated their foraging activity around 09:50-11:30 h of the day and gradually ceased at 17:10 to 18:15 h of the day hour.

Verma and Partap (1994) noticed that total time of foraging (duration of foraging) for single and separate foraging trips of *Apis cerana indica* for cauliflower was 26.9 min for cabbage 23.9 min. Their visits to each and every flower were from 4.3 to 6.7 sec counted during day time for both cabbage and cauliflower; honey bees spend 5 - 8 flowers/min. They also stated that activity of foraging decreased mostly after 05.00 pm of the day.

Chakravarty (2000) had experimented that Italian bee or *Apis mellifera* started their foraging work an hour later compared to *Apis cerana indica* in Mustard (*Brassica napus*.) The time of initiation of foraging was around 08:24 to 09:40 h. While the cessation of foraging of *Apis mellifera* was recorded almost 30 minutes earlier than *Apis cerana indica* in that same crop *sp*.

Chaudhary (2004) reported that *Apis florea* and *Apis dorsata* started their foraging activity of foraging in the morning (07:00-08:00 h) and stopped their activity at evening time of the day (18:00 h). Sajjanar *et al.* (2004) also stated that rock bee started their foraging activity on cucumber flowers at very early morning around 06:00 h, whereas rest all insect pollinators and visitors started activity by 08:00 h.

Alqarni (2006) observed that in case of desert conditions more number of foragers bee left their colonies at 8 am than at 10 am.

Kralj and Fuchs (2006) and Kralj and Fuchs (2010) observed that if the forager bees were infected with diseases and parasites for example as *Nosema sp.* or *Varrao destructor* and that will result in late return to their colonies or in other words increased time to return in their hives.

Reyes-Carrillo *et al.* (2007) observed and informed that huge amount of pollen collection was recorded in the early morning compared to low amounts of pollen collection during afternoon.

Choudhary and Kumar (2007) reported that European or Italian honey bee started their foraging activities early in the morning at 09:05 hours and reached its peak foraging activity during 11:00 to 16:00 and there after it reduced gradually.

Saeed *et al.* (2008) reported that honey bee activity for pollination started at 06:00 h of the day hour and finished by at 13:00 h of the day hour. They also stated that insect pollinators obtained peak activity during 10:00 to 12:00 hr.

Amdam *et al.* (2009) showed that under case with increased vitellogenin levels reduced the activity of brood rearing and witnessed a delay in activity initiation in foraging and death was also common.

Pavana (2010) reported that foraging activity of rock bee or *Apis dorsata* on bitter gourd was started at 06:30 hour of the day.

Mott and Breed (2012) stated that in earlier times commencement of foraging activity (shifting to Out-colony tasks) was mostly found to be hampered by the treatments with bovine insulin hormone.

Mattu *et al.* (2012) informed about the influence of altitude on foraging initiation and cessation time, total duration of foraging and trips and also impact the total number of flowers visited by the bees per minute.

Silva *et al.* (2013) stated that foragers have the ability to recall the exact time of the day hour when the higher food materials are there to take as it was found in case of *Sysirinchium palmifolium* plants and that kind of ability can be correlated with peaks of the foraging activity.

3.2) Foraging rate

Atwal (1970) informed that the foraging rate of *Indian bee*, *little bee* and *rock bee* on mustard with an average value of foraging rate of 12.7, 17.2 and 16.6 flowers per minute respectively.

Benedek (1976) informed that foraging rate of several insect pollinators relies on the floral character like length of the corolla and also different environmental factors like solar radiation, solar intensity, temperature, humidity, etc.

Gilbert (1980) reported that both foraging rate and foraging speed directly depends upon foraging behaviour and structure of the associated flowers.

Gupta *et al.* (1984) informed that *A. mellifera* visited their respective flowers on an average 25.8 and 33.6 flowers /min in morning and evening respectively. Alam *et al.* (1987) informed that bees activity was highly depend upon pollen and nectar flow or availability of the crop. They stated that honey bee activities for foraging was

maximum around 31.8⁰C-34.9⁰C and later the activities of bee gradually decreased as the overall availability of pollen and nectar dried.

Zar (1984) conducted his research and showed that honey bees were counted based on times after their foraging on minimum 10 flowers, later the total number of flowers were visited by a single bee in 1 min was also counted. He categorized bees as *Bombus impatiens*, honey bees another *Bombus sp.* While floral handling times in respect of flower visited by bee per minute were calculated. Obtained data for each single year were calculated statistically by ANOVA and also with Scheffe's multiple comparison tests (this was used due to unequal sample sizes).

Deshmukh *et al.* (1985) found after their successful experiment that the time of foraging varied on safflower slightly among different honey bee species. The value showed that during pollen collection the time duration was 12, 24, 32 seconds and while for nectar foraging it was 13, 21 and 33 seconds were spent on an single flower head by *Apis dorsata*, *Apis cerana*, *Apis florea* respectively.

Seeley *et al.* (1991) observed that foraging rate and activity of bee was much higher when availability of flowers was more and on the other hand less number was recorded when of honey bees differed greatly and with relative humidity it has a negative correlation.

Free (1993) said that the foraging rate or number of flowers visited per bee per minute depends upon several numbers of factors. This factor includes instinctive behaviour of foraging, proboscis length, floral structure specifically the depth of corolla, quantity and types of floral rewards, flower density of a specific variety of the crop grown and lastly the particular time of the day.

Dukas and Visscher (1994) showed that commonly, the skills regarding foraging and the total number of forager or worker bees were found to be increased with age of the colony.

Sharma and Singh (1999) informed that rock bee or *Apis dorsata* visited 4.31 flowers/min and little bee or *Apis florea* visited 2.20 flowers/min. Melendez-Ramirez *et al.* (2002) observed and showed that weather conditions greatly influenced the activity of foraging of bees.

Chandel *et al.* (2004) stated that rock bee (*Apis dorsata*) visited 7.5 flowers /onion flower per visit and it was during the optimum hours of bees foraging activity

(12:00-14:00 h) but when it was compared with *A. cerana* it was only about 5.4 flowers per umbel per visit.

Yucel and Duman (2005) showed that average flowers visited by European honey bee or *Apis mellifera* was 8.0, 13.0, and 4.0 flowers per minute at different day hours like 09:00, 12:00 and 15:00 h, respectively.

Pernal and Currie (2010) informed that a higher foraging rate was observed during afternoon with mean (36.02 foragers/min) while during the early hours of the day or in morning period it was (17.66 foragers/min).

Ali (2011) observed that a higher rate of foraging was found for Yemeni honey bees compared to Carniolan honey bees and it was mainly in the month of June and August and the result was obtained by monitoring different times; 6–7 am, 11–12 am and 4–5 pm.

Kant *et al.* (2013) reported that the rate of foraging of *Apis cerana indica*, *Apis mellifera* and *Episyrphus balteatus* were 15.2, 13.1 and 7.8 respectively on coriander crop.

Tchindebe and Tchuenguem (2014) informed after their successful experiment that honey bee visited 47.12 ± 7.19 flowers per minute.

Tribhuvan (2016) stated that *A. mellifera* showed the most efficient method of foraging behaviour by visiting around 17.14 ± 1.38 number of flowers in 147.5 ± 8.14 seconds on an onion flower.

3.3) Foraging speed

Rymahesvskii (1956) observed and stated that Indian bee or *Apis cerana indica* is much better pollinator in the sub-tropical region compared to European bee or *Apis mellifera*. He also added that this species of honey bee gives more than 30 seconds on each of the apple bloom.

Tanda (1984) informed that *Apis florea* initiated their foraging later and spent much more time 5.9-6.4 sec on mustard (*Brassica juncea*) flowers as compared to *Apis mellifera* who spent 3.0-3.2 sec and it was least in case of *Apis dorsata* 2.2-3.0 sec.

Raj and Rana (1994) stated that during nectar collection *A. mellifera* spent 3.64 seconds per flower heads on the other hand during pollen collection they spent

3.37 sec per flower head and foraging speed of pollen collector bees was little higher compared to the nectar gathering bees.

Rana and Kumar (1999) informed that European honey bee spent much more time during pollen collection (6.6 sec) while for collecting nectar they spent (7.3 sec) followed by Indian honey bee or *Apis cerana indica* they spent 5.1 sec during pollen collection and 6.3 sec during nectar collection of cauliflower.

Sharma and Singh (1999) showed that *Apis dorsata* invests 9.20 sec/flower and *Apis florea* invests 37.99 sec/flower. Kumar and Singh (2004) stated that *Apis mellifera* invests much more time (6.6 sec/ capitulum) on sunflower rather than time spent by *Apis cerana indica*.

Yang *et al.* (2008) observed that generally for the same feeding site the interval of normal foraging is less than 5 min while they also concluded that bees generally expend different duration per flower and it is totally depending on the plant species on which they forage.

Devi (2011) showed that *A. dorsata* exhausted little more time (3.33 seconds/ umbel) on onion flower then followed by *A. mellifera* (2.96 seconds/ umbel), *A. cerana* (2.90) seconds/ umbel) and lastly by *A. florea* (2.65 seconds/ umbel).

Sushil *et al.* (2013) observed that the time spent by *Apis mellifera* per flower was 6.92 for Chinese cabbage, 6.50 for broccoli and for kohlrabi it was 5.54seconds.

3.4) Population dynamics of *Apis mellifera* on mustard bloom-

Abrol (1989) observed that total 20 species of foragers were found on mustard bloom in which 12 families were belongs to hymenopteran and dipteran order. They also noticed that the most common and numerous visitors and important pollinator were *Apis mellifera*, *Halictus sp.*, *Apis cerana indica*, Halictid bees and *Lasioglossum sp.* of *Brassica* crops.

Atmowidi *et al.* (2007) noticed that the diversity of insect pollinators and its impact in seed setting of mustard (*Brassica rapa*) planted in West Java. He observed at least 19 species of insects pollinators helps in pollinating the mustard, and out of them three species *i.e.* *Apis cerana*, *Apis dorsata* and *Ceratina sp.*, showed maximum abundance. The maximum abundance and richness of species of pollinators mostly

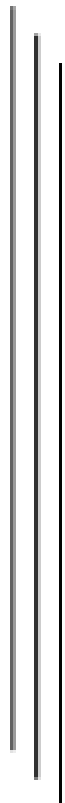
observed at 08.30-10.30 am of the day and the diversity of pollinators was directly was related to the total number of flowering plants.

Singh *et al.* (2018) observed that the overall population dynamics of *Apis spp.* On *Brassica sp.* was counted when maximum the day temperature was (20.0⁰C) and low relative humidity (60.75%) and recorded minimum temperature (15.25⁰C) and high relative humidity (84.33%). They also noted that *Apis mellifera* was the common and abundant pollinators among all the *Apis spp.* They also clarified it that might be due to the location of the experimental plot was near to a place where *Apis mellifera* colonies were present.





CHAPTER - III



MATERIALS AND METHODS



MATERIALS AND METHODS

Experimental site

The whole experiment was conducted in university apiary. Location of the University Apiary is Dr .Rajendra Prasad Central Agricultural University, Pusa (Samastipur), Bihar, 848125 and the latitude is 25.59°N, longitude 85.48°E and altitude 52.98 MSL. The climate mainly maintained by the southwest monsoon and it is generally humid subtropical if we classify according to nature.

Determination of brood survival rate in existing *Apis mellifera* population

1.1) Study on Brood survival rate-

In order to determine the Brood survival rate we had inserted one empty comb in the centre of each ten hives. Total 10 numbers of such bee hives was taken for this research. Each 10 hives contained 10 frames (1 empty frame and 9 already occupied by honey bees). Marking of the eggs laid within 24 hours in such a frame with the help of ordinary steel pins in an area of 25 sq. cm at 4 places was done.

Counting the number of eggs in the marked area was done & then after counting, the combs were returned in to those colonies. Removal of these marked combs after three days was done and observation of the number of eggs hatched in marked area was taken. On the fourth day, the observations of the number of surviving larvae from hatched eggs in the marked area were made and calculation of the brood survival rate of each colony was done on the basis of the formulae.

$$\text{Per cent brood survival rate} = \frac{\text{Number of surviving larvae}}{\text{Total number of eggs laid}} \times 100$$

On the basis of brood survival of the colonies grouping them into three categories was done as:

- i) High brood survival rate (90-99%)
- ii) Medium brood survival rate (80-89%)
- iii) Low brood survival rate (70-79%).

1.2) Study on scattered brood (pepper-pot brood):

Checking the colonies for continuity of brood pattern was done. Colonies with brood diseases and colonies with queens that have good egg-laying behaviour were

carefully examined. Measurements of the scattered brood were done by counting the number of cells in the wire grid covered by healthy broods. Scoring them 1-3 to each colony on basis of their performance was done. The value of 1 indicates a high degree of continuity, whereas a value of 3 indicates an irregular brood pattern.

2. Impact of Brood survival rate on behavioural, economical and resistant traits of *Apis mellifera*.

2.1) Behavioural Characters:

2.1.1) Nest cleaning:

To determine the nest cleaning ability of each colony, observation of the debris content at the bottom board was taken and total 4 observations were recorded at 7 days interval. Then the colonies were designated as

- i. Very clean
- ii. Clean
- iii. Poor cleaning ability

2.1.2) Bee activity:

Observation on the activity of honeybee colonies were done by counting the number of bees leaving the hive (outgoing) and those landing at the entrance (incoming) per minute during different day hours (09:00 to 10:00, 13:00 to 14:00 and 16:00 to 17:00 hours of the day) were taken.

2.2) Economic traits:

2.2.1) Honey stores:

Firstly estimation the honey stores with the help of grid frame was done then converting the number of squares covering honey area into grams by multiplying with 8.06 (considering that an average frame full of honey, contained 1000 g of honey) was done. Total honey yielding from all 10 hives was recorded.



Plate No. 1: Field view of experimental plots

2.2.2) Pollen stores:

Measurements of the pollen stores were done in cm² by counting the number of cells in the wire grid covered by pollens.

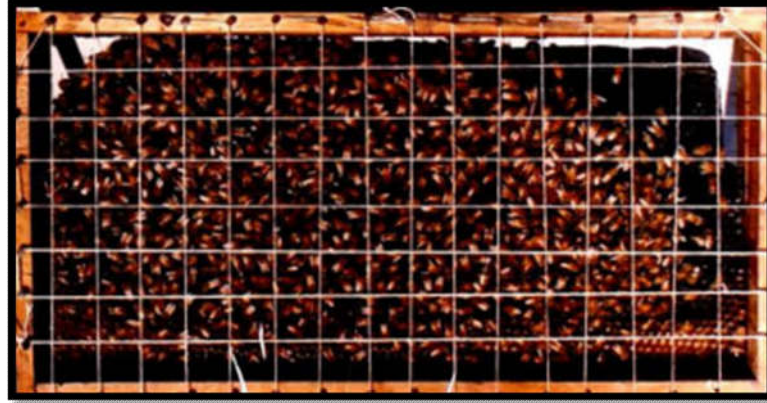


Plate No. 2: Standard grid frame made of iron wire

2.3) Resistance traits:

2.3.1) Hygienic behaviour:

To determine the hygienic behaviour pin killing of 25 capped pupae was done. Killing the pupae in the sealed stage by piercing the caps with sharp needle was done. Observation in the pierced area after 24 hours for removal of pin killed broods was taken. Per cent removal of killed brood was calculated by using the below formula:

$$\text{Per cent removal} = \frac{\text{Number of pin killed pupae removed}}{25} \times 100$$

3) Foraging behaviour of *Apis mellifera*:

Mustard (*Brassica sp.*) crop was taken to study the foraging behaviour of *Apis mellifera* L. To determine the foraging behaviour of *A. mellifera* the following parameters were studied.

3.1) Duration of foraging:

Three hives of *A. mellifera* having almost equal strength was taken for studying the foraging duration of the bees. First exit of a worker bee from the hive was recorded in the morning and last entry was recorded in the afternoon. After last entry of the forager bee, half an hour time was taken for confirmation of last entry. To

determine the duration of foraging on flowers by *A. mellifera*, 10 observations were recorded on different dates at 7 days interval in morning and evening hours. Duration of foraging was calculated as

$$\text{Duration of foraging} = \text{Initiation time (first exit)} - \text{cessation time (last entry)}$$

3.2) Foraging rate:

Number of flowers visited by a bee per minute was recorded on mustard. Number of bees visiting the floral area per square meter was also noted. To determine the foraging rate 5 observations were recorded on different dates at 7 days interval at different day hours (07:00, 09:00, 11:00, 13:00, 15:00 and 17:00 hours of the day). The experiment was replicated 5 times in factorial RBD, keeping 2 factors, date and hour.

3.3) Foraging speed

Time spent (in seconds) by a single worker bee per flower was recorded by eye estimation using stopwatch. To determine the foraging speed 5 observations were recorded on different dates at 7 days interval at different day hours (07:00, 09:00, 11:00, 13:00, 15:00 and 17:00 hours of the day). The experiment was replicated 5 times in factorial RBD, keeping 2 factors, date and hour.

3.4) Population dynamics of *Apis mellifera* on mustard bloom:

The bees were counted using stop watch of android mobile phone. Daily observations were recorded for 5 minutes on mustard bloom at three times *viz.* 09:00 hrs. 12:00 hrs. and 15:00 hrs. The mean of the values obtained at three occasions were taken. The meteorological data pertaining to maximum and minimum temperature (°C), relative humidity (%) and rainfall (mm) were collected from the Meteorological Unit of the RPCAU, Pusa.

Statistical analysis

The statistical analysis was done in factorial RBD. The population of *A. mellifera* was correlated with the environmental parameters by following standard statistical methodologies. Correlation, regression analysis was also done by using OPSTAT programme (Sheoran *et al.*, 1998) so as to determine the role of environmental parameters on their population.



Plate No. 3: Bee boxes used for the experiments



Plate No. 4: Insertion of empty frame in to bee box



Plate No. 5: Frame showing scattered brood pattern

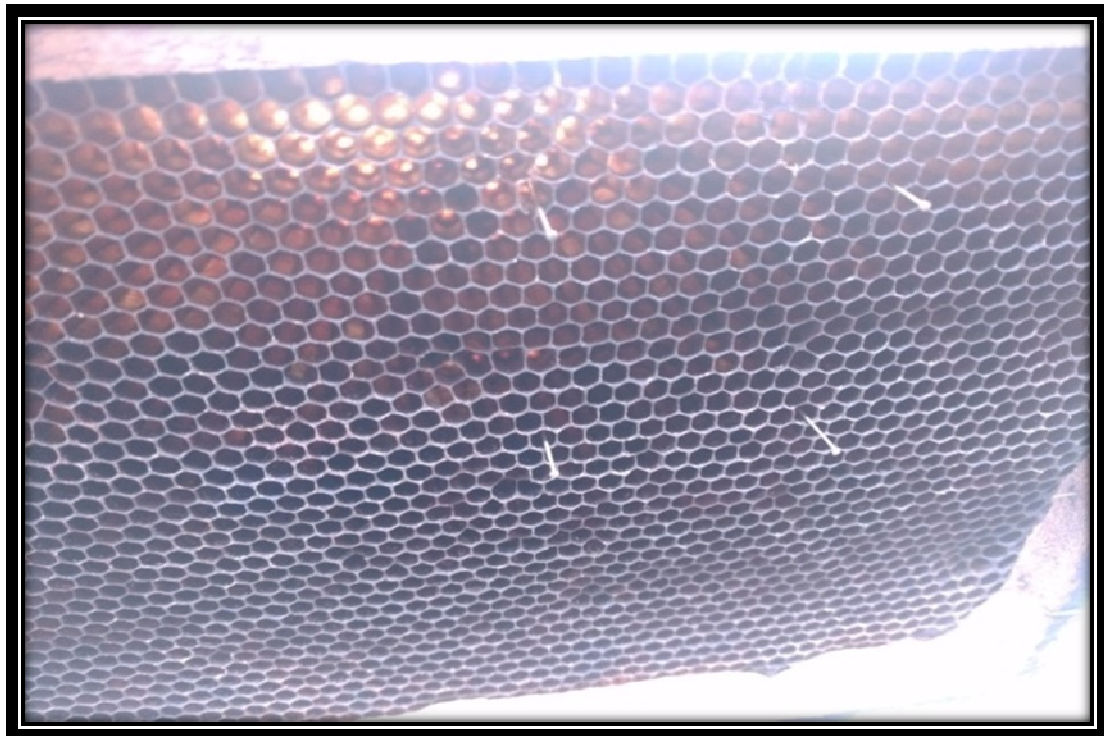
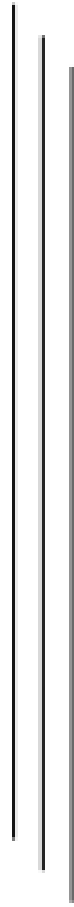


Plate No. 6: Frame showing area marked with steel pin





CHAPTER - IV



RESULTS AND DISCUSSION



RESULTS AND DISCUSSION

The results we got from various aspects of this present research entitled “Studies on brood survival rate and foraging behaviour of *Apis mellifera* L. on mustard” have been elaborately discussed in this particular chapter.

1. Determination of brood survival rate in existing *Apis mellifera* L. population:-

1.1) Study on Brood survival rate-

The studies were carried on “Determination of brood survival rate in existing *Apis mellifera* population” at university apiary RPCAU, Pusa, Bihar during “Rabi” 2020-21 to find out the study on brood survival rate in existing *Apis mellifera* stock. The obtained results of the experiment listed down in table no. 1 and depicted in fig. 1. Total 10 colonies of *Apis mellifera* were used for this experiment and each colony contained total 10 frames, 1 empty frame and remaining 9 already occupied by bees. Brood survival rate was calculated with the following formulae,

$$\text{Per cent brood survival rate} = \frac{\text{Number of surviving larvae}}{\text{Total number of eggs laid}} \times 100$$

From table no. 1, it was obtained that out of 10 colonies were taken for the research 6 colonies (colony no. 3, 5, 6, 7, 8 and 10) showed high brood survival rate, 3 colonies (colony no. 1, 2 and 4) having medium brood survival rate and only 1 colony *i.e.* colony no 9 showed low brood survival rate. The highest brood survival rate was recorded in colony no. 5 where the per cent brood survival was 96.87 as out of mean 32 eggs laid in that particular marked area 31 were able to hatch and turns into healthy larvae. Where as in colony no. 9 out of mean 33 eggs only 25 able to hatch and turned in to healthy surviving larvae and thus its per cent brood survival was 75.75. There was good amount of mite infestation was found in colony no. 9 which is the most probable reason for its low rate of brood survival. When all colonies were taken in to consideration it was found that average brood survival rate of all 10 colonies were 90.72 % which comes under high brood survival rate category.

Improper mating followed by incomplete fertilization is also an important factor responsible for low brood survival rate as the same concept was supported by many scientist like Shaskolsky (1968) observed the distribution theoretically that a

queen honeybee producing the brood with different survival rates which were in relation to the total number of successful mating in population.

1.2) Study of scattered brood (pepper-pot brood):

To study the continuity of brood pattern this experiment was done. Scoring the colonies 1-3 based on their performance was done. The value of 1 indicates a high degree of continuity, whereas a value of 3 indicates an irregular brood pattern while the score 2 denotes medium continuity of brood pattern.

From the study it was revealed that out of 10 colonies, colony no. 1, 3, 5, 6, 7, 8 and 10 showed excellent continuity of brood pattern and colony no. 2 and 4 showed medium continuity with score of 2, while colony no 9 having the worst performance among all and showed very scattered pattern of brood and thereby it scored 3. The scoring was done with the help of a standard iron grid frame and through eye observation. The obtained values were listed done in the table no. 1.

The values in the table 1, suggest that the colonies having high and medium brood survival rate showed good continuity in brood pattern or they showed less scattered ness in brood pattern while colonies having low brood survival rates showed poor pattern or scattered pattern of brood.

There are several factors that determine brood survival of a bee colony. During 1950's it was believed that some of the queens were noted producing scattered pattern of brood after their siblings' semen were inseminated to them (Mackensen, 1951, Hachinohe and Jimbu, 1958). After few years it was concluded that the homozygous eggs which were situated at the sex locus they failed to hatch and worker bees finally removed them. However in 1962, Woyke recorded that those eggs were actually laid by the queens and they were viable in nature.

Table 1: Assessment of brood survival rate and scattered brood pattern in *Apis mellifera* colonies

Colony no.	No. of eggs laid in marked area (25 Sq.cm)	No. of eggs hatched	Larvae survived	Brood survival rate (%)	Category	Score based on brood continuity pattern
1	46	45	39	84.78	Medium brood survival rate	1
2	37	37	33	89.18	Medium brood survival rate	2
3	33	32	31	93.93	High brood survival rate	1
4	39	37	35	89.74	Medium brood survival rate	2
5	32	32	31	96.87	High brood survival rate	1
6	44	44	42	95.45	High brood survival rate	1
7	38	38	35	92.10	High brood survival rate	1
8	45	44	43	95.55	High brood survival rate	1
9	33	33	25	75.75	Low brood survival rate	3
10	41	41	38	92.68	High brood survival rate	1
CD (< 5%)	2.824	3.23	2.188			
CV (%)	5.062	5.81	4.284			
Average	38.8	38.3	35.2	90.72 %		

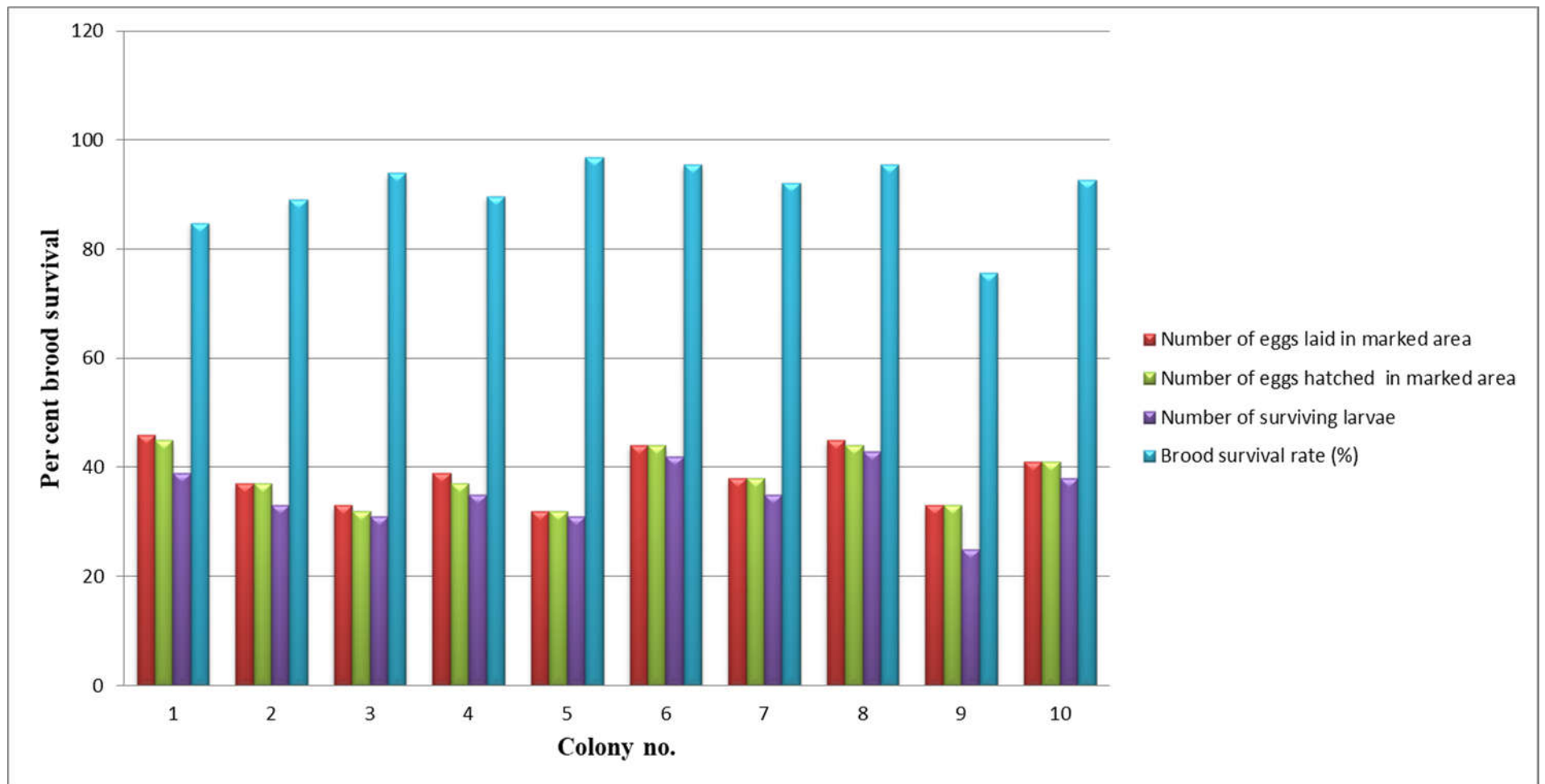


Fig. 1: Brood survival rate of *Apis mellifera* in different colonies

2) Impact of Brood survival rate on behavioural, economical and resistant traits of *Apis mellifera* L.

2.1) Behavioural Characters:

2.1.1) Nest cleaning:

To determine the nest cleaning ability of each colony, observation of the debris content at the bottom board was recorded. 4 observations were taken at 7 days interval. The collected debris was weighted in weighing balance and the obtained results are listed down in table 2 and depicted in fig. 2.

The experiment on nest cleaning ability of *A. mellifera* showed that among all 10 colonies colony no.5 was the most clean colony and its debris content for the 1st, 2nd, 3rd and 4th week was only 1.24, 1.31, 1.29 and 1.87 g respectively and this colony was categorized under very clean colony while the average debris content for the whole month of that colony was only 1.43 g. On the other hand colony no. 9 was found to be the dirtiest colony with their debris content for the 1st, 2nd, 3rd and 4th week was only 2.10, 3.14, 3.23 and 3.76 g respectively and the average weight of debris for the whole month was 3.05 g. Colony no. 9 faced high levels of mite attack and for that reason some bees were died and total debris content was also much more compared to other colonies and categorized under colony with poor cleaning ability and remaining 8 colonies were remarked as clean colony.

It was observed that the weight of the debris content kept increasing as the month proceeds and for almost all colonies the maximum value of debris content was during the 4th week. Now from the experiment it was noticed that the colony no. 5 was very clean colony and this colony witnessed the highest brood survival rate with per cent brood survival rate of 96.87 while on the other side colony no. 9 with the lowest per cent brood survival rate of 75.75 was the dirtiest colony and same trend was found with the remaining colonies.

Table 2: Weight of debris content (g) of different colonies

Colony Number	24.11.2020	01.12.2020	08.12.2020	15.12.2020	Mean	Status
1	1.89	1.87	1.92	2.94	2.15	Clean
2	1.91	1.83	1.89	2.95	2.14	Clean
3	1.63	1.77	1.71	2.72	1.96	Clean
4	1.76	1.79	2.82	2.87	2.31	Clean
5	1.24	1.31	1.29	1.87	1.43	Very clean
6	1.52	1.55	2.59	2.69	2.08	Clean
7	1.71	2.72	2.71	1.74	2.22	Clean
8	1.46	2.45	1.53	3.61	2.26	Clean
9	2.10	3.14	3.23	3.76	3.05	Poor cleaning
10	2.65	1.68	1.75	1.76	1.96	Clean
				CD (<5%)	0.73	
				CV (%)	12.92	

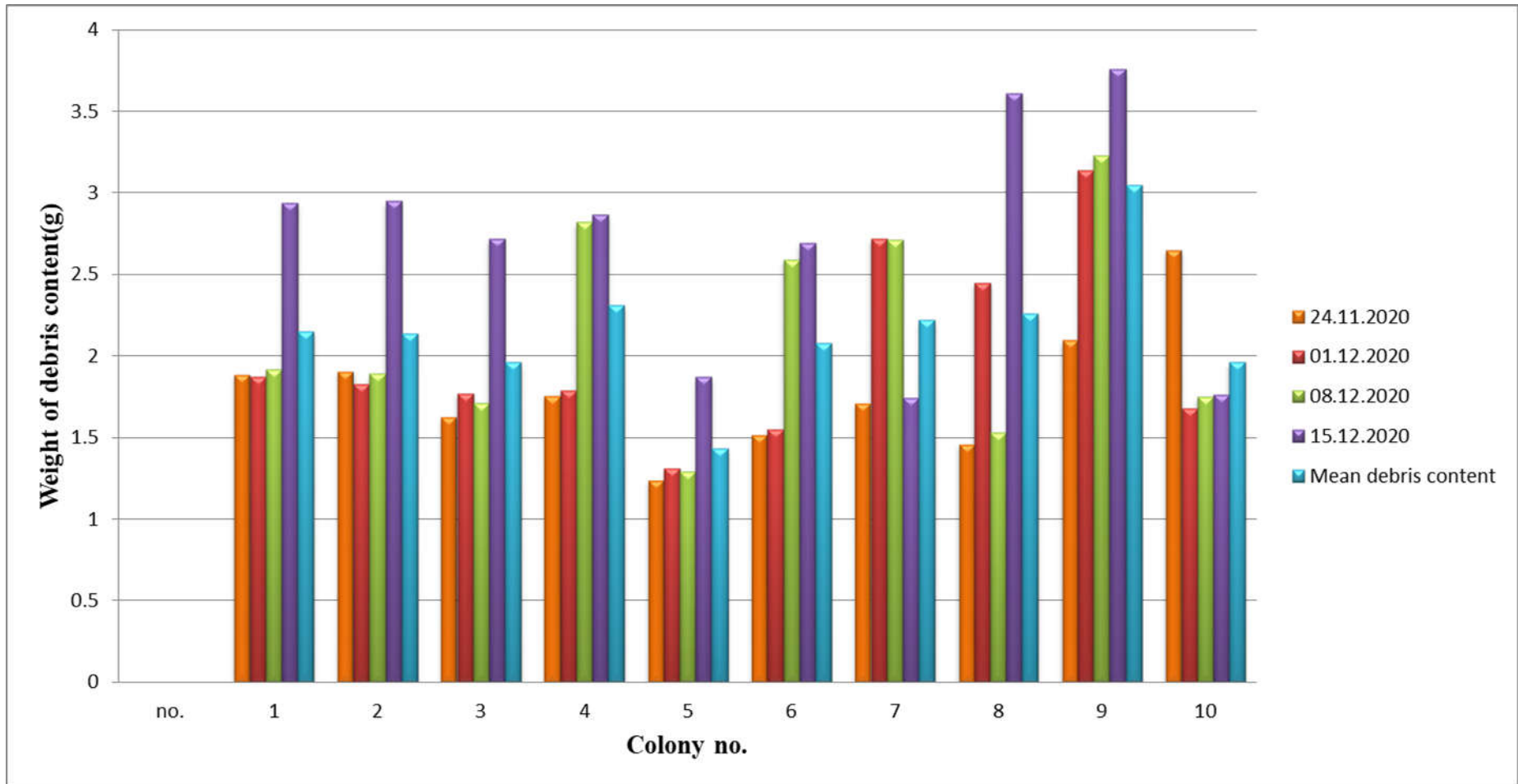


Fig. 2: Weight of debris content (g) of different colonies

2.1.2) Bee activity:

Observation on the activity of honeybee colonies by counting the number of bees leaving the hive (outgoing) and those landing at the entrance (incoming) per minute during different day hours (09:00 to 10:00 hrs., 13:00 to 14:00 hrs. and 16:00 to 17:00 hrs.) was taken for the whole month(01.12.2020 to 31.12.2020) was done.

Android mobile phone was used as stop watch and for each colony 3 observations were taken during different day hours. The data was analysed in factorial RBD with 2 factors in OPSTAT programme. The obtained data regarding bee activity are listed down in table 3.

So from the data of outgoing bees it was concluded that almost all the colonies having good activity except colony no 9. In colony no. 9 the average no. of outgoing bees were 11.33, 9.33 and 6.33 during 09:00 to 10:00 hrs., 13:00 to 14:00 hrs. and 16:00 to 17:00 hrs. respectively. The average no. of outgoing bees for the whole month of colony no. 9 was only 9.00. On the other hand maximum no. of outgoing bees were noticed in colony no. 3 with average no. of outgoing bees were 15.67, 12.00 and 8.67 during 09:00 to 10:00 hrs., 13:00 to 14:00 hrs. and 16:00 to 17:00 hrs. respectively and the average no. of active outgoing bees was 12.11 for the whole month in colony no.3.

While in respect to daytime it was observed that activity or no. of outgoing bees were maximum in early morning that is during 09:00 to 10:00 hrs. with 14.11 average outgoing followed by during 13:00 to 1400 hrs. with 11.79 average outgoing bees and it showed least activity during late hour of the day or during 16:00 to 17:00 hours with only average 7.63 mean no. of bees found to leave their colony. It was observed that bee activity almost completely stopped by 17:30 hrs.

Now in relation to brood survival rate and bee activity it was observed that colony no.9 with the lowest brood survival rate witnessed least no. of active outgoing bees and on the other side colony no. 3 having the 3rd highest brood survival rate, and in other colonies the same trend was observed that colony having high and medium brood survival rate having more no. of active outgoing bees and with low brood survival rate less no of active outgoing bees were noticed.

From the data of incoming bees it was noticed that the lowest active incoming bees were recorded in colony no.9, in colony no. 9 the average no. of incoming bees were only 7.33, 9.00 and 9.33 during 09:00 to 10:00 hrs., 13:00 to 14:00 hrs. and 16:00 to 17:00 hrs. respectively. The average no. of incoming bees for the whole month of colony no. 9 was only 8.56. On the other hand maximum no. of incoming bees were recorded in colony no. 5 with average no. of incoming bees were 10.33, 10.67 and 14.33 during 09:00 to 10:00 hrs., 13:00 to 14:00 hrs. and 16:00 to 17:00 hrs. respectively and the average no. of active outgoing bees was 11.78 for the whole month in colony no.5.

While in respect to day hour point of view it was found that maximum no. of incoming bees were recorded during late hour of the day or during 16:00 to 17:00 hours with average 12.33 no of incoming bees were noticed in all 10 colonies followed by during 13:00 to 1400 hrs. with 10.43 average incoming bees and it showed least activity with only average 8.70 mean no. of bees found to come back to their colony during early hours of the day with the time range of 09:00 to 10:00 hrs.

Now in relation to brood survival rate and bee activity it was observed that colony no.9 having the lowest brood survival rate witnessed least no. of active incoming bees and on the other side colony no. 5 having the highest brood survival rate of 96.87% and in other colonies the same trend was observed that colony having high and medium brood survival rate having more no. of active incoming bees and with low brood survival rate less no of active incoming bees were noticed.

2.2) Economic traits:

2.2.1) Honey stores:

Firstly to measure the economic traits quantitatively of all the colonies a standard grid frame made of iron was made, estimation the honey stores with the help of grid frame was done then converting the number of squares covering honey area into grams by multiplying with 8.06 (considering that an average frame full of honey, contained 1000 g of honey) . Total honey yielding from those selected 10 hives was recorded.

Table 3: Average number of outgoing and incoming bees during different day hours

Period of observation	Colony Number	Outgoing bees during different day hours				Incoming bees during different day hours			
		09:00-10:00 hrs.	13:00-14:00 hrs.	16:00-17:00 hrs.	Average no. of outgoing bees	09:00-10:00 hrs.	13:00-14:00 hrs.	16:00-17:00 hrs.	Average no. of incoming bees
01.12.2020 to 31.12.2020	1	14.77	11.33	6.33	10.78	7.67	9.67	11.33	9.55
	2	13.33	11.00	7.67	10.67	9.67	9.00	11.67	10.11
	3	15.67	12.00	8.67	12.11	9.33	11.33	12.33	11.00
	4	16.33	10.00	9.33	11.89	6.67	10.00	12.33	9.67
	5	15.00	14.33	6.00	11.78	10.33	10.67	14.33	11.78
	6	14.67	12.67	6.00	11.11	10.67	12.00	12.33	11.67
	7	13.00	11.33	9.33	11.22	8.33	10.33	12.00	10.22
	8	14.67	12.00	7.67	11.44	9.67	10.33	14.33	11.44
	9	11.33	9.33	6.33	9.00	7.33	9.00	9.33	8.56
	10	12.33	14.00	9.00	11.78	7.33	12.00	12.33	10.56
	Average	14.11	11.79	7.63	11.17	8.70	10.43	12.23	10.45

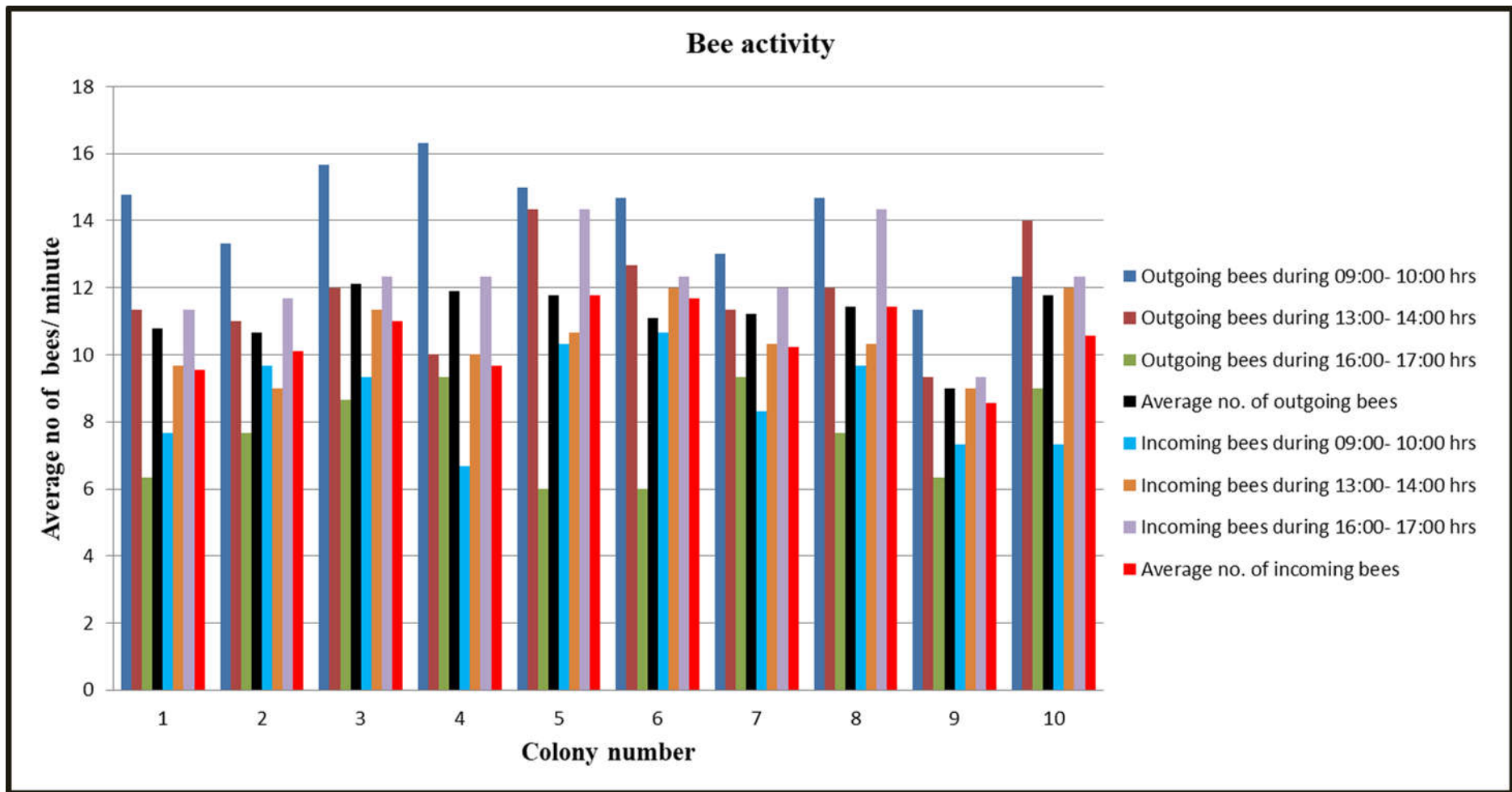


Fig. 3: Average number of outgoing and incoming bees during different day hours

2.2.2) Pollen stores:

Measurements of the pollen stores was done in cm^2 by counting the number of cells in the wire grid covered by pollens were taken. One square of standard grid frame contains 10 cells in to it and area of Single Square of grid frame is 1 square inch and 1 square inch= 6.45 cm^2 . So based on this area of pollen was calculated.

The obtained results of economic traits showed that colonies having high and medium brood survival rates contained more amount of honey and pollen stores. Quantity of honey was maximum in colony no. 5 with 394.94 g as total 49 squares were filled with honey and by multiplying with the factor 8.06 the total quantity of honey was calculated. Some others colonies with higher brood survival rate like colony no. 3, 6, 7, 8 and 10 witnessed 378.82, 346.58, 370.6, 330.46 and 338.49 g of honey respectively. On the other side colony no. 1, 2 and 4 were categorised under medium brood survival rate colony and their yield of honey was 314.34, 298.22 and 322.40 g respectively. While the colony no. 9 that having the lowest brood survival rate among all the colonies able to produce only 249.86 g of honey as only 31 squares were filled with honey of that particular frame inserted in to colony no. 9. While the average honey content for the all 10 colonies was 334.49 g.

The data regarding pollen stores showed the same trend like honey stores as the pollen store was maximum in colony no. 5 with 36.13 cm^2 pollen area as total 52 cells were filled with pollen and by multiplying with 6.45 the total area of pollen in cm^2 was calculated. Some others colonies with higher brood survival rate like colony no. 3, 6, 7, 8 and 10 having also good amount of pollen area with 31.61 cm^2 , 33.55 cm^2 , 27.74 cm^2 , 32.90 cm^2 and 29.03 cm^2 pollen are respectively. On the other side colony no. 1, 2 and 4 were categorised under medium brood survival rate colony and their yield of pollen was 26.45 cm^2 , 29.03 cm^2 and 28.39 cm^2 respectively. While the colony no. 9 that having the lowest brood survival rate among all the colonies able to produce only 25.16 cm^2 pollen area as only 39 cells were filled with honey of that particular frame inserted in to colony no. 9. Average pollen area of all 10 colonies was 26.45 cm^2 .

Table no. 4: Quantity of honey (g) and pollen stores (cm²) in different colonies

Colony Number	Brood survival rate (%)	No. of square filled with honey	Quantity of honey (g) (No of square filled with honey × 8.06)	No. of cells filled with pollen	No. of squares in grid frame covered with pollen	Pollen stores(cm ²) (No. of squares in grid frame covered with pollen × 6.45)
1	84.78	39	314.34	41	4.1	26.45
2	89.18	37	298.22	45	4.5	29.03
3	93.93	47	378.82	49	4.9	31.61
4	89.74	40	322.40	44	4.4	28.39
5	96.87	49	394.94	56	5.6	36.13
6	95.45	43	346.58	52	5.2	33.55
7	92.10	46	370.76	43	4.3	27.74
8	95.55	41	330.46	51	5.1	32.90
9	75.75	31	249.86	39	3.9	25.16
10	92.68	42	338.52	45	4.5	29.03
Average	90.72	41.5	334.49	41	4.1	26.45

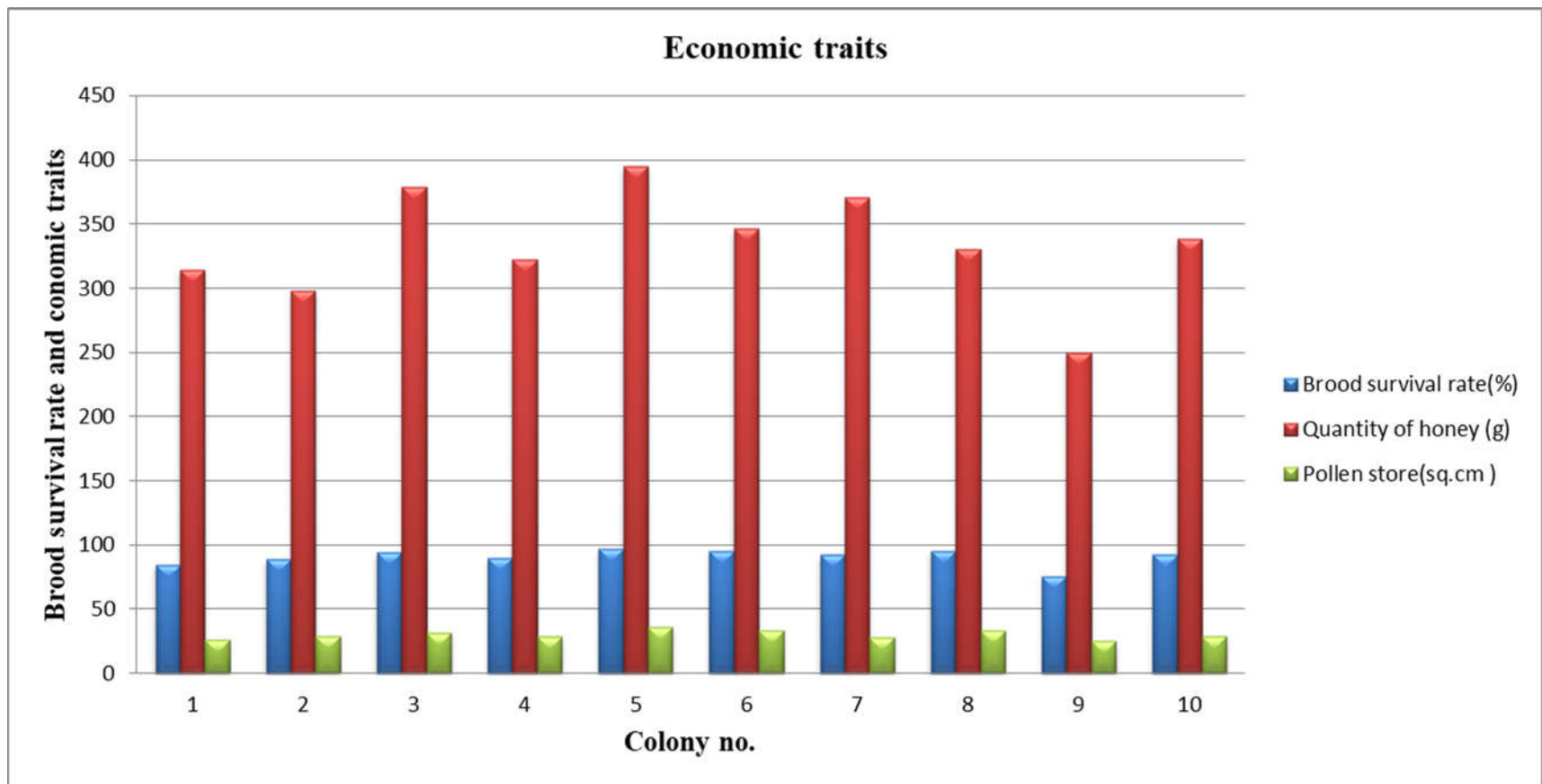


Fig. 4: Quantity (g) of honey and pollen stores (cm²) in different colonies

2.3) Resistance traits:

2.3.1) Hygienic behaviour:

To determine the hygienic behaviour pin killing of 25 capped pupae was done. Killing the pupae in the sealed stage by piercing the caps with sharp steel needle was done. Recording the pierced area after 24 hours to count the no. of dead pupae removed by worker bees and then per cent removal of killed brood was calculated by using the below formula:

$$\text{Per cent removal} = \frac{\text{Number of pin killed pupae removed}}{25} \times 100$$

The obtained data of hygienic behaviour showed that there is significant impact of brood survival rate with resistant traits of *A. mellifera*. As the result showed that colonies having higher rate of survival rates were much efficient to remove the dead pupae and thereby per cent removal was more in those colonies and on the other side colony with low rate of survival were less efficient in removing the dead pupae.

The highest per cent removal of dead pupae was recorded in colony no. 3 as the worker bees of that particular colony were able to remove all 25 dead pupae and thereby per cent removal was 100. While some other colonies like in colony no. 6 the per cent removal were 96 while in colony no. 1, 5 and 8 the value of per cent removal was 92. While in colony no. 9 or the colony with the lowest brood survival rate was able to remove only 19 dead pupae out of 25 and thus per cent removal was only 76. The average per cent removal of dead pupae of all 10 colonies was 90.80%.

Our research is in accordance with the study of Toufailia H *et al.* (2018) where they had quantified removal of freeze-killed and chalkbrood-infected larvae. This was done in open cells and total 20 colonies they used. They also quantified the removal of freeze killed larvae from sealed cells. Categorization of colonies was done and ranged from non-hygienic to fully hygienic (52–100% removal within 2 days). They observed that all larvae killed in open cells were completely removed. That behaviour showed that all colonies, including those with low in hygienic behaviour against dead brood in sealed cells, are highly hygienic against dead brood in open cells and suggests that low hygienic behaviour against dead brood in sealed cells is a trait in its own right. The study also contributes to understand why hygienic behaviour is less common in *A. mellifera*, which is puzzling as it decreased many diseases without adverse effects.

Table 5: Per cent removal of diseased or dead honey bees in different colonies

Colony no.	Per cent brood survival	Pupae killed	Removed pupae (dead)	Removal (%)
1	84.78	25	23	92.00
2	89.18	25	22	88.00
3	93.93	25	25	100.00
4	89.74	25	21	84.00
5	96.87	25	23	92.00
6	95.45	25	24	96.00
7	92.10	25	22	88.00
8	95.55	25	23	92.00
9	75.75	25	19	76.00
10	92.68	25	22	88.00
Average	90.72			90.80

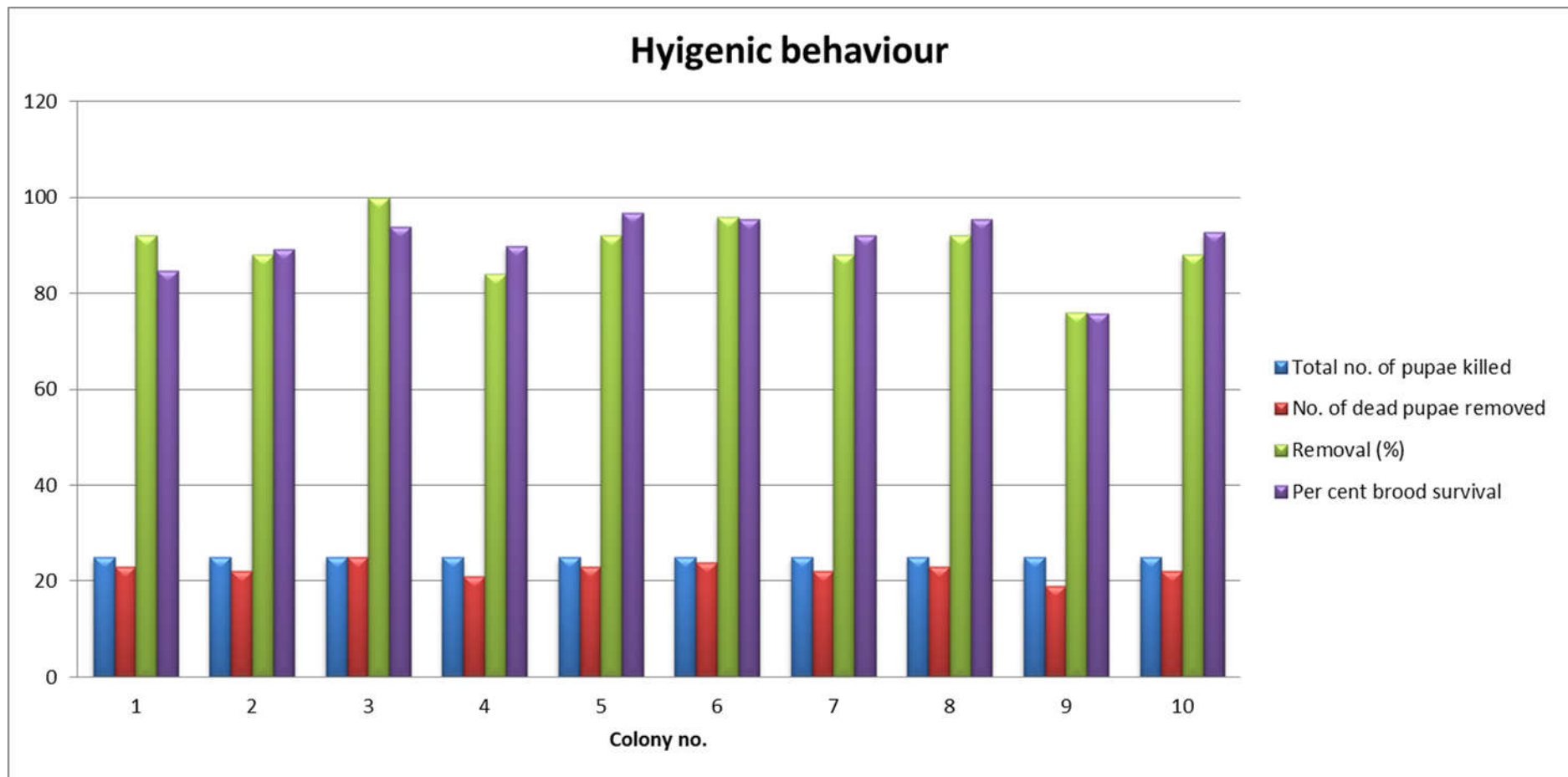


Fig. 5: Per cent removal of diseased or dead honey bees in different colonies

3) Foraging behaviour of *Apis mellifera* L. on mustard:

Mustard (*Brassica sp.*) crop was taken for this experiment to study the foraging behaviour of *Apis mellifera*. So to study the foraging behaviour of *A. mellifera* the following parameters were studied.

3.1) Duration of foraging:

To find out the duration of foraging of *Apis mellifera* on mustard, 10 observations were counted in open eye at 7 days intervals on separate days starting from 11.12.2020 to 12.02.2021 for two times, in morning and evening hours of the day to investigate the initiation time and cessation time of Italian bee on mustard. Initiation time refers to the morning time when *A. mellifera* started their foraging activity and on the other hand cessation time is evening hour when bees stopped their foraging activity.

First exit of a worker bee from the hive was recorded in the morning and last entry was recorded in the afternoon. After last entry of the forager bee, half an hour time was taken for confirmation of last entry.

From the experiment it was obtained that *A. mellifera* started their foraging activities quite earlier and complete cessation in their activity was found in early evening of the day. It was also noticed that duration of foraging kept increasing with increase in flower availability. For *A. mellifera* the duration of foraging was maximum on 12.02.2021 which was 9.35hours and minimum duration of foraging was noticed on 11.12.2020 with 9.03 hours of foraging. While the mean time of initiation time of foraging was 7:28 hrs. and mean cessation time of foraging time was 16.45 pm of the day with average duration of foraging was 9.17 hours.

Table 6: Initiation time and Cessation time of *Apis mellifera* visiting flowers of mustard

Date of observations	Initiation time (hrs.) (First exit)	Cessation time (hrs.) (Last entry)	Duration of foraging (hrs.)
11.12.2020	7:35	16:38	9.03
18.12.2020	7:28	16:35	9.07
25.12.2020	7:36	16:41	9.05
01.01.2021	7:35	16:46	9.11
08.01.2021	7:31	16:41	9.10
15.01.2021	7:25	16:48	9.23
22.01.2021	7:27	16:45	9.18
29.01.2021	7:20	16:49	9.29
05.02.2021	7:21	16:51	9.30
12.02.2021	7:23	16:58	9.35
Average	7.28	16.45	9.17

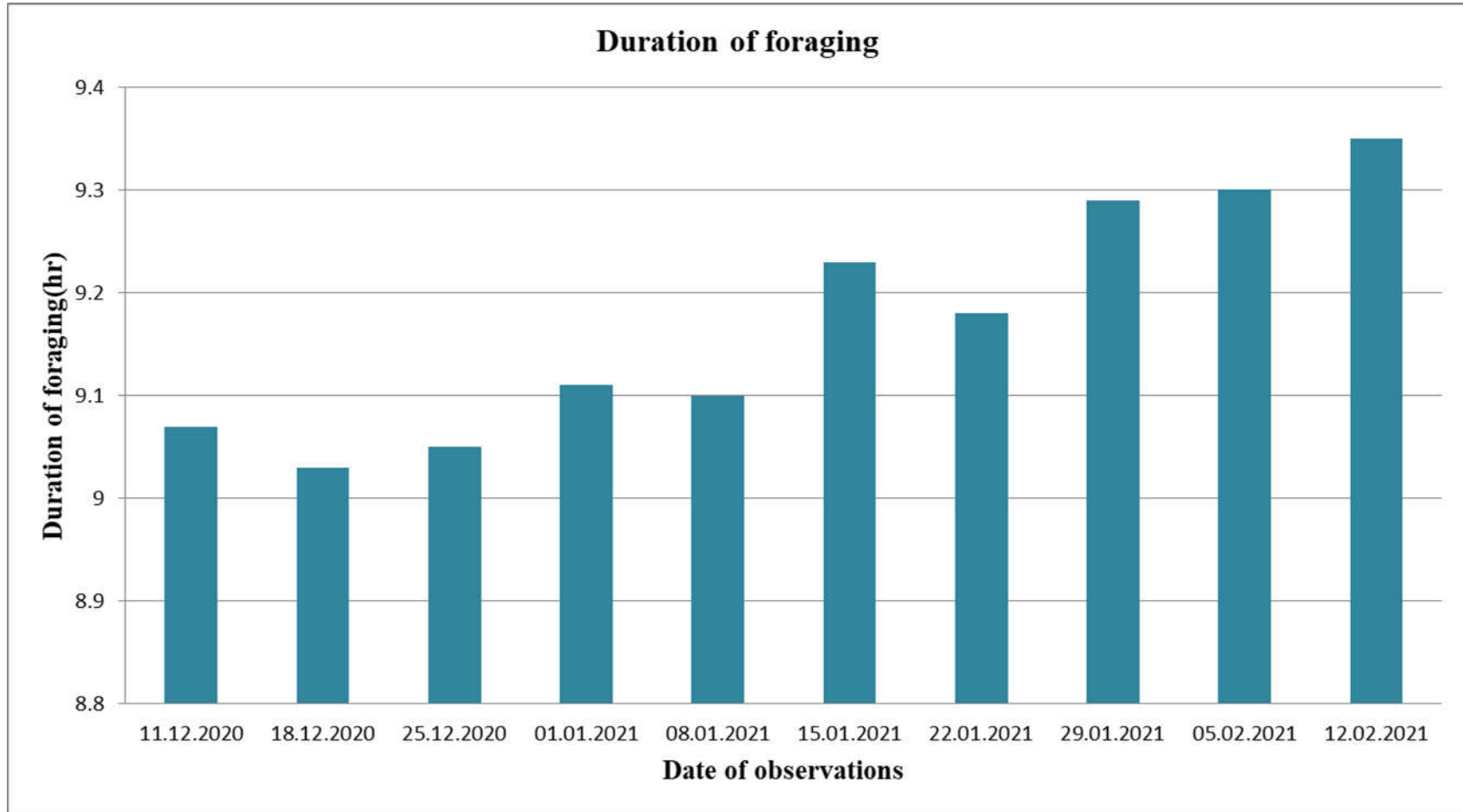


Fig. 6: Duration of foraging of *Apis mellifera* visiting flowers of mustard during different day hours

These findings are quite in accordance with earlier data given by Joshi and Joshi (2010) they stated that, workers of *Apis mellifera* initiated their foraging activity at 7.17 am but they also said that this beginning time can be greatly impacted due to various difference of different region. In another experiment conducted by Singh et al. (2018) showed that *A. mellifera* started foraging earlier (09:36 h) and ceased her foraging late (16:15h) and maximum period of activity (6.35 h) by *A. mellifera* spent time for collecting nectar or pollen in the field of Indian mustard.

3.2) Foraging rate:

Number of flowers visited by a bee per minute was recorded on mustard and this is known as rate of foraging. To determine the foraging rate 5 observations were recorded on different dates at 7 days interval during different day hours (07:00, 09:00, 11:00, 13:00, 15:00 and 17:00 hours of the day).

The foraging rate of *A. mellifera* was experimented during different day hours and the studies showed that average rate of foraging was maximum during 15:00 hours of the day which was 19 flowers/ min followed by 17, 16.44, 10.6 and 0.44 flowers/ min during 13:00hrs, 11:00hrs, 09:00hrs and 17:00hrs respectively while the lowest average rate of foraging was 0.24 flowers/ min during 07:00 hours. It was observed that foraging rate of *A. mellifera* kept increasing as the day hour progressed; the reason behind this was during the early morning hrs. flowers of mustard are fully enriched with pollen and nectar so bees need to visit less no. of flowers to meet their food requirement and on the other side during late hour of the day flowers gets almost exhausted form nectar and pollen and therefore need to visit more no. of flowers to obtain their specific needs of food so foraging rate also get increased.

Average foraging rate of *A. mellifera* was maximum on 07.01.2021 and minimum on 17.12.2020 which was 11.16 and 10.06 flowers/minute respectively.

The obtained data of foraging rate was analysed in factorial RBD with 2 factors date and time through OPSTAT programme.

Table 7: Foraging rate of *Apis mellifera* visiting on mustard flower

Date of observations	Mean number of flowers visited per minute by <i>Apis mellifera</i>						Average
	07:00 hour	09:00 hour	11:00 hour	13:00 Hour	15:00 hour	17:00 hour	
17.12.2020	0.2	11.4	14.2	15.0	19.2	0.4	10.06
24.12.2020	0.2	11.6	18.2	16.2	17.2	0.4	10.63
31.12.2020	0.2	9.6	16.8	16.4	20.6	0.4	10.66
07.01.2021	0.4	11.2	15.4	19.4	20.0	0.6	11.16
14.01.2021	0.2	9.2	17.6	18.0	18.0	0.4	10.56
Average	0.24	10.6	16.44	17.0	19.0	0.44	10.62

Factors	S. Em (\pm)	CD (<5%)	CV (%)
Date	0.33	0.92	11.84
Time	0.36	1.01	
Date x Time	0.80	2.26	

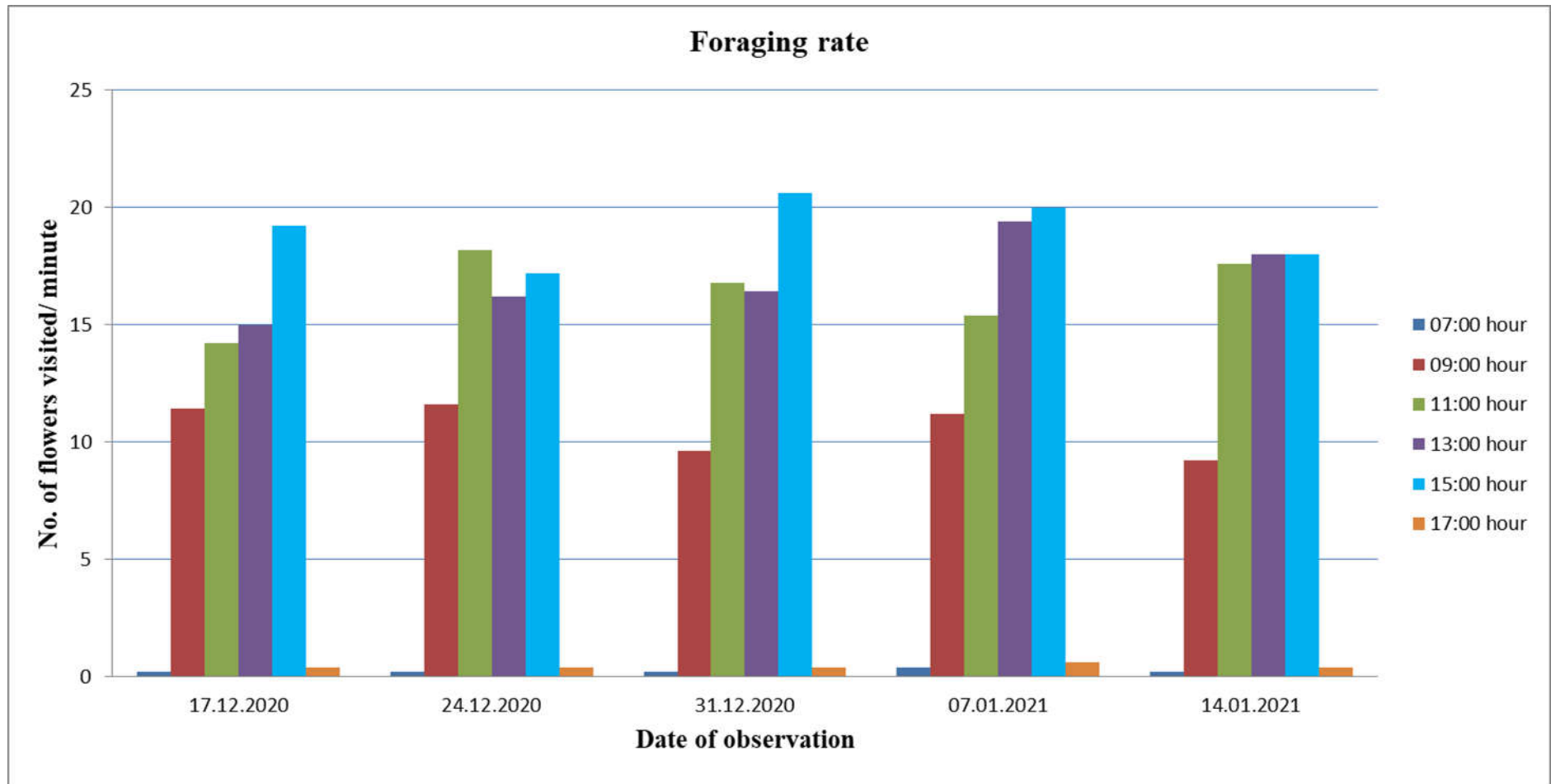


Fig. 7: Foraging rate of *Apis mellifera* visiting on mustard flower

This experiment finding are mostly accordance with previous work where Chakravarty (2000) observed that foraging rate of Indian bee, Italian or European bee, rock bee and little bee was 19.25, 18.80, 16.53 and 21.54 flowers per minute, respectively. Jhajj *et al.* (1996) studied the foraging rate of *Apis* species on raya and brown sarson. In case of raya they noticed that, *A. mellifera* visited 14.06 flowers per minute.

3.3) Foraging speed:

Time spent (seconds) by a single worker bee of *A. mellifera* per flower was recorded by eye estimation using stopwatch of an android mobile phone. To determine the foraging speed 5 observations were recorded on different dates at 7 days interval during different day hours (07:00, 09:00, 11:00, 13:00, 15:00 and 17:00 hrs. of the day). The experiment was replicated five times in factorial RBD, keeping two factors, date and hour.

The obtained result showed that the foraging speed of *A. mellifera* was maximum on 31.12.2020 on that day *A. mellifera* invests average 2.20 seconds on a single flower while the lowest value was recorded on 14.01.2020 where average foraging speed of *A. mellifera* was 1.76 seconds.

Average maximum foraging speed was found during 0900 hours of the day which was 2.94 sec/ flower followed by 2.53, 2.02, 1.99 and 1.51 seconds/ flower during 11:00, 13:00, 15:00 and 17:00 hrs. respectively, on the other side the lowest value was 1.13 sec/ flower obtained during 07:00 hours of the day.

The reason behind this trend in foraging speed was due to the reason that during very early morning at 07:00 hrs. bees were not abundant in the field due to low temperature and fog, then as the temperature started to increase their population was also increased. It was found that foraging speed kept decreasing as the day hour progressed as the flowers of mustard get exhausted from pollen and nectar therefore bees required more no. of flowers to fulfil their food requirement and therefore they spent less time on a single flower and thus foraging speed get decreased.

Table 8: Foraging speed (time spent in seconds) of *Apis mellifera* on mustard flowers

Date of observation	Time spent on flowers (sec.)						Average
	07:00 hour	09:00 hour	11:00 hour	13:00 hour	15:00 hour	17:00 hour	
17.12.2020	1.03	2.85	2.64	1.99	1.93	1.52	1.99
24.12.2020	1.08	2.66	2.62	2.10	2.38	1.77	2.10
31.12.2020	1.39	3.00	2.94	2.29	2.15	1.45	2.20
07.01.2021	1.12	3.72	2.11	2.21	1.52	1.65	2.05
14.01.2021	1.03	2.49	2.37	1.53	1.96	1.19	1.76
Average	1.13	2.94	2.53	2.02	1.99	1.51	2.02

Factors	S. Em (\pm)	CD (<5%)	CV (%)
Date	0.14	0.39	14.32
Time	0.16	NA	
Date x Time	0.35	0.97	

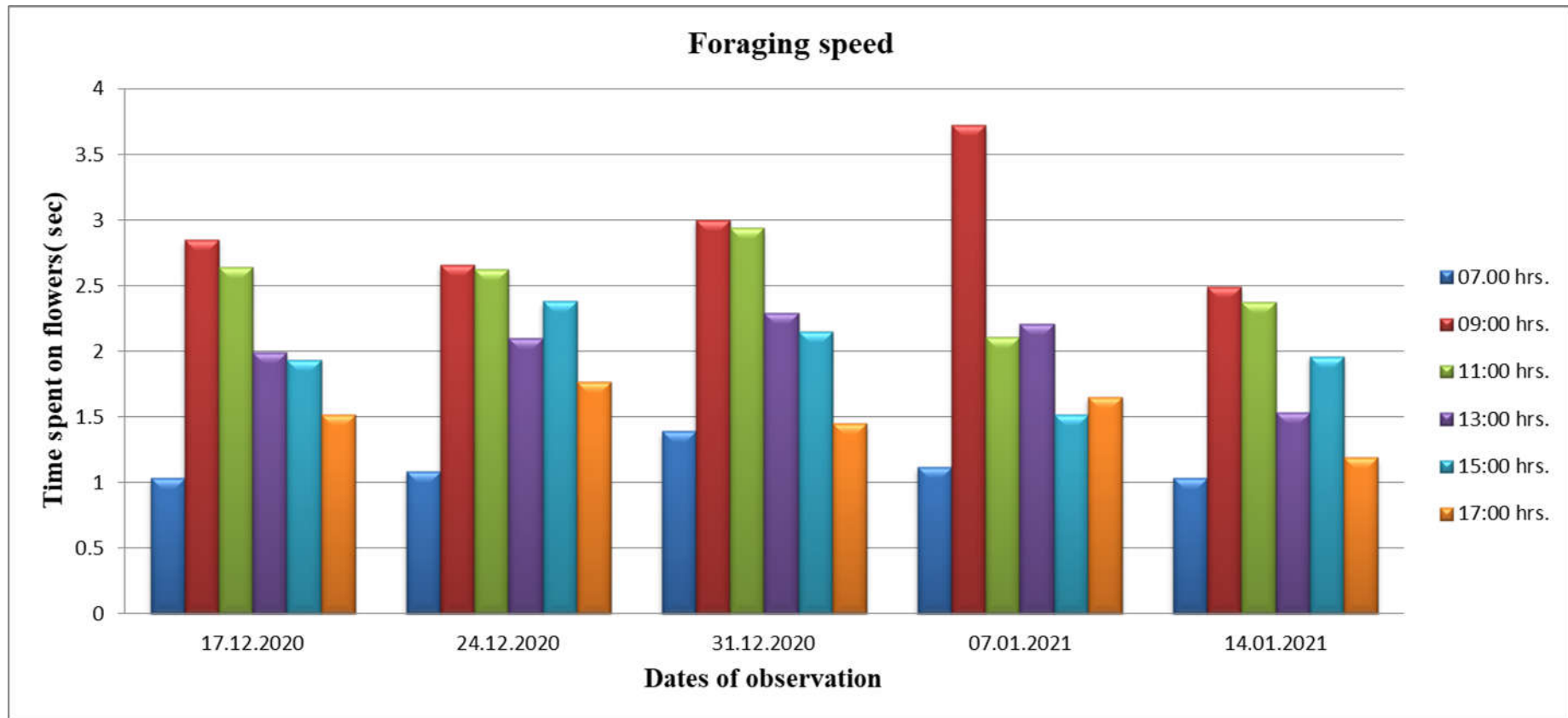


Fig. 8: Foraging speed (time spent in seconds) of *Apis mellifera* on mustard flowers

The findings regarding foraging speed are in consent with the previous records that showed foraging speed depends upon weather parameters and floral rewards availability. Tribhuwan (2016) noticed that *Apis mellifera* showed excellent foraging behaviour by visiting 17.14₋/+1.38 flowers. Nagpal *et al.* (2020) recorded that the mean foraging speed during different hours of the day varied from 2.38 to 3.09 seconds for *A. mellifera* on flowers of *Brassica juncea*. They also observed that foraging speed of *A. mellifera* was minimum (2.38 seconds) during 12:00 to 14:00 hours of the day.

Population dynamics of *Apis mellifera* on mustard bloom:

The bees were counted using stop watch of an android mobile phone. Daily observations were recorded for 5 minutes on mustard bloom at three times *viz.* 09:00 hrs, 12:00 hrs. and 15:00 hrs. in 1 m² randomly selected areas. The mean of the values obtained at three occasions were taken. The meteorological data pertaining to maximum and minimum temperature (°C), relative humidity (%) and rainfall (mm) were collected from the Meteorological Unit of the RPCAU, Pusa.

The data on population dynamics of *A. mellifera* on mustard bloom showed that the maximum mean population was recoded on 03.12.2020 and lowest on the day of 15.01.2021 which was 58.70 and 39.83 respectively. The population of bees on mustard bloom fluctuate with the daily temperature and relative humidity. Data was taken during the whole flowering period and total 10 standard meteorological weeks (SWM) were taken in to consideration for this experiment.

Population of forager bees on mustard flower less during the early hour of the day with mean population of 41.31 as the temperature remains cool during 09:00 hour of the day and maximum population was recorded during 12:00 hour of the day which was 59.40 as the temperature at that time was much favourable during that time. While the population tends to decrease as the temperature falls during late hour of the day and mean population was found 47.36 during 15:00 hour of the day.

Population of *A. mellifera* was maximum on 49th standard week when the average temperature was also highest among all 10 standard weeks and then decrease in mean population was observed during 50th and 51st SWM. Then again increase in mean population was noticed during 52nd and 01st SWM followed by decrease in 02nd

and 03rd SWM, the mean population was lowest in 03rd SWM and the average temperature was also the lowest among all SWM taken in consideration. Then from 04th SWM gradual increase was observed and continued up to 06th SWM.

This findings are in accordance with the recent research of Singh *et al.* (2018), they conducted their experiment on the population dynamics (number of bee visits/minute /m²) of *Apis spp.* on rai and showed that among all *Apis spp.* European honey bee or *A. mellifera* was dominant pollinator. They recorded 13.06 average bees//minute /m²) on mustard flower between 20th November 2016 to 10th December 2016.

Table 9: Population dynamics of *Apis mellifera* on mustard bloom during different day hours

Standard Meteorological Weeks (SMW)	Mean * population dynamics of <i>Apis mellifera</i> on mustard bloom(counted for 5 minutes in 1 m ² randomly selected area)			Average
	09:00 hrs.	12:00 hrs.	15:00 hrs.	
49	47.5	76.75	51.87	58.70
50	42.25	70.5	50.62	54.45
51	37.00	64.85	44.12	48.66
52	43.75	67.37	50.25	53.79
01	44.62	69.35	53.13	55.70
02	36.14	61.57	43.28	47.00
03	35.6	42.3	41.6	39.83
04	39.1	41.5	39.4	40.00
05	39.9	43.3	44.7	42.63
06	47.3	56.5	54.7	52.83
Average	41.31	59.40	47.36	49.36

(* Replication of 3 values)

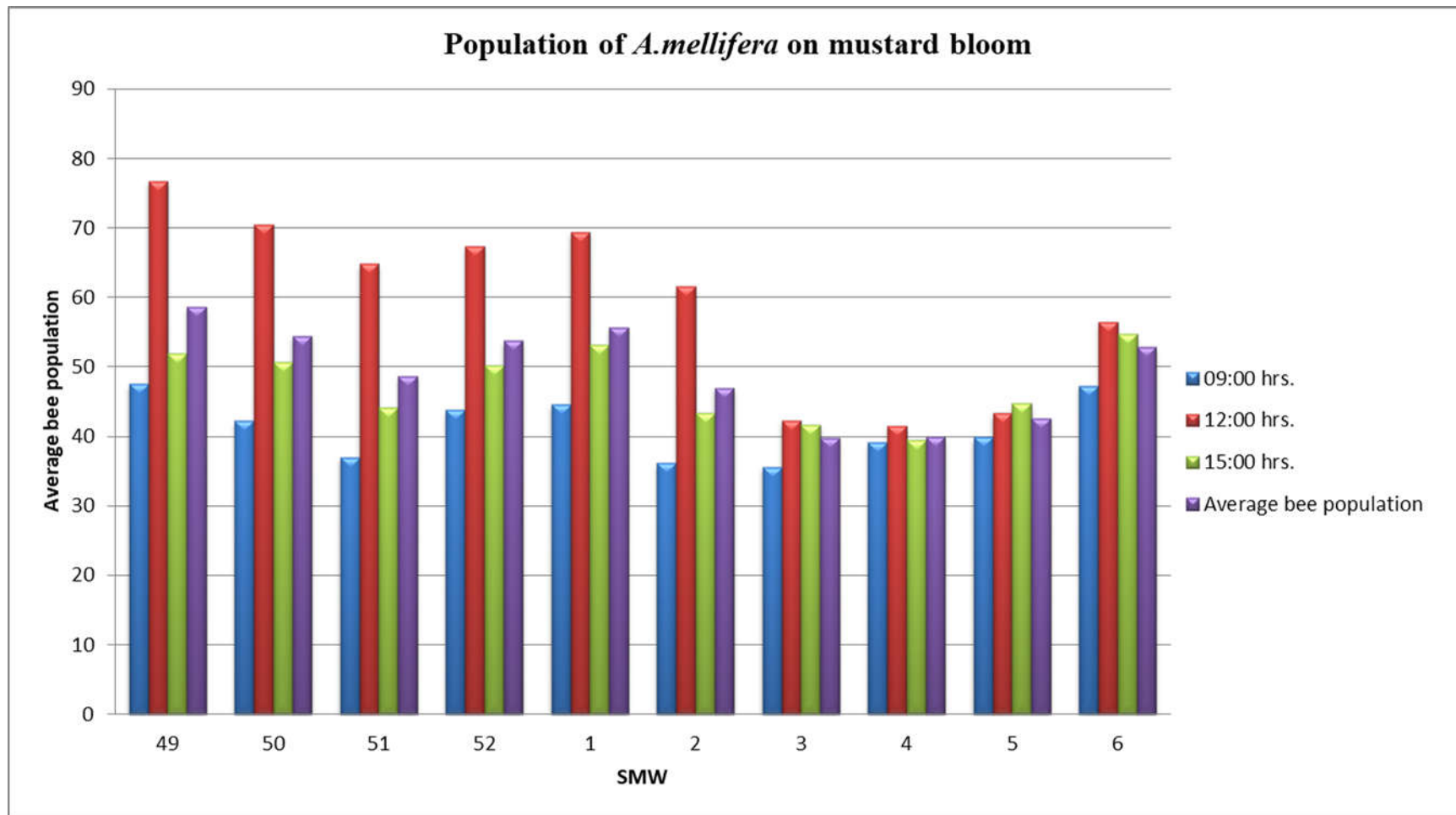


Fig. 9: Population dynamics of *Apis mellifera* on mustard bloom during different day hours

Table 10: Population of *Apis mellifera* in relation with meteorological data

SM W	Bee populati on (no.)	Average temperature (°C)			Average relative humidity (%)			Total rainfa ll (mm)
		Maximu m	Minimu m	Averag e	Mornin g	Evenin g	Averag e	
49	58.70	23.51	12.22	17.87	95.75	76.37	86.06	0
50	54.45	22.68	12	17.34	81.28	72.2	76.74	0
51	48.66	19.84	8.38	14.11	95.71	71	83.35	0
52	53.79	22.88	7.96	15.42	94.25	65.37	79.81	0
01	55.70	24.45	11.24	17.84	94.71	65.71	80.21	0
02	54.43	21.67	10.95	16.31	95.28	68.42	81.85	0
03	39.83	15.60	8.22	11.91	93.71	77.42	85.56	0
04	40	16.94	8.27	12.60	94.71	82.14	88.42	0
05	42.63	19.44	7.2	13.32	94.28	74	84.14	0
06	52.83	24.61	10.68	17.64	90.28	62.85	76.56	0

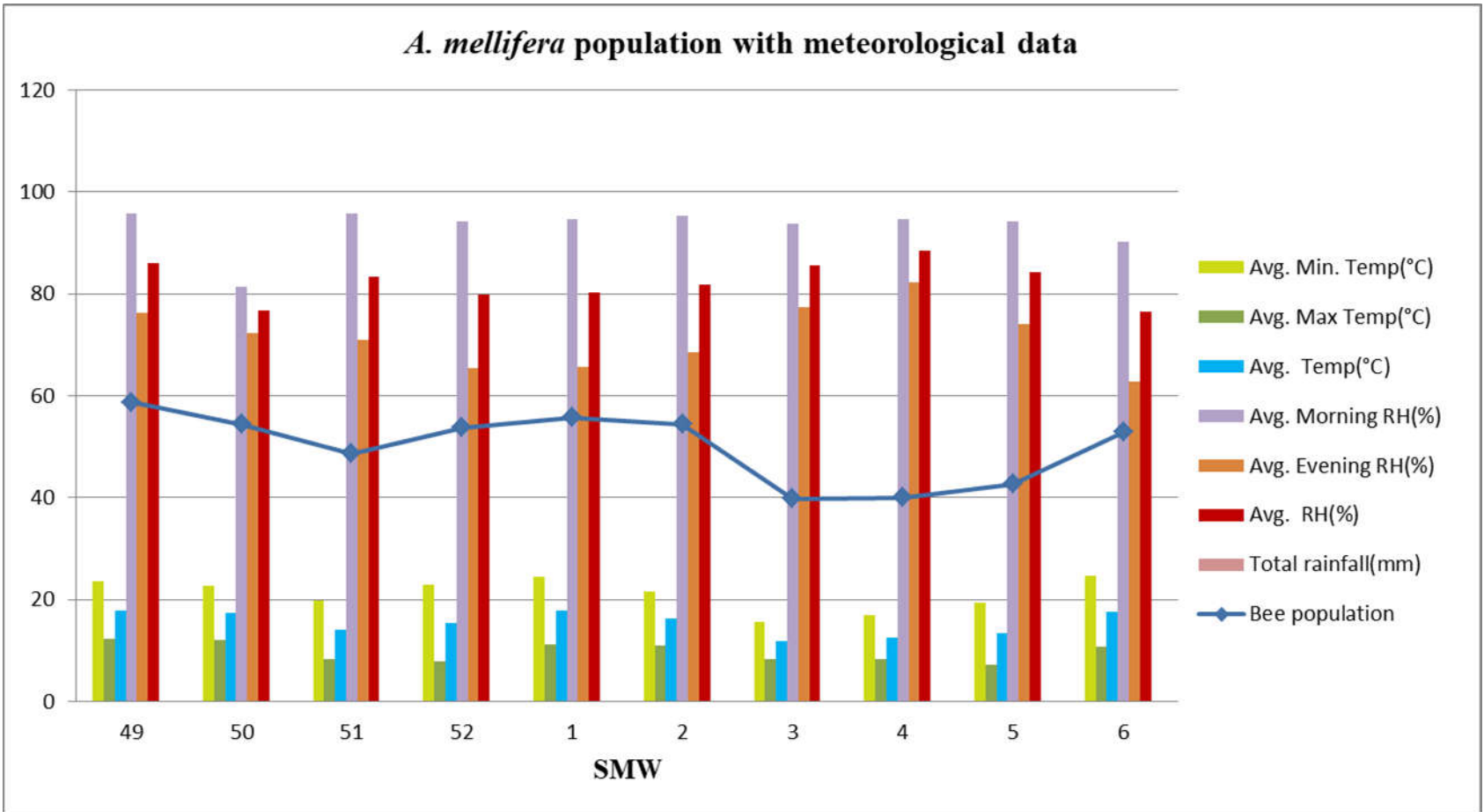


Fig. 10: Population of *Apis mellifera* in relation with meteorological data

Table 11: Correlation coefficient between weather parameters (X) and mean population of *Apis mellifera* on mustard bloom (Y)

Weather parameters	Correlation coefficient (r)
Average maximum Temperature (°C) (X ₁)	0.93 ^{**}
Average minimum Temperature (°C) (X ₂)	0.76 [*]
Average Temperature (°C) (X ₃)	0.94 ^{**}
Average morning relative Humidity (%) (X ₄)	-0.70 [*]
Average evening relative Humidity (%) (X ₅)	-0.18
Average relative humidity. (%) (X ₆)	-0.64 [*]
Total rainfall (mm) (X ₇)	0

Multiple regression equation:

$$Y = 10.56 + 1.19(X_1) + 1.44 (X_2) + 1.33(X_3) + 0.42 (X_4) + 0.15(X_5) - 0.47(X_6)$$

Coefficient of determination (R^2) = 0.86

(**p < 0.01)

(* p < 0.05)

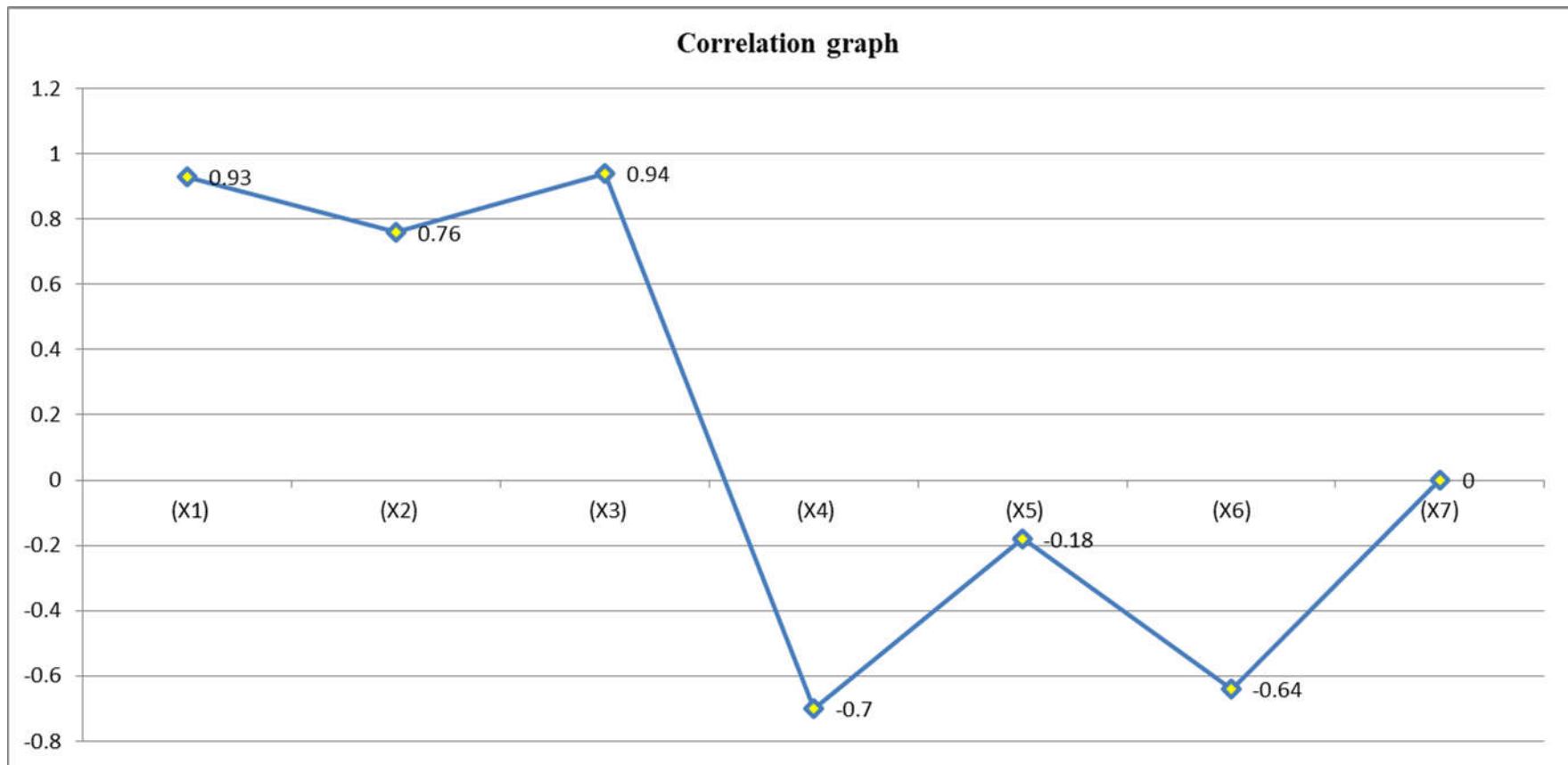


Fig. 11: Correlation coefficient graph between weather parameters (X) and mean population of *Apis mellifera* on mustard bloom (Y)

Correlation of bee population with weather parameters is listed down in table 11 and depicted in fig. 11. It clearly showed that average maximum temperature(°C) and average temperature(°C) showed positive and strong correlation with significant effect while average minimum temperature(°C) showed positive correlation with significant effect on bee population($r = 0.93^{**}$, 0.94^{**} and 0.76^* respectively).

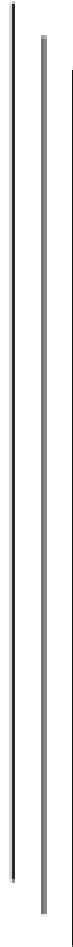
On the other side average morning relative humidity (%) and average relative humidity (%) showed negative correlation with significant effect while average evening humidity showed negative but non-significant effect on bee population on mustard bloom($r = -0.70^*$, -0.64^* and -0.18 respectively). There was no rainfall was recorded during the whole experiment so no correlation was found between total rainfall (mm) and bee population. However all the weather parameters influenced 86 % bee population ($R^2 = 0.86$).

These data are quite similar with the findings of Singh *et al.* (2018) where they have found that the population of different *Apis spp.* On rai was obtained when the maximum temperature was 20.0°C with low humidity 60.75% and on the other side the population was minimum when the recoded temperature was 15.25°C and at high humidity 84.33%. So they concluded that there is positive correlation was there between the *Apis mellifera* population and temperature while with relative humidity they observed negative correlation.





CHAPTER - V



SUMMARY AND CONCLUSIONS



SUMMARY AND CONCLUSION

The whole experiment on “Studies on brood survival rate and foraging behaviour of *Apis mellifera* L. on mustard” was successfully carried out at University Apiary, RPCAU, Pusa, Bihar during “Rabi” 2019- 20.

The experiment was conducted on 10 bee hives and the results showed that out of 10 colonies 1 colony showed low brood survival rate (survival rate 70- 79 %), 3 colonies witnessed medium brood survival rate (survival rate 80-89%) and 6 colonies showed high brood survival rate (survival rate 90- 99 %).

Excellent, medium and poor brood pattern was found in 7, 2 and 1 colonies respectively in the experiment of study on scattered brood pattern.

The study on behavioural traits of *A. mellifera* showed that in case of nest cleaning ability, colonies with high and medium survival rate were much more cleaner compared to colony having low brood survival rate.

Experiment on bee activity was done on the basis of the value of total numbers of outgoing and incoming bees / minute. The obtained data showed that colony no. 9 having lowest number of outgoing bees with average 9 bees / minute. Highest outgoing bees were recorded in colony no. 3 with average 12.11 bees / minute. While in case of day hour most of the bees left the hive during early morning with the time range 09:00 to 10:00 hours followed by 13:00 to 14:00 hours and lastly 16:00 to 17:00 hours.

In case of incoming bees colony no. 5 having the highest value with average 11.78 incoming bee and colony number 9 having the less value with average 8.56 of bees came back to the hive. In respect to day hours most of the honey bees returned to the colony during 16:00 to 17:00 hours followed by 13:00 to 14:00 hours and lastly during 09:00 to 10:00

Total honey content was maximum in colony no. 5 with 394.94 g and lowest in colony no. 9 having only 249.86 g of honey. For pollen content the same trend was followed as colony no. 5 having maximum area of pollen of 36.13 cm² and lowest in colony no. 9 with total area of 25.16 cm² pollen.

The obtained results of hygienic behaviour showed that colony no. 3 having 100 per cent efficiency as their worker bees were able to remove all 25 dead pupae. Colony no. 9 having lowest efficiency of 76% as only 19 out of 25 dead pupae were removed.

Duration of foraging was maximum on 12.02.2021 which was 9.35 hours. While the minimum duration of foraging was recorded on 11.12.2020 with 9.03 hours. Mean initiation time of foraging was very early morning at 7:28 hours and mean cessation time of foraging time was 16.45 hours of the day. The average duration of foraging was 9.17 hours.

The foraging rate of *A. mellifera* was experimented during different day hours and the studies showed that average rate of foraging was maximum during 15:00 hours of the day which was 19 flowers/ min while the lowest average rate of foraging was 0.24 flowers/ min during 07:00 hours. Average foraging rate of *A. mellifera* was maximum on 07.01.2021 and minimum on 17.12.2020 which was 11.16 and 10.06 flowers/minute respectively.

Foraging speed of *A. mellifera* was maximum on 31.12.2020 on that day *A. mellifera* invests average 2.20 seconds on a single flower while the lowest value was recorded on 14.01.2020 where average foraging speed of *A. mellifera* was 1.76 seconds. Average maximum foraging speed was found during 0900 hours of the day which was 2.94 sec/ flower and on the other side the lowest value was 1.13 sec/ flower obtained during 07:00 hours of the day.

Population dynamics of *A. mellifera* on mustard bloom was recorded for 5 minutes in 1m² randomly selected areas from the plot. The observation was counted by using an android smart phone stop watch. Daily observation was taken during the whole flowering period. Data was taken during 3 different day hours i.e. at 09:00 hours, 12:00 hours and 15:00 hours. Maximum mean population was recorded on 03.12.2020 and lowest on the day of 15.01.2021.

The population of *A. mellifera* was correlated with daily minimum temperature, maximum temperature, average temperature, average relative humidity at morning hours, average relative humidity at evening hour, average relative humidity and with total rainfall. The obtained data showed that population of *A.*

mellifera showed positive correlation with daily minimum temperature, maximum temperature, average temperature. With relative humidity at morning hours, relative humidity at evening hour, average relative humidity it had negative correlation. As there was no rainfall during that particular time there was no correlation found between population of *A. mellifera* and average rainfall.

Conclusion

- Colonies having high and medium brood survival rates were much cleaner, more active, showed higher economic and resistant traits and good hygienic behaviour.
- Colony with low brood survival rates was less clean, less active and poor in resistant and hygienic behaviour.
- It was noticed that the duration of foraging kept increasing as the blooming period progressed.
- Foraging rate showed that mean rate of foraging was maximum during 15:00 hours of the day and lowest during 07:00 hours.
- Foraging speed was maximum during 09:00 hours of the day lowest during 07:00 hours of the day.
- Population of *A. mellifera* showed positive and significant correlation with average temperature and negative and significant correlation with average relative humidity.
- There is a significant impact of brood survival rate with both physiological and economical traits of *A. mellifera* therefore to get good profit in terms of economic aspects of apiculture one must go with colonies having high brood survival rates.
- The foraging behaviour and activity of *A. mellifera* is highly depends upon weather parameters.
- So proper planning should be done in accordance with the season and selection of queen from colonies with high brood survival rate will leads to high net return to apiculturists.





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