MELISSOPALYNOLOGY OF INDIAN HONEY BEE (*Apis cerana indica* Fab.) APIARIES IN SOUTHERN KERALA

by

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DECLARATION

I hereby declare that this thesis entitled **"Melissopalynology of Indian honey bee (*Apisceranaindica*Fab.) apiaries in southern Kerala"** is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award of any degree, diploma, fellowship or other similar title, of any other University or Society.

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LIST OF ABREVIATIONS

%	-	per cent
AICRP	-	All India Coordinated Research Project
ANOVA	-	Analysis of Variance
Conc.	-	Concentrated
CoV	-	Co Variance
et al.	-	And others
Fam.	-	Family
Fig.	-	Figure
mm	-	millimeter
ml	-	milliliter
MT	-	Metric Tonnes
nos.	-	Numbers
sp.	-	Species
SD	-	Standard Deviation

INTRODUCTION

1. INTRODUCTION

Beekeeping is an agro based enterprise where bees are utilized to harvest nectar and pollen from the plant sources to produce honey and other hive products. In India, beekeeping covers 40,000 villages and provides part time employment to more than 2,50,000 persons (Sivaram, 2006). More than 5,000 tonnes of honey is produced annually in our country. In India, Kerala produces largest quantity of honey (Nair, 2005). Kerala state, which supports a green cover of wild and cultivated forest trees, conventional food, ornamental and medicinal plants, plantation crops such as coconut, tea, coffee, rubber, cardamom etc. has an immense potential for commercial bee keeping industry (Padmanabhan, 2003). There are about 600,000 bee colonies in the state. An average yield of 20-25 kg honey per colony is being produced in Kerala making the industry a profitable one (Prathapan *et al.*, 2010).

Apis florae Fab., *Apis dorsata* Fab., *Apis cerana* Fab. and *Apis mellifera* Fab. are the species of honey bees distributed in the Kerala state. Of these, *A. cerana* and *A. mellifera* are used for commercial bee keeping. The extra floral nectaries of rubber (*Hevea brasiliensis* Muell. Arg.) which is produced after the commencement of new flushes during January to April serves as a major source of nectar to bees and is being utilized for commercial beekeeping in the state. Apicultural practices can be divided into three seasons under Kerala conditions namely honey flow season, dearth season and brood rearing season. During honey flow season foraging activity of bees will be higher and the honey production also will be increased. Availability of nectar and pollen is scarce during dearth period as the foraging activity will be less due to heavy rain. Brood rearing season is the growth period of honey bees where the queen bee lays more number of eggs there by increases the colony population. Thus pollen is an essential requirement during brood rearing season for bee nutrition.

Higher honey yield from a particular honey bee species does not entail that it is a good source for honey production. Here floral structure, floral display and the bee

behavior of individual species act together so as to offer the sweet liquid for the bee visitor (Nair, 2007). Thus recognition and initial screening of various plants representing potential sources of nectar and pollen for honey bees throughout the year is an important prerequisite for launching apiary industry in any locality (Kalpana and Ramanujam, 1997). There are many endemic plant species with important forage values, which are good geographical markers. (Chaturvedi and Temsunungla, 2008).

Palynology is the study of pollen grains, spores and of other biological materials that can be studied by means of palynological techniques (Hyde and Williams, 1944). Melissopalynology is the one of the applied branch of palynology that deals with the microscopic analysis of the pollen contents of honey and pollen loads from a locality. It will provide reliable information regarding the floral types which serves as major / minor nectar and / or pollen sources for the honey bees (Peterson and Bryant, 2011; Bryant and Jones, 2001; Lieux, 1975). Further, these studies reveal various types of unifloral honey that can be obtained in different seasons. Informations on floral sources will help the bee keepers to frame proper management practices during dearth periods (Ramanujam and Khatija, 1991). Pollen analysis in honey can provide information on geographical and botanical origin of honey to a certain extent. Moreover, pollen analysis provide some important information about honey extraction and filtration, fermentation (Russman, 1998), some kinds of adulteration (Kerkvliet *et al.*, 1995) and hygienic aspects such as contamination with mineral, dust and starch grains (Louveaux *et al.*, 1978).

Melissopalynological studies, thus aids the beekeepers to formulate their seasonal bee management schedules particularly for migration of colonies to different floral sources by identifying the floral diversity of that specified area. This can enhance the honey production and also the quality of honey. A comprehensive knowledge about bee botany and phenology enables the bee keepers to identify plants in the local area, which are significant contributors to the bee colony build up/honey flow. Thereby they can recognize the key pollen resources for the critical brood

rearing and dearth period of the honey bee. Location specific melissopalynological studies in combination with scientific bee management practices pave a way for higher yield and better quality honey.

India is rich with diversified flora providing abundant quality of nectar and pollen to the bees. The very first attempt in the study of honey plant resources in Kerala was made with the co-ordinate activities of Khadi Village Industries Corporation (KVIC) in 1959, the commission made a preliminary survey on the bee flora of Karnataka and Kerala and listed very few plants as bee forage in these two states. Later pollen analysis of 76 different samples of Indian honeys from various floristic zones of India was made by Nair (1964).

Honey obtained from our state represents a large array of diversified flora but scientific information about this bee flora is limited. Since such knowledge is localized as it depends on local vegetation, location wise scientific knowledge on pollen and nectar sources are highly indispensable. However, the location based study conducted in Kerala state has limited information on the seasonal variation (Nair, 2007).

Under these circumstances, the present study was carried out with the following objectives:

- To identify the potential pollen and nectar sources of honey bee across the seasons in southern Kerala.
- To estimate the contribution of each bee flora during all the three seasons.
- To determine the frequency of occurrence of each pollen types across the three seasons.
- To cluster the locations with similar floristic composition based on the presence or absence of pollen types.

REVIEW OF LITERATURE

2. REVIEW OF LITERATURE

Kerala is blessed with rich floral diversity and congenial climatic conditions which offers immense scope for bee keeping in the state. The state has been contributing 70 per cent of the annual production of honey in India till 1991 (Jacob *et al*., 1992). *Apis cerana indica*, known as Indian honey bee or Asian hive bee is the most popular honey bee species used for commercial bee keeping in Kerala. Taxonomically Indian honey bee belongs to the family Apidae under the order Hymenoptera. Apidae has three subfamilies: Xylocopinae, Nomadinae and Apinae. The subfamily Apinae has nineteen tribes including Apini, Meliponini and Bombini. The tribe Apini contains only one genus, *Apis* (Michener, 2000).

In this chapter, effort has been made to review of the available literature pertaining to the different nectar and pollen source of *A. cerana indica* in different seasons.

2.1 BEEKEEPING IN INDIA

Scientific bee keeping in India was started at the end of nineteenth century. The first attempt in India to keep *Apis cerana* bees in movable frame hives to enhance maneuverability were made in 1880 in Bengal and in 1883-84 in Punjab and Kullu Valley but with little success. In South India, Rev. Newton started beekeeping training and trained a number of rural folks during 1911-17 and also devised a hive for *A. cerana* now named after him (*Newton hive*) for Indian climatic conditions. Beekeeping work in earnest was taken up in Travancore in 1917 and in Mysore in 1925. The recommendation of *Royal Commission on Agriculture* (*1928*) for developing cottage industries gave a boost to beekeeping in rural India. Beekeeping work afterwards in real earnest was taken up in Madras in 1931, in the Punjab in 1933, in Coorg in 1934 and in Uttar Pradesh in 1938. In 1938-39, beekeepers of India

organized themselves and founded *All India Beekeepers Association*. Afterwards, Indian Council of Agricultural Research (ICAR) established the first Beekeeping Research Station in the Punjab in 1945 and six years later at Coimbatore in Tamil Nadu (Sivaram, 2003). Since 1950, ICAR has been funding various research projects on beekeeping. In 1980, ICAR started All India Coordinated research Project (AICRP) on Honey Bees Research and Training which at present known as AICRP on Honey Bees and Pollinators has 15 Co-ordinating centres and 2 voluntary centres throughout the country with administrative centre at Haryana Agricultural University Campus at Hisar in Haryana state.

2.2 BEEKEEPING IN KERALA

Scientific beekeeping in Kerala dates back to 1824, when a European Missionary Fr. Newton introduced the principles of movable frame hives, smokers, bee knife and honey extractors. Along with Khadi commission, regional khadi and village industries boards were established in different states including Kerala. Through their nodal agencies and regional centres, financial and technical assistance were provided to local beekeepers and Bee Keeper's Co-operative Societies were established (Padmanabhan, 2003).

Migratory bee keeping with Indian bees, *A. cerana indica* formed the basis of the apiculture industry of the state. Detection of rubber as a rich source of extra floral nectar during seventies gave a big boost to the beekeeping industry in the state. About 50,000 beekeepers and their families depend on this industry for their livelihood. However, rubber could provide nectar only during honey flow season (January to April). Flora around the apiary serves a major role in the growth and development of honey bees especially during the brood rearing season (October to December) (Nair, 2007).

2.3 HONEY AND POLLEN

Usefulness of a flora depends upon the quantity of food and energy provided by the flora which satisfies the energy requirement of bees. According to Lin *et al.* (1990), carbohydrates are provided by nectar whereas proteins by the pollen which is essential for the growth and development of honey bees.

The various chemical components of honey includes carbohydrates as the major portion (about 82%) and proteins comprising a number of enzymes and eighteen free amino acids, of which proline is the most abundant one. Pollen consists of protein (25 %), vitamins, fats, oils, pigments and resins which accounts for 1-14 % of dry mass. Pollen is also a rich source of minerals such as phosphorous, potassium magnesium, calcium and iron (Mishra, 1995).

According to Bryant and Jones (2001) pollen can be incorporated into honey in a number of ways; through behaviour of honey bees during foraging and storing of honey, by anemophilous plants and by the action of apiarists. Honey bees visit flowers of diverse plant species, foraging for nectar and pollen grains, thus enabling fertilization as well as conservation of biodiversity. When bees collect nectar from flowers, they obtain some quantity of pollen from the flower of the plant. After the nectar has been converted into honey in the hive, some of the pollen remains in the honey (Engel *et al.*, 2005).

2.4 MELISSOPALYNOLOGY AND ITS IMPORTANCE IN BEEKEEPING

Melissopalynology is the branch of palynology that studies pollen and spores in honey (Ebenezer and Olugbenga, 2010). According to the 1868 edition of Paxtons Botanical Dictionary, both melissa and melitta means a bee. For over 100 years the

literature pertaining to the study of pollen in honey has been termed in several ways, including melissopalynology, mellittopalynology and melitopalynology (Bryant and Jones, 2001).

Pollen and nectar availability to foraging bees fluctuated with time of the year and flowering of different species of plants (Free, 1970). The nectar yielding plants contributing to nectar/honey flow are specific to different areas and they have micro-regional habitat. Even in rich floral areas, continuous succession of nectar yielding plants throughout the year is lacking. Bees might face protracted dearth period and in certain areas, they may not have flora to accumulate sufficient honey stores.

For successful beekeeping, it is desirable to have crops that flower one after the other and thus provides a sequence of flowering for a continuous supply of food to bees all the year around within the surrounding area of an apiary (Free, 1970; Akrathanakal, 1987). Generally more than one species are planted and choice can be made in favour of these species that can provide nectar and pollen to the bees. Recognition and initial screening of various bee plants representing potential sources of nectar and pollen for honey bees throughout the year is an important prerequisite for launching apiary industry in any locality (Kalpana and Ramanujam, 1989). This paved way to melissopalynological studies which are essential to ensure the plant species availed by bees.

2.5 METHODS IN MELISSOPALYNOLOGICAL ANALYSIS

There are different types of methods for preparing samples from honey in melissopalynology; ranging from acetolysis to KOH treatment. Louveaux *et al.* (1970) proposed different methods of honey analysis with and without acetolysis. It makes the pollen grains transparent and thus helps the pollen identification in the honey and pollen samples. However, the method of analysis recommended by

International Commission for Bee Botany (ICBB) remains as a well established method involved in routine honey analysis and it was considered adequate for verifying the pollen spectrum of a honey sample with declared botanical and geographical origin (Louveaux *et al*., 1978; Jones and Bryant, 2001; Der *et al*., 2004).

2.6 LOCATION BASED STUDY

Palynological investigations of honey and pollen load samples from different parts of India (Nair, 1964; Sharma and Nair, 1965; Suryanarayana and Thakar, 1966; Sharma, 1970; Suryanarayana *et al*., 1981; Shah and Shah, 1989; Kalpana and Ramanujam, 1996; Lakshmi and Suryanarayana, 1997) stressed the importance of location based studies for the elucidation of nectar and pollen sources of different honey bee species in different areas. But most of the studies are concentrated on Northern and North eastern plains of Indian Sub-continent. Exceptions were regarding the studies in Andhra Pradesh, Karnataka and Tamil Nadu. This indicates the lack of location based studies in Southern Peninsula, especially Kerala state.

2.7 POLLEN DENSITY

Quantitative melissopalynological analysis is based mainly on the interpretation and calculation of sediment-honeydew-pollen ratio in honey samples and the relationship between these co factors and their effects on honey classification. Ramos and Ferreras (2006) calculated pollen density of thirty one honey samples from Canary Islands and found that six honeys were unifloral and twenty five honey samples as multifloral. The pollen density ranged from 1042 grains/ml of honey to 24478 grains/ml with an average of 7471 grains/ml. Sa-otera *et al.* (2006) also reported that the pollen density of honey samples collected from North West Spain ranged 17834 - 53450 grains/ml of honey indicating that the honey is moderately rich in pollen.

Wide variation in pollen density of honey samples was shown in most of the studies. The quantitative analysis of honey samples collected from Tamil Nadu recorded that the pollen content ranged from 10000 – 77000 grains/ ml (Singh and Suryanarayana, 2002) whereas pollen density of honey samples from Annamalainagar ranged from 2464 to 4826 grains/ml (Hariprasad *et al.*, 2006). Pollen density of 10 honey samples from Karnataka was found to be ranged from 4000 to 80000 grains/ml (Bhargava *et al.*, 2009). Absolute pollen count of the 10 honey samples from Ahmednagar also reported the same which ranged from 48000 to 1250000 grains /ml (Kolhe *et al.*, 2011). Similarly wide variation in pollen count from honey samples of different locations of Himalaya were reported by Tiwari *et al.* (2012) in which pollen density ranged from 500-5500 grains/ml.

Studies conducted by Nair (2005) revealed that the pollen density of honey samples collected from the apiaries were far less than those from the natural hives. He pointed that this may be due to the preference of bees in apiaries to rubber honey as the apiaries were concentrated on rubber plantation. Seasonal variation in pollen density was reported by Chaturvedi and Temsunungla (2009) where the pollen density of honey samples collected during winter season (January-February) was found to be higher than that of autumn season (August-September).

2.8 POLLEN IDENTIFICATION AND CHARACTERIZATION

Earlier studies on the honey bee pollen loads made in India by Deodikar (1964); Phadke (1964); Suryanarayana and Thakar (1966) were based on the colour shade of an individual pollen loads. Later, the pollen loads were analysed on the basis of pollen morphological character by Sharma (1970); Chaturvedi (1976); Chaudhari (1977); Garg and Nair (1994). They have listed important bee foraging plants of different regions in India.

According to Barth (2004) pollen grain and their morphological characters led to the investigation of the species or the taxa of their origin, as well as the quantity that is indicative of properties. Hence pollen taxonomy is a prerequisite to compare the pollen present in honey samples with special reference to melissopalynological investigation. Pollen from the different flowers has specific shape, size and ornamentation. The apertures or furrows (colpi) in the pollen grain along with pores are a major criterion for the identification of pollen types.

Campo *et al.* (1979) described morphology of *Tabernaemontana* pollen with 3-4 colporate, isopolar, sub-olate to prolate, oval shape with curved to straight sides. Previous investigations made on honey bee pollen loads of Bhimal region (near Nainital) have elucidated 24 families of significant bee forage plants of spring season (Garg and Nair, 1994). According to Kuriakose (2007) pollen grains of the plants under the family Papilionaceae were consistently aperturate and are zonal in position and the tribe is euripalynous with colporate, colporodiate, and porate aperture. Shubharani *et al.* (2013) studied the pollen morphology of sixty eight flowering plants collected from Western Ghats of Karnataka. The results revealed that pollen morphology varied among different plant species and occurred in varying shapes and forms. Species belonging to family Asteraceae pollen are spinolous spherical in shape while that of family Malvaceae are echinate and of family Myrtaceae are colporate and prolate.

Sivaram (2006) reported about honey flora of *A. cerana, A. dorsata* and *A. florae* in Karnataka state. The study revealed that more than 340 plant species of both cultivated and wild ones are useful to honey bees as food sources. The genera like *Syzigium* sp., *Cassia* sp., *Citrus, Pongamia, Azadirachta indica* A. Juss, *Albezia, Brasssica, Areca, Cocos nucifera* L., *Guizotia* sp., *Helianthus* sp., *Lagerstroemia* sp., *Polinathus* sp., *Sapindus* sp. and *Tecoma* sp. are some of the important plants which were identified from honey samples. Nair (2005) reported that the honey and pollen

samples collected from different agro ecological situations of Thiruvananthapuram with respect to honey bee species showed the presence of 58 different species.

Tiwari *et al.* (2012) reported that most of the *A. cerana* honey sample collected from Garhwal Himalaya was multifloral which contains plant species like *Alnus nepalensis* D., *Sarcococca coriacea* (Hook.), *Chenopodium album* L., *Fagopyrum dibotrys* (D. Don) Hara, *Impatiens* sp., *Citrus* sp., plants belonging to families Asteraceae, Rosaceae and Poaceae. Pollen analysis of honey samples from Western Ghats of Karnataka revealed that *Syzygium, Acacia* sp., *Terminalia belerica, Pongamia pinnata* (L.), *Eucalyptus* sp., *A. catechu* L. and *Mangifera indica* L. were the main pollen types (Ramnath and Venkataramegowda, 2012).

2.9 DISTRIBUTION OF POLLEN TYPES IN HONEY AND POLLEN SAMPLES

Percentage distribution of pollen types in honey/pollen samples exhibit the contribution of each plant species visited by bees for nectar and pollen sources. Pollen spectrum, constructed based on these percentages, is an index of the vegetation surrounding that place at that time. From this quantitative pollen data, potential information on the plant species which acts as the most important and minor source to bees can be obtained. According to Louveaux *et al.* (1978) pollen spectra can be used for determining the geographical origin of honey since it consists of flora of that particular region from where the honey collected.

Yifeng *et al.* (2006) conducted melissopalynological analysis of *A. cerana* honey samples from Qinglan mangrove area in China. The pollen spectra constructed based on pollen percentages revealed that *Mimosa pudica* L. (89.14%) pollen was the predominant source of nectar and pollen and members of family Araceae were the important minor (3-15 %) pollen types, where as those of family Arecaceae were minor (<3 %) pollen type.

The palynological investigation of the pollen spectra of 21 honey and pollen samples of *A. cerana indica* from wild habitats of Western Ghats revealed the characteristic deciduous forest elements in abundance in the honey samples, which enabled them in labeling deciduous forest honeys as *Terminalia* honey, *Lagerstroemia* honey, *Lannea* honey, *Bombax* honey, *Olea* honey and *Elaeocarpus honey* (Nair, 2007). She also reported that diverse spectral combination of different honey bee species provides valuable information about the floral preference of different bee species with differential foraging behaviour. Apart from these information, examination of percentage of pollen content helps in ascertaining the honey quality which is adulterated or not (Chauhan and Singh, 2010).

Reddy and Reddy (2008) conducted pollen analysis of *A. cerana* honey samples collected from Khamam district of Andhra Pradesh and found that 10 samples were unifloral and one sample was multifloral type. Pollen analysis was carried out by Bhargava *et al.* (2009) on ten honey samples from Chickmagalur district of Karnataka and the predominant pollen types identified were *Coffea* sp. and *Cocos nucifera*. The pollen morphotypes recorded were *Eucalyptus globules* Esser, Lora L., *Coffea Arabica* L., *Cocos nucifera, Hevea brasiliensis, Coriandrum sativum*, *Areca catechu, Syzigium cumini* (L.), *Nerium oleander* L., plants of families, Fabaceae and Poaceae.

Cherian *et al.* (2011) reported that pollen spectra of honey sample collected from Nagpur represented the pollen types such as *Melia azadirachta* L., *Millingtonia hortensis* Linn. and members from the family Asteraceae which was absent in the pollen load. Pollen analysis of *A. cerana indica* and *A. dorsata* honey samples collected from Coorg district of Karnataka state was undertaken by Shubharani *et al.* (2012). Predominant pollen type was found to be members of family Fabaceae (>45 %), secondary dominant pollen types (16-45 %) were *Coffea* sp., *C. nucifera, Aster* sp., *Syzigium* sp., *Terminalia* sp., *Brassica* sp. and *Croton* sp. Important minor pollen

types (3-15 %) were *Ageratum conyzoides* L., *Dahlia* sp., *Eucalyptus* sp., *Eupatorium odoratum* (L.), *Dalbergia sissoo* Roxb. ex DC. and minor types(< 3%) were *Jasminum* sp., *Cinnamomum* sp., *Santalum album* L., *Sapindus laurifolia* Vaha. and *Tribulus terrestris* L.

2.10 SEASONAL VARIATION OF POLLEN TYPES

The flowering plants of several plant families are blooming at different time intervals of the year which results in fluctuation of pollen and nectar availability to foraging bees (Free, 1970). It has been observed that in Islamabad, at the full bloom season *Brassica campestris* L. and *Callistemon citrinus* (Curtis) are exploited by the Indian bees most, but as the season changes bees tend to be attracted to *Silybum marianum* (L.) Gaertn., *Taraxicum officinalis* Weber., *Eucalyptus camaldulensis* Den. and *Citrus arvensis* (Noor *et al.*, 2009).

According to Ramanujan *et al.* (1992), *Borassus flabellifer* L. was found in high frequency in honey samples of *A. cerana indica* collected during summer season from Andhra Pradesh which can be considered as an important and reliable source of nectar. Jhansi *et al.* (1992) reported that *Lagerstroemia parviflora* Roxb., *Crotalaria juncea* L. and *Schleichera oleosa* Lour. were the chief nectar sources of *A. cerana indica* in the deciduous forests areas of Vishakapatanam district during late summer season. *Borassus flabellifer*, *Hygrophila* sp., *Crotalaria juncea, Cucumis* sp., *Phylanodiflora* and *Momordica charantia* L. represents the main bee plants of the agricultural tracts of the Guntur district during summer (Jhansi *et al.*, 1994).

Analysis of 16 honey samples collected during the honey flow season from different regions of Tamil Nadu state showed that the bulk of honey supply was from plant genera *Tamarindus, Psidium guajava* L., *Hevea brasiliensis*, *Santalum album* and *Cocos nucifera* pollen which were over represented in ten honey samples and were considered as a major pollen source for bees (Singh and Suryanarayana, 2002).

Another melissopalynological study was conducted by Chaturvedi and Temsunungla (2008) on the *A. cerana* honey samples collected from Kupza village of Nagaland and they concluded that presence of monocot pollen grains are more in the autumn honey samples than the winter honey samples which was due to more anthesis of monocot during autumn season than in winter.

Nair (2005) conducted palynological investigations of honey and pollen loads from *A. cerana indica*, *A. dorsata*, *A. florae* and *Trigona iridipennis* hives at various localities of Trivandrum district. The results revealed the presence of pollen types of *Bombax ceiba*, *Dillenia pentagyna* Roxb., *Aporosa lindliyana* (Wt.) Bail., *C. nucifera* and *Peltophorum pterocarpum* (DC.) Backer during the honey flow seasons of the tropical ecosystem in Kerala.

Mukhopadhyay (2007) reported that *Rosa* sp., *Ageratum conyzoides* L., *Rubus ellipticus* Smith., *Prunus* sp., *Millettia pulchra* Kurz., *Trifolium repens* L., *Primula* sp., *Schima wallichii* (DC.) Korth., *Fragaria* sp., *Potentilla* sp. was found to be the major source of *A. cerana* during summer and *A. conyzoides*, *S. wallichii*, *Sedum multicaule* Wall. Ex Lindl. (during autumn) and *Aristolochia* sp., *Rosa* sp. during winter season eastern Himalayan part of West Bengal.

Attiri (2010) also conducted the melissopalynological studies of different honey samples of *A. cerana indica* collected from different blocks of Chamba district during autumn and early winter. Pollen analysis of honey samples revealed the following predominant sporomorphs, *Fagopyrum* sp., *Plectranthus* sp., *Saliva* sp. *Rosaceae* sp. and *Bauhinia sp.* Chakraborti and Bhattacharya (2011) reported that pollen spectra revealed the contribution of each pollen types and it varies with seasons. Pollen of *Syzygium* sp. and *Cassia* sp. was found to be the predominant pollen grains which showed the importance of wild plant where as *Helianthus* sp. was

found to be predominant during midsummer season, which is an important cultivated plant.

Palynological study of honey collected from Kamrup reserve forests of Assam during different seasons reported that honey bee preferred *Coriandrum sativum* and *Brassica nigra* (L.) during winter season followed by the gradual increase in secondary pollen types namely *Coriandrum, Eupatorium, Mimosa, Melia, Syzygium, Butea monosperma* Lam. Kuntze etc. during the successive season (Bera *et al.*, 2007).

Dixit *et al.* (2011) analysed eighteen honey samples procured from summer and winter season from Goalpara district of Assam and the results showed that honey bee preferred the plant *Salmalia malabaricum* (DC.) in summer and *Brassica campestris* in winter season.

2.8. CLUSTER ANALYSIS

Valle *et al.* (2007) reported that honey samples of five regions obtained after the cluster analysis in different districts from Argentina showed difference in pollen depending upon the regions and the natural variability of honey samples makes it very difficult to define borders between the different honey regions. It was also reported that honey samples from region 1, 3 and 4 were unifloral while those from region 2 and 5 were multifloral. Similarly Silva *et al.*, 2012 also reported that similarity analysis among the honey samples from 5 regions of Brazil resulted in grouping the samples into region I, II and II whereas region II was with lowest pollen diversity and region III was with highest pollen diversity.

MATERIALS AND METHODS

3. MATERIALS AND METHODS

The present study on 'Melissopalynology of Indian honey bee (*Apis cerana indica* Fab.) apiaries in southern Kerala' was conducted at AICRP on honey bees and pollinators, Department of Agricultural Entomology, College of Agriculture, Vellayani and based on the apiaries of bee keepers (Indian bee) from Thiruvananthapuram and Kollam during 2011-13.

3.1 SELECTION OF APIARIES

The apiaries were selected from two districts of southern Kerala, Thiruvananthapuram and Kollam (Fig. 1 & 2). Among the three physiographic zones, namely the highland, midland and lowland (Nair *et al.*, 2007), bee keeping is prominent in midland and highland and therefore the present study was limited to midland and upland regions only. Number of apiaries present in midland is more as compared to upland, hence ten apiaries from midland and five apiaries from upland were selected for this study. The apiaries which had minimum ten hives were selected (Plate. 1) for the study. The details of the locations from which the apiaries were selected are given in Table 1.

Table 1. Physiographic details of various areas for collection of honey and pollen load samples of *A. cerana indica* in Thiruvananthapuram and Kollam districts

Sl. No.	District	Midland/ Upland	Location	Latitude	Longitude
1.	Thiruvananthapuram	Midland	Kodankara (ML-1)	8^0 21 N	77^0 6 E
2.	Thiruvananthapuram	Midland	Malayam (ML-2)	$8^0$28N	77^0 1 E
3.	Thiruvananthapuram	Midland	Kulappada (ML-3)	$8^0$35 N	77^0 3 E
4.	Thiruvananthapuram	Midland	Pullampara (ML-4)	8^0 41 N	76^0 57 E
5.	Thiruvananthapuram	Midland	Kilimanoor (ML-5)	8^0 46 N	76^0 53 E

6.	Kollam	Midland	Kadakkal (ML-6)	8^0 49 N	76^0 55 E
7.	Kollam	Midland	Veliyam (ML-7)	8^0 54 N	76^0 46 E
8.	Kollam	Midland	Ummannoor (ML-8)	8^0 56 N	76^0 49 E
9.	Kollam	Midland	Karikkam (ML-9)	8^0 57 N	76^0 50 E
10.	Kollam	Midland	Pathanapuram (ML-10)	9^0 5 N	76^0 51 E
11.	Thiruvananthapuram	Upland	Aryanad (UL-1)	8^0 34 N	77^0 5 E
12.	Thiruvananthapuram	Upland	Tholicode (UL-2)	8^0 38 N	77^0 3 E
13.	Thiruvananthapuram	Upland	Chennanpara (UL-3)	8^0 40 N	77^0 5 E
14.	Kollam	Upland	Kulathupuzha (UL-4)	8^0 54 N	77^0 3 E
15.	Kollam	Upland	Rosemala (UL-5)	8^0 55 N	77^0 10 E

3.2 SAMPLING

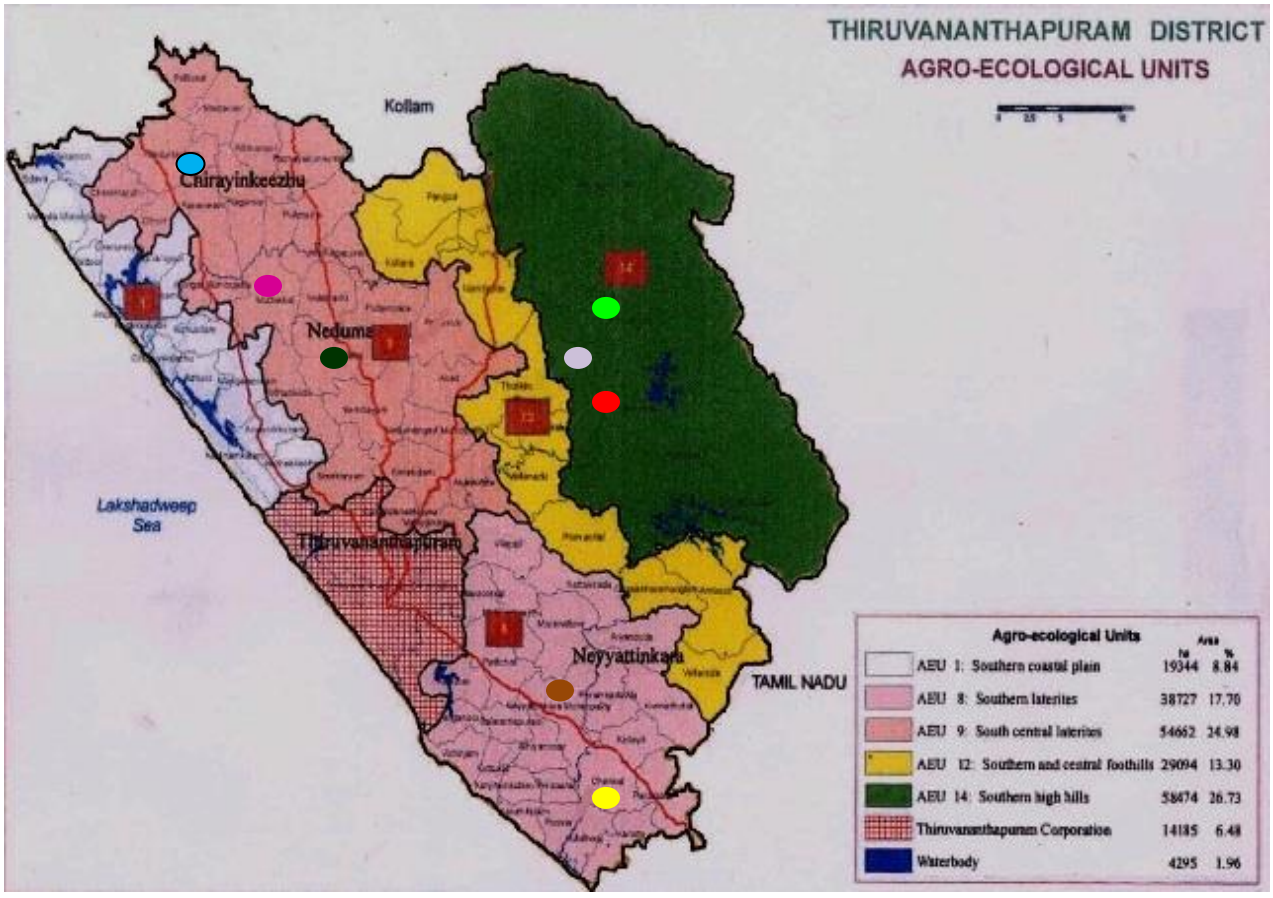
Three samples each of honey and pollen load were collected from the selected apiaries during three seasons namely; honey flow season (January to April), dearth season (June to September) and brood rearing season (October to December) which are prevalent seasons in Kerala. Three hives were selected randomly from apiaries. Samples of honey and pollen load were taken from a single hive. Altogether 270 samples were collected.

3.2.1 Collection of honey samples

The honey sample (3ml) was collected directly from the comb cells with the aid of graduated filler to glass vials having 5 ml capacity (Plate 2.), labeled and numbered for further investigation. All the honey samples were raw and unprocessed.

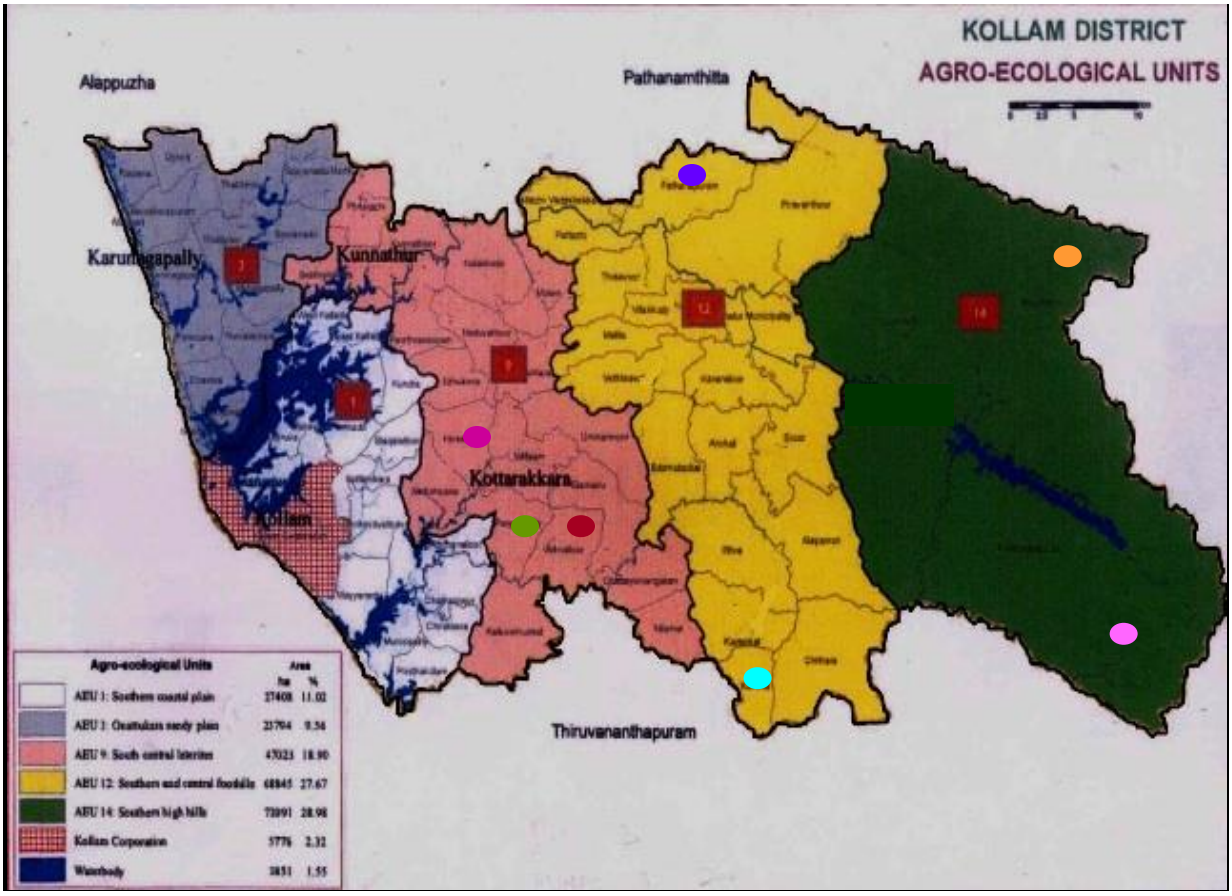
3.2.2 Collection of pollen load

Pollen loads were collected from the pollen storage cells of the honey comb (Plate 2.). Selection of pollen loads were made by considering colour and position in



Kilimanoor
Malayam
Chennanpara
Pullampara
Kodankara
Aryanad
Kulappada
Tholicode

Fig. 1 Locations selected from Thiruvananthapuram district



Karikkam
Pathanapuram
Rosemala
Veliyam
Kadakkal
Kulathupuzha
Ummannoor

Fig. 2 Locations selected from Kollam district

Plate 1. Indian bee apiaries at the locations, Malayam and Kodankara

Plate 2.Collection of honey samples and pollen loads

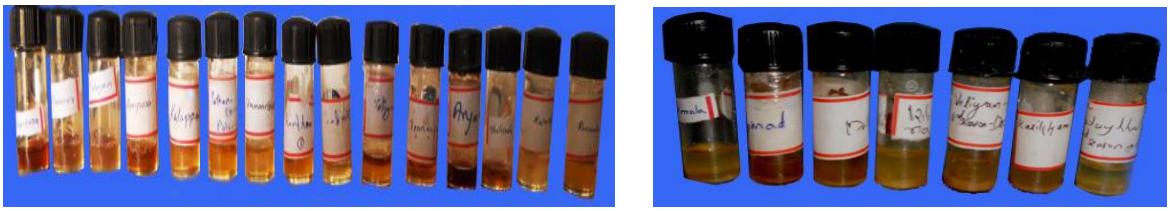


Plate 3. Collected honey and pollen samples

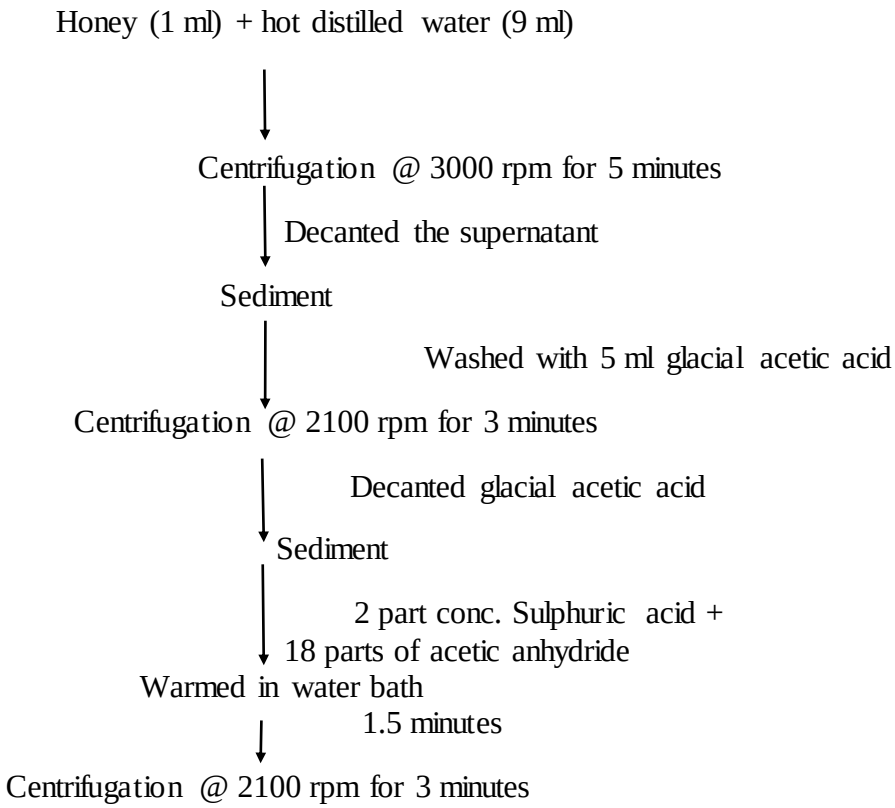
the comb cells. Samples were collected with the help of sterilized forceps and the collected pollen samples were fixed in 70 per cent ethyl alcohol, labeled for further studies.

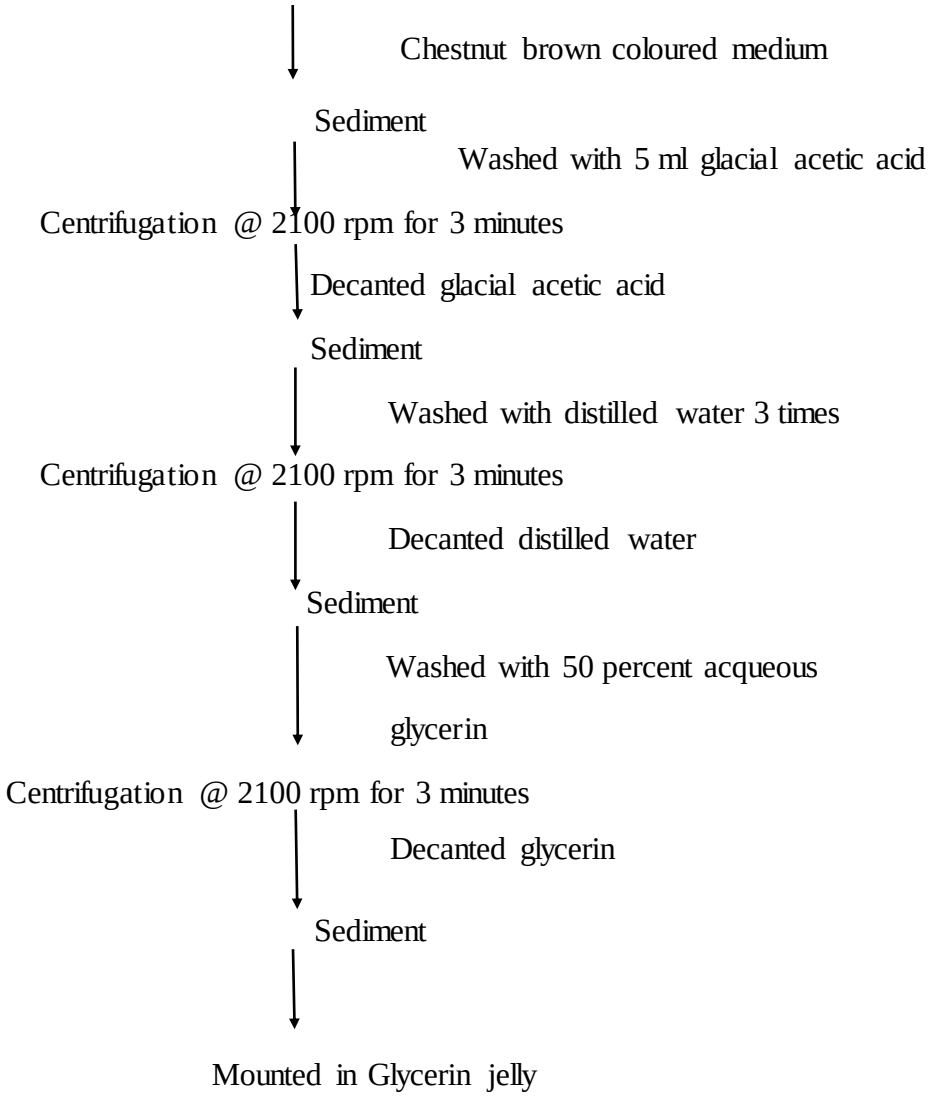
3.3 LABORATORY ANALYSIS

The collected honey and pollen load samples (Plate 3.) were stored in room temperature and was subjected to pollen analysis (acetolysis and pollen slide preparation). Acetolysis was done mainly to remove the fine cellulose materials present in pollen grains thus providing better visibility for palynomorphs based on the procedures of Erdtman (1960) with a 5 minute decrease in heating time.

3.3.1 Laboratory analysis of honey samples

Procedure for analysis of honey sample is outlined below:





3.3.2 Laboratory analysis of pollen load

The pollen sample suspended in ethyl alcohol (70%) (1 ml) was taken in a centrifuge tube and centrifuged at 2100 rpm for 3 minutes. The supernatant was decanted off. The samples were subjected to acetolysis by following the same procedure as that of 3.3.1.

3.3.3 Preparation of permanent slides

Permanent slides of pollen were prepared by mounting the pollen sediment on glycerin jelly. Reagents required for preparation of mounting medium are the following:

Gelatin - 5 g
Water - 30 ml
Glycerine - 35 ml
Phenol crystals - 3g

Gelatin was dissolved in boiled water (100^0C) with continuous stirring. The other components were added to it and then filtered hot and was kept in a closed bottle. Phenol crystals were added as a preservative to avoid growth of fungal elements. For the preparation of permanent slides, pollen sediment was taken on a pallet of glycerin jelly and transferred to the centre of the slides of 75 x 25 mm size having 1.35 mm thickness. After being warmed slightly, the melted jelly with pollen sediment was covered by 18 mm size cover glass. The cover glass was later sealed with paraffin wax.

3.4 DETERMINATION OF POLLEN DENSITY

The total number of pollen grains per ml in each honey sample was recorded. Observation on the pollen count from pollen slides was taken under the magnification of 400 X using the compound microscope. From each sample four slides were prepared. The number of pollen grains from five fields of each slide was counted. Pollen density was estimated by multiplying the pollen count from five fields with conversion factor (1.94). Data was subjected to ANOVA after square root transformation (Sahu and Das, 2009) to compare between seasons, between locations,

between upland and midland and locations within each season. Completely Randomised Design was followed for ANOVA.

3.5 POLLEN CHARACTERIZATION

The pollen grains of different types obtained from each sample were identified based on the reference slides and the characters were described based on the terminology used in 'the glossary of spore terminology'. The pollen grain was described based on their characteristics such as position of aperture, types of aperture and exine ornamentation as detailed in Table 2.

Table 2. Morphological characters of pollen grains (Nair, 1964)

Pollen characters	Particulars			
Position of aperture	Polar (Proximal)	Polar (Distal)	Zonal	Global
Aperture types	Monolete Trilete Hilate	Sulcate Ulcerate	Colpate Porate	Rugate Forate
Exine ornamentation	Pilate, Areolate, Foveolate, Retipilate, Scrobiculate, Fossulate, Rugulate, Striate, Psilate, Granulose, Spinose, Baculate, Clavate, Gemmate, Verrucate, Punctate			

3.6 MICROPHOTOGRAPHY

The microphotographs of pollen grains were taken using the compound microscope with image analyzer at 1000 X magnification.

3.7 POLLEN TYPES AND PERCENTAGE OF POLLEN TYPES

Percentage of particular pollen type in each sample from different locations was determined using the formula

Percentage of pollen types in each sample =

$$\frac{\text{Number of pollen grains of a particular pollen type}}{\text{Total number of pollen grains}} \times 100$$

3.7.1 Estimation of pollen class frequency types

From the recorded pollen count of each honey and pollen sample, the pollen types were classified according to the number of pollen grains in each sample (Louveaux *et al.,* 1978) which includes:

Predominant pollen - more than 45 per cent of the pollen grains counted
Secondary pollen - 16-45 per cent
Important minor pollen - 3-15 per cent
Minor pollen - less than 3 per cent

Pollen spectra were constructed based on these percentages. From this quantitative pollen data, potential information on the plant species which acts as the most important and minor source to bees was obtained.

3.7.2 Frequency of occurrence of pollen types

From the pollen count of each honey and pollen sample, frequency of occurrence of pollen types was estimated and was categorized into different classes.

The frequency classes (Louveaux *et al.*, 1978) include:

Very frequent – more than 50 per cent
Frequent - 20-50 per cent
Infrequent - 10 – 20 per cent
Rare - less than 10 per cent

3.8 SEASONAL VARIATION

The occurrence of pollen types during the three seasons was recorded based on the data obtained after the analysis of samples collected from different apiaries.

3.9 CORRELATION COEFFICIENT BETWEEN POLLEN DENSITY AND POLLEN TYPES

The correlation coefficient between pollen types (x) and pollen density (y) was calculated (Pearson's Correlation Coefficient) (Gupta and Kapoor, 2001) using the formula:

$$r_{yx} = \frac{Cov\,(xy)}{SDx \times SDy}$$

where as,

Cov is the Covariance of x and y

SD_x is the standard deviation of x

SD_y is the standard deviation of y

3.10 CLUSTER ANALYSIS

The palynological data recorded for individual samples from different floristic locations was subjected to cluster analysis (Sahu and Das, 2009) to obtain similarity index between locations. If a particular pollen type is present in the honey sample from a location, it is coded as (1), and the absence is coded as (0). The two-way binary data matrix of locations x pollen types was subjected to cluster analysis to determine similarity matrix and group the locations within each cluster and to generate the dendrogram.

RESULTS

4. RESULTS

The salient results of the study on "Melissopalynology of Indian honey bee (*Apis cerana indica* Fab.) apiaries in southern Kerala" is presented below under the following heads.

4.1 POLLEN DENSITY

Studies on the pollen density (pollen grains per ml of honey sample) recorded over the three seasons from fifteen locations of southern Kerala is given in Table 3.

4.1.1 Pollen density between locations during the dearth season

There was significant difference in mean pollen density of locations within the seasons. In dearth season, maximum mean pollen density was recorded from Kulathupuzha (965. 32 grains/ml) which was on par with Pullampara (803. 42 grains/ml) while the lowest mean pollen density was recorded in Ummannoor (28.09 grains/ml). Both these locations Kulathupuzha and Pullampara showed high significant difference in mean pollen density with the remaining locations. Similarly Pathanapuram (493. 64 grains/ml) and Kulappada (455. 19 grains/ml) were on par but highly significant from all other locations. Rosemala (317. 39 grains/ml), Kilimanoor (242. 93 grains/ml) and Karikkam (218. 39 grains/ml) were on par with each other and was significantly different from all the remaining locations. There was no significant difference in mean pollen density between the locations: Chennanpara (133. 69 grains/ml), Kodankara (100.46 grains/ml), Aryanad (57. 45 grains/ml), Tholicode (47. 96 grains/ml), Kadakkal (48. 39 grains/ml), Malayam (43. 24 grains/ml), Veliyam (29. 54 grains/ml) and Ummannoor (28. 09 grains/ml).

4.1. 2 Pollen density between locations during the brood rearing season

There was significant difference in mean pollen density between the locations during brood rearing season. Maximum mean pollen density during brood

Table 3. Location wise pollen density (mean no. of pollen grains/ml) of honey samples over three seasons.

Locations	Dearth season	Brood rearing season	Honey flow season	Mean
Kodankara	100.46 (10.02)	1957.83 (44.25)	1845.37 (42.96)	1301.22 (32.409)
Malayam	43.24 (6.56)	210.49 (14.51)	405.14 (20.13)	219.62 (13.737)
Kulappada	455.19 (21.34)	803.18 (28.34)	254.68 (15.96)	504.35 (21.878)
Pullampara	803.42 (28.35)	364.46 (19.09)	776.79 (27.87)	648.22 (25.102)
Kilimanoor	242.93 (15.59)	450.63 (21.23)	83.55 (9.14)	259.04 (15.318)
Kadakkal	48.39 (6.96)	216.63 (14.72)	427.60 (20.68)	230.87 (14.118)
Veliyam	29.54 (5.43)	71.38 (8.45)	296.32 (17.21)	132.41 (10.366)
Ummannoor	28.09 (5.30)	806.55 (28.40)	508.79 (22.56)	447.81 (18.752)
Karikkam	218.39 (14.78)	1561.64 (39.52)	772.10 (27.79)	850.71 (27.361)
Pathanapuram	493.64 (22.22)	1690.44 (41.12)	2132.23 (46.18)	1438.77 (36.503)
Mean(mid land)	246.32 (13.66)	813.24 (25.96)	750.26 (25.05)	-
Aryanad	57.45 (7.58)	133.64 (11.56)	371.69 (19.29)	187.59 (12.806)
Tholicode	47.96 (6.93)	589.72 (24.28)	314.29 (17.73)	317.32 (16.312)
Chennanpara	133.69 (11.56)	285.05 (16.88)	311.94 (17.66)	243.56 (15.369)
Kulathupuzha	965.32 (31.07)	565.21 (23.77)	166.58 (12.91)	565.70 (22.583)
Rosemala	317.39 (17.82)	750.96 (27.40)	299.82 (17.02)	456.06 (20.748)
Mean (upland)	304.36 (14.99)	774.86 (20.78)	292.86 (16.92)	-
Mean (seasons)	294.69 (14.10)	794.05 (24.24)	521.56 (22.34)	-
Midland Vs Upland	S*	S**	S**	-
CD $_{(0.05)}$ for M x U	1.15	1.35	1.12	-
CD $_{(0.05)}$ for Locations	1.91			
CD $_{(0.05)}$ for Seasons	0.84			
CD $_{(0.05)}$ Locations within seasons	3.50			

Figures in parentheses are values after square root transformation.

rearing season was recorded from the location, Kodankara (1957. 83 grains/ml) which was on par with Pathanapuram (1690. 44 grains/ml). Lowest mean pollen density was recorded in honey samples from Veliyam (71. 38 grains/ml). Karikkam (1561. 64 grains./ml) was significantly different from all other locations. Mean pollen density of Ummannoor (806. 55 grains/ml), Kulappada (803. 18 grains/ml) and Rosemala (750. 96 grains/ml) were on par while Rosemala was also found to be on par with Tholicode (589. 72 grains/ml). Mean pollen density of Tholicode (589. 72 grains/ml) was also on par with Kulathupuzha (565. 21 grains/ml) and Kilimanoor (450. 63 grains/ml). Both Kilimanoor (450. 63 grains/ml) and Pullampara (364. 46 grains/ml) were on par but Kilimanoor was found to be significantly different with Chennanpara (285. 05 grains/ml) and Malayam (210. 49 grains/ml) but Kadakkal (216. 63 grains/ml) was on par with Pullampara. Even though Pullampara was on par with Chennanpara, it was significantly different from Kadakkal and Malayam.

4.1.3 Pollen density between locations during the honey flow season

Mean pollen density in honey flow season also recorded significant variations between the locations. Maximum mean pollen density in honey flow season was recorded in Pathanapuram (2132. 23 grains/ml) and Kodankara (1845. 37 grains/ml) which were on par. The location Pullampara with a mean pollen density of 776. 79 grains/ml was significantly different from all other locations. The lowest mean pollen density was recorded in Kilimanoor (83. 55 grains/ml). Pathanapuram (2132. 23 grains/ml) and Kodankara (1845. 37 grains/ml) showed no difference in mean pollen density to each other and they differed significantly from all other locations. Pullampara (776. 79 grains/ml) and Karikkam (772. 10 grains/ml) were on par and significantly differed from all other locations. No significant difference was recorded from the locations, Ummannoor, Kadakkal, Malayam, Aryanad, Tholicode, Chennanpara, Veliyam, Rosemala Kulappada and Kulathupuzha with a mean pollen density ranging from 166. 58 to 508.79 grains/ml.

4.1.4 Pollen density between the locations of midland and upland regions

Considering the midland and upland locations, significant difference in mean pollen density was recorded between the locations and also within the locations. Significantly high mean pollen density was obtained in the location Pathanapuram (1438.77 grains/ml) followed by Kodankara (1301.22 grains/ml), Karikkam (850.71 grains/ml) and Pullampara (648.22 grains/ml) while the lowest mean pollen density was recorded in Veliyam (132.41 grains/ml). The mean pollen density of the locations: Kulathupuzha, Kulappada and Rosemala was 565.70, 504.35, 456.06 grains/ml respectively which were on par. Ummannoor was significantly different from all other locations with a mean pollen density of 447.81 grains/ml while the locations, Tholicode (317.32 grains/ml), Chennanpara (243.56 grains/ml) and Kilimanoor (259.04 grains/ml) were on par.

4.1.5 Pollen density between midland and upland physiographic regions

Studies on the pollen density recorded over the three seasons from fifteen locations of southern Kerala revealed that there was significant difference between the midland and upland physiographic regions. Maximum mean pollen density was recorded in midland during the brood rearing season (813.24 grains/ml) and honey flow season (750.26 grains/ml) whereas maximum mean pollen density was recorded in dearth season (304.36 grains/ml).

4.1.6 Pollen density of locations between the three seasons

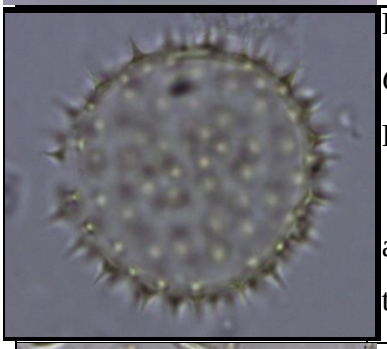
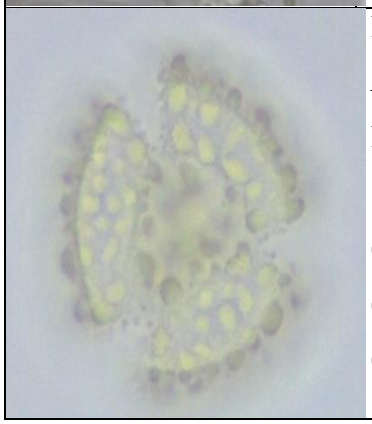
There was significant difference in mean pollen density during the three seasons. Maximum mean pollen density was observed in brood rearing season (794.05 grains/ml) followed by honey flow season (521.56 grains/ml) and dearth season (294.69 grains/ml).

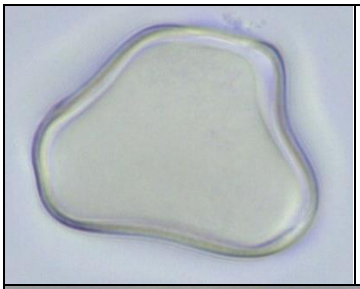
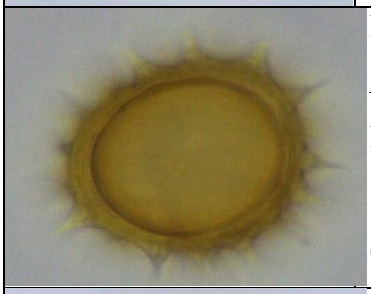
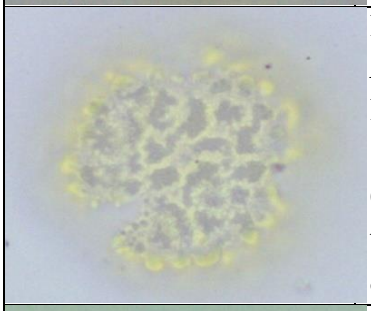
4.2 POLLEN CHARACTERIZATION

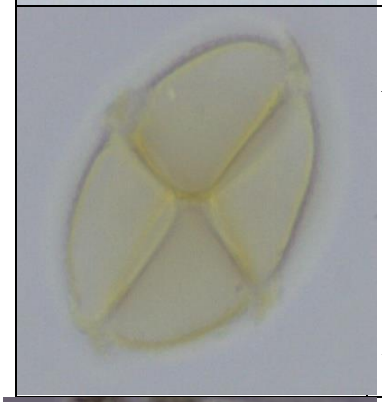
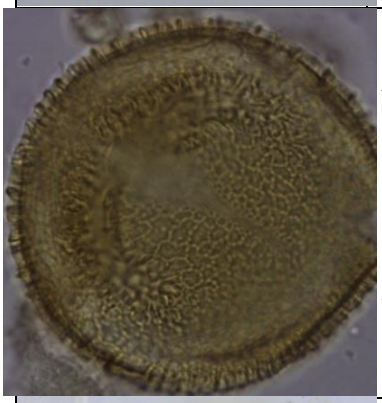
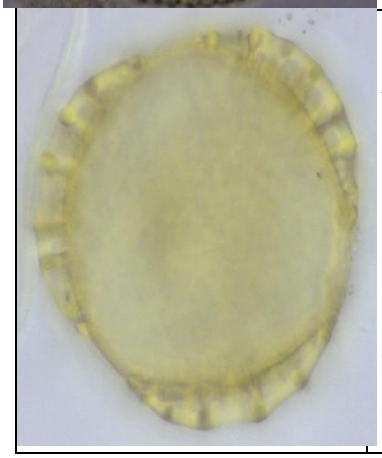
Pollen analysis of honey and pollen samples resulted in the recognition of 69 pollen types over all the three seasons. The pollen grains obtained from the samples by analysis were identified to generic, species and family level. The identified pollen types and its characteristics are detailed below in Fig. 3.

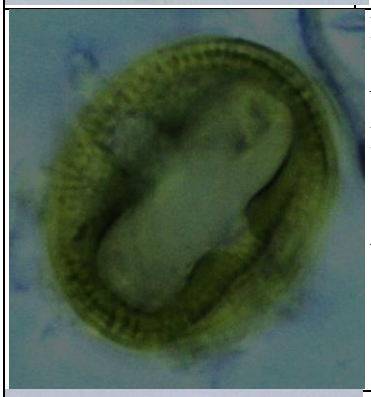
Fig. 3. Morphological description of individual palynomorphs with photos

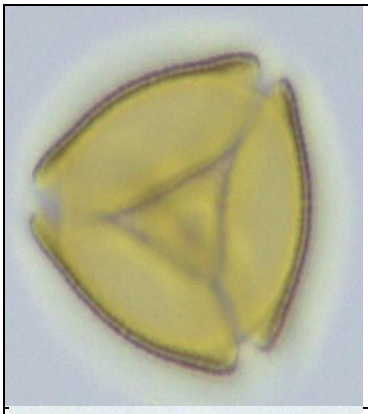
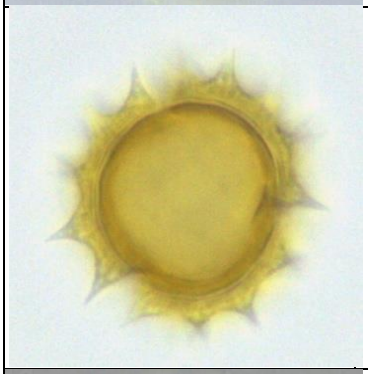
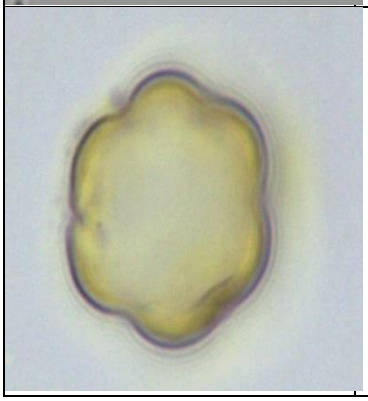
Pollen type	Pollen morphology
	P-1 *Acacia auriculiformis* A. Cunn. (Australian wattle) Fam. Mimosaceae Pollen grains in polyads, polyads 12-celled, spherical, polar outline circular, equatorial outline elliptic-obtuse-plane, polypantoporate, pori circular, ektexine psilate.
	P-2 *Bombax* sp. Fam. Bombacaceae Pollen grains in monads, tricolporate pollen with wide colpus, polar and equatorial outline circular, psilate pollen grain with thick ektexine and endexine.
	P-3 *Borassus flabellifer* Roxb. (Palmyra palm) Fam. Mimosaceae Pollen grains in monads**,** tricolpate pollen grain, ektexine foveolate and puntum were present in between and ektexine almost as thick as endexine.

	P-4 *Cocos nucifera* L. (Coconut) Fam. Arecaceae Pollen grains in monads, pollen monocolpate, colpus with rounded ends, ektexine faintly reticulate ektexine is almost as thick as endexine, endexine smooth.
	P-5 *Colocasia esculenta* L. (Taro) Fam. Araceae Pollen grains in monads, spheroidal in equatorial and polar view, ektexine echinate, spines tapering at the tips.
	P-6 *Cullenia exarillata* Robyns. (Wild durian) Fam. Bombacaceae Pollen grains in monads, polar outline circular and equatorial outline elliptic, tricolporate with very short ectoaperture, ektexine thicker than endexine with granules scattered on the surface.
	P-7 *Dillenia pentagyna* Roxb. (Dillenia) Fam. Dilleniaceae Pollen grains in monads, polar outline circular and equatorial outline elliptic, trizonocolpate, colpi narrowly elliptic, wide at equator, sides tapering and tips acute, ektexine verrucate.

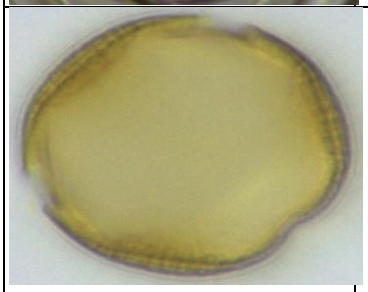
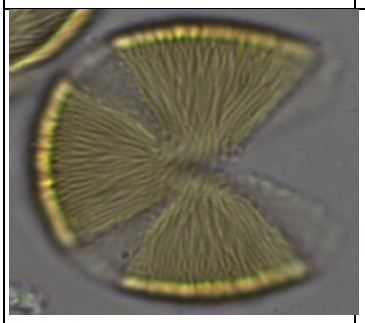
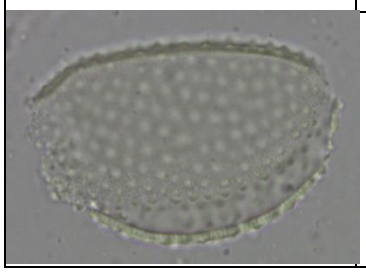
	P-8 *Elaeis guineensis* Jacq. (Oil palm) Fam. Palmae Pollen grains in monads, polar outline triangular-obtuse-concave and equatorial outline elliptic, trizonoporate, psilate pollen grain.
	P-9 *Hibiscus rosasinensis* Linn. (Shoe flower) Fam. Malvaceae Pollen grains in monads, polar and equatorial outline circular, pantoporate and echinate pollen type.
	P-10 *Hopea ponga* dennst. (Elapongu) Fam. Dipterocarpaceae Pollen grains in monads, polar outline triangular; obtuse-convex and equatorial outline elliptic, trizonocolpate with wide colpus, sides tapering towards apocolpia to acute tips, ektexine reticulate.
	P-11 *Impatiens* sp. Fam. Balsaminaceae Pollen grains in monads, polar outline quadrangular; obtuse-convex and equatorial outline elliptic, tetrazonoporate, ektexine reticulate with homobrochi.
	P-12 *Mangifera indica* L. (Mango) Fam. Anacardiaceae Pollen grains in monads, prolate in equatorial view, tricolporate grain, ektexine reticulate and thicker than endexine.

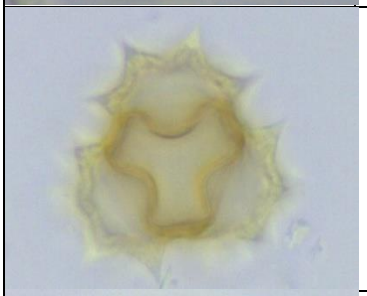
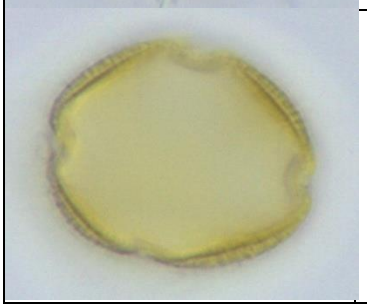
	P-13 *Mimosa pudica* L. (Sensitive plant) Fam. Mimosaceae Pollen grains in tetrads, tetrads spherical, polar outline circular, equatorial outline is quadrangular-obtuse-plane, tetrapantoporate, pori circular, ektexine psilate.
	P-14 *Mimosa invisa* (Gaint sensitive plant) Fam. Mimosaceae Pollen grains in tetrads, tetrads elliptic, prolate spheroidal, exine surface tuberculated, polar outline circular and equatorial outline quadrangular-obtuse-plane, tetrapantoporate, pori circular, psilate pollen.
	P-15 *Passiflora foetida* (Passion fruit) Fam. Passifloraceae Pollen grains in monads, polar outline triangular-obtuse-convex and equatorial outline elliptic, trizonocolporate, ektexine reticulate, reticulum lumen large.
	P-16 *Peltophorum pterocarpum* (Golden flame) Fam. Fabaceae Pollen grains in monads, polar outline circular and equatorial outline circular to elliptic, trizonocolporate, colpi narrowly elliptic, sides tapering, tips acute, margins thick, ora circular to oblate, ektexine reticulate, endexine smooth, lumina irregularly polygonal to isodiametric.

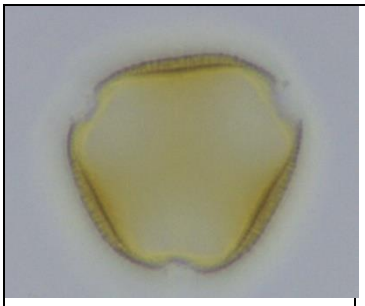
	P-17 *Poeciloneuron paucifolium* Bedd. Fam. Clusiaceae Pollen grains in monads, polar and equatorial outline circular, tricolporate pollen, ektexine fainltly microreticulate, ektexine thicker than endexine.
	P-18 *Psidium guajava* L. (Guava) Fam. Myrtaceae Pollen grains in monads, triangular in polar view, oblate spheroidal in equatorial view, trizonocolporate, oralalongate, psilate pollen.
	P-19 *Rosa* sp. Fam. Rosaceae Pollen grains in monads, elliptic in equatorial view, trizonocolporate, ektexine reticulate.
	P-20 *Spilanthus calva* D. C. (Tooth ache plant) Fam. Asteraceae Pollen grains in monads, polar and equatorial outlines circular, trizonocolporate with elliptic colpi, sides abruptly tapering towards the poles, tips acute, ora narrowly elliptic, ektexine echinate.

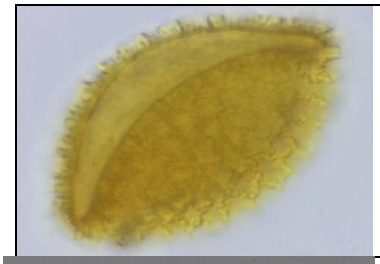
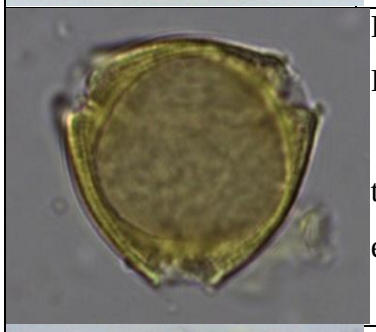
	P-21 *Schleichera oleosa* (Lour.) Oken. (Kusum tree) Fam. Sapindaceae Pollen grains in monads, polar outline triangular-acute convex to circular, equatorial outline elliptic, trizono para syncolporoidate, parasyncolpia triangular acute-concave, ektexine reticulate.
	P-22 *Tagetus erecta* L. (Big marigold) Fam. Asteraceae Pollen grains in monads, equatorial outline circular, trizonocolporate, ektexine echinate.
	P-23 *Tabernaemontana gamblei* (Crape jasmine) Fam. Apocynaceae Pollen grains in monads, tetrazonocolporate, prolate in polar view and circular in equatorial view, ektexine thicker and bulged outward on the four sides, psilate pollen grain and endexine smooth.
	P-24 *Terminalia paniculata* Roth. (Maruthu) Fam. Melastomaceae Pollen grains in monads, polar outline triangular-obtuse-convex and equatorial outline elliptic, trizonocolporate with pseudocolpi in between, ektexine psilate and almost as thick as endexine.

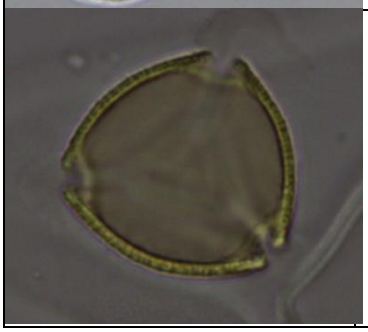
	P-25 *Tridax procumbens* L. (Coat buttons) Fam. . Asteraceae Pollen grains in monads, polar and equatorial outlines circular, tetrazonocolporate, colpi elliptic, sides abruptly tapering towards the poles and sides tapering to acute tips, ektexine echinate, echinae narrowly triangular in outline.
	P-26 *Trichilia connaroides* Wight & Arm. (Peelimaram) Fam. Meliaceae Pollen grains in monad, circular in equatorial view and quadrungular in polar view, tetrazonocolporate, ektexine granulated, bulged and thicker than endexine.
	P-27 *Muntingia calabura* L. (Bird cherry) Fam. Rosaceae Pollen grains in monads, triporate pollen, pores very small in size, ektexine granulated, endexine smooth.
	P-28 *Antigonon sp.* Fam. Polygonaceae Pollen grains in monads, tricolpate pollen, pollen grains having reticulate ornamentation with thicker ektexine than endexine.

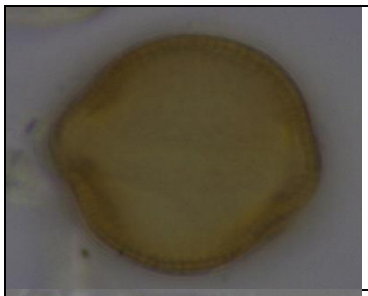
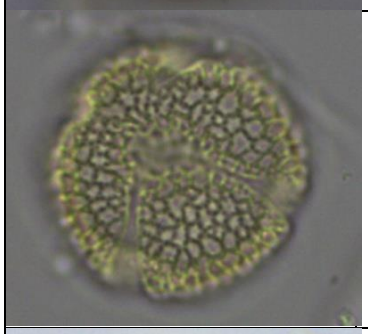
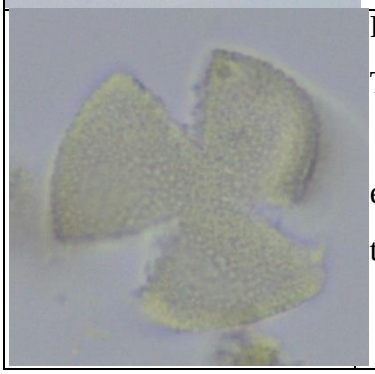
	P-29 Fam. Acanthaceae Pollen in monads, trizonocolporate, equatorial outline circular, ektexine reticulate.
	P-30 Fam. Amaranthaceae Pollen in monads, circular in equatorial and polar view, tricoplorate grain, psilate pollen grain.
	P-31 Fam. Anacardiaceae Pollen grains in monads, trizonocolporate with wide colpus, ektexine faintly reticulate and thicker than endexine.
	P-32 Fam. Anacardiaceae Pollen grains in monads, tricolpate grain, colpus wide and tapering inward, ektexine with striate ornamentation, ektexine is almost as thick as endexine.
	P-33 Fam. Arecaceae Pollen grains in monads, ektexine foveolate.

	P-34 Fam. Arecaceae Pollen grains in monads, monocolpate, psilate grain with thick ektexine.
	P-35 Fam. Asteraceae Pollen grains in monads, polar outline triangular - obtuse – convex, trizonocolporate, grains lacunate and sculpturing echinate, ektexine foveolated with fenestrate ornamentation and thicker than endexine.
	P-36 Fam. Asteraceae Pollen grains in monads, equatorial and polar outline circular. Trizonocolporate. Echinolophate grain with cavea.
	P-37 Fam. Asteraceae Pollen grains in monads, equatorial and polar outline circular, trizonocolporate grain, echinolophate grain.
	P-38 Fam. Brassicaceae Pollen grains in monads, equatorial outline prolate spheroidal, tetrazonocolpate, ektexine faintly reticulate.

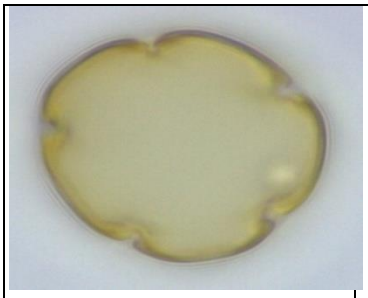
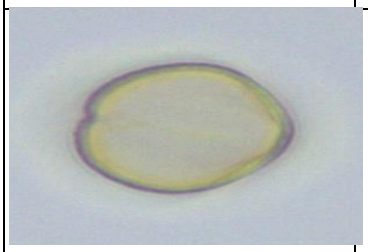
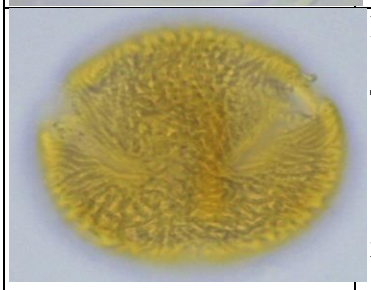
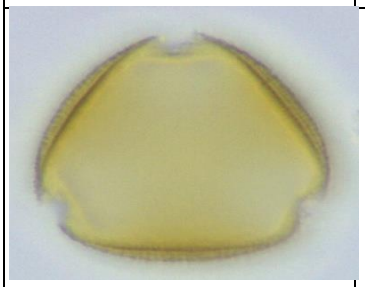
	P-39 Fam. Celastraceae Pollen grains in monads, polar and equatorial outline circular, tricolporate with pantoapertural thinning, ektexine microreticulate.
	P-40 Fam. Convolvulaceae Pollen grains in monads, oblate in polar and equatorial outline. 8-zonocolporate, ektexine granulated with reticulate ornamentation.
	P-41 Fam. Convolvulaceae Pollen grains in monads, pentazonocolporate, ektexine with rugulate ornamentation.
	P-42 Fam. Cyperaceae Pollen grains in monads, monoporate, polar out line ellipsoidal, equatorial outline convex triangular, ektexine and endexine smooth.
	P-43 Fam. Elaecarpaceae Pollen grains in monads, polar outline circular; Equatorial outline elliptic, trizonocolporoidate, colpi faint and narrowly elliptic, sides tapering, exine psilate.

	P-44 Fam. Euphorbiaceae Pollen grains in monads, monocolpate pollen, colpus with pointed ends, ektexine regulate.
	P-45 Fam. Fabaceae Pollen grains in monads, tricolporate grain, colpus short, polar and equatorial outline circular, psilate pollen with thick ektexine.
	P-46 Fam. Fabaceae Pollen grains in monads, polar outline circular, equatorial outline prolate, trizonoporate pollen, pores periporate, psilate pollen.
	P-47 Fam. Flacourtiaceae Pollen in monads, trizonocolporate, colpus slightly tapering, equatorial outline circular, ektexine thicker than endexine, psilate pollen grain.
	P-48 Fam. Lamiaceae Pollen grains in monads, polar and equatorial outline circular, tricolporate grain, colpus short, psilate pollen with thick ektexine.

	P-49 Fam. Meliaceae Pollen grains in monads, tetrazonocolporate, equatorial and polar outline circular, psilate grain with thick ektexine.
	P-50 Fam. Myrsinaceae Pollen grains in monads, trizonocolporate, ektexine faintly reticulate with thick ektexine than endexine.
	P-51 Fam. Myrsinaceae Pollen in monads, tetrazonocolporate, quadrangular in polar view and circular in equatorial view, psilate pollen grain
	P-52 Fam. Myrtaceae Pollen grains in monads, polar outline triangular, trizonocolporate, psilate pollen grain with thick ectexenie.
	P-53 Fam. Myrtaceae Pollen grains in monads, circular in equatorial view, trizonocolporate grain, ektexine microreticulate.

	P-54 Fam. Oleaceae Pollen in monads, trizonocolporate, ektexine reticulate with heterobrochi, endexine smooth.
	P-55 Fam. Oleaceae Pollen grains in monads, tetrazonocolporate, ektexine reticulate with large lumen.
	P-56 Fam. Sapindaceae Pollen grains in monads, trizonocolporate, psilate grain.
	P-57 Fam. Sapindaceae Pollen grains in monads, trizonocolporate, psilate grain.
	P-58 Type 1 Pollen grains in monads, polar outline and equatorial outline circular, trizonocolporate, aperture triangular in shape, ektexine grano rugulate.

	P-59 Type 2 Pollen Grains in monads, polar outline circular, 5-zono colporate, sculpturing microreticulate.
	P-60 Type 3 Pollen grains in monads. Equatorial and polar outline spheroidal, trizonocolporate grain, colpus widely opened, ektexine verrucate, endexine smooth.
	P-61 Type 4 Pollen in monads, monocolporate, oblate in equatorial view. Ektexine granulated with reticulate ornamentation.
	P-62 Type 5 Pollen grains in monads, trizonocolporate, prolate in equatorial view, ektexine faintly foveolate, endexine smooth, ektexine thicker than endexine.
	P-63 Type 6 Pollen in monads, trizonocolporate, colpus wide, ektexine verrucate.

	P-64 Type 7 Pollen grains in monads, pentazonoporate, equatorial and polar outline spheroidal, psilate pollen grain with thicker ektexine.
	P-65 Type 8 Pollen in monads, triporate, ektexine and endexine with same thickness, ektexine psilate.
	P-66 Type 9 Pollen grains in monads, tetraporate, quadrangular in polar view, ektexine and endexine with same thickness, psilate pollen grain.
	P-67 Type10 Pollen grains in monads, tricolporate grain, circular in polar and equatorial view, ektexine with striato reticulate ornamentation.
	P-68 Type 11 Pollen grains in monads, tricolporate pollen, triangular in polar view and circular in equatorial view, ektexine faintly reticulate.

P-69

Type12

Pollen grains in monads, tricolporate pollen, circular in polar and equatorial view, psilate pollen grain.

4.3. PERCENTAGE OF POLLEN TYPES IN EACH SAMPLE

Percentage of each pollen types in honey and pollen samples from fifteen locations during the three seasons namely dearth (June to September), brood rearing (October to December) and honey flow (January to April) was worked out to record the contribution of each identified plant species to individual locations.

4.3.1 Distribution of pollen types in the honey samples during dearth season (June – September) in different locations

Honey samples collected during dearth season showed that the number of pollen types identified ranged from 1 to 4 at various locations (Table 4). A total of 14 pollen types were recorded from the honey samples during dearth season over fifteen locations which included *C. nucifera, M. pudica*, *M. diplotrica*, *T. paniculata*, *P. foetida*, *C. esculenta*, *M. calabura,* members of the families Lamiaceae, Elaeacarpaceae, Myrsinaceae (P-51), Fabaceae (P-46) and 3 unidentified pollen (Type-8, Type-9 and Type-12). Maximum number of pollen types was identified (4) from locations Kulappada, Pathanapuram and Rosemala. Only one pollen type (*C. nucifera)* was identified from the location Malayam (100 %). Two types of pollen were recorded from the locations Kodankara, Kadakkal, Veliyam, Ummannoor, Karikkam, Aryanad and Tholicode. Three pollen types were identified from Pullampara, Kilimanoor, Chennanpara and Kulathupuzha.

Distribution of pollen types revealed that *C. nucifera* (49.42 – 100.00 %) was the predominant pollen types in the honey samples from 10 locations (Kodankara, Malayam, Kadakkal, Veliyam, Ummannoor, Karikkam, Pathanapuram, Aryanad, Tholicode and Chennanpara) followed by *M. pudica* (58.22 – 98.56 %) in three locations (Kulappada, Pullampara and Kulathupuzha), *T. paniculata* (60.37 %) in Rosemala and an unidentified pollen (Type-12) (76.80 %) in Kilimanoor. Even

Table 4. Location wise distribution of pollen types in honey samples during dearth season (June to September)

Locations	Pollen types (nos.)	Percentage of pollen types collected from different locations			
		Predominant pollen	Secondary Pollen	Important minor pollen	Minor pollen
Kodankara	2	*C. nucifera* (69.23)	*M. pudica* (30.77)	-	-
Malayam	1	*C. nucifera* (100.00)		-	-
Kulappada	4	*M. pudica* (58.22)	*C. nucifera* (28.69)	*C. esculenta* (7.17) *M. calabura* (5.91)	-
Pullampara	3	*M. pudica* (98.56)	-	-	*C. nucifera* (0.24) F: Myrsinaceae (P-51) (1.20)
Kilimanoor	3	Type-12 (76.80)	*C. nucifera* (16.00)	Type -9 (7.20)	-
Kadakkal	2	*C. nucifera* (88.00)		*M.pudica* (12.00)	-
Veliyam	2	*C. nucifera* (80.00)	F: Fabaceae (P-46) (20.00)	-	-
Ummannoor	2	*C. nucifera* (80.00)	*M. pudica* (20.00)	-	-
Karikkam	2	*C. nucifera* (75.22)	Type-12 (25.66)	-	-
Pathanapuram	4	*C. nucifera* (49.42)	*M. pudica* (44.75)	F: Elaecarpaceae (3.11)	*P. foetida* (2.72)
Aryanad	2	*C. nucifera* (90.00)	-	*M. pudica* (10.00)	-
Tholicode	2	*C. nucifera* (68.00)	*M. pudica* (32.00)	-	-
Chennanpara	3	*C. nucifera* (72.97)	*M. pudica* (18.92)	Type-8 (8.12)	-
Kulathupuzha	3	*M. pudica* (96.21)	-	*C. nucifera* (3.19)	F: Lamiaceae (0.60)
Rosemala	4	*T. paniculata* (60.37)	*M. pudica* (22.56)	*M. diplotrica* (7.32) *C. nucifera* (9.15)	-

though *M. pudica* was one of the predominant pollen types in some samples, it was also recognized as secondary pollen in samples from Kodankara, Ummannoor, Tholicode, Chennanpara and Rosemala. Other secondary pollen types recorded during this season were of *C. nucifera* in Kulappada (28.69 %) and Kilimanoor (16.00 %), plant species belonging to the family Fabaceae (P-46) (20.00 %) in Veliyam and an unidentified pollen (Type-12) in Karikkam (25.66 %). Pollen types coming under the category of minor pollen, were not recorded from Kodankara, Malayam, Ummannoor, Pullampara, Veliyam, Karikkam and Tholicode. Important minor pollen types which observed in other locations included *C. esculenta* (7.17 %) and *M. calabura* (5.91 %), member of the family Flacourtiaceae (5.91 %) from Kulappada, an unidentified pollen (Type-9) (7.20 %) in Kilimanoor, *M. pudica* from Kadakkal (12.00 %) and Aryanad (10.00 %), an unidentified pollen (Type-8) in Chennanpara (8.12 %), plant species from the family Elaecarpaceae (3.11 %) in Pathanapuram, *M. diplotrica* (7.32 %) in Rosemala and *C. nucifera* in both Kulathupuzha (3.19 %) and Rosemala (9.15 %). *C. nucifera* (0.24 %) and pollen type belonging to family Myrsinaceae (P-51) (1.20 %) was recorded as the minor pollen type from Pullampara. Other minor pollen types recorded were, *P. foetida* (2.72 %) from Pathanapuram and members of the family Lamiaceae (0.60 %) from Kulathupuzha.

4.3.2 Distribution of pollen types in pollen samples during dearth season (June to September) in different locations

Distribution of pollen types in each pollen sample collected in dearth season from various locations is given in Table 5. Analysis of pollen loads showed the presence of 20 pollen types from 15 locations. Maximum number of pollen types was recorded in Kulathupuzha (7) while the least number (2) from the locations: Kodankara, Aryanad and Chennanpara. Three types of pollen was recorded from the pollen samples of Kulappada, Pullampara, Kilimanoor, Veliyam and Ummannoor.

Table 5. Location wise distribution of pollen types in pollen loads during dearth season (June to September)

Locations	Pollen types (nos.)	Percentage of pollen types collected from different locations			
		Predominant pollen	Secondary Pollen	Important minor pollen	Minor pollen
Kodankara	2	*M. pudica* (80.02)	*C. nucifera* (19.98)	-	
Malayam	5	*M. pudica* (58.64)	*C. nucifera* (29.73)	*H. rosasinensis* (8.84)	*A. auriculiformis* (2.68) *M. calabura* (0.12)
Kulappada	3	*M. pudica* (62.44)	*C. nucifera* (29.86)	*C. esculenta* (7.69)	-
Pullampara	3	*M. pudica* (89.31)	-	*C. nucifera* (10.69)	F: Myrsinaceae (P-55) (0.04)
Kilimanoor	3	*C. nucifera* (86.20)	-	F: Convolvulaceae (P-41) (13.28)	Type-8 (0.52)
Kadakkal	4	*C. nucifera* (82.67)	-	F: Myrtaceae (P-52) (5.33) F: Oleaceae (P-54) (11.46)	*M. pudica* (0.54)
Veliyam	3	*M. pudica* (54.67)	*C. nucifera* (29.14) F: Asteraceae (P-35) (16.2)	-	-
Ummannoor	3	*M. pudica* (76.87)	*C. nucifera* (22.7)	-	F: Fabaceae (P-45) (0.43)
Karikkam	4	*M. pudica* (68.15)	*C. nucifera* (30.19)	-	F: Anacardiaceae (P-31) (0.66) F: Lamiaceae (0.99)
Pathanapuram	4	-	*C. nucifera* (34.09) *M. pudica* (22.16) *Impatiens* sp. (40.06)	*H. rosa-sinensis* (3.69)	-
Aryanad	2	*C. nucifera* (86.45)	-	*M. pudica* (13.55)	-
Tholicode	5	*M. pudica* (85.05)	-	*C. nucifera* (7.21) Type-9 (5.68)	*A. auriculiformis* (1.81) F: Convolvulaceae (P-41) (0.25)
Chennanpara	2	*M. pudica* (80.92)	*C. nucifera* (19.08)		
Kulathupuzha	7	*M. pudica* (63.87)	-	*C. nucifera* (4.64) *A. auriculiformis* (11.09) F: Flacourtiaceae (12.59)	F: Myrtaceae (P-53) (2.69) F: Convolvulaceae(P-41) (2.55) Type-9 (2.55)
Rosemala	5	*C. nucifera* (39.62) *A. auriculiformis* (48.07)	-	*M. pudica* (6.33) *Impatiens* sp. (4.57)	F: Elaecarpaceae (1.41)

Pollen samples from the locations Kadakkal, Karikkam and Pathanapuram recorded 4 number of pollens where as five pollen types was recorded from the locations, Tholicode and Rosemala. Pollen types of plants belonging to 18 families were recorded from pollen loads during dearth season. Of these, Myrtaceae and Mimosaceae contribute maximum number of pollen types whereas maximum percentage contribution was made by plants belonging to families, Arecaceae and Mimosaceae. *M. pudica* (54.67 – 89.31 %) and *C. nucifera* (39.62 – 86.45 %), were the predominant pollen type in most of the pollen samples whereas in Rosemala, *A. auriculiformis* (48.07 %) was found to be the predominant type.

C. nucifera (19.08 – 34.09 %) and *M. pudica* (22.16 %) was also recorded as secondary pollen types in Pathanapuram. Other secondary pollen constituted pollen types from the family Asteraceae (P-35) (16.20 %) from Veliyam and *Impatiens* sp. (40.06 %) from Pathanapuram. Important minor pollen types recorded were *H. rosasinensis* (8.84 %), *C.esculenta* (7.69 %), *C.nucifera* (4.64 – 10.69 %), *A. auriculiformis* (11.09 %), *Impatiens* sp. (4.57 %) pollen of the families Convolvulaceae (P-41)(13.28 %), Myrtaceae (P-52) (5.33 %), Oleaceae (P-54) (11.46 %), Flacourtiaceae (12.59 %) and an unidentified pollen (Type-9) (5.68 %). Minor pollen constituted *A. auriculiformis* (1.81 – 2.68 %), *M. calabura* (0.12 %), *M. pudica* (0.54%), Pollen Type-8 (0.52 %), plants of families Fabaceae (P-45) (0.43 %), Myrsinaceae (P-55) (0.04 %), Anacardiaceae (P-31) (0.66 %), Lamiaceae (0.99%), Convolvulaceae (P-41) (0.25%), Myrtaceae (P-53) (2.69 %), Elaeocarpaceae (1.41%) and pollen type-9 (2.55 %).

4.3.3 Distribution of pollen types in honey sample during brood rearing season (October to December) in different locations

Analysis of honey samples collected in brood rearing season given in Table 6. revealed the presence of pollen from 16 different plant species. Maximum number of pollen types (6 nos.) was recorded in samples collected from Pathanapuram and

Table 6. Location wise distribution of pollen types in honey samples during brood rearing season (October to December)

Locations	Pollen types (nos.)	Percentage of pollen types collected from different locations			
		Predominant pollen	Secondary Pollen	Important minor pollen	Minor pollen
Kodankara	4	F: Cyperaceae (93.65)	-	*C. nucifera* (4.85)	*M. pudica* (0.69) *T. erecta* (0.79)
Malayam	4	-	*M. pudica* (39.64) F: Cyperaceae (42.34)	*S. calva* (14.41) *M. calabura* (3.60)	-
Kulappada	2	*M. pudica* (98.79)	-	-	*C. nucifera* (1.20)
Pullampara	4	*A. auriculiformis* (57.98)	*M. pudica* (17.02) *C. nucifera* (17.55)	F: Asteraceae (P-37) (7.45)	-
Kilimanoor	4	-	F: Asteraceae (P-37) (21.98) *M. pudica* (32.76) *C. nucifera* (34.91)	*A. auriculiformis* (10.34)	-
Kadakkal	5	*M. pudica* (63.96)	*C. nucifera* (18.02)	*T. erecta* (3.60) *M. diplotrica* (6.31) *S. calva* (8.11)	-
Veliyam	2	*M. pudica* (84.62)	*S. calva* (15.38)	-	-
Ummannoor	3	*M. pudica* (95.67)		*C. nucifera* (3.36)	*M. diplotrica* (0.96)
Karikkam	5	*S. calva* (51.77)	*M. pudica* (32.92)	*C. nucifera* (13.59)	*T. procumbens* (0.86) F: Lamiaceae (0.86)
Pathanapuram	6	*M. pudica* (65.75)	-	F: Cyperaceae (12.90) *M. diplotrica* (15.53)	*C. nucifera* (0.03) *Rosa* sp. (1.48) F: Asteraceae (P-37) (1.60)
Aryanad	2	*C. nucifera* (66.67)	*M. pudica* (33.33)	-	-
Tholicode	5	-	*M. pudica* (30.07) *C. nucifera* (33.01) *Rosa* sp. (33.01)	-	F: Sapindaceae (P-56) (0.98) *B. flabellifer* (2.94)
Chennanpara	6	*M. pudica* (63.51)	-	*M. diplotrica* (6.76) F: Asteraceae (P-36) (10.81) *C. nucifera* (14.86)	*A. auriculiformis* (2.03) F: Cyperaceae (2.03)
Kulathupuzha	3	*M. pudica* (59.18)	F: Asteraceae (P-37) (39.12)		F: Cyperaceae (1.70)
Rosemala	5	*M. pudica* (62.53)	*T. paniculata* (18.35)	*M. diplotrica* (7.24) *C. nucifera* (11.11)	*S. calva* (0.78)

Chennanpara while least pollen types (2 nos.) were observed from samples in Kulappada, Veliyam and Aryanad. Pollen of three different plant species was recorded from the locations, Ummannoor and Kulathupuzha. Honey samples from the locations Kodankara, Malayam, Pullampara and Kilimanoor showed the presence of four types of pollen where as the samples from Kadakkal, Karikkam, Tholicode and Rosemala recorded the presence of pollen of five different plant species.

C. nucifera was recorded as the predominant pollen type (66.67 %) in honey samples from Aryanad also the secondary pollen type in Pullampara (17.55 %), Kilimanoor (34.91 %), Kadakkal (18.02 %) and Tholicode (33.01 %). *C. nucifera* was also recorded as important minor pollen type in Kodankara (4.85 %), Ummannoor (3.36 %), Karikkam (13.59 %), Chennanpara (14.86 %), Rosemala (11.11 %) and as minor pollen type in Kulappada (1.20 %) and Pathanapuram (0.03 %).

Other than *C. nucifera*, pollen grain of *M. pudica* (59.18 – 98.79 %), *A. auriculiformis* (57.98 %), *S. calva* (51.77 %) were also recorded as predominant pollen during brood rearing season. No secondary pollen was recorded from Kodankara, Kulappada, Pathanapuram and Chennanpara. Secondary pollen types observed in other locations were *M. pudica* (17.02-39.64 %), *S. calva* (15.38 %) in Veliyam, member of family Asteraceae (P-37) (21.98 – 39.12 %) in Kilimanoor and Kulathupuzha and *T. paniculata* (18.35 %) in Rosemala. Pollen grains of *C. nucifera* (3.36-14.86 %), *A. auriculiformis* (10.34 %), *M. diplotrica* (6.31-15.53 %), *S. calva* (8.11- 14.41 %), *T. erecta* (3.60%) and the members of the families Asteraceae (P-37) (7.45 – 10.81%), Cyperaceae (12.90 %) constituted the important minor pollen types from various locations. *M. pudica* (0.69 %), *T. erecta* (0.79%), *C. nucifera* (0.03-1.20%), *M. diplotrica* (0.96%), *T. procumbens* (0.86 %), *Rosa* sp. (1.48 %), *B. flabellifer* (2.94 %), *A. auriculiformis* (2.03 %), *S. calva* (0.78 %) and plant species of families Lamiaceae (0.86 %), Cyperaceae (1.70- 2.03 %), Asteraceae

(P-37) (1.60 %) and Sapindaceae (P-56) (0.98 %) were also recorded as minor pollen type in some locations. No minor pollen types were observed from Malayam, Pullampara ,Kilimanoor ,Kadakkal and Veliyam.

4.3.4 Distribution of pollen types in pollen loads during brood rearing season (October to December) in different locations

Analysis of pollen loads collected in brood rearing season (Table 7.) revealed that out of the 30 pollen types recorded, the pollen loads from Pathanapuram had maximum number (9) of pollen types. Least number (4) of pollen grains was recorded from Aryanad which is located in upland region. Pollen loads from Kilimanoor and Kadakkal recorded the presence of five different types of pollen where as that of Malayam, Pullampara, Ummannoor, Karikkam, Chennanpara and Kulathupuzha recorded six types of pollen from pollen loads. Seven types of pollen was recorded from the pollen loads collected from Kulappada and veliyam where as the locations Kodankara and Rosemala showed the presence of 8 different pollen types. Family Asteraceae was found to be contributing more number of pollen types (5) followed by Mimosaceae (3) and Arecaceae (2).

The predominant pollen types recorded were *M. pudica*, *S. calva*, *C. nucifera* and members of F. Cyperaceae. *M. pudica* (45.41 – 76.31 %) was predominantly found in samples from Kulappada, Pullampara, Kilimanoor, Kadakkal, Ummannor, Pathanapuram and Kulathupuzha whereas plants of family Cyperaceae (75.51-78.79 %) occurred predominantly from Kodankara and Malayam. *S. calva* was the predominant pollen type observed in samples from Veliyam (52.23 %) while predominance of *C. nucifera* (48.00 %) was recorded from Aryanad. Secondary pollen grains recorded in different locations constitute *M. pudica*, *C. nucifera*, *S. calva*, *M. diplotrica*, *A. auriculiformis* and plants belonging to families Asteraceae (P-36) and Cyperaceae. *M. pudica* was recorded from Kodankara (15.73 %), Veliyam

Table 7. Location wise distribution of pollen types in pollen loads during brood rearing season (October to December)

Locations	Pollen types (nos.)	Percentage of pollen types collected from different locations			
		Predominant pollen	Secondary Pollen	Important minor pollen	Minor pollen
Kodankara	8	F: Cyperaceae (75.51)	*M. pudica* (15.73)	*T. erecta* (3.90)	*C. nucifera* (2.39) *C. exarillata* (0.12) F: Sapindaceae (P-56) (1.04) Type-4 (0.64) Type-11 (0.69)
Malayam	6	F: Cyperaceae (78.79)	-	*C. nucifera* (6.21) *M. pudica* (3.42) *S. calva* (5.08) *P. guajava* (3.57)	*A. auriculiformis* (2.93)
Kulappada	7	*M. pudica* (52.22)	Type-2 (32.59)	F: Cyperaceae (10.61)	*C. nucifera* (2.12) *M. diplotrica* (1.23) *S. calva* (0.07) F: Elaecarpaceae (0.53)
Pullampara	6	*M. pudica* (57.82)	*S. calva* (32.21)	*C. nucifera* (5.92)	*M. diplotrica* (1.84) F: Elaecarpaceae (1.01) Type-4 (1.21)
Kilimanoor	5	*M. pudica* (76.31)	*S. calva* (17.91)	*C. nucifera* (3.69)	*A. auriculiformis* (1.41) *T. erecta* (0.67)
Kadakkal	5	*M. pudica* (60.04)	*C. nucifera* (38.8)	-	*S. calva* (0.58) F: Anacardiaceae (P-31) (0.26) F: Cyperaceae (0.32)
Veliyam	7	*S. calva* (52.23)	*M. pudica* (33.74)	*C. nucifera* (9.60)	*A. auriculiformis* (1.09) F: Fabaceae (P-45) (0.50) F: Cyperaceae (2.01) F: Myrtaceae (P-52) (0.84)
Ummannoor	6	*M. pudica* (47.42)	*S. calva* (36.28)	*C. nucifera* (4.52) *M. diplotrica* (9.84)	F: Cyperaceae (1.68) F: Convolvulaceae (P-41) (0.27)
Karikkam	6	-	*M. pudica* (42.84)	*C. nucifera* (20.57) *S. calva* (18.92) F: Asteraceae (P-37) (8.04) F: Acanthaceae (7.24)	*M. diplotrica* (2.40)
Pathanapuram	9	*M. pudica* (45.41)	*M. diplotrica* (21.42) *S. calva* (15.93)	*C. nucifera* (3.01) F: Myrtaceae(P-52) (9.02)	F: Fabaceae (P-45) (0.45) F: Cyperaceae (2.24) F: Celastraceae (1.14) Type-2 (1.38)

Table 7 continued

Locations	Pollen types (nos.)	Percentage of pollen types collected from different locations			
		Predominant pollen	Secondary Pollen	Important minor pollen	Minor pollen
Aryanad	4	*C. nucifera* (48.00)	*M. pudica* (29.33)	*M. diplotrica* (14.66) F: Cyperaceae (8.02)	-
Tholicode	9	-	*C. nucifera* (30.57) *M. pudica* (24.68) F: Cyperaceae (19.50)	*P. guajava* (14.83) *B. flabellifer* (4.42)	*A. auriculiformis* (2.49) *M. diplotrica* (0.91) *T. procumbens* (2.23) F: Euphorbiaceae (0.36)
Chennanpara	6	-	*C. nucifera* (20.84) *M. pudica* (41.48) F: Asteraceae (P-36) (24.35)	*M. diplotrica* (12.02)	F: Convolvulaceae (P-40) (0.88) F: Sapindacea (P-56) (0.44)
Kulathupuzha	6	*M. pudica* (68.44)	*S. calva* (25.21)	-	*C. nucifera* (0.75) F: Oleaceae (P-54) (1.92) *M. diplotrica* (1.88) F: Cyperaceae (1.79)
Rosemala	8	-	*C. nucifera* (32.15) *A. auriculiformis* (34.71) *S. calva* (15.17)	*M. pudica* (4.29) *M. diplotrica* (8.57) F: Arecaceae (P-33) (3.79)	F: Lamiaceae (1.07) *P. paucifolium* (0.25)

(33.74 %), Karikkam (42.84 %), Tholicode (24.68 %) and Chennanpara (41.48 %) while *C. nucifera* from Tholicode(30.57 %), Chennanpara (20.84 %) and Rosemala (32.15 %). *S. calva* was recorded as secondary pollen type from Pullampara (32.21 %), Kilimanoor (17.91 %), Pathanapuram (15.93 %), Kulathupuzha (25.21 %) and Rosemala (15.17 %). Pollen types of family Cyperaceae were also found as secondary pollen from samples of Tholicode (19.50 %). *M. diplotrica* from Pathanapuram (21.42%), *A. auriculiformis* (34.71%) and family Asteraceae (P-36) (24.35%) from Rosemala were the other secondary pollen types recorded.

Important minor pollen recorded were *C. nucifera* (3.01- 20.57%), *M. diplotrica* (8.57-14.66%), *M. pudica* (3.42 – 4.29%), *T. erecta* (3.90%), *P. guajava* (3.57 – 14.83 %), *B. flabellifer (*4.42%), members of the families Cyperaceae (8.02-10.61%), Asteraceae (P-37) (8.04%), Acanthaceae (7.24%), Arecaceae (P-33) (3.79%) and Myrtaceae (P-52) (9.02%). Minor pollen types recorded from fifteen locations included *C. nucifera* (0.75 – 2.39 %), *C. exarillata* (0.12 %), *A. auriculiformis* (1.09 – 2.93 %), *M. diplotrica* (0.91-1.88 %), *S. calva* (0.07 – 0.58 %), *T. procumbens* (2.23 %), *P. paucifolium* (0.25 %), members of families Sapindaceae (P-56) (0.44 – 1.04 %), Elaecarpaceae (0.53-1.41 %), Anacardiaceae (P31) (0.26 %), Cyperaceae (0.32 – 2.34 %), Myrtaceae (P-52) (0.84 %), Convolvulaceae (P-41) (0.27-0.88 %), Fabacaee (P-45) (0.45 %), Celastraceae (1.14 %), Euphorbiaceae (0.36 %), Oleaceae (P-54) (1.92 %), Lamiaceae (1.07 %) and three unidentified pollen types, Type-4 (0.64-1.21 %), Type-11 (0.68 %) and Type-2 (1.38 %).

4.3.5 Distribution of pollen in honey sample during honey flow season (January to April) in different locations

Maximum pollen types (26) were recorded during honey flow season from all the fifteen locations (Table 8). Kodankara and Ummannoor had maximum number (8) of pollen types while Kilimanoor had the least (2). Kulappada,

Table 8. Location wise distribution of pollen types in honey samples during honey flow season (January to April)

Locations	Pollen types (nos)	Percentage of pollen types collected from different locations			
		Predominant pollen	Secondary Pollen	Important minor pollen	Minor pollen
Kodankara	8	F: Cyperaceae (53.73)	*C. nucifera* (38.61)	Type-10 (6.72)	*M. pudica* (1.47) *H. ponga* (0.42) *Bombax* sp. (0.84) *Rosa* sp. (2.62) Type -11 (0.42)
Malayam	6	-	*C. nucifera* (39.71)	*M. pudica* (11.01) F: Cyperaceae (12.44) *P. guajava* (11.96) F: Sapindaceae (P-57) (6.70)	Type-6 (18.18)
Kulappada	3	*M. pudica* (47.33)	*C. nucifera* (41.22)	F: Oleacaea(P-54)(11.45)	-
Pullampara	3	*M. pudica* (45.50) F: Cyperaceae (48.50)	-	*C. nucifera* (6.00)	-
Kilimanoor	2	*C. nucifera* (66.67)	*M. pudica* (33.33)	-	-
Kadakkal	5	F: Cyperaceae (49.32)	*C. nucifera* (18.55) *M. pudica* (19.46)	*A. auriculiformis* (6.33) F: Arecaceae (P-34) (6.33)	-
Veliyam	5	*M. pudica* (78.21)	-	*C. nucifera* (3.21) F: Cyperaceae (5.77) *Rosa* sp.(7.69) *M. indica* (4.49)	-
Ummannoor	8	*C. nucifera* (43.89)	F: Cyperaceae (24.43)	*M. pudica* (12.60) F: Arecaceae (P-34) (8.40) F: Myrtaceae (P-53) (5.34)	*Rosa* sp. (1.53) *T. gamblei* (2.29) Type-5 (1.53)
Karikkam	5	*C. nucifera* (65.16)	F: Cyperaceae (25.56)	*M. pudica* L. (4.26) Type-11 (4.01)	*M. diplotrica* (1.00)
Pathanapuram	6	*M. pudica* (92.19)	-	F: Cyperaceae (4.36)	*C. nucifera* (1.91) *A. auriculiformis* (0.27) *M. diplotrica* (0.91) F: Elaecarpaceae (0.36)
Aryanad	6	*C. nucifera* (51.02)	F: Cyperaceae (19.38)	*M. pudica* (9.18) *A. auriculiformis* (7.14) F: Amaranthaceae (4.08) Type-11 (9.18)	*S. calva* (1.81) Type-2 (0.61)

Table 8 continued

Locations	Pollen types (nos.)	Percentage of pollen types collected from different locations			
		Predominant pollen	Secondary Pollen	Important minor pollen	Minor pollen
Tholicode	7	-	F: Celastraceae (33.33) *M. pudica* (29.70)	*C. nucifera* (12.73) *P. guajava* (6.67) F: Cyperaceae (15.15)	-
Chennanpara	4	*C. nucifera* (50.93)	*M. pudica* (22.36)	*Rosa* sp. (11.18) Type-8 (15.53)	-
Kulathupuzha	3	-	*C. nucifera* (37.21) *M. pudica* (27.91) F: Cyperaceae (34.88)	-	-
Rosemala	6	-	*M. pudica* (24.16) F: Sapindaceae (P-57) (18.12) Type-6 (29.53)	*C. nucifera* (13.42) F: Cyperaceae (10.74) *D. pentagyna* (4.03)	-

Pullampara and Kulathupuzha recorded the presence of 3 number of pollen types. Honey samples from Chennanpara recorded the presence of 4 number of pollen types. Pollen of five different plant species were recorded from Kadakkal, Veliyam and Karikkam where as honey samples from Malayam, Pathanapuram, Aryanad and Rosemala showed the presence of 6 pollen types. Only the location Tholicode showed seven types of pollen from honey samples.

C. nucifera was the predominant pollen type in honey samples from five locations with a percentage contribution ranging from 43.89 – 66.67 % followed by *M. pudica* with a percentage contribution ranging from 45.50 – 92.19 %. Other predominant pollen present was plant species from family Cyperaceae in Kodankara (53.73 %), Pullampara (48.5 %) and Kadakkal (49.32 %). Secondary pollen constituted *C. nucifera* (18.55 – 41.22 %), *M. pudica* (19.46 – 33.3 %), plant species from the family Cyperaceae (19.38 – 34.88 %), Celastraceae (33.33 %), Sapindaceae (P-57) (18.12 %) in Tholicode, and an unidentified pollen (Type-6) in Rosemala (29.53 %). There was no pollen recorded as secondary in honey samples from Pullampara, Veliyam and Pathanapuram.

Important minor pollen types included *M. pudica* (4.26 – 12.60 %) in Malayam, Ummannoor, Karikkam and Aryanad, *C. nucifera* in Pullampara (6 %) and Rosemala (13.42 %), *Rosa* sp. (7.69-11.18 %) in Veliyam and Chennanpara and *M. indica* (4.47 %) in Veliyam, *A. auriculiformis* in Kadakkal (6.33 %) and Aryanad (7.14 %). Important minor pollen recorded from Malayam included *M. pudica* (11.01 %), *P. guajava* (11.96%), members of the families Cyperaceae (12.44%) and Sapindaceae (P-57) (6.70 %). Pollen types of family Arecaceae (P-34) constituted the important minor pollen in Kadakkal (6.33%). *C. nucifera* (3.21%), plants belonging to family Cyperaceae (5.77%), *Rosa* sp. (7.69%) and *M. indica* (4.49%) were recorded as important minor pollen from Veliyam. Important minor pollen recorded from Aryanad included *M. pudica* (9.18%), *A. auriculiformis* (7.14 %), members of family Amaranthaceae (4.08%) and an unidentified pollen (Type-11)

(9.18 %). Minor pollen recorded was *M. pudica* (1.47%), *H. ponga* (0.42 %), *Bombax* sp. (0.84 %), *Rosa* sp. (1.53 – 2.62 %), *T. gamblei* (2.29%), *M. diplotrica* (0.91-1.00 %) , *A. auriculiformis* (0.27 %), *S. calva*(1.81 %), members of family Elaecarpaceae (0.36 %) and four unidentified pollen, Type – 11 (0.42 %), Type – 6 (18.18 %), Type – 5 (1.53%) and Type 2 (0.61%).

4.3.6 Distribution of pollen types in pollen loads during honey flow season (January to April) in different locations

A total of 38 pollen types were recorded from pollen loads collected during honey flow season from fifteen apiary sites. Distribution of different pollen types to each sample was detailed in Table 9. Maximum number of pollen types was recorded in Rosemala (12) while minimum number of pollen grains was recorded in Veliyam (2). Pollen loads from the Kadakkal recorded the presence of 3 different types of pollen. 4 different types of pollen were recorded from Aryanad, Tholicode and 5 different types of pollen were recorded from Kodankara, Pullampara and Ummannoor. Locations, Kilimanoor and Chennanpara had six number of pollen types where as seven number of pollen types was recorded from the pollen loads of Malayam, Kulappada and Karikkam. Pathanapuram had ten number of pollen types and Kulathupuzha recorded the presence of 8 number of pollen types.

Pollen analysis of pollen loads collected during honey flow season revealed that *C. nucifera*, *M. pudica*, *D. pentagyna* and plant species from family Cyperaceae were the predominant pollen type. Of these pollen grains of *C. nucifera* was recorded predominant in the locations Kodankara (86.11 %), Malayam (48.82 %), Kilimanoor (71.71%), Kadakkal (86.99 %), Veliyam (98.44 %), Ummannoor (62.20 %), Karikkam (54.43 %), Tholicode (52.99 %), Chennanpara (49.08 %) and Kulathupuzha (67.13 %). *D. pentagyna* was found to be predominant in Rosemala (50.75 %). Apart from these, pollen grain in members of the family Cyperaceae was found as predominant type in Pullampara (66.36 %).

Table 9. Location wise distribution of pollen types in pollen loads during honey flow season (January to April)

Locations	Pollen types (nos.)	Percentage of pollen types collected from different locations			
		Predominant pollen	Secondary Pollen	Important minor pollen	Minor pollen
Kodankara	5	*C. nucifera* (86.11)	-	F: Cyperaceae (9.33)	F: Anacardiaceae (P-31) (1.34) *M. indica* (1.04) Type-11(2.19)
Malayam	7	*C. nucifera* (48.82)	*M. pudica* (43.56)	-	*P. foetida* (0.54) *S. calva* (1.09) F: Cyperaceae (1.09) F: Meliaceae (2.09) F: Amaranthaceae (2.81)
Kulappada	7	-	*M. pudica* (34.73) *C. nucifera* (44.73)	*M. diplotrica* (5.38) *S. calva* (4.84) *T. erecta* (3.96) F: Meliaceae (4.29)	*T. paniculata* (2.09)
Pullampara	5	F: Cyperaceae (66.36)	*M. pudica* (24.76)	*C. nucifera* (4.50) F: Celastraceae (3.88)	*T. gamblei* (0.50)
Kilimanoor	6	*C. nucifera* (71.71)	*T. connaroides* (19.22)	F: Anacardiaceae (P-32) (4.43)	*T. gamblei* (2.27) F: Myrtaceae (P-52) (1.62) Type-7 (0.76)
Kadakkal	3	*C. nucifera* (86.99)	-	*M. pudica* (9.57) *T. gamblei* (3.44)	-
Veliyam	2	*C. nucifera* (98.44)	-	-	*M. pudica* (1.06)
Ummannoor	5	*C. nucifera* (62.2)	F: Oleaceae(P-55) (17.54)	*T. gamblei* (4.64) *M. pudica* (13.04)	F: Myrtaceae (P-52) (2.58)
Karikkam	7	*C. nucifera* (54.43)	F: Cyperaceae (26.82)	*M. pudica* (3.24) F: Fabaceae (P-45) (3.17) F: Celastraceae (9.73)	*S. calva* (1.37) *H. rosasinensis* (1.23)
Pathanapuram	10	-	Fam. Celastraceae (34.83) *M. diplotrica* (17.07) *C. nucifera* (19.18)	*M. pudica* L. (12.24) Fam. Meliaceae (3.13) Fam. Cyperaceae (8.78)	*H. rosa-sinensis* (1.63) *Antigonon* sp. (1.56) *T. paniculata* (0.82) Type-1 (0.75)
Aryanad	4	-	*C. nucifera* (28.05) F: Cyperaceae (29.43) F: Celastraceae (36.59)	*A. auriculiformis* (5.93)	-

Table 9 continued

Locations	Pollen types (nos.)	Percentage of pollen types collected from different locations			
		Predominant pollen	Secondary Pollen	Important minor pollen	Minor pollen
Tholicode	4	*C. nucifera* (52.99) *M. pudica* (45.29)	-	-	*M. diplotrica* (1.26) *S. calva* (0.46)
Chennanpara	6	*C. nucifera* (49.08)	*M. pudica* (38.75)	*A. auriculiformis* (4.62) F: Cyperaceae (5.27)	F: Anacardiaceae (P-31) (1.83) Type 7 (0.44)
Kulathupuzha	8	*C. nucifera* (67.13)	-	*T. gamblei* (8.22) *P. pterocarpum* (4.18) *E. guineensis* (10.72) F: Myrtaceae (P-53) (5.99)	*M. pudica* (1.25) *H. ponga* (2.23) *Impatiens* sp. (0.28)
Rosemala	12	*D. pentagyna* (50.75)	F: Sapindaceae (P-57) (24.63)	*M. pudica* (4.55) F: Cyperaceae (P-57) (14.83)	*C. nucifera* (0.11) F: Fabaceae (P-45) (1.34) F: Elaecarpaceae (1.39) *S. oleosa* (0.54) F: Brassicaceae (0.48) Type-3 (0.16) Type-8 (0.54) Type-12 (0.86)

Secondary pollen constituted *C. nucifera* in Kulappada (44.73 %), Pathanapuram (19.18 %) and Aryanad (28.05%), *M. pudica* in Malayam (43.56 %), Kulappada (34.73 %), Pullampara (24.76 %) and Chennanpara (38.75 %), members of the family Cyperaceae (26.82-29.43 %), *T. connaroides* in Kilimanoor (19.22 %), members of the family Oleaceae (P-55) in Ummannoor (17.54%), plants of family Celastraceae (34.83 %) and *M. diplotrica* (17.07 %) in Pathanapuram and members of family Sapindaceae (P-57) in Rosemala (24.63 %).

Important minor pollen observed during honey flow season was plant species belonging to the family Cyperaceae which was recorded from Kodankara, Pathanapuram, Chennanpara and Rosemala in 9.33, 0.27, 8.78 and 14.83 percent respectively. *M. diplotrica* (5.38 %), *S. calva* (4.84 %), *T. erecta* (3.96 %) and pollen types of family Meliaceae (4.29 %) were recorded from Kulappada whereas *C. nucifera* (4.50 %) and members from family Celastraceae (3.88 %) were the important minor pollen from Pullampara. Members of family Anacardiaceae (P-32) (4.43 %), which was the only important minor pollen recorded from Kilimanoor. *M. pudica* (9.57%) and *T. gamblei* (3.44 %) was recorded from Kadakkal whereas *T. gamblei* (4.64 %) and *M. pudica* (13.04 %) was also recorded from Ummannoor as important minor pollen. The important minor pollen types found in samples from Karikkam was *M. pudica* (3.24 %), plant species belonging to family Fabaceae (P-45) (3.17 %) and family Celastraceae (9.73 %). *M. pudica* (12.24 %), plants from family Meliaceae (3.13 %) and family Cyperaceae (8.78 %) was the important minor pollen types recorded from Pathanapuram whereas *A. auriculiformis* (5.93 %) was recorded from Aryanad. The other important minor pollen recorded were *A. auriculiformis* (4.62 %) and member of the family Cyperaceae (5.27 %) in Chennanpara, *T. gamblei* (8.22 %), *P. pterocarpum* (4.18 %), *E. guineensis* (10.72 %) and family Myrtaceae (P-53) (5.99 %) in Kulathupuzha, *M. pudica* (4.55 %) and plant species of family Cyperaceae (14.83 %) in Rosemala. There were no important minor pollen type recorded in Malayam,Veliyam and Tholicode.

Maximum number of minor pollen types was recorded from Rosemala (8). Minor pollen types recorded were from plants belonging to family Anacardiaceae in Kodankara (1.34 %) and Chennanpara (1.83 %), *M. indica* (1.04 %) and an unidentified pollen Type-11 (2.19 %) in Kodankara. *S. calva* from Malayam (1.09 %), Karikkam (1.37 %) and Tholicode (0.46 %). Pollen grains of family Cyperaceae were recorded from Malayam (1.09 %). Apart from these plants of family Meliaceae (2.09 %), family Amaranthaceae (2.81 %) and *P. foetida* (0.54 %) were recorded as minor pollen from Malayam. Pollen grain of *T. paniculata* was recorded from Kulappada (2.09 %) and Pathanapuram (0.82 %). *T. gamblei* (0.50-2.2 7%), member of family Myrtaceae (P-52) (1.62 %) and an unidentified pollen (Type-7) (0.76 %) was recorded as minor pollen types from Kilimanoor . Pollen Type -7 (0.44 %) from Chennanpara and members of family Myrtaceae (P-52) (2.58 %) was also recorded as a minor pollen. *M. pudica* was also found to be the minor pollen types in Veliyam (1.06%) and Kulathupuzha (1.25%). Minor pollen types recorded from Pathanapuram were *H. rosa-sinensis, Antigonon* sp., *T. paniculata* and an unidentified pollen Type – 1 (0.75 %). Pollen of *M. pudica* (1.25 %), *H. ponga* (2.23 %) and *Impatiens* sp. were recorded as minor pollen types in Kulathupuzha whereas *C. nucifera* (0.11 %), *S. oleosa* (54 %), plants of family Fabaceae (P-45) (1.34 %), family Brassicaceae (0.48 %) and three unidentified pollen types (Type-3) (0.16 %), Type-8 (0.54 %) and Type-12 (0.86 %) from Rosemala.

4.3.7 Pollen spectra of honey and pollen samples

Pollen spectra were constructed based on the percentage of pollen types in each honey and pollen samples from fifteen locations. (Fig. 4. a, b, c, d, e and f).

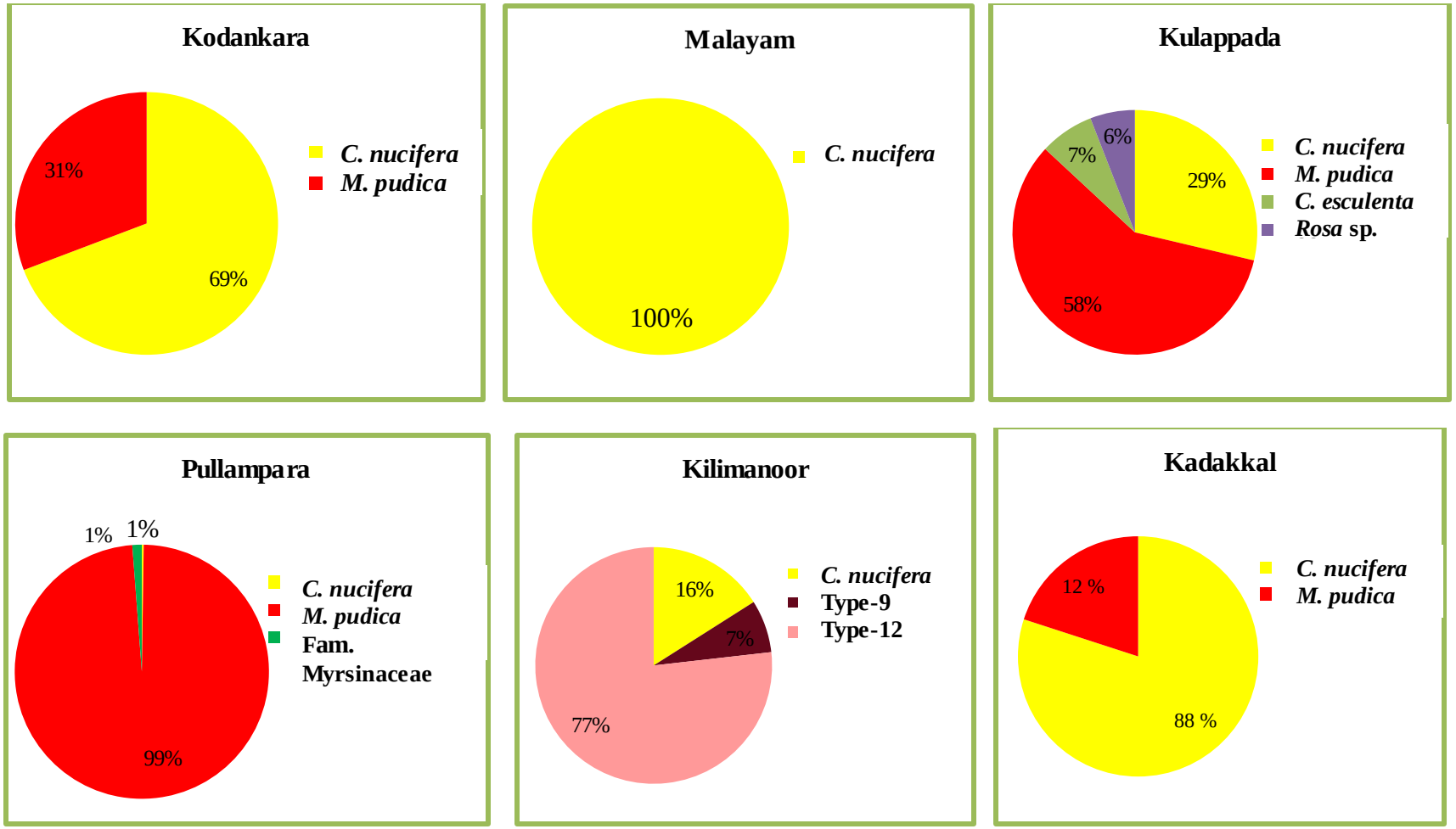


Fig.4. a. Pollen spectra of honey samples during dearth season

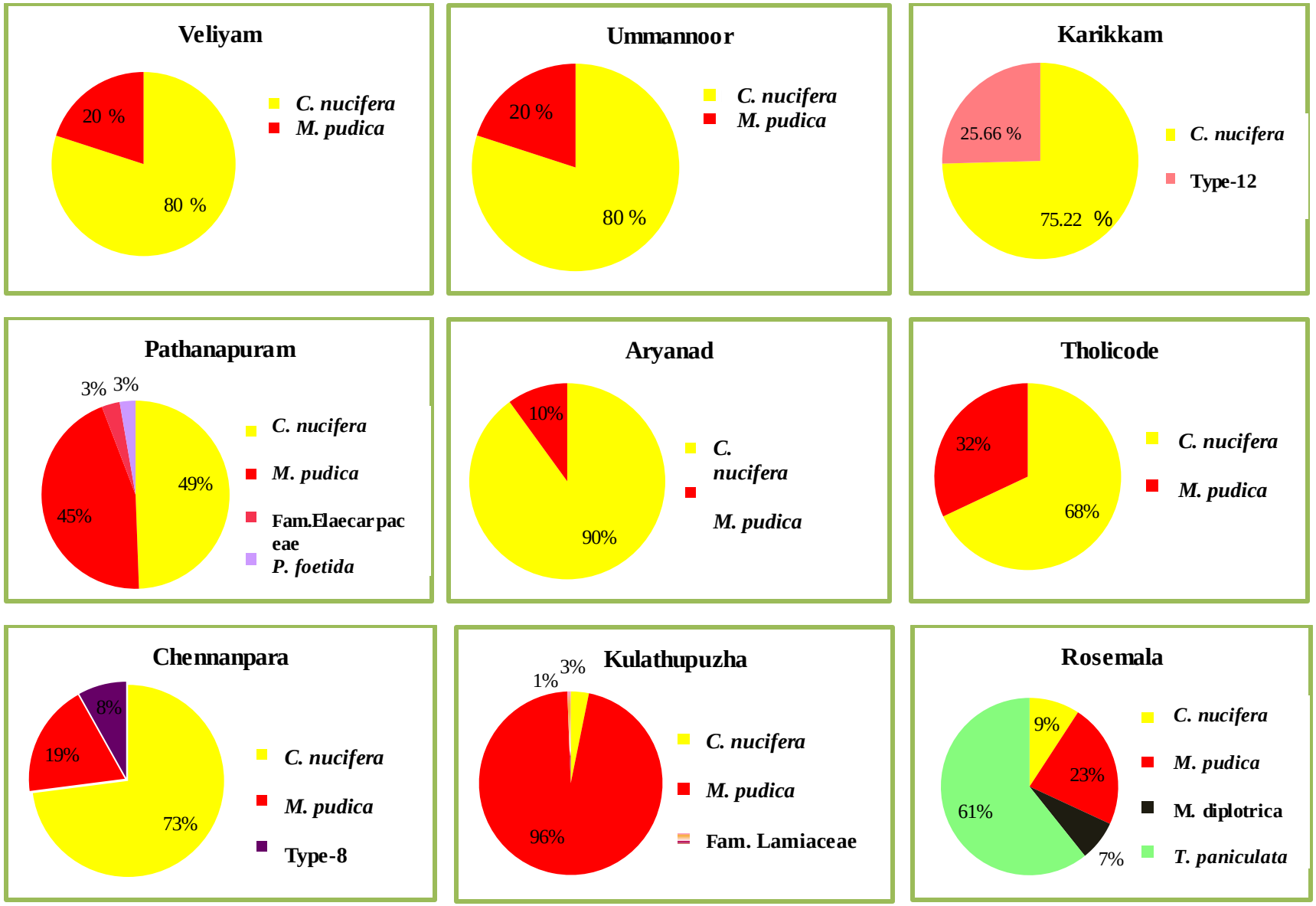


Fig-4. A.continued

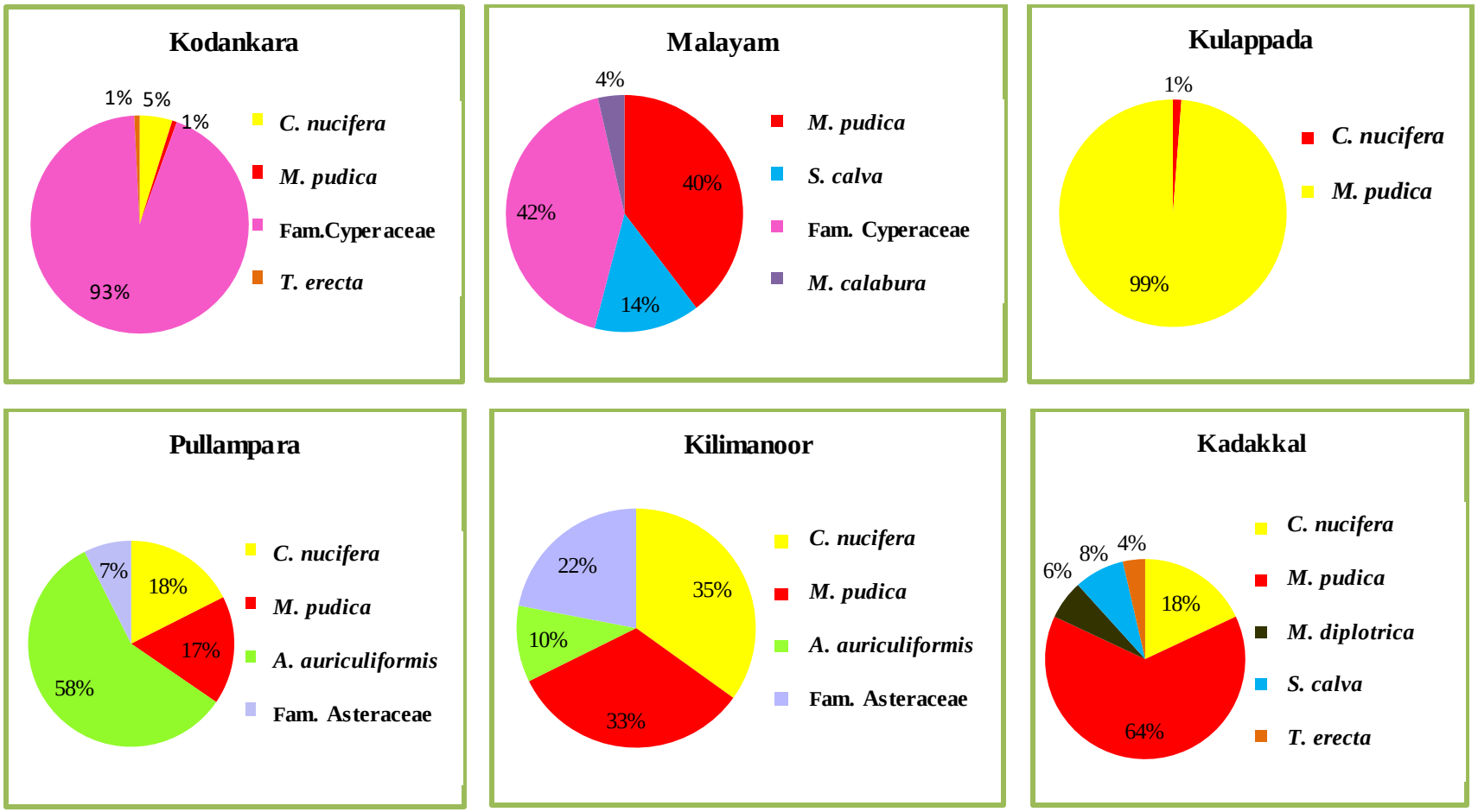


Fig. 4. b. Pollen spectra of honey samples during brood rearing season

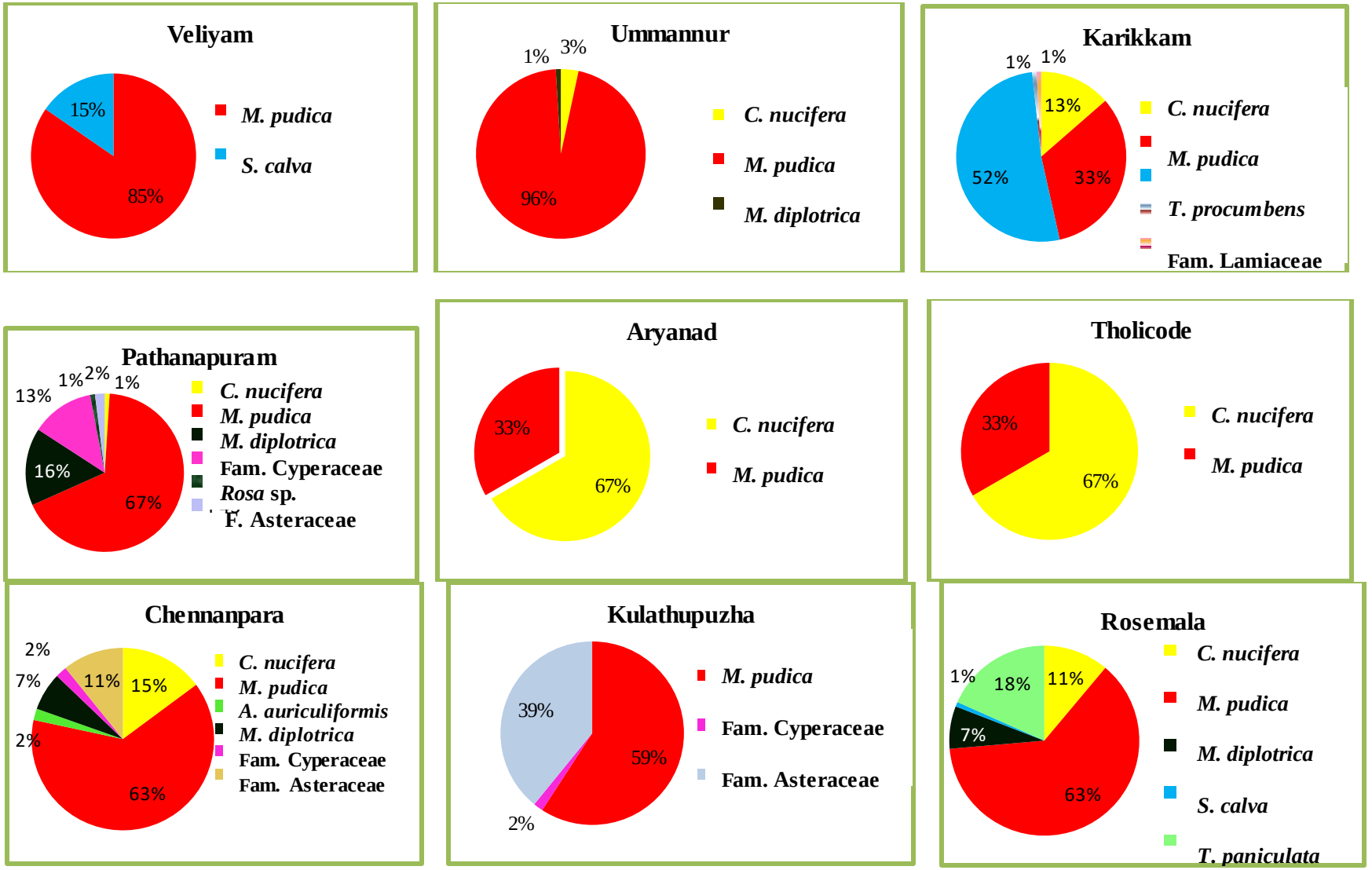


Fig. 4. b. continued

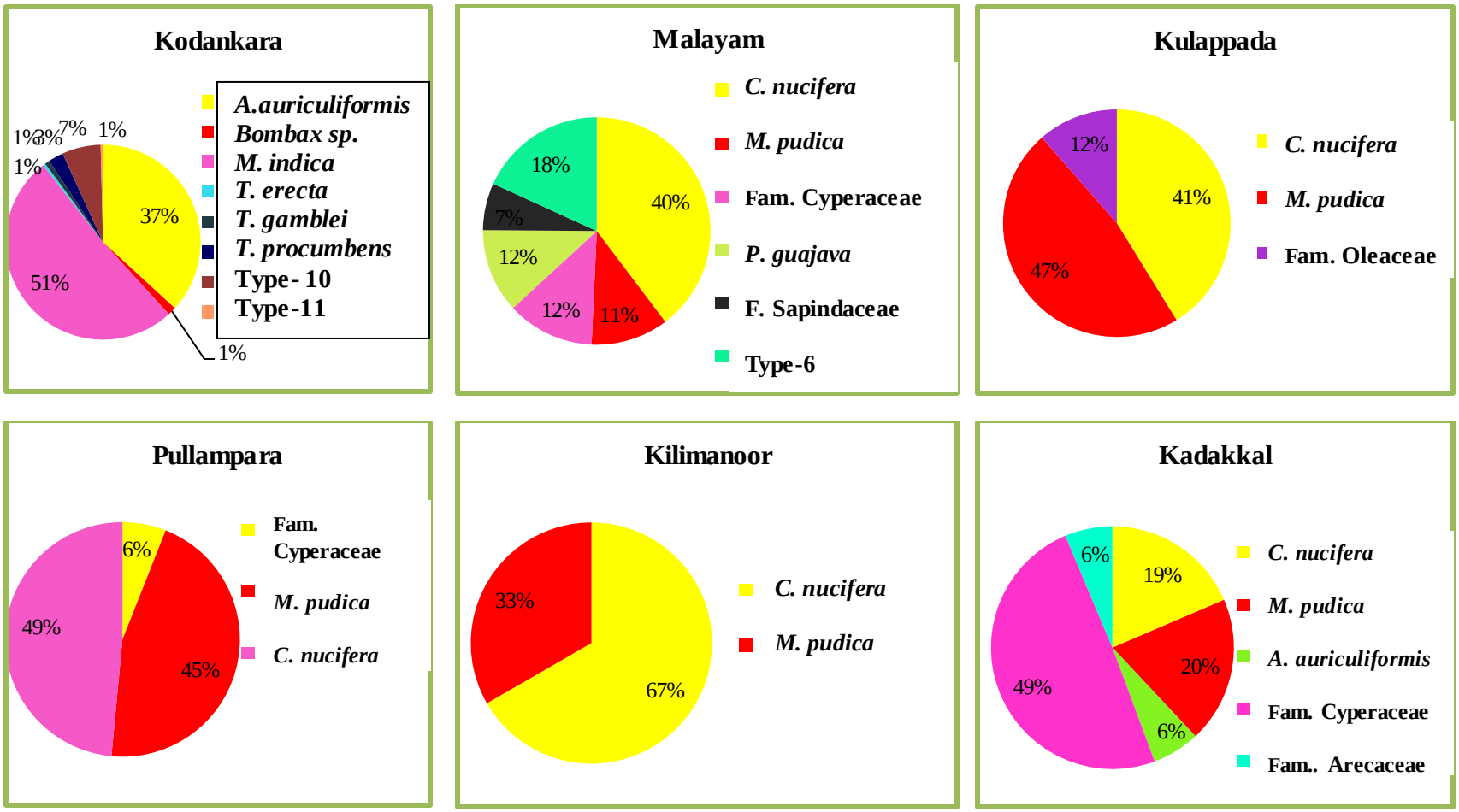


Fig. 4. c. Pollen spectra of honey samples during honey flow season

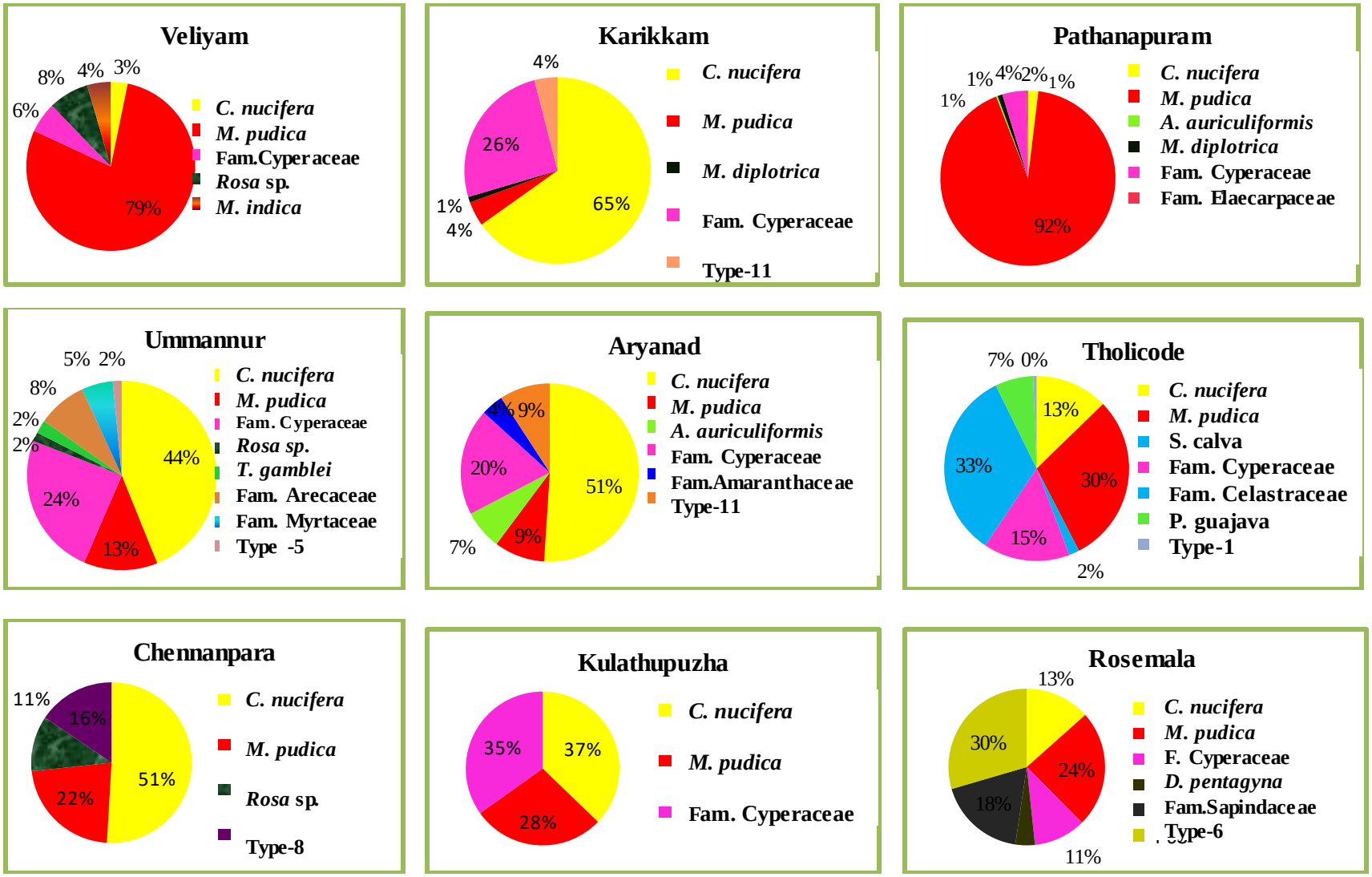


Fig.4. c. continued

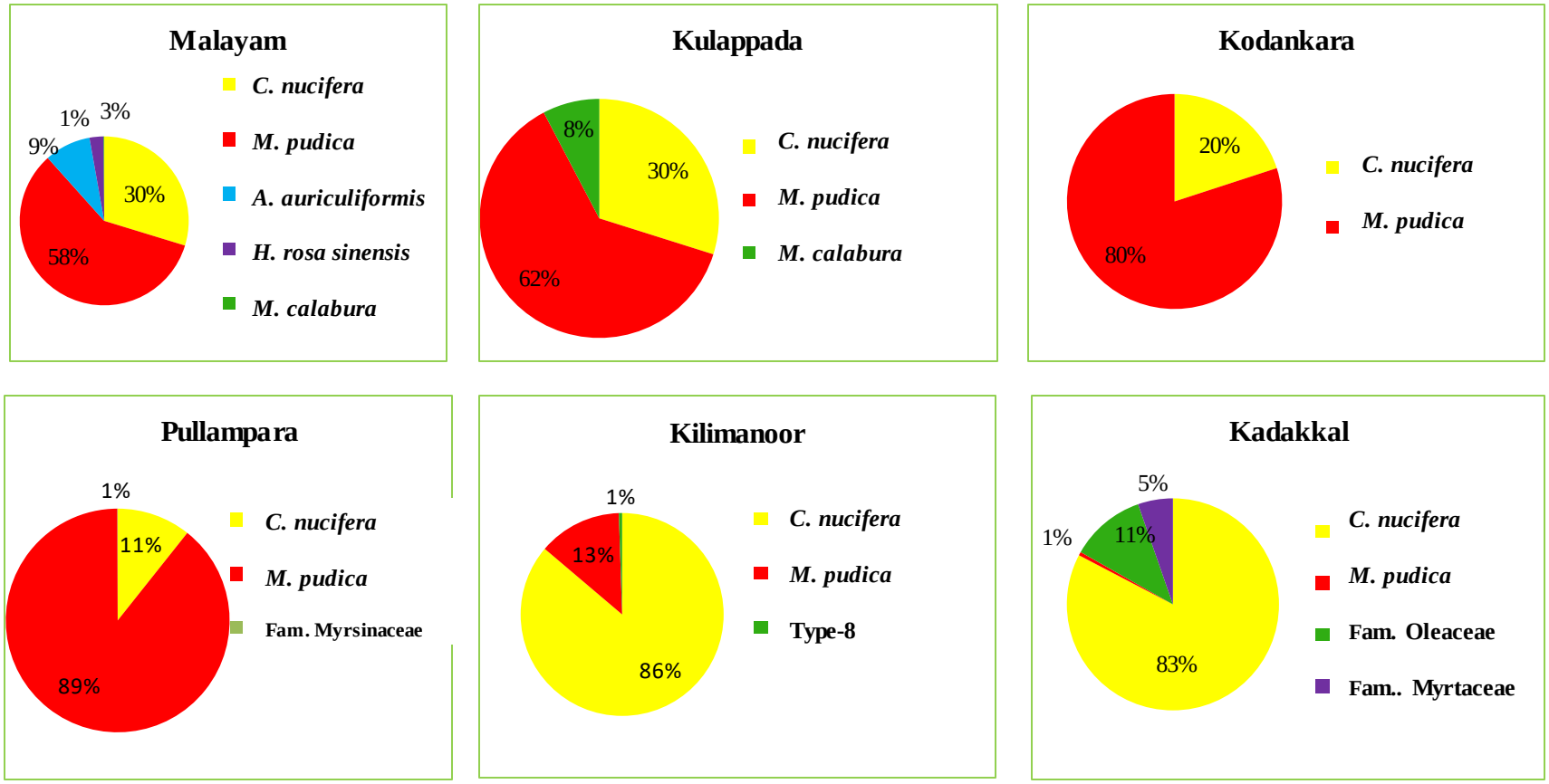


Fig. 4. d. Pollen spectra of pollen loads during dearth season

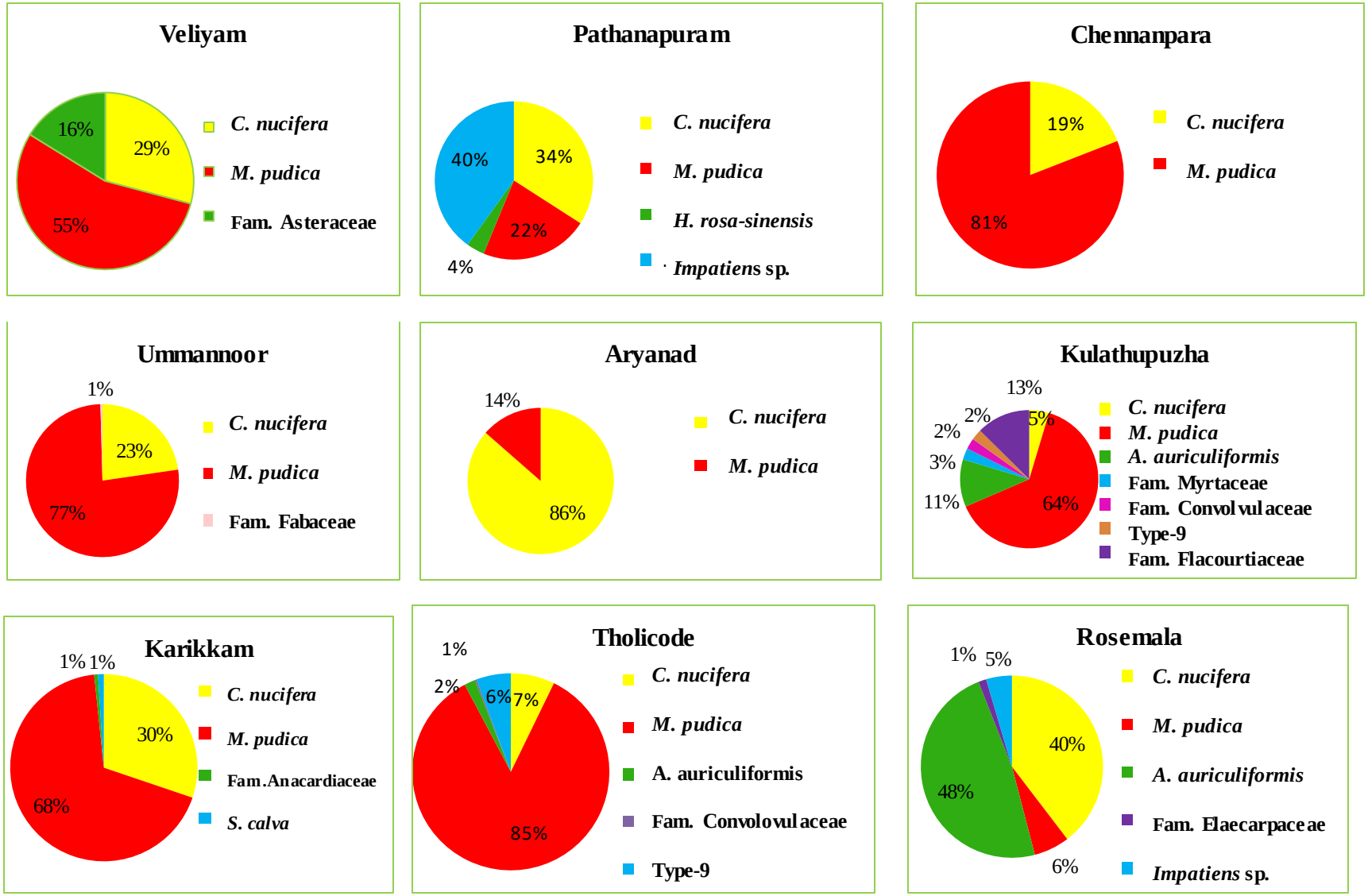


Fig. 4. d. continued

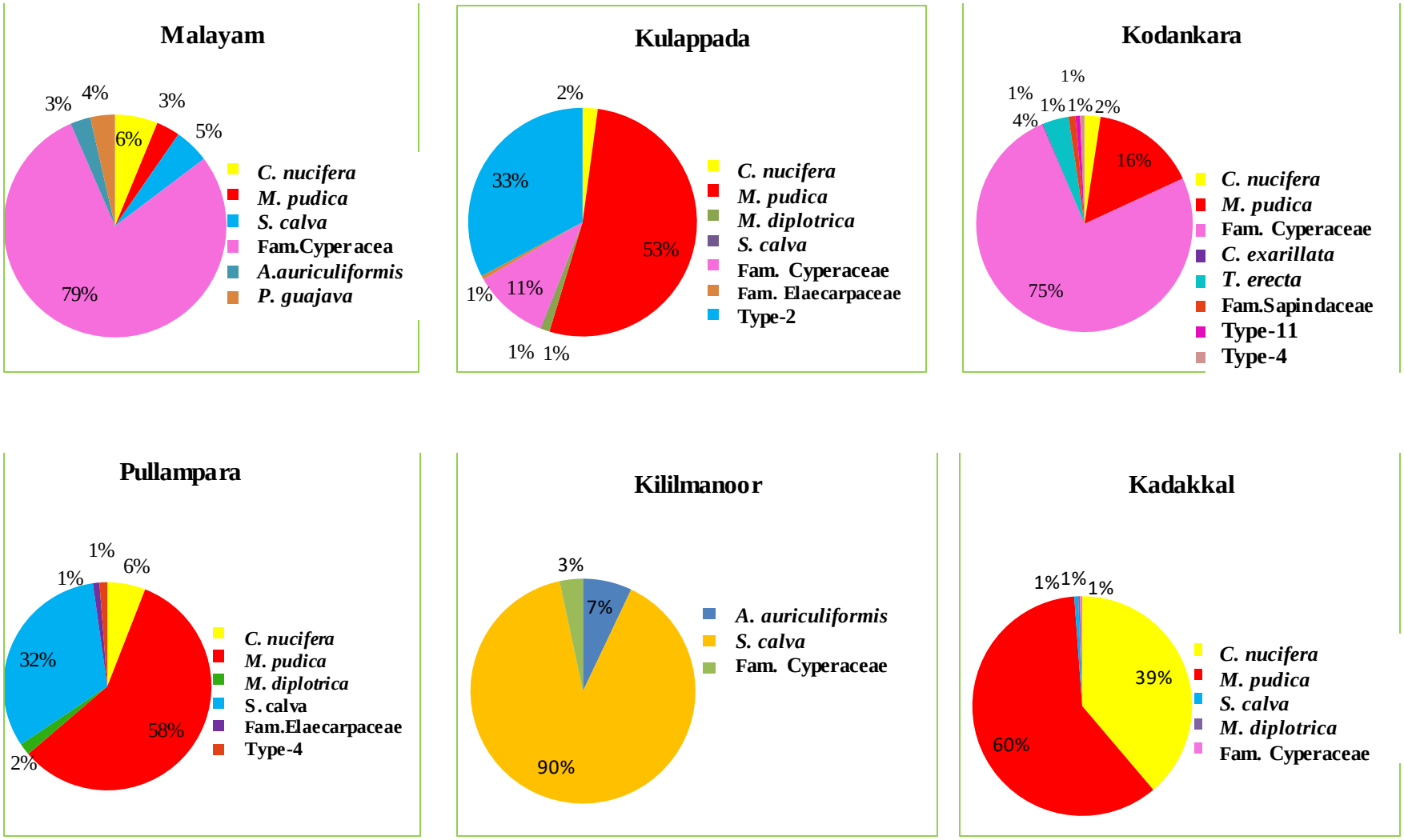


Fig.4. e. Pollen spectra of pollen loads during brood rearing seaso

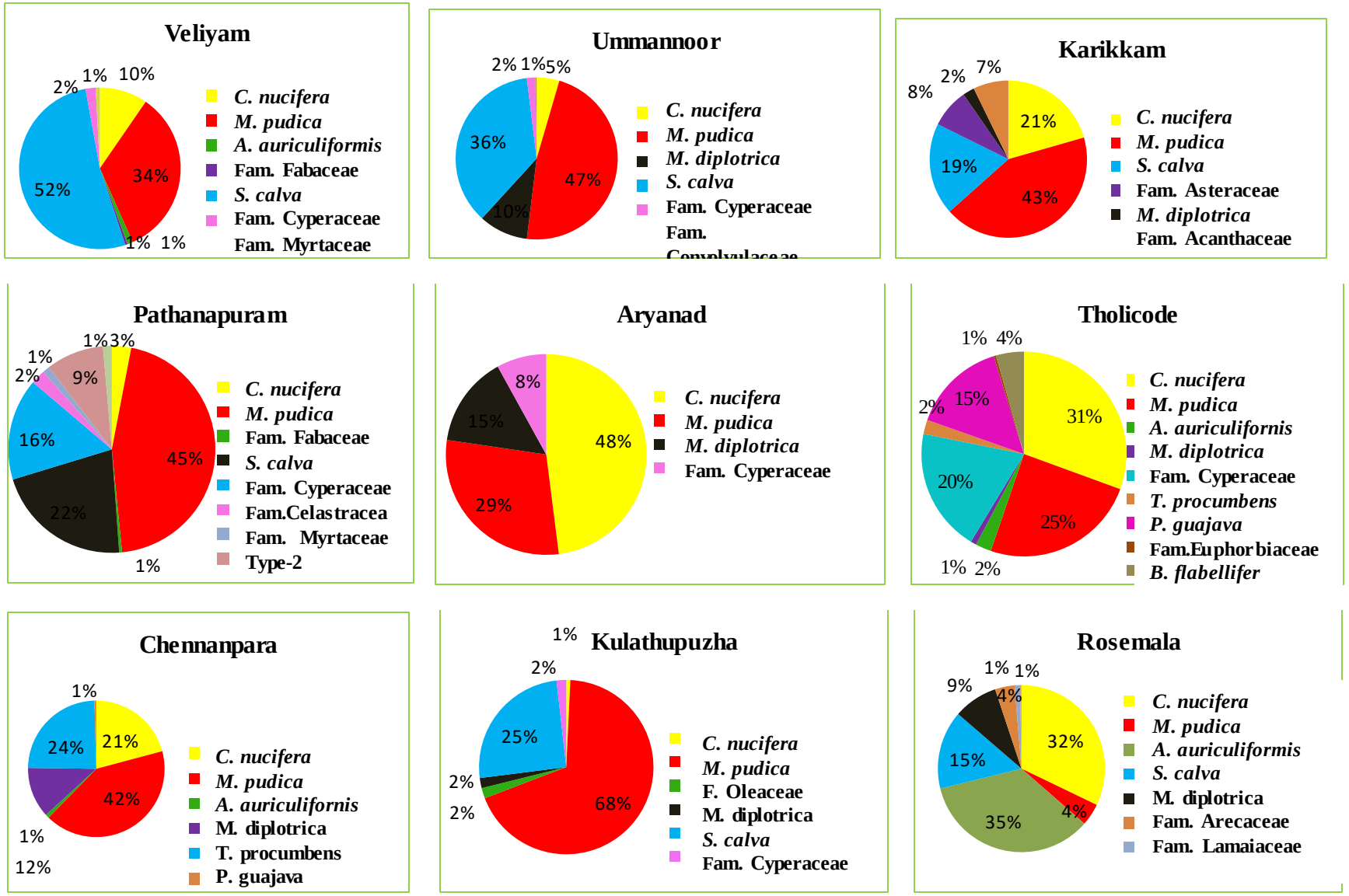


Fig. 4. e. continued

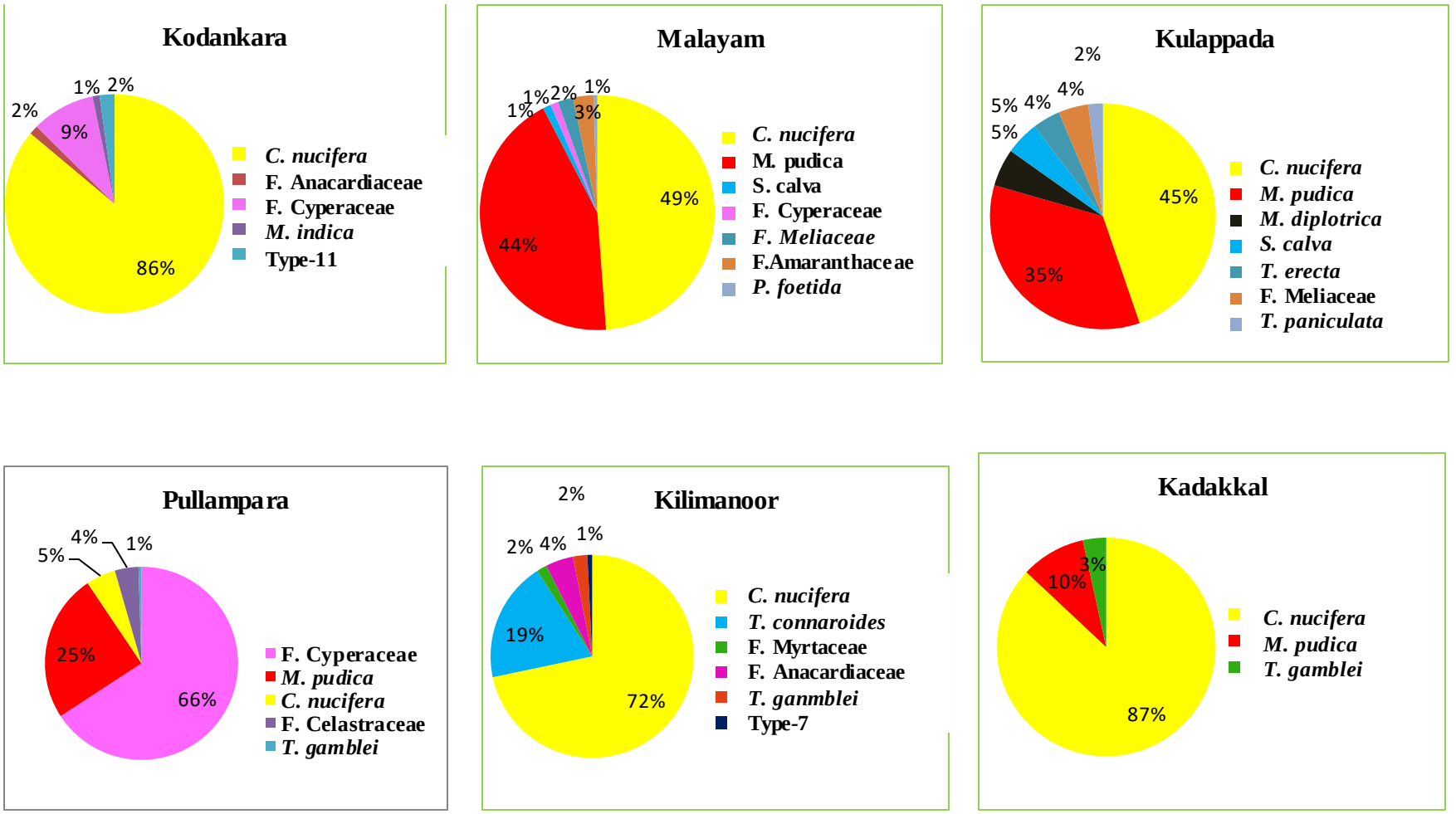


Fig. 4. f. Pollen spectra of pollen loads during honey flow seaso

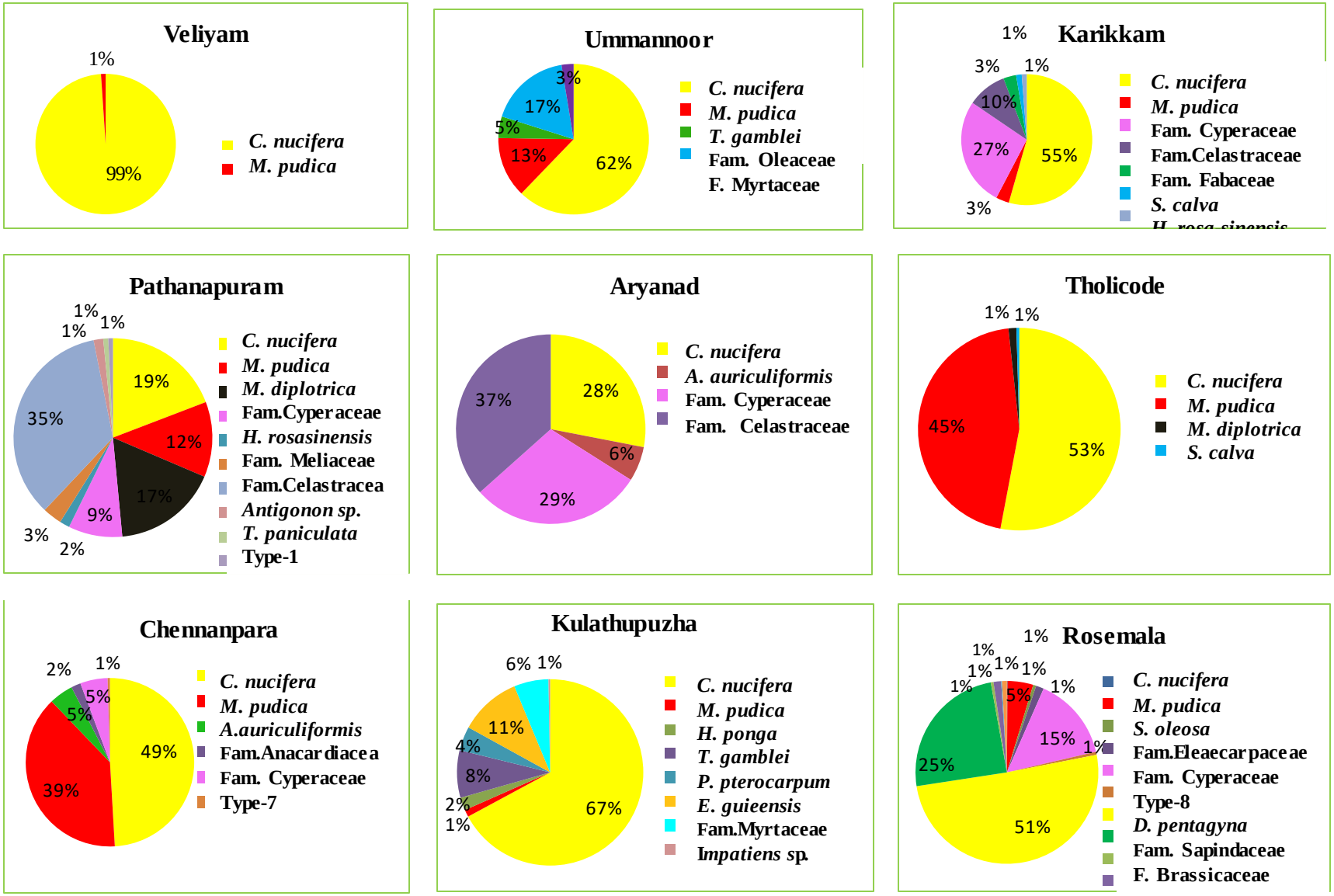


Fig.4. f. continued

4.4 DETERMINATION OF FREQUENCY OF OCCURRENCE OF POLLEN TYPES

A total of 69 pollen types were recognized from honey and pollen loads during all the seasons.

4.4.1 Frequency of occurrence of pollen types in honey samples in three seasons from various locations

Frequency of occurrence of each pollen types in honey samples of fifteen locations revealed that pollen types of *C. nucifera* and *M. pudica* occurred very frequently in all the three seasons. These pollen types were recognized from more than eight locations (Table 10).

No pollen types were recorded under the category 'frequent' (pollen types which occurred in more than 4-7 locations) during dearth season while an unidentified pollen (Type-12) was recorded as infrequent pollen (pollen types which occurred in 2 locations). Pollen types of *C. esculenta, M. calabura, P. foetida, T. paniculata, M. diplotrica,* and members of the families Lamiaceae, Myrsinaceae (P-51), Fabaceae (P-46), Elaecarpaceae, Type-8 and Type-9 occurred rarely.

Pollen types of *A. auriculiformis, M. diplotrica, S. calva*, and the plant species belonging to the families Asteraceae (P-37) and Cyperaceae were found to occur frequently (pollen types which occurred in more than 4-7 locations) during brood rearing season whereas *T. erecta, Rosa* sp., *B. flabellifer* and pollen grain from the plants in Asteraceae (P-36) occurred infrequenty. The rare pollen types (pollen types which occurred only in 1 locations) recorded during this season were *M. calabura, T. procumbens, T. paniculata* and plant species from families Lamiaceae and Sapindaceae (P-56).

Table 10. Frequency of occurrence of different pollen types in honey samples over the three seasons

Seasons	Very Frequent	Frequent	Infrequent	Rare
Dearth season	*C. nucifera* *M. pudica*	-	Type-12	*C. esculenta* *M. calabura* *M. diplotrica* *P. foetida* *T. paniculata* Fam. Lamiaceae Fam. Myrsinaceae (P-51) Fam. Fabaceae (P-46) Fam. Elaecarpaceae Type-8 Type-9
Brood rearing season	*C. nucifera* *M. pudica*	*A. auriculiformis* *M. diplotrica* *S. calva* Fam. Asteraceae (P-37) Fam. Cyperaceae	*B. flabellifer* *Rosa* sp. *T. erecta* Fam. Asteraceae (P-36)	*M. calabura* *T. procumbens* *T. paniculata* Fam. Lamiaceae Fam. Sapindaceae (P-56)
Honey flow season	*C. nucifera* *M. pudica* Fam. Cyperaceae	*A. auriculiformis* *Rosa* sp. Type-11	*M. diplotrica* *P. guajava* Fam. Arecaceae (P-34) Fam. Sapindaceae(P-57) Type-6	*Bombax* sp. *D. pentagyna* *H. ponga* *M. indica* *S. calva* *T. gamblei* Fam. Myrtaceae (P-53) Fam. Amaranthaceae Fam. Celastraceae Fam. Elaecarpaceae Fam. Oleaceae (P-54) Type-1 Type-5 Type-8 Type-10

In honey flow season, pollen types from family Cyperaceae also occurred very frequently apart from that of *A. auriculiformis, C. nucifera* and *M. pudica*. *Rosa* sp., and an unidentified pollen (Type-11) were found to occur frequently while pollen types of *M. diplotrica, P. guajava,* plants belongs to families Arecaceae (P-34), Sapindaceae (P-57), and an unidentified pollen type (Type-6) occurred infrequently. The rare pollen types were *Bombax* sp., *D. pentagyna, H. ponga, M. indica, S. calva, T. gamblei,* plants of families Amaranthaceae, Celastraceae,

Elaecarpaceae, Myrtaceae (P-53), Oleaceae (P-54), and four unidentified pollen types (Type 1, Type 5, Type 8 and Type 10) during honey flow season.

4.4.2 Frequency of occurrence of pollen types in pollen loads over all the three seasons

Frequency of occurrence of each pollen types in pollen loads during the three seasons is illustrated in Table 11. Pollen types of *C. nucifera* and *M. pudica* occurred very frequently in the pollen loads collected during all the seasons. Apart from these, pollen types which belong to the category 'very frequent' were *M. diplotrica* and *S. calva* during brood rearing season and a member of family Cyperaceae during honey flow season.

A. auriculiformis was found to be the frequently occurring pollen types in both dearth season and brood rearing season while in honey flow season it was recorded as the 'infrequent' pollen type. Other frequently occurring pollen types were of plant species belonging to families Convolvulaceae (P-41) during dearth season, Cyperaceae during brood rearing season and *M. diplotrica, S. calva, T. gamblei,* plants of families Celastraceae, Meliaceae during honey flow season. During dearth season pollen types of *H. rosasinensis,* I*mpatiens* sp. and an unidentified pollen (Type -9) occurred infrequently while that of *P.guajava, T. erecta* and member of families Elaecarpaceae, Fabaceae (P-45), Myrtaceae (P-52), Sapindaceae (P-56) and two unidentified pollen types (Type-2 and Type-4) occurred in brood rearing

Table 11. Frequency of occurrence of different pollen types in pollen loads over the three seasons

Seasons	Very frequent	Frequent	Infrequent	Rare
Dearth season	*C. nucifera* *M. pudica*	*A. auriculiformis* Fam. Convolvulaceae (P-41)	*H. rosa-sinensis* *Impatiens* sp. Type-9	*C. esculenta* *M. calabura* Fam.Anacardiaceae (P-31) Fam.Asteraceae (P-35) Fam.Elaecarpaceae Fam.Fabaceae (P-44) Fam.Flacourtiaceae Fam.Lamiaceae Fam.Myrsinaceae (P-50) Fam.Myrtaceae (P-52) Fam.Myrtaceae (P-53) Fam.Oleaceae (P-54) Type-8
Brood rearing season	*C. nucifera* *M. pudica* *M. diplotrica* *S. calva*	*A. auriculiformis* Fam.Cyperaceae	*P.guajava* *T. erecta* Fam.Elaecarpaceae Fam.. Fabaceae (P-45) Fam.Myrtaceae (P-52) Fam.Sapindaceae (P-56) Type-2 Type-4	*B. flabellifer* *C. exarillata* *P. paucifolium* *T. procumbens* Fam.Acanthaceae Fam.Anacardiaceae (P-31) Fam.Arecaceae (P-33) Fam. Asteraceae (P-36) Fam.Asteraceae (P-37) Fam.Celastraceae Fam.Convolvulaceae (P-40) Fam.Convolvulaceae (P-41) Fam.Euphorbiaceae Fam.Lamiaceae Fam.Oleaceae (P-54) Type-11
Honey flow season	*C. nucifera* *M. pudica* Fam.Cyperaceae	*M. diplotrica* *S. calva* *T. gamblei* Fam.Celastraceae Fam.Meliaceae	*A. auriculiformis* *H. rosa-sinensis* *T. paniculata* Fam.Anacardiaceae (P-31) Fam.Myrtaceae (P-52)	*Antigonon* sp. *D. pentagyna* *E. guineensis* *H. ponga* *Impatiens* sp. *M. indica* *P. foetida* *P. pterocarpum* *S. oleosa* *T. connaroides* *T. erecta* Fam.Amaranthaceae Fam.Anacardiaceae (P-32) Fam.Brassicaceae Fam.. Elaecarpaceae Fam.Fabaceae (P-45) Fam.Myrtaceae (P-53) Fam.Oleaceae (P-55) Fam.Sapindaceae (P-57) Type-1 Type-3 Type-7 Type-8 Type-11 Type-12

season. Honey flow season had infrequently occurring pollen types *A. auriculiformis, H. rosasinensis, T. paniculata,* and the pollen types from the families Anacardiaceae (P-31) and Myrtaceae (P-52).

Pollen grains of *C. esculenta, M. calabura,* members of families Anacardiaceae (P-31), Asteraceae (P-35), Elaecarpaceae, Fabaceae (P-44), Flacourtiaceae, Lamiaceae, Myrsinaceae (P-50), Myrtaceae (P-52 & P-53), Oleaceae (P-54) and two unidentified types (Type-8) occurred rarely during dearth season whereas pollen types of *B. flabellifer, C. exarillata, P. paucifolium, T. procumbens,* and plants belonging to the families Acanthaceae, Anacardiaceae (P-31), Arecaceae (P-33), Asteraceae (P-36 & P-37), Celastraceae, Convolvulaceae (P-40 & P-41), Euphorbiaceae, Lamiaceae, Oleaceae (P-54) and an unidentified pollen type (Type-11) occurred in brood rearing season. During honey flow season, pollen types of *Antigonon* sp., *D. pentagyna, E. guineensis, H. ponga , Impatiens* sp., *M. indica, P. foetida, P. pterocarpum, S. oleosa, T.connaroides, T. erecta* and members of families Amaranthaceae, Anacardiaceae (P-32), Brassicaceae, Elaecarpaceae, Fabaceae (P-45), Myrtaceae (P-53), Oleaceae (P-55), Sapindaceae (P-57) and six unidentified pollen types (Type-1, 3, 7, 8, 11 and 12) were recorded as rare pollen types.

C. nucifera and *M. pudica* were the most frequently occurring pollen types in all the three seasons, whereas *A. auriculiformis* was the frequently occurring pollen grain in both dearth and brood rearing seasons. Other than this, very frequently occurred pollen types were from *M. diplotrica, S. calva,* plants under the family Cyperaceae during brood rearing season and family Cyperaceae in honey flow season. Frequently occurred pollen types during honey flow season were *S. calva, T. gamblei* and plants belonging to Celastraceae family.

4.5 SEASONAL VARIATIONS OF POLLEN TYPES IN VARIOUS LOCATIONS

The seasonal variation of pollen types in various locations are given in table 12. There was substantial difference in pollen types identified from 15 locations over the seasons. Maximum number of pollen types (46 nos.) was recorded in honey flow season followed by brood rearing season (33 nos.) and dearth season (26 nos.). Pollen types which were present in samples of all the three seasons included *A. auriculiformis, C. nucifera, M. pudica, M. diplotrica, T. paniculata* and each pollen type from members of the families Oleaceae (P-54), Fabaceae (P-45), Anacardiaceae (P-31), Myrtaceae (P-52) and Elaecarpaceae. Pollen types of *H. rosasinensis*, *Impatiens* sp., *P. foetida* and two unidentified pollen (type-8 and type-12) were recorded in samples collected during both dearth and honey flow season. Pollen types observed in samples during both dearth and brood rearing season were *M. calabura*, plants from the families Convolvulaceae (P-41) and Lamiaceae while *P.guajava, Rosa* sp., *S.calva, T.erecta,* from the plants of families Celastraceae, Cyperaceae and an unidentified (Type 11) occurred during brood rearing and honey flow season.

Other pollen types which occurred during dearth season were *C. esculenta*, plants of the families Flacourtiaceae, Fabaceae (P-45 & P-46), Asteraceae (P-35), two unidentified pollen (Type-2 and Type-9) and two pollen types from the family Myrsinaceae (P-50 & P-51) while those occurred in brood rearing season included pollen grains of *B. flabellifer, C. exarillata, P. paucifolium, T. procumbens* and from plants belonging to family Acanthaceae, Arecaceae (P-33), two pollen types of Asteraceae (P-36 & P-37), Convolvulaceae (P-40 & P-41), Euphorbiaceae, Sapindaceae (P-56) and two unidentified pollen grains (Type 2 and Type 4). Pollen grains which were found exclusively during honey flow season was pollen types of *Antigonon* sp., *Bombax* sp., *D. pentagyna, E. guineensis, H. ponga, M. indica, P. pterocarpum, S. oleosa, T. gamblei, T. connaroides,* each members of familis Anacardiaceae (P-31 & P-32), Amaranthaceae, Arecaceae (P-34), Brassicaceae, Meliaceae, Oleaceae (P-54 & P-55), Sapindaceae (P-57) and six unidentified pollen (Type-1, 3, 5, 6, 7 and 10).

Table 12. Seasonal occurrence of pollen types

Sl. No.	Dearth season	Brood rearing season	Honey flow season
1.	*A. auriculiformis*	*A. auriculiformis*	*A. auriculiformis*
2.	*C. nucifera*	*B. flabellifer*	*Antigonon* sp.
3.	*C. esculenta*	*C. nucifera*	*Bombax* sp.
4.	*H. rosa-sinensis*	*C. exarillata*	*C. nucifera*
5.	*Impatiens* sp.	*M. diplotrica*	*D. pentagyna*
6.	*M. diplotrica*	*M. pudica*	*E. guineensis*
7.	*M. calabura*	*M. calabura*	*H. rosa-sinensis*
8.	*M. pudica*	*P. paucifolium*	*H. ponga*
9.	*P. foetida*	*P. guajava*	*Impatiens* sp.
10.	*T. paniculata*	*Rosa* sp.	*M. diplotrica*
11.	F. Asteraceae (P-35)	*S. calva*	*M. pudica*
12.	F. Anacardiaceae (P-31)	*T. erecta*	*M. indica*
13.	F. Convolvulaceae (P-41)	*T. procumbens*	*P. foetida*
14.	F. Elaecarpaceae	*T. paniculata*	*P. pterocarpum*
15.	F. Fabaceae (P-45)	F. Acanthaceae	*P. guajava*
16.	F Fabaceae (P-46)	F. Anacardiaceae(P-31)	*Rosa* sp.
17.	F. Flacourtiaceae	F. Arecaceae (P-33)	*S. oleosa*
18.	F. Lamiaceae	F. Asteraceae (P-36)	*S. calva*
19.	F. Myrsinaceae (P-50)	F. Asteraceae (P-37)	*T. gamblei*
20.	F. Myrsinaceae (P-51)	F. Celastraceae	*T. erecta*
21.	F. Myrtaceae (P-52)	F. Cyperaceae	*T. paniculata*
22.	F. Myrtaceae (P-53)	F. Convolvulaceae (P-40)	*T. connaroides*
23.	F. Oleaceae (P-54)	F. Convolvulaceae (P-41)	F. Anacardiaceae (P-31)
24.	Type-8	F. Elaecarpaceae	F. Anacardiaceae (P-32)
25.	Type-9	F. Euphorbiaceae	F. Arecaceae (P-34)
26.	Type-12	F. Fabaceae (P-45)	F. Amaranthaceae
27.	-	F. Lamiaceae	F. Brassicaceae
28.	-	F. Myrtaceae (P-52)	F. Celastraceae
29.	-	F. Oleaceae (P-54)	F. Cyperaceae
30.	-	F. Sapindaceae (P-56)	F. Elaecarpaceae
31.	-	Type-2	F. Fabaceae (P-45)
32.	-	Type-4	F. Meliaceae
33.	-	Type-11	F. Myrtaceae (P-52)
34.	-	-	F. Myrtaceae (P-53)
35.	-	-	F. Oleaceae (P-54)
36.	-	-	F. Oleaceae (P-55)
37.	-	-	F. Sapindaceae (P-57)
38.	-	-	Type-1
39.	-	-	Type-3
40.	-	-	Type-5
41.	-	-	Type-6
42.	-		Type-7
43.	-	-	Type-8
44.	-	-	Type-10
45.	-	-	Type-11
46.	-	-	Type-12

4.6 CORRELATION COEFFICIENT BETWEEN POLLEN DENSITY AND POLLEN TYPES

Statistical analysis on the correlation between pollen density and pollen types of honey samples during the tree seasons revealed that there was no significant positive correlation between pollen density and pollen types in dearth season (0.03) and brood rearing season (0.12). A negative but was not statistically significant colleration was recorded during honey flow season (-0.06).

4.7 CLUSTER ANALYSIS

Cluster analysis of different locations based on the presence or absence of pollen types recorded from individual samples resulted in four major clusters (Fig. 5). Pullampara, Aryanad, Karikkam and Tholicode were grouped under cluster 1 whereas the locations Veliyam, Chennanpara and Kodankara under cluster 2. Cluster 3 comprised of the locations Kadakkal, Ummannoor, Kilimanoor and Kulathupuzha while cluster 4 had Malayam, Kulappada, Pathanapuram and Rosemala (Table 13).

Cluster analysis of different locations showed that there was not much difference in floristic composition of midland and upland physiographic region and also between the two districts, Thiruvananthapuram and Kollam. Clusters 1 consists only one location from Kollam district, Karikkam. It was observed that the floristic composition of locations, Pullampara and Karikkam which was from the midland region is similar with that of the upland region, Ayanad and Tholicode. In cluster 2, of the three locations, Veliyam and Kodankara were from midland region whereas Chennanpara was from upland area. Chennanpara and Kodankara belong to Thiruvananthapuram district but Veliyam belongs to Kollam district. In cluster 3, Kadakkal, Ummannoor and Kilimanoor were from the midland while Kulathupuzha was from upland area. Among these four locations, Kadakkal, Ummannoor and Kulathupuzha belongs to Kollam district while Kilimanoor to Thiruvananthapuram district. The locations, Malayam and Kulappada (midland region) of Thiruvananthapuram district; Pathanapuram (midland) and Rosemala (upland) of Kollam district were grouped under Cluster 4.

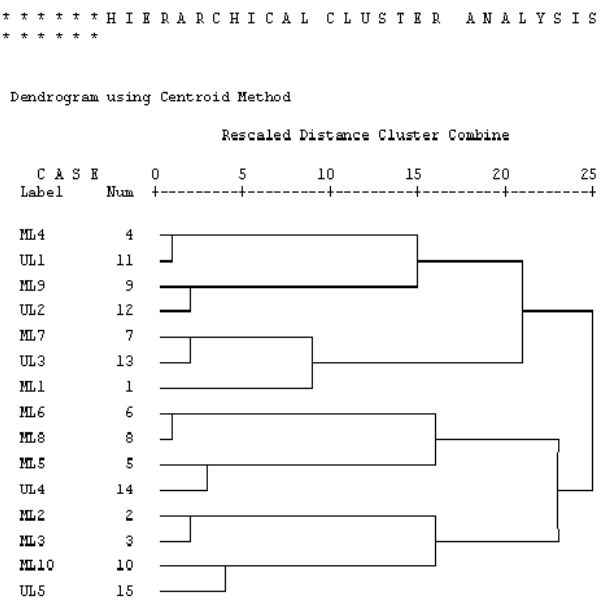


Fig. 5 Dendrogram showing cluster analysis of locations based on pollen types

Table 13. Distribution of locations based on cluster analysis of locations

Cluster No.	Location			
Cluster I	Pullampara	Aryanad	Karikkam	Tholicode
Cluster II	Veliyam	Chennanpara	Kodankara	-
Cluster III	Kadakkal	Ummannoor	Kilimanoor	Kulathupuzha
Cluster IV	Malayam	Kulappada	Pathanapuram	Rosemala

DISCUSSION

5. DISCUSSION

Investigations were conducted at the Department of Agricultural Entomology, College of Agriculture, Vellayani to identify the potential pollen and nectar sources of Indian honey bee across the seasons in southern Kerala during the period 2011-2013. In this chapter, attempt is being made to discuss salient experimental findings.

The study included pollen analysis of honey samples collected during the three seasons namely, dearth season, brood rearing season and honey flow season.

5.1 POLLEN DENSITY OF HONEY SAMPLES

In the present study, significant variation in mean pollen density of honey samples (Table 3.) was recorded between locations within the seasons, between midland and highland andalso between seasons.

Considering the physiographic zones, maximum mean pollen density was recorded from both the midland and upland during the brood rearing season where as during the other seasons where the lowest mean pollen density was recorded during dearth season in midland and honey flow season in the upland. The increased pollen density during dearth season in upland indicates either the foraging activity of bees must be higher in upland compared to midland or there may be the variation in raining time between the two lands. Increased pollen density from honey samples of forested highland of Thiruvananthapuram district was also reported by Nair (2005).

Pollen density was also found to be greatly varied in all the three seasons. This might be due to the difference in floral diversity and foraging activity of honey bees with seasons which indicates the influence of climate on the foraging behavior of bees.

Among the three seasons, honey samples during brood rearing season showed maximum mean pollen density whereas dearth season recorded lowest mean pollen density. Pollen being an essential ingredient for bee nutrition, brood development and for maintenance of a healthy bee colony; is rich in proteins, amino acids, carbohydrates, vitamins and hormones. In order to meet its dietary requirements of brood the foraging bees will collect ample quantity of pollen which results in higher pollen density during brood rearing season. The importance of pollen as major source of protein to the brood has been also reported from the studies conducted by Lin *et al.*1990; Garg, 1996; Sadia *et al.* 2008.

During dearth season, because of the rain honey bees could not forage the wide range of plants for nectar and pollen which leads to the low pollen density in honey samples collected in the present study. This is in accordance with the conclusions derived by Chaturvedi and Temsunungula (2008) where he suggested that due to rainy season in Nagaland honey bees could not forage the wide range of plants for nectar and pollen during autumn season but during the onset of winter season these bee collect pollen and nectar from a wide range of plants which reflects in the honey samples of winter season. Apart from that, beekeepers follow the practice of providing sugar syrup as artificial food during dearth season which in turn decreases the chance of getting pollen into honey. The situation gets altered as the season changes, especially during the honey flow season (January-April) where the bees collect pollen and nectar from a wide range of plants which is reflected in the honey samples.

The variation in pollen density between the locations and locations within the seasons might be due to the diversity and richness of bee flora in different locations and also the preference of bees with locations. Similar conclusions have been reported by Sa-otera*et al.* 2006; Patnaik*et al.* 2008; Bhargava *et al.* 2009; Shubharani*et al.* 2012 and which confirmed that number of pollen grains per ml of

honey appeared to be varying from place to place depending upon the plant diversity of an area.

Among the locations, Pathanapuram recorded significantly high mean pollen density. It was followed by Kodankara, Karikkam and Pullampara while the lowest mean pollen density was recorded in Veliyam. This indicates that the location Pathanapuram is enriched with abundant flora with maximum diversity of plants, rather than rubber compared to other locations. Apart from these, the quantity of pollen collected by the foraging bees from preferred plants will be high. Lowest pollen concentration in Veliyam indicates the poor pollen diversity around that location.

5.2 POLLEN IDENTIFICATION AND CHARACTERIZATION

Pollen grains identified by analysis from samples of fifteen locations enabled to determine the botanical variations of the respective source area. Pollen grains of different plants were present in each honey and pollen load samples and the number of plants involved in honey production varied in different samples. The pollen grains identified in the samples exhibited different shapes and sizes and also had various types of aperture and exine thickness.

A detailed characterization of all the honey and pollen samples from different agro ecological situations showed the preference of honey bees to 69 different species of plants which were found to be mostly from the native species. This can be confirmed from the studies conducted by Nair (2005) who reported that inspite of the abundance of exotic, ornamental species; bees showed a strict avoidance of these exotic food resources and preferred native species like *C. nucifera, M. pudica, Elaeocarpusserratus*etc in wild habitats of Western ghats. The identified plants in this study included local flora such as *C. nucifera, M. pudica, M. diplotrica, T. erecta, A. auriculiformis*etc. Along with local plants like *C. nucifera*, weeds like *M. pudica*, wild plant species such as *D. pentagyna, H. ponga, S. oleosa, C.exarillata*were also

recorded. The presence of wild plant pollens may be because of the nearness of apiaries to forested areas in highlands. This was also confirmed by Chakraborti and Bhattacharya (2011) where he reported the presence of wild plants such as *Syzygium* sp., *Eucalyptus* sp., *Cassia* sp. and *Terminalia* sp. in honey samples from West Bengal and Orissa.

Though most of the apiaries are concentrated around the rubber plantations, no rubber pollen was present in any of the sample collected during the three seasons. This is because of the fact that honey bees collect nectar from the extra floral nectaries of rubber so the chance of occurrence of rubber pollen in honey samples is very less. This finding was in line with that of Kumar and Jagtap (2005) who reported that *Heveabrasiliensis* will be under represented in the samples due to the production of the nectar in the extra floral nectaries on the petioles of newly emerging leaves.

In the present study, 24 pollen types were identified up to species level which comprised of 13 trees, six herbs, two shrubs and one climber. Tree species was found to be dominant (Fig. 3) which included trees such as *C. nucifera, A. auriculiformis, C. exarillata, D. pentagyna H. ponga, S. oleosa, M. indica,P. pterocarpum, E. guineensis, P. guajava, T. paniculata* and *B. flabellifer*. This is in accordance with Mishra (1995) which suggested that trees present large number of flowers as compared to bushes, shrubs and crop plants unless the latter are growing in large continuous areas. Similar findings were reported by Sharma (2011) who reported that bees highly prefers trees followed by herbs and Shubharani*et al.* 2012 which reported the dominance of tree species (53.84 %) from samples of Coorg honey.

Preference of honey bees to plants with different habit (trees, herbs, shrubs) also indicated that height of the plant is not a barrier to bees for collecting pollen and nectar as they are found to visit both the tall trees (*C. nucifera*) and small plants (*M. pudica).* Results of the studies conducted by Adekanbi and Ogundipe (2009) also reported that height of the trees is not a barrier for bees as they are found to visit

Tridaxprocumbens, which are a few centimeters in height, along with tall trees such as *E. guineensis* in regions of Nigeria. Thus the bees satisfy their dietary requirements from the preferred sources in and around the apiary irrespective of the plant height.

Fifty seven plants identified up to family level were found to be distributed among 32 families. Of these, the pollen types from the families of Asteraceae, Mimosaeae and Arecaceae were best represented in honey and pollen load samples. Maximum contribution of pollen types was from plants belonging to the family, Asteraceae which indicates the preference of honey bees to these plants for nectar and pollen sources. Similar findings were reported by Sharma, 1969; Chaubal, 1976; Garg, 1996; Chaturvedi and Temsunungula, 2008; Bhargava *et al*., 2009 reported that most of the pollen grains in honey belong to Asteraceae where as Chandra and Sharma (2011) reported this family as minor source in Uttar Pradesh.

5.3 PERCENTAGE OF POLLEN TYPES

Percentage of pollen types varied among the samples of various locations and also between the seasons.

Microscopic analysis of honey and pollen samples collected during honey flow season (Table 8 and 9) revealed that Pathanapuram and Rosemala had maximum number of pollen types. This shows that the rich and diversified natural flora of these areas which blooms during the honey flow season is easily accessible to the bees. The pollen spectra of the samples from Pathanapuram displayed varied vegetation which according to Ige and Apo (2007) more the pollen type, the more the source of nectar collection and more richness of honey.

Pollen grains of *C. nucifera* and *M. pudica*was found to be dominant during the three seasons. The relevance of plant species like *C.nucifera, M. pudica, Bombax*sp. have also been reported in the Indian honeys by Sharma, 1970;

Nair, 2005 and Bera*et al*. 2007). Apart from the contribution by these species, pollen spectrum in the present study also reflected the dominance of wild plants in some locations. Pollen spectrum was also well represented with local flora like *H. rosasinensis, T. erecta*and *T. procumbens*which may be present in the apiaries in rubber plantation. Predominance of *C. nucifera* and *M. pudica* in honey and pollen samples was also reported by Nair (2005) and Silva *et al*.(2012).

In addition to this, intermittent encounter of pollen of *S. calva*, *T. paniculata*, *A. auriculiformis* plant species belonging to families Asteraceae, Cyperaceaeetc suggests that they are the secondary source of nectar and pollen in most of the locations. This finding was in line with that of Chauhan and Singh (2010) who reported the importance of *T. paniculata* as secondary pollen.

During brood rearing season and honey flow season, honey bees showed a great preference to anemophilous plants, those of Cyperaceae, Arecaceae and *Borassus* which indicates that there is no direct correlation between the pollen collection by bees. Even nectar less anemophilous plants can also be frequented by honey. This finding was in line with that of Sharma (1970) and Jhansi *et al*.(1994) who reported that honey bee prefers highly anemophilous plants as their pollen source. Simialr findings were also reported by Chaturvedi, 1976; Jhansi and Ramanujam, 1990; Garg, 1996. However the plant species like *H. ponga*, *P. pterocarpum* and members of the families Convolvulaceae, Anacardiaceae, Sapindaceae, Fabaceae and Acanthaceae were also preferred as minor source of pollen and nectar in the present study.

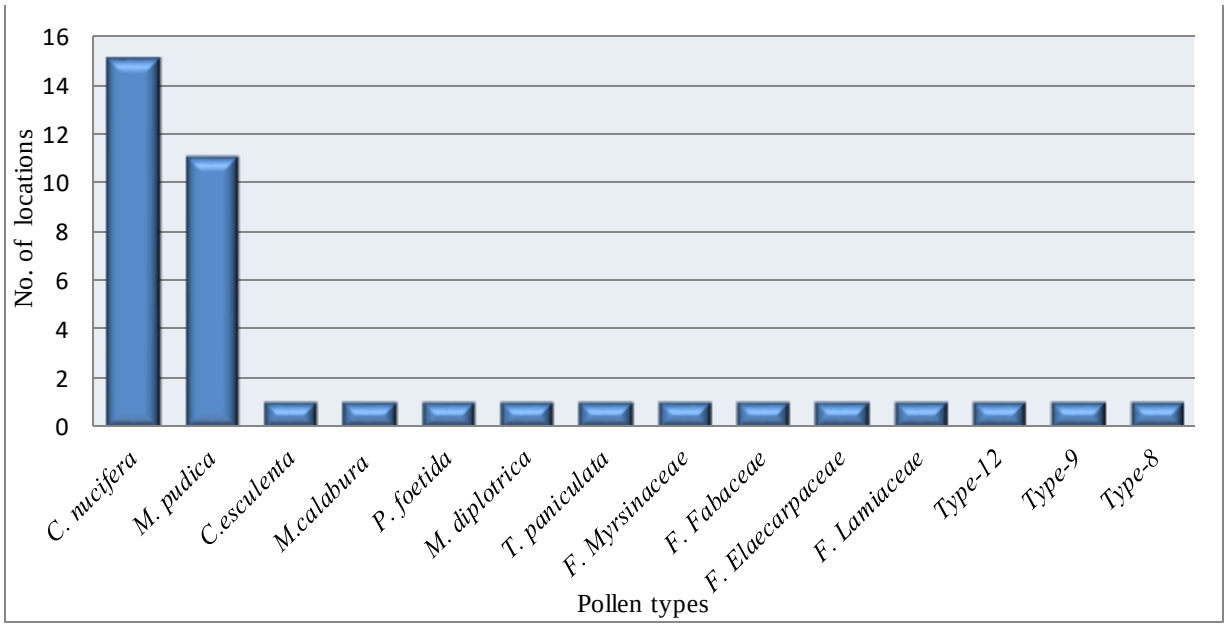
The critical analysis of pollen spectra of honey and pollen samples (Fig. 4)from fifteen locations showed diversity in resource preference by Indian honey beeduring the three seasons. The analysis of spectral combination of different honey bees revealed that almost all the samples of *A. ceranaindica* are multifloral in nature which may be due to its differential resource preference. Pollen spectra of

A. ceranaindica showed high preference to *M. pudica* and *C. nucifera*. Similar findings were also reported by Yifeng*et al.* 2006; Nair, 2005 and Scinthia, 2009 where the*A. ceranaindica*was foundto dependhighly on *C. nucifera* and *M. pudica.*

5.4 FREQUENCY OF OCCURRENCE

Of the total 69 pollen types identified, 10 plant species (*A. auriculiformis, C. nucifera, M. pudica, M. diplotrica, T. paniculata*and each pollen types from members of the families Oleaceae, Fabaceae, Anacardiaceae, Elaecarpaceae and Myrtaceae) were of either medium or major importance as source of bee forage. These pollen types occurred frequently in all the three seasons (Fig. 6, 7 and 8)which indicated as a good choice for bee forage. Pollens of *C. nucifera* and *M. pudica*occurred very frequently during all the three seasonswhile the pollens of *M. diplotrica* and Cyperaceae family during brood rearing seasonand honey flow season.

The frequency of occurrence of pollen types varied with different locations. Of these, *C. nucifera, M. pudica, M. diplotrica* and *A. auriculiformis,* are quite common in these locations. This might be the reason for frequent occurrence of these pollen types during all the seasons. *C. nucifera, M. pudica, M. diplotrica* are found to bloom throughout the year which confirms its availability throughout the seasons. However, *A. auriculiformis* which blooms during the honey flow seasonand*T. paniculata* during dearth season was also found to be the common taxa in three seasons. It is also striking to note that some locations is familiar for its large number of plants like *Antigonon*sp., *M. indica*and *M. calabura*, but their pollen is rarely encountered in the honey samples inspite of the regular visit of bees during this period. Similar findings were also reported by Chakraborti and Bhattacharya (2011).



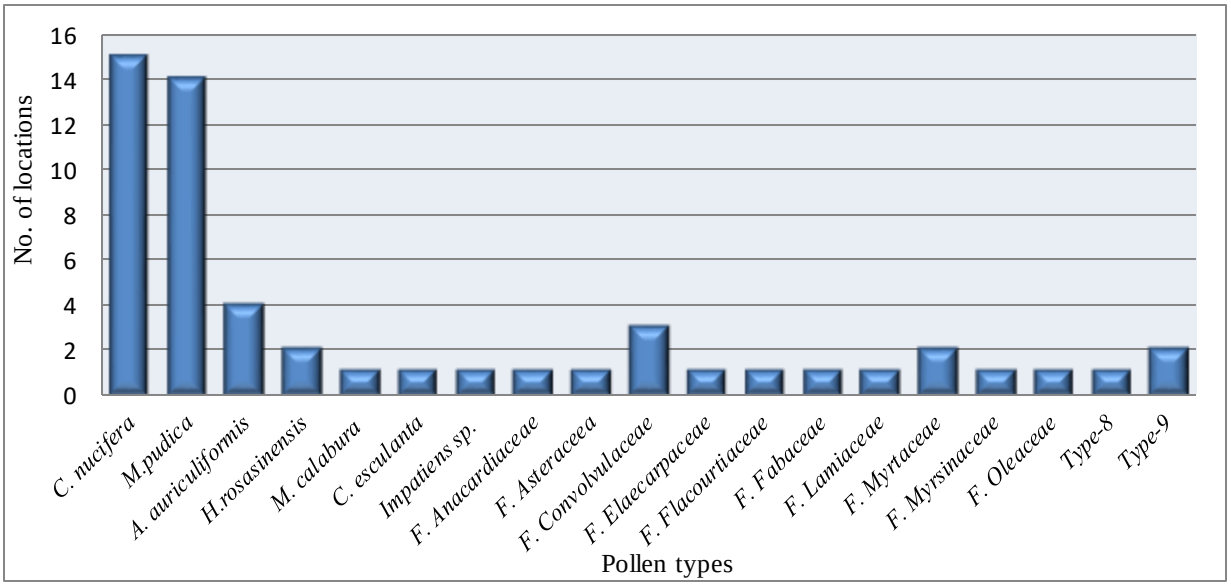


Fig. 6Frequency of occurrence of pollen types in honey and pollen samples duringdearth season

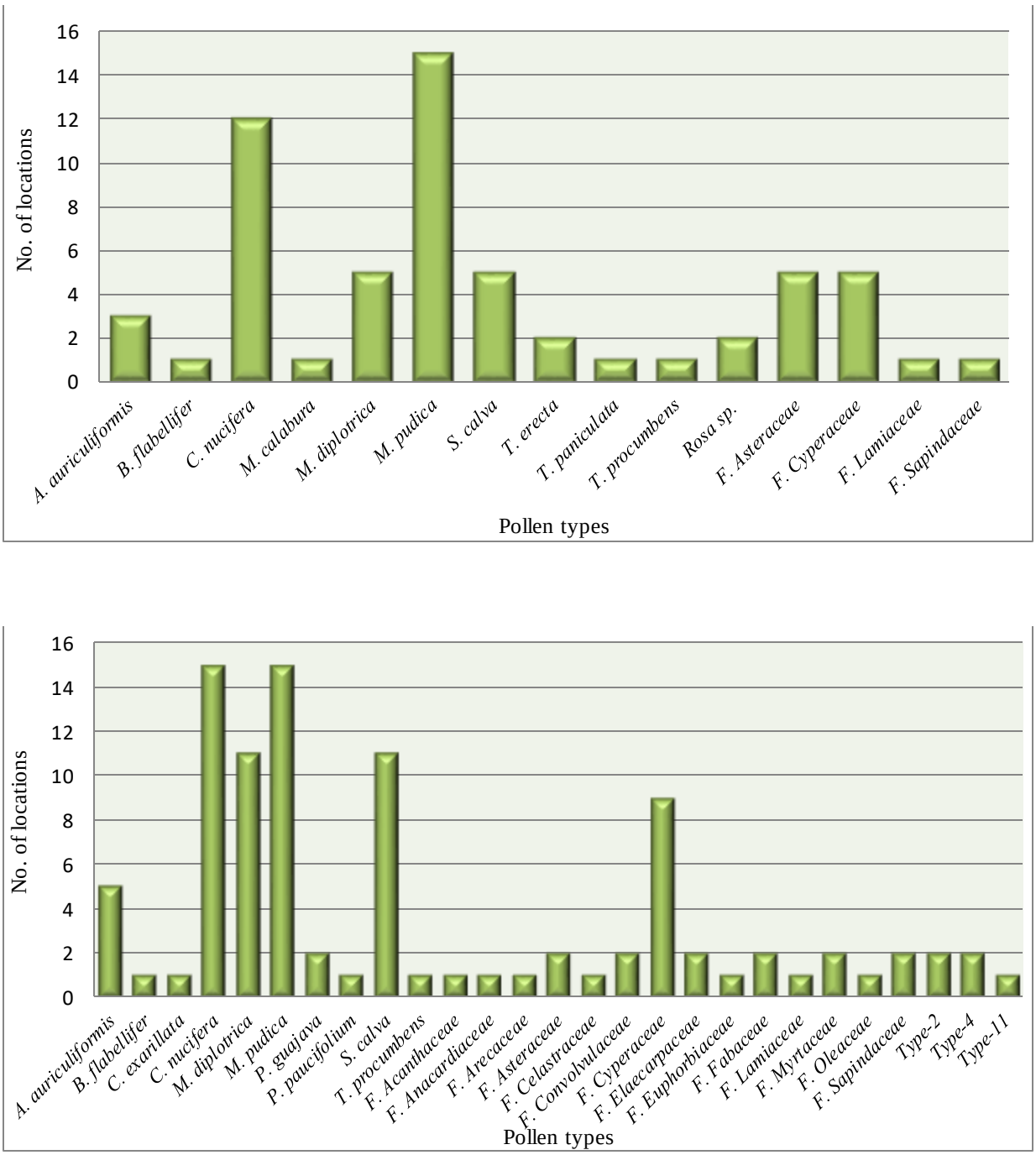
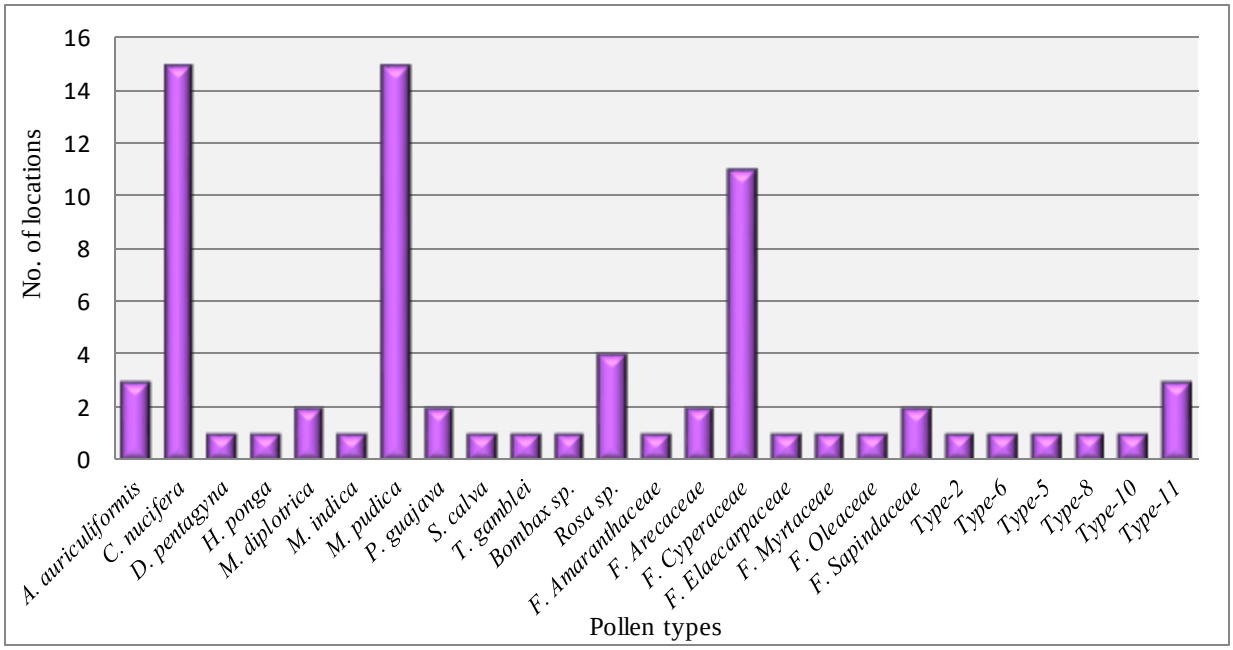


Fig. 7Frequency of occurrence of pollen types in honey and pollen samples duringbrood rearing season



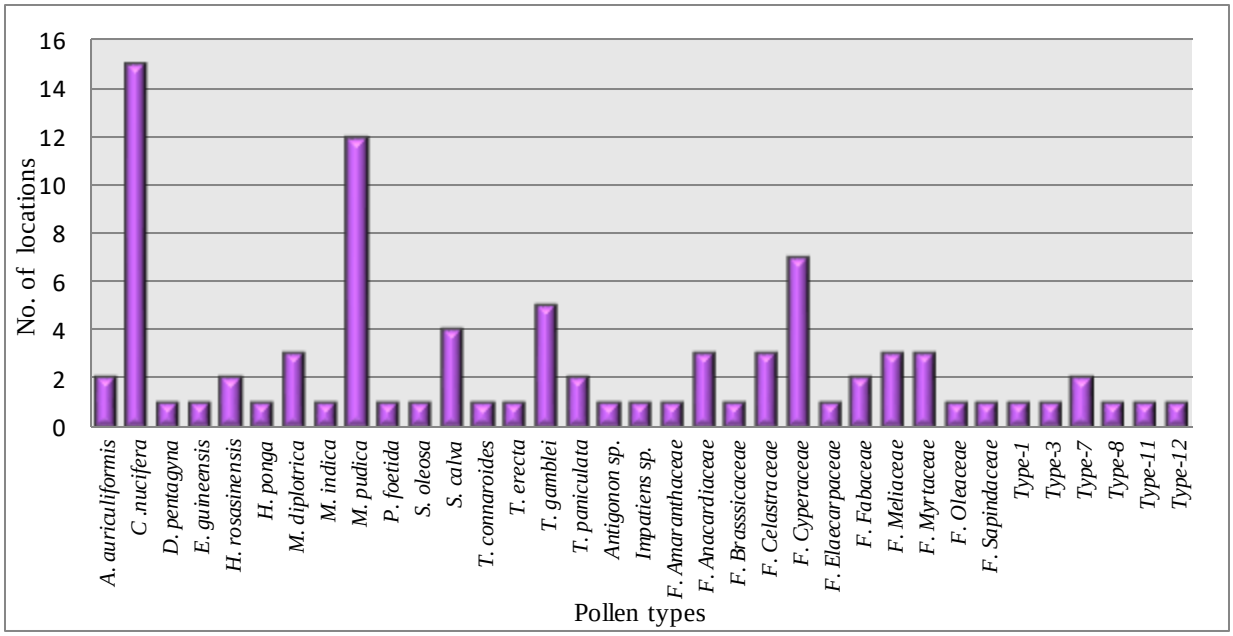


Fig.8Frequency of occurrence of pollen types in honey and pollen samples duringhoney flow season

5.5 SEASONAL VARIATION

The study of the honey samples and pollen loads provide ample evidence of the honey bee flora of the locality during the three seasons. Number of pollen types found to be varying in different seasons(Table 12). This may be due to the difference in available flora during that time. This variation in dietary preference of honey bees with different seasons were also reported by Dixit *et al.*, 2011.

Maximum number of pollen types was obtained during honey flow season followed by brood rearing season and dearth season. This quantitative variation in the presence of pollen grains during the three seasons might be due to the diversity of flowering plants and their period of anthesis in and around the apiary site. It seems that during dearth season because of rains, honey bees could not forage the wide range of plants for the nectar and pollen but with the onset of honey flow season these bees collect nectar and pollen from a wide range of plants which is reflected in the samples of honey flow season.

The presence of 69 pollen types indicated that the richness of forage to the bees in the absence of any one dominant bee flora. *C. nucifera* and *M. pudica* was found to be occurred in three seasons. The data on flowering plants indicated the availability of forage to the beesthroughout the year. Similar observations have been made by Kumar and Jagtap (2005) and Joshi and Lakshmi(2010) who reported the abundance of pollen types from different regions of Maharashtra.

5.6 CLUSTER ANALYSIS

Cluster analysis of fifteen locations indicated (Table 13) that differences among the apiaries are not well defined. In the present study it was noted that some locations under midland and upland, coming under two districts have been grouped into same cluster which indicates that demarcation of locations based on the botanical origin of honey is not possible because of the overlapping of different bee flora from

all the locations. This is because floristic composition is more or less similar to all locations which are irrelevant to physiographic regions or districts. This is in accordance with the findings of Valle *et al.*, 2007 who stated that because of the natural variability of honey it is difficult to define borders between the different honey regions.

The palynological investigation thus revealed the contribution of 69 plant species for the nectar and pollen requirement of the bees from fifteen locations during the three seasons. Critical analysis revealed that 24 different species contributed critical resources during the three seasons. Out of 69 pollen types recognized, *C. nucifera*and *M. pudica*were the predominant (>45 %) as well as the frequently occurring pollen types during all the three seasons. Pollen combination of individual samples indicated a wide variability in resource preference and resource sharing among the bees.

The present observation helps us to make some conclusions over some plant species as the major bee foraging plants. Utilisation of these critical resources during brood rearing season and honey flow season could enhance the colonial sustenance and honey production. The observations may enhance the scope for agricultural activity by exploring regional floristic resource for bee foraging and honey production throughout the year.

SUMMARY

6. SUMMARY

The study entitled 'Melissopalynology of Indian honey bee (*Apis cerana indica* Fab.) apiaries in southern Kerala ' was conducted at Department of Agricultural Entomology, College of Agriculture, Vellayani and in the apiaries of bee keepers (Indian bee) from Thiruvananthapuram and Kollam districts during the period 2011- 2013.

Samples were collected from fifteen apiaries with minimum ten hives from two districts of southern Kerala, Thiruvananthapuram and Kollam. Ten apiaries were selected from the midland and five apiaries from the highland purposively. Three samples each of honey and pollen load were collected from the selected apiaries during three seasons namely; honey flow season (January to April), dearth season (June to September) and brood rearing season (October to December).

Observations were recorded on pollen density of honey samples, pollen identification and characterization, percentages of pollen types, pollen frequency classes, frequency of occurrence and seasonal variation of pollen types. The data collected were analysed, presented in tables and figures discussed in previous chapters. The findings of the study are summarised below.

- Mean pollen density of honey samples were found significantly different between the seasons, locations and locations within the seasons. Maximum mean pollen density was recorded in midland during the brood rearing season (813.24 grains/ml) and honey flow season (750.26 grains/ml) whereas maximum mean pollen density was recorded in dearth season (304.36 grains/ml) in upland.

- Mean pollen density were found significantly different was between the midland and upland physiographic regions. Maximum mean pollen density was recorded from both the midland and upland during the brood rearing season where as the lowest

mean pollen density was recorded in dearth season from midland and during honey flow season in the upland.

- Among the locations, Pathanapuram (1438.77 grains/ml) of midland physiographic region had maximum mean pollen density while Veliyam (132.41grains/ml) in midland recorded lowest mean pollen density.

- Maximum mean pollen density during dearth season was recorded from Kulathupuzha (965.32 grains/ml) while the lowest mean pollen density was recorded in Ummannoor (28.09 grains/ml). During brood rearing season, maximum mean pollen density was recorded from the location, Kodankara (1957.83 grains/ml) while the lowest mean pollen density was recorded in honey samples from Veliyam (71.38 grains/ml). Maximum mean pollen density in honey flow season was recorded in Pathanapuram (2132.23 grains/ml) while the lowest mean pollen density was recorded from honey samples of Kilimanoor (83.55 grains/ml).

- A total of 69 plants were recorded as foraging plants by bees from the study area (15 locations) over the three seasons. Twenty four pollen types were identified up to species level, four only up to genus level and twenty nine up to family level. Twelve pollen types were unidentified.

- Identified plant species (24) were *Cocos nucifera, Mimosa pudica, Acacia auriculiformis, Mimosa diplotrica, Spilanthes calva, Elaeis guineensis, Cullenia exarillata, Tagetes erecta, Tridax procumbens, Hibiscus rosa-sinensis, Dillenia pentagyna, Hopea ponga, Schleichera oleosa, Tabernaemontana gamblei, Trichilia connaroides, Mangifera indica, Muntingia calabura, Peltophorum pterocarpum, Poeciloneuron paucifolium, Psidium guajava, Terminalia paniculata, Borassus flabellifer, Passiflora foetida and Colocasia esculenta*. Plants identified up to genus level include *Bombax* sp., *Rosa* sp., *Impatiens* sp. and *Antigonon* sp.

- Pollen spectra revealed that though the percentage of pollen types varied among the samples of various locations, maximum contribution to honey samples during dearth and honey flow season was made by *C. nucifera* while in brood rearing season, from *M. pudica.*

- *C. nucifera* and *M. pudica* were the predominant (more than 45 per cent) as well as the frequently occurring pollen types during all the three seasons.

- Seasonal occurrence of pollen types revealed that maximum number of pollen types obtained during honey flow season (46 nos.) followed by brood rearing season (33 nos.) and dearth season (26 nos.).

- Frequency of occurrence of each pollen types in honey and pollen samples of fifteen locations revealed that pollen types of *C. nucifera* and *M. pudica* occurred very frequently (more than 50 percent) in all the three seasons and ten pollen types (*C. nucifera*, *M. pudica, A. auriculiformis, M. diplotrica, T. paniculata,* plants belonging to families Oleaceae, Fabaceae, Anacardiaceae, Elaecarpaceae and Myrtaceae) were seen during all the seasons.

- In the present study cluster analysis was carried out based on the presence or absence of pollen types. Fifteen locations were grouped into four clusters based on the presence or absence of 69 pollen types. Pullampara, Aryanad, Karikkam and Tholicode were included in Cluster 1 where as the locations Veliyam, Chennanpara and Kodankara were grouped into cluster 2. Locations, Kadakkal, Ummannoor, Kilimanoor and Kulathupuzha were grouped into cluster 3 and Malayam, Kulappada, Pathanapuram and Rosemala into cluster 4.

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7. REFERENCES

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MELISSOPALYNOLOGY OF INDIAN HONEY BEE (*Apis cerana indica* Fab.) APIARIES IN SOUTHERN KERALA

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ABSTRACT

The present investigation on "Melissopalynology of Indian honey bee (*Apis cerana indica* Fab.) apiaries in southern Kerala" was conducted at Department of Entomology, College of Agriculture, Vellayani during 2012-2013. The objectives were to study the potential pollen and nectar sources of Indian honey bee across the seasons in southern Kerala. Fifteen apiaries with a minimum of ten hives were selected purposively from midland and upland physiographic regions of Thiruvananthapuram and Kollam districts. Three samples each of honey and pollen load was collected randomly from the selected apiaries at three seasons *viz.* honey flow season (January to April), dearth season (June to September) and brood rearing season (October to December). The collected samples were analyzed in the laboratory and the pollen types were identified and quantified.

Mean pollen density of honey samples were found significantly different between the seasons, locations and locations within the seasons. Maximum mean pollen density was recorded in brood rearing season followed by honey flow season and dearth season. Among the locations, Pathanapuram of midland physiographic region had maximum mean pollen density while Veliyam recorded lowest mean pollen density.

A total of 69 plants were recorded as foraging plants by bees from the study area (15 locations) over the three seasons. Twenty four pollen types were identified up to species level, four up to genus level and twenty nine up to family level. Twelve pollen types were unidentified. Identified plant species were *Cocos nucifera, Mimosa pudica, Accacia auriculiformis, Mimosa invisa, Spilanthes calva, Elaeis guineensis, Cullenia exarillata, Tagetes erecta, Tridax procumbens, Hibiscus rosasinensis, Dillenia pentagyna, Hopea ponga, Bombax* sp., *Schleichera oleosa, Rosa* sp., *Tabernaemontana gamblei, Trichilia connaroides, Mangifera indica, Microtropis stocksii, Peltophorum pterocarpum, Poeciloneuron paucifolium, Psidium guajava, Terminalia paniculata, Impatiens* sp., *Borassus flabellifer, Passiflora foetida and Colocasia esculenta.*

Pollen spectra revealed that though the percentage of pollen types varied among the samples of various locations, maximum contribution throughout the season was made by *M. pudica* and *C. nucifera*.

Seasonal occurrence of pollen types revealed that maximum number of pollen types obtained during honey flow season (46 nos.) followed by brood rearing season (33 nos.) and dearth season (26 nos.). Nine pollen types (*C. nucifera, M. pudica, A. auriculiformis, M. invisa,* and plants belonging to families Oleaceae, Fabaceae, Anacardiaceae, Elaecarpaceae and Myrtaceae) were commonly seen during all the seasons. Of these 9 types, *C. nucifera* and *M. pudica* were the predominant (>45 %) as well as the frequently occurring pollen types during all the three seasons. Fifteen locations were grouped into four clusters based on the presence or absence of 69 pollen types.

Thus, the palynological investigation of honey and pollen samples collected from Indian bee apiaries of Thiruvananthapuram and Kollam districts revealed the presence of 69 different foraging plants for their colonial sustenance and honey production.