**RESEARCH ARTICLE** 



# Pollen morphology and taxonomic implications of bee-foraged plant taxa using microscopic techniques

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Abstract This study aimed to explore the systematic details of the Fabaceae family and their melliferous features. Pollen morphological diversity was studied using light microscopy (LM) and scanning electron microscopy (SEM) techniques. In this study, 15 fabaceous melliferous plants were analyzed. Systematically, all the Fabaceae pollen had no spines, psilate sculpturing, tricolporate, polyads, monads, prolate-spheroidal, and oblate-spheroidal shapes. 85.55 µm polar diameter was the highest in Albizia lebbeck (L.) Benth, the minimum was observed as 13.84 µm in Alhagi maurorum Medik. The maximum fertility rate observed was 88% in Leucaena leucocephala (Lam.) de Wit, whereas the minimum observed was 74% in Vicia sativa L. All the Fabaceous plants were identified as melliferous due to the consistent visits of honeybees. Exploration of bee flora helps identify honey's botanical and

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M. Ahmad e-mail: mushtaqflore@hotamil.com geographical origin and production. The study area is suitable for beekeeping businesses and honey production because of its maximum floral diversity. This research is very informative for beekeepers and apiculturists to manage their beekeeping businesses and professions. The bee floral plants should be cultivated to increase honey production.

**Keywords** Fabaceae  $\cdot$  Bee flora  $\cdot$  Honey  $\cdot$  Bannu  $\cdot$  Systematics

### Introduction

The family Fabaceae comprises approximately 700 genera and 19,000 species of herbaceous, shrubby, arboreal, and climbing plants distributed worldwide (Lewis et al. 2005; Mans et al. 2022). It belongs to the

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I. Ullah University of Chinese Academy of Sciences, Beijing 100049, China third-largest angiosperm family and is ecologically and economically significant (Kahraman et al. 2014). The science of palynology known as melissopalynology examines the botanical and geographic origins of honey through microscopic examination of honey sediments and pollen that bees collect (Ullah et al. 2024). The family Fabaceae is considered euripolinic and has a significant variation in pollen 12n grain morphology (Khan et al. 2020). Polyades, psilate, tricolporate, oblate-spheroidal, prolate-spheroidal, and monads features are used for Fabaceae classification (Yousaf et al. 2022). Different types of pollen grains were previously studied to distinguish the fabaceous flora from each other using micromorphological features (Kahraman et al. 2017). The analysis of pollen morphology provides strong evidence for taxonomic classification at various hierarchy levels (Bapir & Galalaey 2023).

Melliferous plants and honeybees maintain a vital symbiotic interaction, as the flowers of the former serve as an important food supply for them. Bees and plants work together to generate pollen and nectar, which gives plants the nutrition they need to survive and helps with pollination, which is essential for plant fertilization. While nectars mostly include carbohydrates, pollen grains are a rich source of proteins, amino acids, lipids, sterols, vitamins, and minerals that promote the robust health and production of both plants and bees.

The species of family Fabaceae are found in tropical and temperate areas (Xu et al. 2017). Most of the plants are xerophytes and have multiple significances. The tap root system and herbaceous, woody stems are present that climb by tendrils. The leaf system is stipules: alternate, palmate, pinnate, and simple. Fabaceae have indehiscent or dehiscent legumes. Pollen grains of fabaceous plants have diverse sizes and indicate their diagnostic features to address their taxonomic identification (Khan et al. 2020). The ornamentation features are to regulate, perforate, and reticulate (Lashin 2006).

Honeybees seem to be the most widely used pollinators in nature, visiting flowers on average 13% of the time, yet visiting just 5% of plant species (*Apis mellifera*) (Tomczyk et al. 2023). The Fabaceae family has polliniferous features frequently visited by honeybees, indicating its melliferous characteristics. Melliferous floral species have unique pollen types that determine the botanical source of honey and its significance for honey quality control and its product (Nabila et al. 2022). Melliferous plants also provide botanical sources and geographical origin (Nascimento et al. 2015). The study of the insect-bee flora relationship is most interesting, and honeybees are the most important pollinating facilitators (Saavedra-Carhuatacto et al. 2014). Palynomorphology concerns honeybee plant diversity's geographical and botanical origin and is important for honey purity and the honeykeeping business (Ahmad et al. 2019). Different pollen morphological features, such as size, shape, ornamentation, and sculpturing, are associated with the pollination mode of honeybees (Saavedra-Carhuatacto et al. 2014).

In a hive, worker bees are divided into distinct types and assigned specific tasks. These tasks include building the comb, collecting nectar and pollen from flowers, making honey, cleaning and cooling the hive, and other tasks. The flower is the most important part of honeybees' lives. Selected plant families blossom at different times and intervals throughout the year (Ghorab et al. 2021). Plant blooming depends upon the soil condition, vegetation habitat, and climatic factors that may change for some nectar plants (Laal-lam & Chenchouni 2024).

The most recent products created by beekeeping have been a wide range of goods, mostly food and medicine. In terms of social, cultural, and economic benefits, beekeeping is a valuable pastime that benefits individuals and communities (Hinton et al. 2020). Exploration of flowering duration and blooming is essential for beekeeping management (Fassil et al. 2022). Among all these flowering families, the Fabaceae is considered the dominant family for honeybee visits and has mellifluous features (Khan et al. 2024). Different species of Fabaceae have sources of nectar and pollen for honeybees (Khan et al. 2024). A total of eleven species were investigated as melliferous plants of the Fabaceae family (Khan et al. 2024).

This research focuses on investigating the geographical and botanical origins of honey and bees. Field observation represents the maximum visit of honeybees to Fabaceae plants, and there is no research or observation about the Fabaceous melliferous checklist. Additionally, this study will solve the taxonomic issues of the Fabaceae family and the floristic composition around apiaries. The beekeeper should gain knowledge of bee floral diversity and suitable places for apiaries to improve the production of honey. This project aims to investigate the diversity of Fabaceous honeybee plants, their taxonomic characteristics, and aspects of botanical sources and bees, using light microscopy (LM) and scanning electron microscopy (SEM) techniques to evaluate the authenticity of honey.

### Material and method

### Study area

A total of 15 different honey-bee plants were collected from various localities in the division of Bannu, Pakistan. Although there is much flora across the area, no one has ever considered the possibility of identifying some melliferous plant taxa. For beekeepers, this study represents a substantial potential taxa (plant) source for honeybee foraging in the area (Fig. 1).

### Fabaceous plant identification

In April 2024, a series of field expeditions were scheduled to collect and identify nectariferous species. The current collection contains 15 Fabaceae species (both cultivated and wild). Visual inspection and discussions with beekeepers and knowledgeable locals verified the presence of plants as honeybee flora. Plants were dried, pressed, and then preserved in the PMAS Arid Agriculture University Rawalpindi, Department of Botany, Plant Taxonomy Lab before being identified. Plant species correctly named scientifically by a taxonomist of high caliber were assigned accession numbers and submitted to the Taxonomy Lab. Additional confirmation of plant names were obtained from the Pakistan Flora and International Plant Naming Index (Ullah et al. 2021).

### Light microscopic (LM) analysis

Fabaceous melliferous plant pollen was examined under a light microscope using the flowers collected during the fieldwork. The pollen grain slide was



Fig. 1 Map of the study area

prepared with little modification to the method of (Ullah et al. 2021; Majeed et al. 2020). Glycerin jelly was used to stain pollen to clarify its morphology and quantitative data. A cover slip was used to conserve the slide for future research. The slide and cover slip were mounted with nail polish. After preparation, the slides were inspected with the light microscope and microphotograph (40X) using an attached camera.

Scanning electron microscopic (SEM) analysis

The methods were applied to get pollen grains ready for the SEM examination. We used Scanning Electron Microscopy (SEM) to perform a detailed analysis of the exine thickness, porosity, and appealing coating. After acetolysis, pollen grains were dissolved in 90% ethanol and subjected to SEM analysis. Subsequently, gold was attached to a palladium-precoated metallic rod and assembled using metallic stubs. Using a scanning electron microscope (SEM) of the (JEOL JSM5910) model that is in the CRL of the Physics Department, University of Peshawar, Pakistan.

# Pollen sterility and fertility

The ratio of pollen sterility and fertility was determined by the method of division (Ullah et al. 2018). S represents sterility, and F represents fertility.

 $F = F/F + S \times 100.$  $S = S/F + S \times 100.$ 

# P/E ratio

The following formula was used to determine the P/E ratio by Ahmad et al. (2018) methodology: P/E Ratio = P/E.

In the given formula, P represents the polar diameter, and E represents the equatorial diameter (Figs. 2, 3, 4).

# Data analysis

The statistical program SPSS 16.00 was used to determine the analysis and quantify the net means of each sample. The symbol for values is mean (minimum – maximum)  $\pm$  SE.

### Results

A total of 15 Fabaceous Melliferous species were palynolomorphologically analyzed using LM and SEM. Both types of variations including qualitative and quantitative features of all plant taxa under investigation were noted. These features address the taxonomic problems and honey-bee flora's botanical and geographical origin (Table 1).

Acacia catechu (L.f.) Willd: Pollen was 40.17  $\mu$ m in the view of polar and equatorial size 36.35 m  $\mu$ m. The exine thickness was 1.92  $\mu$ m, and 1.10 was noted as the P/E ratio. Colpi, Pores, and Spines are absent. Sterility and fertility were noted at 78 and 22 (%). Structurally, it was Medium, slightly circular, Prolate-Spheroidal, Polyads, and Scabrate ornamentation. Tables 2 and 3, Fig. 5.

Acacia nilotica (L.) Delile: Pollen was  $45.17 \mu m$ in the view of polar and equatorial size  $48.92 \mu m$ . The exine thickness was  $1.76 \mu m$ , and 0.92 was noted as the P/E ratio. Colpi, Pores, and Spines are absent. Sterility and fertility were noted at 83 and 17 (%). Structurally, it was Prolate-Spheroidal, Medium, Polyads, Square to Pentagonal, and Scabrate ornamentation. Tables 2 and 3, Fig. 5.

Albizia lebbeck (L.) Benth.: Pollen was 85.55  $\mu$ m view of polar and equatorial size 89.050  $\mu$ m. The exine thickness was 2.92  $\mu$ m, and 0.96 was noted as the P/E ratio. Colpi, there is no Pores, and Spines present. Sterility and fertility were noted at 81 and 19 (%). Structurally, it was Polyads, Large, Scabrate, and Prolate-Spheroidal. Tables 2 and 3, Fig. 5.

**Delbergia sissoo** Roxb.: Pollen was 35.70  $\mu$ m view of polar and equatorial size 38.09  $\mu$ m. 2.09  $\mu$ m were noted as exine thickness and 0.93 were noted as P/E ratio. Colpi length was 12.03  $\mu$ m and width 10.5  $\mu$ m, Pores length was 4.02  $\mu$ m and width 4.68  $\mu$ m. Spines are absent. Sterility and fertility were noted at 82 and 18 (%). Structurally, it was Medium, Circular, Oblate spheroidal, Tricolporate, Reticulate ornamentation and monades. Tables 2 and 3, Fig. 5.

*Leucaena leucocephala* (Lam.) de Wit: Pollen was 55.80  $\mu$ m view of polar and equatorial size 49.31  $\mu$ m. 49.31  $\mu$ m were noted as exine thickness and 1.13 were noted as P/E ratio. Colpi length was 23.71  $\mu$ m and width 11.13  $\mu$ m. Pores and Spines are absent. Sterility and fertility were noted at 88 and 12 (%). Structurally, it was Pslite-Scabrate, Medium, Fig. 2 Fabaceae melliferous plants **a** *Acacia catechu* (L.f.) Willd **b** *Acacia nilotica* (L.) Delile **c** *Albizia lebbeck* (L.) Benth **d** *Alhagi maurorum* Medik. **e** *Cicer arietinum* L. **f** *Delbergia sissoo* Roxb



monads, Tricolporate and Sub prolate. Tables 2 and 3, Fig. 6.

**Parkinsonia aculeate** L.: The Pollen was 35.94  $\mu$ m given a polar and equatorial size of 36.28  $\mu$ m. The exine thickness was 2.31  $\mu$ m, and 0.98 was noted as the P/E ratio. Colpi length was 6.30  $\mu$ m and width 5.10  $\mu$ m. Pores and Spines are absent. Sterility and fertility were noted at 81 and 19 (%). Structurally, it was medium, circular, Oblate-spheroidal, Tricolporate, Pslite ornamentation, and monads. Tables 2 and 3, Fig. 6.

**Prosopis cineraria** (L.) Druce: Pollen was 21.17  $\mu$ m view of polar and equatorial size 26.80  $\mu$ m. 2.56  $\mu$ m were noted as exine thickness

and 0.79 were noted as P/E ratio. Colpi length was 7.71  $\mu$ m and 6.70  $\mu$ m width. Pores and Spines are absent. Sterility and fertility were noted at 84 and 16 (%). Structurally, it was Small, Triangular, sub-prolate, Tricolporate, Pslite ornamentation, and monads. Tables 2 and 3, Fig. 6.

**Prosopis juliflora** (SW.) DC: Pollen was 33.39  $\mu$ m view of polar and equatorial size 32.30  $\mu$ m. 2.70  $\mu$ m were noted as exine thickness and 1.09 were noted as P/E ratio. Colpi length was 8.71  $\mu$ m and width 13.60  $\mu$ m. Pores and Spines are absent. Sterility and fertility were noted at 77 and 23 (%). Structurally, it was Small, Triangular,





Sub-Prolate, Tricolporate, Pslite ornamentation, and monads. Tables 2 and 3, Fig. 6.

**Trifolium alexandrinum L.:** Pollen was 34.20  $\mu$ m view of polar and equatorial size 36.26  $\mu$ m. 1.71  $\mu$ m were noted as exine thickness and 0.94 were noted as P/E ratio. Colpi length was 5.45  $\mu$ m and 5.88  $\mu$ m width. Pores and Spines are absent. Sterility and fertility were noted at 79 and 21 (%). Structurally, it was Medium, Circular, Spheroidal, Bicolporate, Pslite ornamentation and monads. Tables 2 and 3, Fig. 7.

*Trifolium pratense* L. L: Pollen was  $33.21 \mu m$  view of polar and equatorial size  $35.21 \mu m$ .  $1.21 \mu m$  were noted as exine thickness and 0.96 were noted as P/E ratio. Colpi length was  $3.86 \mu m$  and  $4.86 \mu m$ 

width. Pores and Spines are absent. Sterility and fertility were noted at 77 and 23 (%). Structurally, it was Pslite, Medium, Tricolporate, monads, and Oblate spheroidal. Tables 2 and 3, Fig. 7.

Alhagi maurorum Medik.: Pollen was 13.84  $\mu$ m view of polar and equatorial size 15.10  $\mu$ m. 1.89  $\mu$ m were noted as exine thickness and 0.91 were noted as P/E ratio. Colpi length was 4.10  $\mu$ m and width 3.67  $\mu$ m. Pores and Spines are absent. Sterility and fertility were noted at 78 and 22 (%). Structurally, it was Small, Circular, Oblate spheroidal, monocolporate, Pslite ornamentation and monads. Tables 2 and 3, Fig. 5.

Fig. 4 Fabaceae melliferous plants a Trifolium alexandrinum L. b Trifolium pratense L. L. c Vicia sativa L



*Vicia sativa* L.: Pollen was 35.84  $\mu$ m view of polar and equatorial size 36.93  $\mu$ m. 2.11  $\mu$ m were noted as exine thickness and 0.97 were noted as P/E ratio. Colpi length was 5.54  $\mu$ m and width 4.19  $\mu$ m. Pores and Spines are absent. Sterility and fertility were noted at 74 and 26 (%). Structurally, it was Medium, Triangular, Oblate spheroidal, Tricolporate, Pslite ornamentation, and monads. Tables 2 and 3, Fig. 7.

*Cicer arietinum* L.: Pollen was 16.26  $\mu$ m view of polar and equatorial size 15.17  $\mu$ m. 1.47  $\mu$ m were noted as exine thickness and 1.07 were noted as P/E ratio. Colpi length was 4.70  $\mu$ m and width 2.10  $\mu$ m. Pores and Spines are absent. Sterility and fertility were noted at 81 and 19 (%). Structurally, it was Small, Triangular, prolate-spheroidal, Tricolporate, Pslite ornamentation, and monads. Tables 2 and 3, Fig. 5.

**Melilotus indica** (L.): Pollen was 23.18  $\mu$ m view of polar and equatorial size 22.50  $\mu$ m. 1.47  $\mu$ m were noted as exine thickness and 1.03 were noted as P/E ratio. Colpi length was 5.46  $\mu$ m and A total of 15 Fabaceous Melliferous species were palynolomorphologically analyzed using LM and SEM. Diversity and variation are noted in all plant species' qualitative and quantitative features. These features address the taxonomic problems and the honey-bee flora's botanical and geographical origin.

Acacia catechu (L.f.) Willd: Pollen was 40.17  $\mu$ m in the view of polar and equatorial size 36.35  $\mu$ m. The exine thickness was 1.92  $\mu$ m, and 1.10 was noted as the P/E ratio. Colpi, Pores, and Spines are absent. Sterility and fertility were noted at 78 and 22 percent, respectively. Structurally, it had medium, slightly circular, prolate-spheroidal, polyads, and scabrate ornamentation. Tables 2 and 3, Fig. 5.

Acacia nilotica (L.) Delile: Pollen was 45.17  $\mu$ m in the view of polar and equatorial size 48.92  $\mu$ m. The exine thickness was 1.76  $\mu$ m, and 0.92 was noted as the P/E ratio. Colpi, Pores, and Spines are absent. Sterility and fertility were noted at 83 and 17 percent, respectively. Structurally, it was prolate-spheroidal, medium, polyads, square to Pentagonal, and scabrate ornamentation. Tables 2 and 3, Fig. 5.

Albizia lebbeck (L.) Benth.: Pollen was 85.55  $\mu$ m given polar and equatorial sizes of 89.050  $\mu$ m. The exine thickness was 2.92  $\mu$ m, and 0.96 was noted as the P/E ratio. Colpi, there are no pores or spines present. Sterility and fertility were noted at 81 and 19 (%), respectively. Structurally, it

| S.No | Fabaceae plants                  | Status            | family   | Habit       | Herbarium codes | Collector name                 | Collection Date |
|------|----------------------------------|-------------------|----------|-------------|-----------------|--------------------------------|-----------------|
| 1    | Prosopis juliflora<br>(SW.)      | Nature Wild       | Fabaceae | Shrubby     | MG-01           | Khushdil Khan                  | 13 April 2024   |
| 2    | Trifolium pratense<br>(L.)       | Nature Cultivated | Fabaceae | Herbaceous  | MG-02           | Khushdil Khan,<br>Shabir Ahmad | 25 April 2024   |
| 3    | Sesbania sesban (L.)             | Nature Cultivated | Fabaceae | Shrubby     | MG-03           | Khusdil Khan                   | 28 April 2024   |
| 4    | Cicer arietinum L                | Nature Cultivated | Fabaceae | Shrubby     | MG-04           | Shabir Ahmad, Ateef<br>Ullah   | 22 April 2024   |
| 5    | Leucaena leucoceph-<br>ala (L.)  | Nature Cultivated | Fabaceae | Tree nature | MG-05           | Khushdil Khan,<br>Shabir Ahmad | 20 April 2024   |
| 6    | Alhagi maurorum<br>Medik         | Nature Wild       | Fabaceae | Herbaceous  | MG-06           | Shabir Ahmad, Ateef<br>Ullah   | 29 April 2024   |
| 7    | Prosopis cineraria<br>(L.)       | Nature Wild       | Fabaceae | Shrubby     | MG-07           | Khushdil Khan                  | 21 April 2024   |
| 8    | Acacia catechu (L.f.)<br>Willd   | Nature Wild       | Fabaceae | Tree nature | MG-08           | Khushdil Khan                  | 19 April 2024   |
| 9    | Trifolium alexandri-<br>num (L.) | Nature Cultivated | Fabaceae | Herbaceous  | MG-09           | Khushdil Khan                  | 30 April 2024   |
| 10   | Acacia nilotica (L.)             | Nature Wild       | Fabaceae | Tree nature | MG-10           | Khushdil Khan                  | 26 April 2024   |
| 11   | Parkinsonia aculeate<br>L        | Nature Wild       | Fabaceae | Herbaceous  | MG-11           | Khushdil Khan                  | 18 April 2024   |
| 12   | Vicia sativa L                   | Nature Wild       | Fabaceae | Herbaceous  | MG-12           | Khushdil Khan                  | 22 April 2024   |
| 13   | <i>Delbergia sissoo</i><br>Roxb  | Nature Cultivated | Fabaceae | Tree nature | MG-13           | Khushdil Khan                  | 21 April 2024   |
| 14   | Melilotus indica (L.)            | Nature Wild       | Fabaceae | Herbaceous  | MG-14           | Khushdil Khan                  | 14 April 2024   |
| 15   | Albizia lebbeck (L.)             | Nature Cultivated | Fabaceae | Tree nature | MG-15           | Shabir Ahmad, Ateef<br>Ullah   | 29 April 2024   |

Table 1 Status of wild/cultivated, family, habitat, herbarium codes, and collection date of Fabaceous melliferous taxa

was polyads, large, scabrate, and prolate-spheroidal. Tables 3 and 2, Fig. 5.

**Delbergia sissoo** Roxb.: Pollen was 35.70  $\mu$ m view of polar and equatorial size 38.09  $\mu$ m. 2.09  $\mu$ m was noted as exine thickness, and 0.93  $\mu$ m was noted as P/E ratio. Colpi length was 12.03  $\mu$ m and width 10.5  $\mu$ m, Pores length was 4.02  $\mu$ m and width 4.68  $\mu$ m. Spines are absent. Sterility and fertility were noted at 82 and 18 (%), respectively. Structurally, it was medium, circular, oblate spheroidal, tricolporate, reticulate ornamentation, and monades. Tables 2 and 3, Fig. 5.

*Leucaena leucocephala* (Lam.) de Wit: Pollen was 55.80  $\mu$ m view of polar and equatorial size 49.31  $\mu$ m. 49.31  $\mu$ m was noted as exine thickness, and 1.13 was noted as P/E ratio. Colpi length was 23.71  $\mu$ m and width was 11.13  $\mu$ m. Pores and spines are absent. Sterility and fertility were noted at 88 and 12 percent, respectively. Structurally, it was Pslite-Scabrate, Medium, Monades, Tricolporate, and Subprolate. Tables 2 and 3, Fig. 6.

**Parkinsonia aculeate** L.: The pollen was 35.94  $\mu$ m given the polar and equatorial size of 36.28  $\mu$ m. The exine thickness was 2.31  $\mu$ m, and 0.98 was noted as the P/E ratio. Colpi length was 6.30  $\mu$ m and width 5.10  $\mu$ m. Pores and spines are absent. Sterility and fertility were noted at 81 and 19 (%), respectively. Structurally, it was medium, circular, oblate-spheroidal, tricolporate, pslite ornamentation, and monades. Tables 2 and 3, Fig. 6.

**Prosopis cineraria** (L.) Druce: Pollen was 21.17  $\mu$ m given polar and equatorial sizes of 26.80  $\mu$ m. 2.56  $\mu$ m were noted as exine thickness, and 0.79 were noted as P/E ratio. Colpi length was 7.71  $\mu$ m and 6.70  $\mu$ m width. Pores and spines are absent. Sterility and fertility were noted at 84 and 16 percent, respectively. Structurally, it was small,

| Table | 2 Pollen quantitati             | ve features of fabac   | cous mellifero                 | us flora              |              |              |             |                |               |                              |                              |
|-------|---------------------------------|------------------------|--------------------------------|-----------------------|--------------|--------------|-------------|----------------|---------------|------------------------------|------------------------------|
| S.No  | Fabaceae Mel-<br>liferous flora | Polar diameter<br>(PD) | Equatorial<br>diameter<br>(ED) | Thickness of<br>Exine | Ratio of P/E | Colpi Length | Colpi Width | Length of Pore | Width of Pore | Fertility<br>percent-<br>age | Sterility<br>percent-<br>age |
| 1     | Sesbania sesban<br>(L.)         | 24.99 µm               | 24.93 μm                       | 1.09 µш               | 1.02         | 7.05 µm      | 5.29 µm     | Nil            | Nil           | 85%                          | 15%                          |
| 2     | Cicer arietinum L               | 16.26 µm               | 15.17 µm                       | 1.47 µm               | 1.07         | 4.70 μm      | 2.10 μm     | Nil            | Nil           | 81%                          | 19%                          |
| 33    | Leucaena leuco-<br>cephala (L.) | 55.80 μm               | 49.31 µm                       | 2.51 µm               | 1.13         | 23.71 µm     | 11.13 µm    | Nil            | Nil           | 88%                          | 12%                          |
| 4     | Trifolium prat-<br>ense (L.)    | 33.21 µm               | 35.21 µm                       | 1.21 µm               | 0.96         | 3.86 µm      | 4.86 µm     | Nil            | liN           | 77%                          | 23%                          |
| 5     | Vicia sativa L                  | 35.84 μm               | 36.93 µm                       | 2.11 μm               | 0.97         | 5.54 µm      | 4.19 µm     | Nil            | Nil           | 74%                          | 26%                          |
| 9     | <i>Delbergia sissoo</i><br>Roxb | 35.70 μm               | 38.09 µm                       | 2.09 µm               | 0.93         | 12.03 µm     | 10.50 µm    | 4.020 µm       | 4.68 µm       | 82%                          | 22%                          |
| ٢     | Prosopis juliflora<br>(SW.)     | 33.39 µm               | 32.30 μm                       | 2.70 µm               | 1.09         | 8.71 µm      | 13.60 µm    | Nil            | liN           | 77%                          | 23%                          |
| ×     | Parkinsonia<br>aculeate L       | 35.94 μm               | 36.28 µт                       | 2.31 µm               | 0.98         | 6.30 µm      | 5.10 µm     | Nil            | liN           | 81%                          | 19%                          |
| 6     | Prosopis ciner-<br>aria (L.)    | 21.17 μm               | 26.80 µm                       | 2.56 µт               | 0.79         | 7.71 µm      | 6.70 µm     | Nil            | liN           | 84%                          | 16%                          |
| 10    | Trifolium alexan-<br>drinum L   | 34.20 μm               | 36.26 μm                       | 1.71 µm               | 0.94         | 5.45 µm      | 5.88 µm     | Nil            | Nil           | 79%                          | 21%                          |
| 11    | Acacia nilotica<br>(L.)         | 45.17 μm               | 48.92 µm                       | 1.76 µm               | 0.92         | Absent       | Absent      | Nil            | liN           | 83%                          | 17%                          |
| 12    | Alhagi maurorum<br>Medik        | 13.84 µm               | 15.10 µm                       | 1.89 µт               | 0.91         | 4.10 µm      | 3.67 μm     | Nil            | Nil           | 78%                          | 22%                          |
| 13    | Albizia lebbeck<br>(L.)         | 85.55 μm               | 89.050 µm                      | 2.92 µm               | 0.96         | Absent       | Absent      | Nil            | Nil           | 81%                          | 19%                          |
| 14    | <i>Melilotus indica</i><br>(L.) | 23.18 µm               | 22.50 µm                       | 1.23 µm               | 1.03         | 5.46 µm      | 6.61 µm     | Nil            | Nil           | 86%                          | 14%                          |
| 15    | Acacia catechu<br>(L.f.) Willd  | 40.17 µm               | 36.35 µm                       | 1.92 µm               | 1.10         | Absent       | Absent      | Nil            | Nil           | 78%                          | 22%                          |

| S.No | Fabaceae<br>members                 | Polar Shape                  | Size of<br>Pollen | Dispersal<br>unit | Type of<br>Pollen  | Symmetry          | Pollen<br>surface  | Polarity    | Exine<br>Sculpturing |
|------|-------------------------------------|------------------------------|-------------------|-------------------|--------------------|-------------------|--------------------|-------------|----------------------|
| 1    | Sesbania<br>sesban<br>(L.)          | Triangular<br>Slightly       | Medium<br>Size    | Monad             | Tricol-<br>poarate | Radial            | Gemmate            | Isopolar    | Scabrate             |
| 2    | Cicer arieti-<br>num L              | Triangular<br>shape          | Small Size        | Monad             | Tricolporate       | Radial            | Psilate            | Isopolar    | Psilate              |
| 3    | Alhagi<br>maurorum<br>Medik         | Circular<br>shape            | Small Size        | Monad             | Monocolpo-<br>rate | Asymmetri-<br>cal | Psilate            | Heteropolar | Psilate              |
| 4    | <i>Delbergia</i><br>sissoo<br>Roxb  | Circular<br>shape            | Medium<br>Size    | Monad             | Tricolporate       | Radial            | Psilate            | Isopolar    | Reticulate           |
| 5    | Trifolium<br>alexandri-<br>num L    | Circular<br>shape            | Medium<br>Size    | Monad             | Bicolporate        | Asymmetri-<br>cal | Psilate            | Heteropolar | Psilate              |
| 6    | Parkinsonia<br>aculeate L           | Circular<br>shape            | Medium<br>Size    | Monad             | Tricolporate       | Asymmetri-<br>cal | Psilate            | Heteropolar | Psilate              |
| 7    | Prosopis<br>juliflora<br>(SW.)      | Triangular<br>shape          | Small Size        | Monad             | Tricolporate       | Radial            | Psilate            | Heteropolar | Psilate              |
| 8    | Trifolium<br>pratense<br>(L.)       | Triangular<br>Slightly       | Medium<br>Size    | Monad             | Tricolporate       | Radial            | Psilate            | Isopolar    | Psilate              |
| 9    | Albizia leb-<br>beck (L.)           | Pentagonal<br>to Square      | Large Size        | Polyad            | Polyads            | Radial            | Psilate            | Isopolar    | Scabrate             |
| 10   | Vicia sativa<br>L                   | Triangular<br>shape          | Medium<br>Size    | Monad             | Tricolporate       | Radial            | Psilate            | Isopolar    | Psilate              |
| 11   | Leucaena<br>leucoceph-<br>ala (L.)  | Slightly<br>circular         | Medium<br>Size    | Monad             | Tricolporate       | Radial            | Psilate            | Isopolar    | Pslite-<br>Scabrate  |
| 12   | Acacia<br>nilotica<br>(L.)          | Square to<br>Pentago-<br>nal | Medium<br>Size    | Polyad            | Polyads            | Radial            | Psilate            | Heteropolar | Scabrate             |
| 13   | Melilotus<br>indica (L.)            | Rounded<br>Slightly          | Small Size        | Monad             | Tricolporate       | Asymmetri-<br>cal | Micro-<br>echinate | Heteropolar | Reticulate           |
| 14   | Prosopis<br>cineraria<br>(L.)       | Triangular<br>shape          | Small Size        | Monad             | Tricolporate       | Asymmetri-<br>cal | Psilate            | Heteropolar | Psilate              |
| 15   | Acacia cat-<br>echu (L.f.)<br>Willd | circular<br>Slightly         | Medium<br>Size    | Polyad            | Polyads            | Radial            | Gemmate            | Isopolar    | Scabrate             |

Table 3 Pollen qualitative features of fabaceous melliferous flora

triangular, sub-prolate, tricolporate, pslite ornamentation, and monads. Tables 2 and 3, Fig. 6.

**Prosopis juliflora** (SW.) DC: Pollen was 33.39  $\mu$ m view of polar and equatorial size 32.30  $\mu$ m. 2.70  $\mu$ m were noted as exine thickness, and 1.09 was noted as the P/E ratio. Colpi length was 8.71  $\mu$ m and width was 13.60  $\mu$ m. Pores and spines are absent. Sterility and fertility were noted at 77 and 23 percent,

respectively. Structurally, it was small, triangular, sub-prolate, tricolporate, pslite ornamentation, and monads. Tables 2 and 3, Fig. 6.

*Trifolium alexandrinum* L.: Pollen was 34.20  $\mu$ m view of polar and equatorial size 36.26  $\mu$ m. 1.71  $\mu$ m was noted as exine thickness, and 0.94  $\mu$ m was noted as P/E ratio. Colpi length was 5.45  $\mu$ m and 5.88  $\mu$ m width. Pores and spines are absent. Sterility and

Fig. 5 a and b Acacia catechu (L.f.) Willd, c and d Acacia nilotica (L.) Delile, e and f Albizia lebbeck (L.) Benth., g and h Alhagi maurorum Medik., i and j Cicer arietinum L



fertility were noted at 79 and 21 (%), respectively. Structurally, it was medium, circular, spheroidal, bicolporate, psilate ornamentation, and monads. Tables 2 and 3, Fig. 7.

*Trifolium pratense* L. Pollen was 33.21  $\mu$ m because of polar and equatorial size 35.21  $\mu$ m. 1.21  $\mu$ m was noted as exine thickness, and 0.96  $\mu$ m was noted as P/E ratio. Colpi length was 3.86  $\mu$ m and 4.86  $\mu$ m width. Pores and spines are absent. Sterility

and fertility were noted at 77 and 23 percent, respectively. Structurally, it was Pslite, Medium, Tricolporate, Monads, and Oblate Spheroidal. Tables 2 and 3, Fig. 7.

Alhagi maurorum Medik.: Pollen was 13.84  $\mu$ m view of polar and equatorial size 15.10  $\mu$ m. 1.89  $\mu$ m were noted as exine thickness, and 0.91 was noted as the P/E ratio. Colpi length was 4.10  $\mu$ m and width 3.67  $\mu$ m. Pores and spines are absent. Sterility and



**Fig. 6 a** and **b** Delbergia sissoo Roxb. **c** and **d** Leucaena leucocephala (Lam.) de Wit **e** and **f** Melilotus indica (L.), **g** and **h** Parkinsonia aculeate L. **i** and **j** Prosopis cineraria (L.) Druce

fertility were noted at 78 and 22 percent, respectively. Structurally, it was small, circular, oblate spheroidal, monocolporate, pslite ornamentation, and monads. Tables 2 and 3, Fig. 5.

*Vicia sativa* L.: Pollen was 35.84  $\mu$ m view of polar and equatorial size 36.93  $\mu$ m. 2.11  $\mu$ m was noted as exine thickness, and 0.97  $\mu$ m was noted as P/E ratio. Colpi length was 5.54  $\mu$ m and width was 4.19  $\mu$ m. Pores and spines are absent. Sterility and fertility were noted at 74 and 26 percent, respectively.

Structurally, it was medium, triangular, oblate spheroidal, tricolporate, pslite ornamentation, and monads. Tables 2 and 3, Fig. 7.

*Cicer arietinum* L.: Pollen was 16.26  $\mu$ m view of polar and equatorial size 15.17  $\mu$ m. 1.47  $\mu$ m were noted as exine thickness, and 1.07 were noted as P/E ratio. Colpi length was 4.70  $\mu$ m and width 2.10  $\mu$ m. Pores and spines are absent. Sterility and fertility were noted at 81 and 19 (%), respectively. Structurally, it was small, triangular, prolate-spheroidal,

Fig. 7 a and b Prosopis juliflora (SW.) DC., c and d Sesbania sesban (L.), e and f Trifolium alexandrinum L., g and h Trifolium pratense L. L., i and j Vicia sativa L



tricolporate, pslite ornamentation, and monads. Tables 2 and 3, Fig. 5.

**Melilotus indica** (L.): Pollen was 23.18  $\mu$ m in polar and equatorial sizes of 22.50  $\mu$ m. 1.47  $\mu$ m was noted as exine thickness, and 1.03 was noted as P/E ratio. Colpi length was 5.46  $\mu$ m and width 6.61  $\mu$ m. Pores and spines are absent. Sterility and fertility were noted at 86 and 14 percent, respectively. Structurally, it was small, slightly rounded, oblate-spheroidal,

tricolporate, with reticulate ornamentation and monads. Tables 2 and 3, Fig. 6.

Sesbania sesban (L.): Pollen was 24.99  $\mu$ m view of polar and equatorial size 24.93  $\mu$ m. 1.09  $\mu$ m was noted as exine thickness, and 1.02  $\mu$ m was noted as P/E ratio. Colpi length was 7.05  $\mu$ m and width 5.29  $\mu$ m. Pores and spines are absent. Sterility and fertility were noted at 85 and 15 percent, respectively. Structurally, it was medium, slightly triangular, slightly ovate, tricolporate, scabrate ornamentation, and monads. Tables 2 and 3, Fig. 7.

#### Discussion

The pollen morphology of Fabaceoeus melliferous plant species has heterogeneous characteristics (Khan et al. 2020; Ozbek et al. 2014). In the studied samples, all pollen was investigated as heterogeneous, which was supported by previous literature. A total of 15 Fabaceoeus melliferous plant species were observed as having melliferous characteristics, which is supported by Khan et al. (2024). Colpi length, pore length, and exine sculpturing are important in distinguishing between taxa of melliferous Fabaceoeus species. This finding of our results is according to El-Sayed et al. (2010) and Lashin (2006). In the Fabaceae family, most pollen grains are observed as triangular and circular, which is supported by Lashin (2006). A total of 15 Fabaceae members were observed as bee flora because of their maximum diversity and polliniferous nature, as supported by Khan et al. (2024).

The maximum polar diameter recorded was 85.55 µm in Albizia lebbeck (L.) Benth, while the minimum was observed at 13.84 µm in Alhagi maurorum Medik. 89.050 µm were noted as the highest equatorial diameter in Albizia lebbeck (L.) Benth, while the minimum was noted as 15.10 in Alhagi maurorum Medik. Such pollen variation was observed in Fabaceae family members from Pakistan, supporting the Fabaceoeus melliferous plant diversity (Khan et al. 2020). 2.92 µm highest exine thickness were observed in Albizia lebbeck (L.) Benth as. The highest P/E ratio was noted as 1.10 in Acacia catechu (L.f.) Willd, while the minimum was in Prosopis cineraria (L.) Druce as 0.79. The highest P/E ratio was observed at 1.46 by Lashin (2006), supporting the current results of Melliferous Fabaceae plants.

Colpi and pores were noted as absent in *Acacia nilotica*, *Acacia catechu*, and *Albizia lebbeck*, and spines are absent in all Fabaceae plant species, indicating their diagnostic features. Spines were noted as absent in plant pollen in the Fabaceae family, supporting the current results (Khan et al. 2020). The highest colpi length was noted in *Leucaena leucocephala* at 23.71 µm, while the minimum in *Trifolium pratense* L. L. was 3.86 µm. *Delbergia sissoo has* only

porate pollen. The highest fertile pollen percentage was noted in *Leucaena leucocephala* (Lam.) de Wit at 88%, while the minimum was in *Vicia sativa* L. at 74%. This fertility rate is justified by Kahraman et al. (2013). The maximum sterility rate was observed in *Vicia sativa* L. at 26%, and the minimum was in Leucaena leucocephala (Lam.) de Wit at 12%.

The pollen size was medium in nine Fabaceoeus melliferous species, small in five, and large in only Albizia lebbeck (L.) Benth. The polar view of pollen is mostly triangular and slightly circular. Antonio-Domingues et al. (2018) supported these pollen features in the Fabaceae family. The pollen shape is mostly observed as prolate-spheroidal and oblatespheroidal in the Fabaceae family. The pollen type was observed as polyads in only Acacia nilotica, Acacia catechu, and Albizia lebbeck, and tricolpoarate was observed in ten Fabaceae samples. Scabrate sculpture was observed in Acacia nilotica, Acacia catechu, and Albizia lebbeck, and the rest of the Fabaceae Melliferous have psilate sculpture. The pollen dispersal unit as polyads was observed in Acacia nilotica, Acacia catechu, and Albizia lebbeck, and the monads in the rest of the Fabaceae plants. The features of pollen morphology in the Fabaceae family are supported by Bravo-Chinguel et al. (2014) and Aye & Lin (2020).

All the Fabaceoeus plants were observed as melliferous plants because honey-bees frequently visited them (Figs. 1, 2, 3). A total of eleven Fabaceae plants were observed as nectariferous, as supported by the current study and results (Khan et al. 2024). The study of the melliferous plant helps identify the floral and geographical origin of honey and its production (Noor et al. 2016). Our results also elaborate on the potential of such plants for identifying and exploring honey production and managing the beekeeping business. Most of the tropical flora are considered honey bee plants in the study area, and the collected and studied Fabaceoeus plants belong to a tropical area (Sharma et al. 2010). The diversity of pollen in the Fabaceae family observed in the studied samples shows interest in honey bee visits to such plants, as supported by Ciucure and Geană (2019). Maximum diversity was observed in the Fabaceae family as melliferous plants in the studied area. The Fabaceae members are considered to have the best melliferous characteristics, as supported by our results (Khan et al. 2022).

The diversity of honeybee plants around bee apiaries influences honey composition and quality. Local individuals and beekeepers visualization and observation of honeybees visits to bee flora verified the confirmation of honey bee plants as a source of nectar and pollen (Saklani & Mattu 2020). All 15 Fabaceoeus melliferous studied plants were observed foraging by bees in the fieldwork and plant collection. The study area is suitable for beekeeping businesses and honey production because of its maximum floral diversity. All the bee floral diversity and flowering periods of blooming are most significant to manage by keeping the profession in the study area.

### Conclusion

The present research effort evaluated 15 plant species that have been identified in the study area and recognized as honeybee flora. The size, shape, aperture features, and exine sculpturing of the pollen samples from the analyzed flora contributed vital data for precisely determining the melliferous species. Furthermore, this work extends to the previous study on honeybee flora and clarifies the palyno-morphological description of the species in the studied area. Our present study suggests that the environment is suited to beekeeping in the area. Therefore, in order to boost the quantity and quality of honey produced, care must be taken to preserve the current melliferous flora and cultivate new species. For the honey market to grow properly, the government should give beekeepers access to modern equipment and training.

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#### Declarations

**Conflict of interest** The authors declare no competing interests.

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