ANNALI DI BOTANICA
Ann. Bot. (Roma), 2024, 14: 21–34
annalidibotanica.uniroma1.it | E-ISSN 2239-3129 | ISSN 0365-0812

INSECT POLLINATORS AND PLANT INTERACTIONS: A TAXONOMIC IMPLICATIONS OF POLLEN MORPHOLOGICAL FEATURES IN MELLIFEROUS PLANTS

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(Received 20 Febr 2024; received in revised form 8 Apr 2024; accepted 18 Apr 2024)

Abstract – The current study was the first report on pollen morphology of 17 species belonging to 11 different families that are particularly involved in nectar and pollen collection by honeybees from southern Pakistan using microscopic techniques. The plants were collected after being foraged by honeybees, identified, and studied. Flowering periods, habits and localities of plants were examined. The pollen grains were then passed through the process of acetolysis, measured and both quantitative and qualitative characteristics evaluated. The investigated pollen grains varied in size, shape, colpi/pore and exine sculpturing. The most common type of pollen examined was tricolporate and among the shapes was oblate spheroidal one. Three types of exine sculpturing were observed, psilate, reticulate and echinate. The recorded pollen fertility ranged from 67 to 93 percent, showing that the considered plants are well established in the area. *Brassica campestris* L. *Asphodelus tenuifolius* Cav. and *Astragalus hamosus* L. resulted in the most visited species by honeybees. Our results showed that the pollen morphology of the melliferous plants in the study varies largely. The principal component analysis did not show clusters in relation to specific pollination syndromes and only some strictly phylogenetically related species were close in the graph. For this reason, the pollen grain characters can be considered sufficiently diversified to be used as markers for species identification in melissopalynological analyses. The present study plays a vital role in taxa identification, floral calendar preparation, and production of honey in the study area. This evidence may provide the possibility of evaluating the geographical provenance of honey from melissopalynological analysis at least in southern Pakistan.

Keywords: taxonomic implications; honeybees; melliferous plant; nectars; palynology.

Introduction

Melliferous plants of a region are the species which are visited by honeybees to collect substances used in honey production. Honeybees visit flowers having higher amounts of nectars and pollen grains of better quality (Ahmad & Aslam, 2002). Nectars are the important source to honeybees for their foraging activities, hive formations and honey productions. Seasons have a great impact on melliferous flora; plants blooming in the spring have a comparatively lower nectar yield than in summer (Ion et al., 2018). The melliferous potential of the plants depends also on ecological characteristics of an area. The qualitative and quantitative features of honey depend on the plants from which honeybees collect pollen and nectars. Consequently, the economic activity of beekeepers is directly related to the flowering periods of a particular area. In Southern Pakistan beekeepers arrange their business of honey annually two

doi: 10.13133/2239-3129/18336

times, one in autumn and another in spring. In melliferous plants pollination occurs through honeybees which increase the annual production of fruits, vegetables, and oils (Gallai et al., 2009). For this reason, an important data is the determination of the flowering periods of different plants. Melissopalynological studies contribute to determining the floral and geographical origin of bees and honey (Agwu & Uwakwe, 19921; Abou-Shaara, 2019).

Pollen grains are fine particles produced in the anthers of a stamen in flowering plants containing the male gametophyte in seed plants. The exine represents the outer and intine is the inner layer of the pollen grain wall. Exine represents various sculpturing patterns, pores, spines and colpi. The field of palynology is not only limited to taxonomy, but it has a lot of applications in other fields such as medicine, forestry, biochemistry, forensics, pharmacy, chemotaxonomy, and systematic botany. Melissopalynology is one of the subcategories of palynology dealing with pollen used in honey production, possible adulterations, foraged of plants by honeybees, its habitat, and the quality of honey (Ashfaq et al., 2018).

Pakistan is considered as a very famous origin of high-quality honey. The investigated area includes both plain and hilly areas. Most common crops of the study area are wheat, gram (chickpea), dates, mango, and maize. The country has a long summer with maximum rainfall occurring in months of July and August. The most represented families of the present study are Asteraceae followed by Poaceae, Fabaceae and Brassicaceae. The climate of the area is semi-arid with hot summer and cool winter seasons (Khan et al., 2020). The most important economic activity of people is agriculture.

Some researchers (Pound et al., 2018; Ahmad et al., 2019) gave basic knowledge of pollen morphology about the melliferous flora of different localities. The aim of the present study was to identify melliferous plants for the first time from southern Pakistan, making its link with plant taxonomic studies, spreading its knowledge among local peoples, describing pollen morphology, its uses in honey formations, and protecting of plants from overgrazing. Such data are important to be able to identify pollen grains in honey samples as the main tool to ascertain the provenance of commercialized honey (Shaheen et al., 2017; Shakeel et al., 2019). Pollen morphological studies aid in the taxonomic and systematic identification of species belonging to the same genera. The melliferous species in the study area were very diverse and important from an evolutionary point of view. Exine thickness, pollen size and shape are the important features useful in taxonomic studies of different plants. Palynology is an important field related to many fields of genetics, pharmacology, evolutionary biology, and plant ecology. The aim of current research work is to identify and explore the bee forage plants for the first time in the study area, making their link with plant taxonomic studies, spreading their knowledge among local people, describing pollen morphology, its uses in honey formations, and conserving the vegetation.

Materials & Method

Field survey

A two-week field trip was arranged during the start of March 2019–April 2020 to the southern districts of Khyber Pakhtunkhwa, Punjab, and Sindh. All such plants were collected and photographed, which are foraged by honeybees. The plants were pressed and dried in newspapers. The specimens were identified with the available species at the Herbarium of (ISL) Quaid-i-Azam University, the flora of Pakistan, and online available literature. During the field trip, interviews were conducted with beekeepers living and working in the study area.

The species collected, identified and analyzed were *Daucus carota* L., *Launaea procumbens* (Roxb.) Ramayya and Rajagopal, *Plantago amplexicaulis* Cav., *Medicago monantha* (C.A. Mey) Trautv., *Cousinia minuta* Boiss., *Convolvulus arvensis* L., *Dactyloctenium aegyptium* (L.) Willd., *Vicia sativa* L., *Fumaria indica* (Hausskn.) Pugsley, *Sisymbrium irio* L., *Malcolmia cabulica* (Boiss.) Hook. f. and Thomson, *Arnebia hispidissima* (Lehm.) A. DC., *Astragalus hamosus* L., *Hypecoum pendulum* L., *Brassica campestris* L., *Asphodelus tenuifolius* Cav. and *Triticum aestivum* L. (Tab. 1). Some of the plants are anemophilous but are visited by bees for pollen collection to feed their young ones due to the unavailability of highly melliferous plants in the study area.

Slides preparation and its microscopic studies

For the microscopical investigation pollen was treated with glycerin jelly for staining purposes. Following the method of (Ashfaq et al., 2018) the percentage of pollen fertility was determined. For different parameter of each species pollen readings were taken. P/E ratios were determined by dividing polar diameter of pollen over equatorial diameter.

For microscopic studies florets were removed from flowers and anthers were separated. The anthers were put on the slide and then treated using acetolysis method (Erdtman et al., 1963). The anthers were crushed so that pollen could come out from the anthers of flowers and debris was removed with the help of camel brush. The slides were then ready for further morphological study using light microscopy.

Table 1. Botanical names, family, collector, locality, and voucher number of collected plants.

S.no	Plant species	Family	Collector	Locality	Voucher no.
1	*Daucus carota* L	Apiaceae	Shabir Ahmad and Shakeel Ahmad	Paniala/ Dera Ismail Khan	SA-QAU-136
2	*Launaea procumbens* (Roxb) Ramayya and Rajagopal	Asteraceae	Shabir Ahmad and Ashfaq Khan	Domail/ Bannu	SA-QAU-125
3	*Plantago amplexicaulis* Cav	Plantaginaceae	Shabir Ahmad and Ashfaq Khan	Darya Khan/ Bhakkar	SA-QAU-178
4	*Medicago monantha* (C.A.Mey) Trautv	Fabaceae	Shabir Ahmad and Shakeel Ahmad	Paniala/ Dera Ismail Khan	SA-QAU-146
5	*Cousinia minuta* Boiss	Asteraceae	Shabir Ahmad and Shakeel Ahmad	Kot Chatta /Dera Ghazi Khan	SA-QAU-154
6	*Convolvolus arvensis* L.	Convolvulaceae	Shabir Ahmad	Gul Imam/ Tank	SA-QAU-143
7	*Dactyloctenium aegyptium* (L.) Willd	Poaceae	Shabir Ahmad and Asif Kamal	Bazar/ Paharpur	SA-QAU-160
8	*Vicia sativa* L	Fabaceae	Shabir Ahmad	Kharmatu/ Kohat	SA-QAU-98
9	*Fumaria indica* (Hausskn.) Pugsley	Fumariaceae	Shabir Ahmad and Muhammad Saqib	Paharpur/ Dera Ismail Khan	SA-QAU-114
10	*Sisymbrium irio* L.	Brassicaceae	Shabir Ahmad and Ashfaq Khan	Kotri/ Jamshoro	SA-QAU-185
11	*Malcolmia cabulica* (Boiss.) Hook. f. and Thomson	Brassicaceae	Shabir Ahmad and Khushdil Khan	Jarma/ Kohat	SA-QAU-137
12	*Arnebia hispidissima* (Lehm.) A. DC.	Boraginaceae	Shabir Ahmad and Asif Kamal	Bahadur Khel/ Karak	SA-QAU-161
13	*Astragalus hamosus* L	Fabaceae	Shabir Ahmad and Abdul Waheed	Abdul Khel/ Lakki Marwat	SA-QAU-155
14	*Hypecoum pendulum* L.	Papaveraceae	Shabir Ahmad and Uzma bibi	Gul Imam/ Tank	SA-QAU-140
15	*Brassica campestris* L	Brassicaceae	Shabir Ahmad and Ashfaq Khan	Shah Alam/ Tank	SA-QAU-138
16	*Asphodelus tenuifolius* Cav	Asphodelaceae	Shabir Ahmad	Nawabshah city/ Nawabshsah	SA-QAU-151
17	*Triticum aestivum* L	Poaceae	Shabir Ahmad and Asif Kamal	Shehbaz Khel/ Lakki Marwat	SA-QAU-122

Statistical analysis

Quantitative characters of pollen comprising polar diameter, equatorial diameter, colpi length, colpi width, spine length, spine width and exine thickness were measured. Consecutive five readings for every parameter of each pollen were noted and their mean, minimum, maximum values and standard deviation of each character were calculated using software SPSS Statistics 20. In tables values are showed as Mean (Min-Max) ± SE. (Tab. 2 and Tab. 3). 15 coded characters (those that showed variation) were used for preparing a matrix used as input for the Principal Component Analysis with the software PAST 3.16 (Hammer et al., 2001).

Results

The pollen morphology of 17 bee forage plants were investigated showing variations in pollen size and shapes. The variation in colpi length and width is represented in Fig. 1, while the variation in exine thickness is shown in Fig. 2. The general aspects of the plants, particularly of the flowers are shown (Fig. 3 and Fig. 4). The quantitative and qualitative features of pollen were described in Tab. 2 and Tab 3, and also shown in Fig. 5 and Fig. 6. Fertility rates of pollen were shown in Tab. 4 showing the stability of species in the study area.

Variations were observed in the bee flora of the study area (Tab. 2). *M. monantha, C. minuta,* showed large size pollen grains. *C. arvensis, F. indica, A. tenuifolius* and *T. aestivum* presented medium size grains, while *D. carota, L. procumbens, P. amplexicaulis, D. aegyptium, V. sativa, S. irio, M. cabulica, A. hispidissima, A. hamosus, H. pendulum* and *B. campestris* showed small sized pollen grains. The shape of the observed pollen grains was oblate-spheroidal, subprolate, prolate-spheroidal and oblate. *D. carota, L. procumbens, V. sativa, M. cabulica, A. hamosus, B. campestris* and *T. aestivum* showed oblate spheroidal shape. *A. tenuifolius, S. irio, D. aegyptium* had prolate-spheroidal shape. *P. amplexicaulis* presented subprolate shape and *A. hispidissima* an oblated one.

A. tenuifolius had the longest colpi length: 44.19 µm, while *P. amplexicaulis* had a minimum of 3.78 µm. *C. arvensis* had a maximum of colpi width: 21.33 µm and *L. procumbens* had the minimal width of 3.00 µm. Colpi noted in species of the study area were tricolporate and triporate. *D. aegyptium*, *T. aestivum* showed pollen grains without colpi but with a pore. *P. amplexicaulis* and *T. aestivum* were the species owning pollen grains with maximal P/E (1.18), while *Arnebia hispidissima* had the lowest P/E (0.71).

The exine thickness reached the highest value of 3.06 µm in *B. campestris* pollen grains, while *A. hamosus* had the minimal value of 0.75 µm. *L. procumbens* was the only species showing pollen grains with spines 1.89 µm long and 1.71 µm wide. The spines of *L. procumbens* pollen grains are an important feature for its identification (Ahmad et al., 2013). *C. minuta* pollen showed no spines on its exine, contrary to what was observed (Zafar et al., 2007). Both exine thickness and spine length are important features for taxa identifications.

Psilate and reticulate exine sculpturing was observed in the investigated species. *L. procumbens* showed an echinated type of pollen. *S. irio*, *M. cabulica, B. campestris* owned reticulate types of exine ornamentations. *A. hamosus* and *T. aestivum* had psilated pollen. The pollen fertility in all species was always over 67%, with *B. campestris* having the highest pollen fertility (93%), while *T. aestivum* had the lowest value (67%).

According to the field trip visits and interviews taken with local people and beekeepers in the study area, most of the honey in the spring season was produced by honeybees

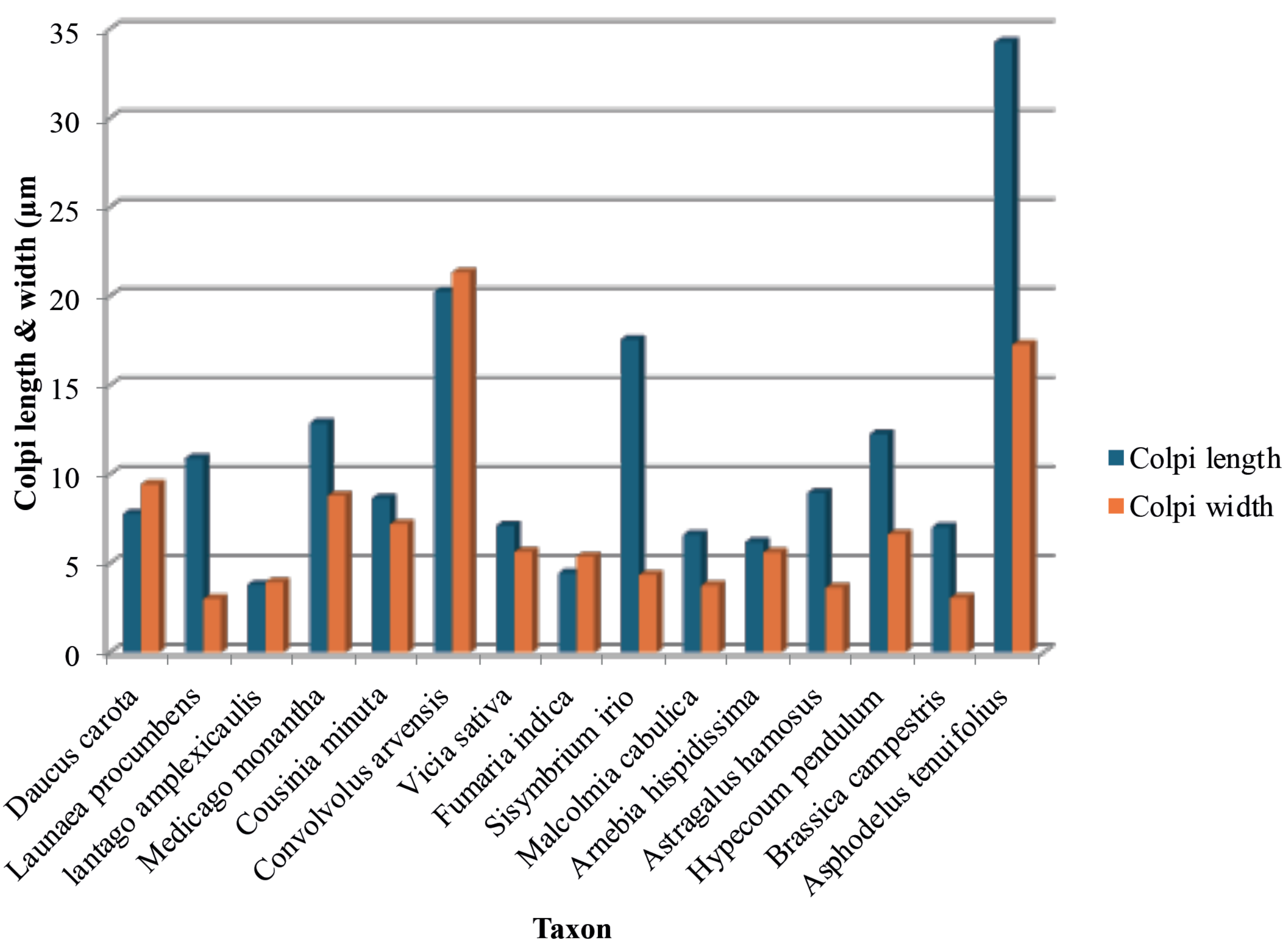

Figure 1. Variations in colpi length and width of melliferous plants from Southern Khyber Pakhtunkhwa, Pakistan.

visiting to *B. campestris*, *A. tenuifolius* and *A. hamosus*. In the field trip, it was observed that honeybees collect mostly pollen and nectar from herbaceous plants including weeds. It was also observed that in the study area honeybees visit white, yellow, purple, and pinkish, light-brown-colored flowers. The most frequently visited species had white and yellow flowers. The pollen grain characters were used to produce a matrix of morphological characters used as input for PAST software. The principal component analysis was shown in Fig. 7. The species was well distributed on the graph with *S. irio* in a particularly isolated position, apparently mainly due to the particularly small colpi length.

Discussion

A total of 17 plants belonging to 11 different families were analyzed through microscopic analysis. Brassicaceae and Fabaceae were the dominant families of the study area during the season of spring, with three species each. Asteraceae and Poaceae (this last family inserted for comparison, not including nectar producing plants but producing pollen) were represented by 2 species, while the rest of the families (Apiaceae, Asphodelaceae, Papaveraceae, Fumariaceae, Boraginaceae, Convolvulaceae and Plantaginaceae) were represented by a single species. Several scientists from different regions of the world reported about local honeybee flora (Bista & Shivakoti, 2001). Ting, (1961) studied Umbelliferae in America. The pollen of Brassicaceae has been studied (Erdtman, 1965). Flowering period of plants in both spring and autumn plays a very important role for honeybees regarding their foraging activities and honey production in southern Pakistan (Latif et al., 2019; Khan et al., 2022).

In the seventeen melliferous plants analyzed from southern Pakistan, a total of five different types of pollen i.e monoporate, monocolpate, tricolpate, tricolporate and pantaporate were reported. The pollen grain shapes examined were oblate-spheroidal, subprolate, prolate-spheroidal and oblate. Apart from pollen types and shapes, it also varies in pores, spines, and sculpturing. Spines found

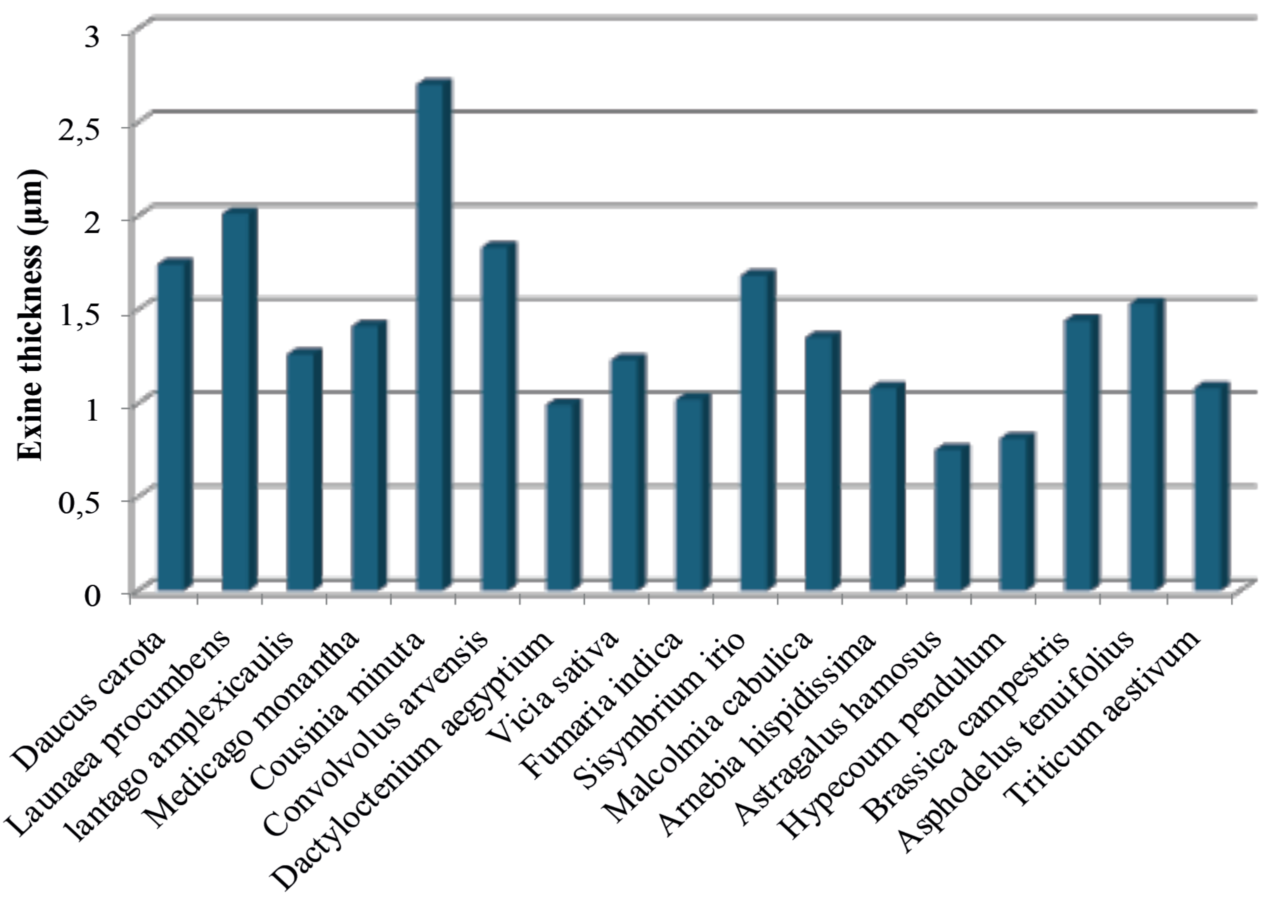

Figure 2. Variations in exine thickness of melliferous plants from Southern Khyber Pakhtunkhwa, Pakistan.

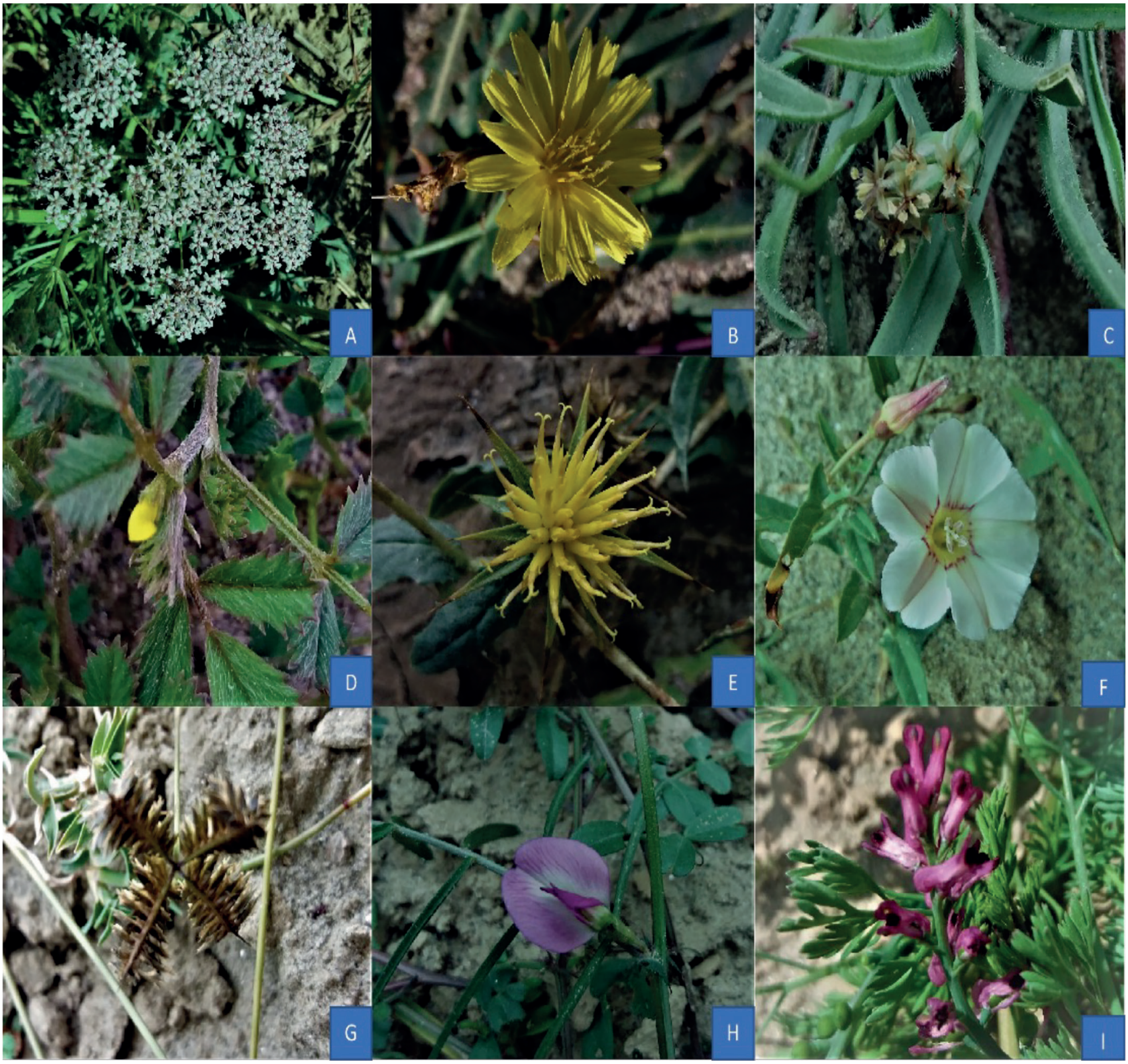

Figure 3. Plants Collected- (A) *Daucus carota* L (B) *Launaea procumbens* (Roxb) Ramayya and Rajagopal (C) *Plantago amplexicaulis* Cav (D) *Medicago monantha* (C.A.Mey) Trautv (E) *Cousinia minuta* Boiss (F) *Convolvulus arvensis* L. (G) *Dactyloctenium aegyptium* (L.) Willd (H) *Vicia sativa* L (I) *Fumaria indica* (Hausskn.) Pugsley

on the surface of pollen were very rare and most pollen studies have found them to be small. The average fertility percentage of the pollen here recorded was 83.03 % with the highest fertility at 89 % and the smallest 67 %. Such data would confirm the predominance of sexual reproduction in the considered species, at least in the geographical region of the study (Johnson, 1975). The high rate of pollen fertility is evidence that the plants are stable and actively reproducing in the study area.

The current study is the first contribution from southern Pakistan about the plants visited by honeybees for pollen and nectar formation. Khan et al., (2012) determined the melliferous potential and medicinal uses of honey in the current study area, but the investigation was only preliminary. The identification of melliferous species helps in developing pollen literature, which is useful for increasing honey production in coming the seasons. Pound et al., (2018) revealed the role of *Brassica* species for honey productions

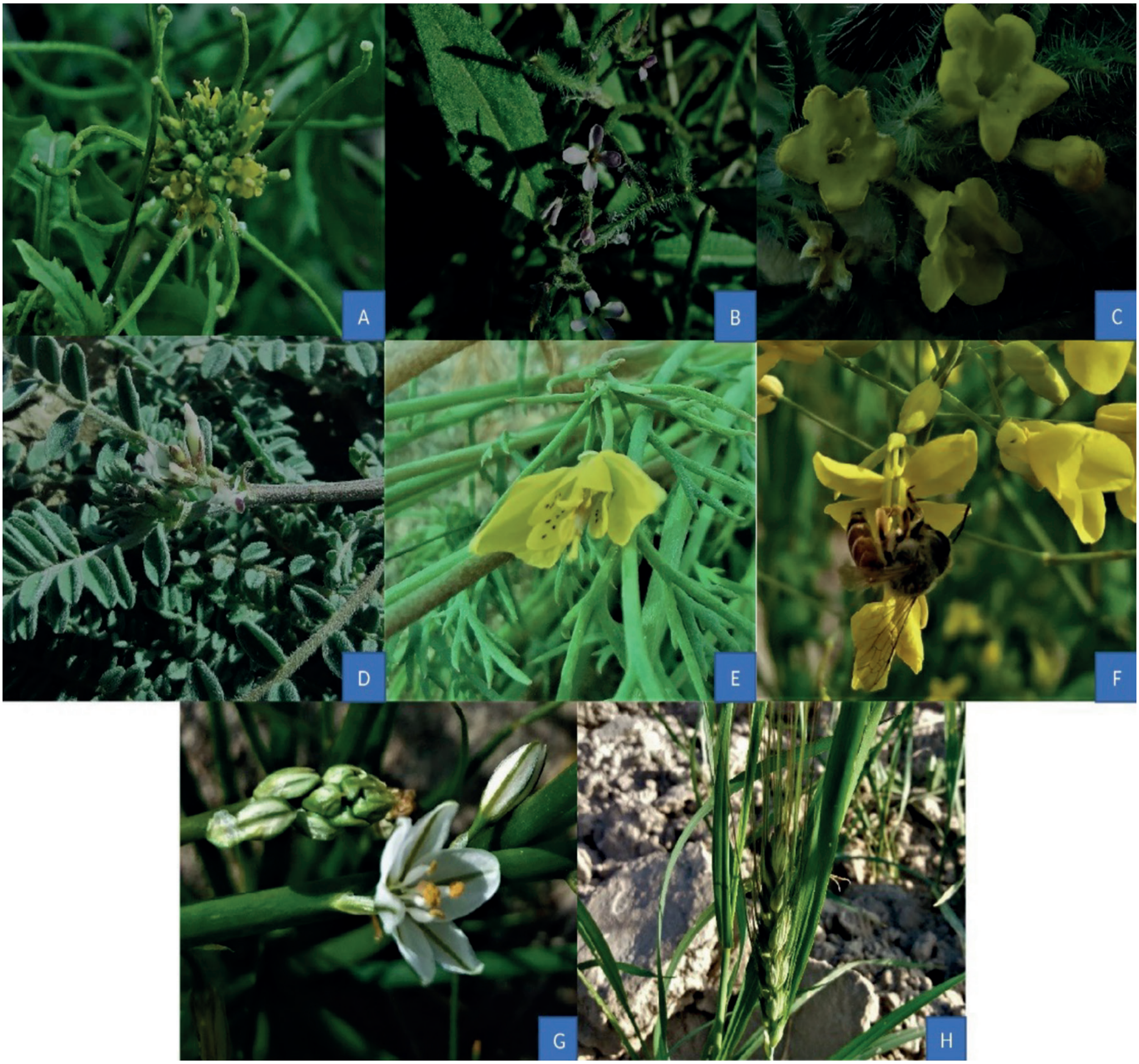

Figure 4. Plants Collected- (A) *Sisymbrium irio* L. (B) *Malcolmia cabulica* (Boiss.) Hook. f. and Thomson (C) *Arnebia hispidissima* (Lehm.) A. DC. (D) *Astragalus hamosus* L (E) *Hypecoum pendulum* L. (F) *Brassica campestris* L (G) *Asphodelus tenuifolius* Cav (H) *Triticum aestivum* L.

in Ponteland, UK. Palynological studies of the study area showed that pollens of the study area are very diverse in nature showing varies of pollen size and shapes which are very helpful for the purpose of identification (Perveen et al., 2004; Yedomonhan et al., 2012).

Different parameters of pollen containing polar diameter, equatorial diameter, colpus length, colpus width, spine length, spine width, number of spines, pores, fertile and sterile pollen are vital features to distinguish one species from another. Pollen examined in current study varies in size and shape. Tricolporate is one of the most common types of pollen (Muller, 1970). *Plantago amplexicaulis* is the only species showing pantaporate pollen.

Pollen data is essential for species identification that are pollinated by honeybees (Yedomonhan et al., 2012). The current microscopic study plays a vital role in the description of pollen morphology (Ashfaq et al., 2018). It has been proved that these results are very helpful for taxonomic identification. The PCA analysis did not show any evident clustering related to the pollination syndrome, since anemophylous plants were quite distantly positioned in the graph. More evident was the clustering of phylogenetically related species, such as *A. hamosus*/*V. sativa* (Fabaceae) or the couple *M. cabulica/B. campestris* (Brassicaceae). For this reason, we can consider the pollen characters useful for melissopalynological identification. Pollen morphological study is very useful in finding out many taxonomic and systematic problems. Some species are known for having other pollinators besides bees and that may influence the general floral structure.

A very wide range of variations were observed using microscopic techniques for selected species from the study area, showing that the field of palynology plays a vital role in the evolutionary studies of plant species. Our light microscopy results show similarity to the previously reported knowledge from different regions of Pakistan (Ahmad et al., 2019; Ashfaq et al., 2018). Pollen size varies from small to large, monocolpate to tricolporate, subprolate to oblate-spheroidal and reticulate to psilate. *B. campestris, M. cabulica, S. irio* have reticulate exine sculpturing, *L. procumbens* have echinated and the rest of the pollen have psilated exine ornamentations. Pollen morphological studies in the present study link the foraging behaviors of honeybees with specific plants. The results of the present study concluded that pollen

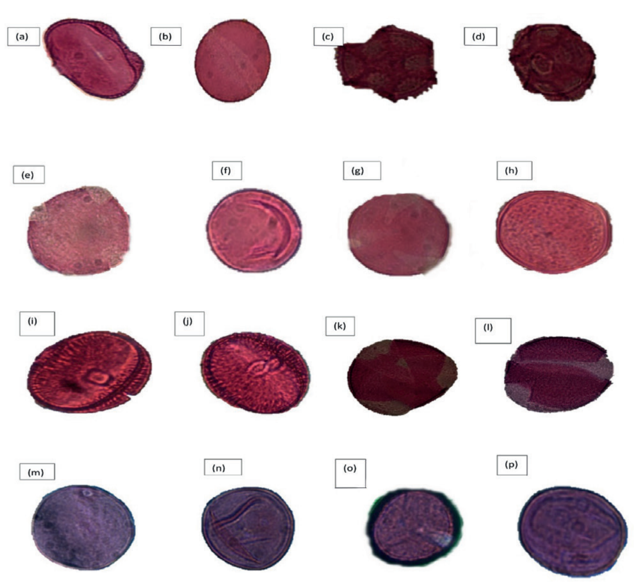

Figure 5. Light microscopy pollen micrographs- *Daucus carota* L (a) Polar view (b) Equatorial view; *Launaea procumbens* (Roxb) Ramayya and Rajagopal (c) Polar view (d) Equatorial view; *Plantago amplexicaulis* Cav (e) Polar view (f) Equatorial view; *Medicago monantha* (C.A.Mey) Trautv (g) Polar view (h) Equatorial view; *Cousinia minuta* Boiss (i) Polar view (j) Equatorial view; *Convolvolus arvensis* L. (k) Polar view (l) Equatorial view; *Dactyloctenium aegyptium* (L.) Willd (m) Polar view (n) Equatorial view; *Vicia sativa* L (o) Polar view (p) Equatorial view.

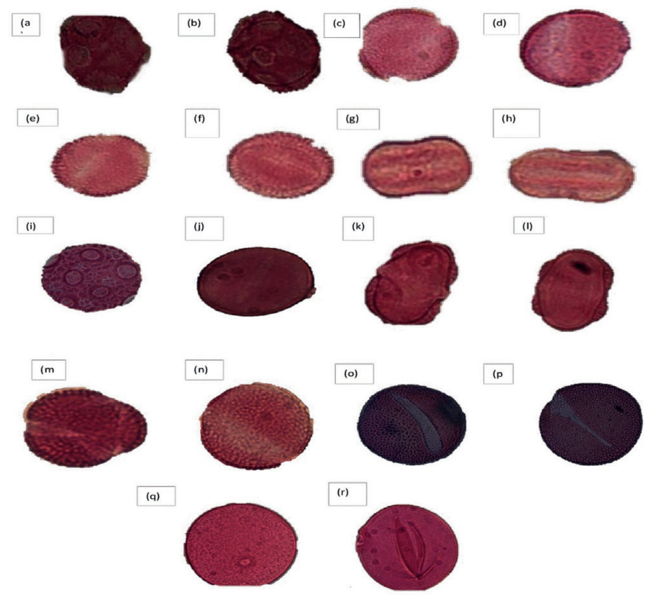

Figure 6. Light microscopy pollen micrographs- *Fumaria indica* (Hausskn.) Pugsley (a) Polar view (b) Equatorial view; *Sisymbrium irio* L. (c) Polar view (d) Equatorial view; *Malcolmia cabulica* (Boiss.) Hook. f. and Thomson (e) Polar view (f) Equatorial view; *Arnebia hispidissima* (Lehm.) A. DC. (g) Polar view (h) Equatorial view; *Astragalus hamosus* L (i) Polar view (j) Equatorial view; *Hypecoum pendulum* L. (k) Polar view (l) Equatorial view; *Brassica campestris* L (m) Polar view (n) Equatorial view; *Asphodelus tenuifolius* Cav (o) Polar view (p) Equatorial view; *Triticum aestivum* L. (q) Polar view (r) Equatorial view.

morphology of melliferous species plays a vital role in plant identification and floral calendar preparation.

D. carota is considered largely pollinated by Diptera (Ahmad & Aslam, 2002). The same can be said about *L. procumbens* (Sajjad et al., 2010). Surprisingly, the observed measures in pollen grains were quite different from those with the polar diameter of 22.44 **µm** in this investigation vs. 34.65 **µm and the width of colpi (3 µm vs. 8.45 µm). We can hence assume that a relatively high degree of intraspecific variation is present, possibly based on the geographical location of the investigated population of the taxa** (Ahmad & Aslam, 2002). Also, the anemophilous *P. amplexicaulis* had pollen measures variable in the current analysis with respect to (Fierascu et al., 2021). Evident differences were observed also in *V. sativa* with respect to the observations of (Khan et al., 2020). *S. irio* was showed in isolated position in the PCA. Our results do not allow us to give a final explanation, but Ara et al., (2019) observed that honeybees feed in this species both on nectar and pollen, and the isolated position may depend on specific adaptation of the pollen grain to this type of nutrition. Also, *A. hispidissima* is an isolated position and this species is known to be used as a source of both nectar and pollen (Abou-Shaara, 2019).

Conclusion

The current study was carried out to identify melliferous plants visited by honeybees using pollen morphology during the seasons of spring and autumn. Variations investigated in the qualitative and quantitative features of pollen showed sufficient changes to be considered useful markers for melissopalynological analysis. This investigation revealed that a complete set of characters is helpful for the identification of species in the field of plant taxonomy and systematics. *C. arvensis* had the largest polar diameter, measuring 51.72 µm and an equatorial diameter 44.43 µm. *M. cabulica* has the smallest polar diameter, 12.69 µm and an equatorial diameter of 13.86 µm. Variations in exine sculpturing have very distinguishing features in plant systematic and biodiversity. The current study was conducted for the first time in the study area to analyze bee foraged species from various localities of the study area, aiming at a better knowledge of melliferous species among local beekeepers and their conservation in the study area. The observed intraspecific variation of pollen grains of the same species from different geographical areas is an indication that these data may also be useful to assess the provenance of honey.

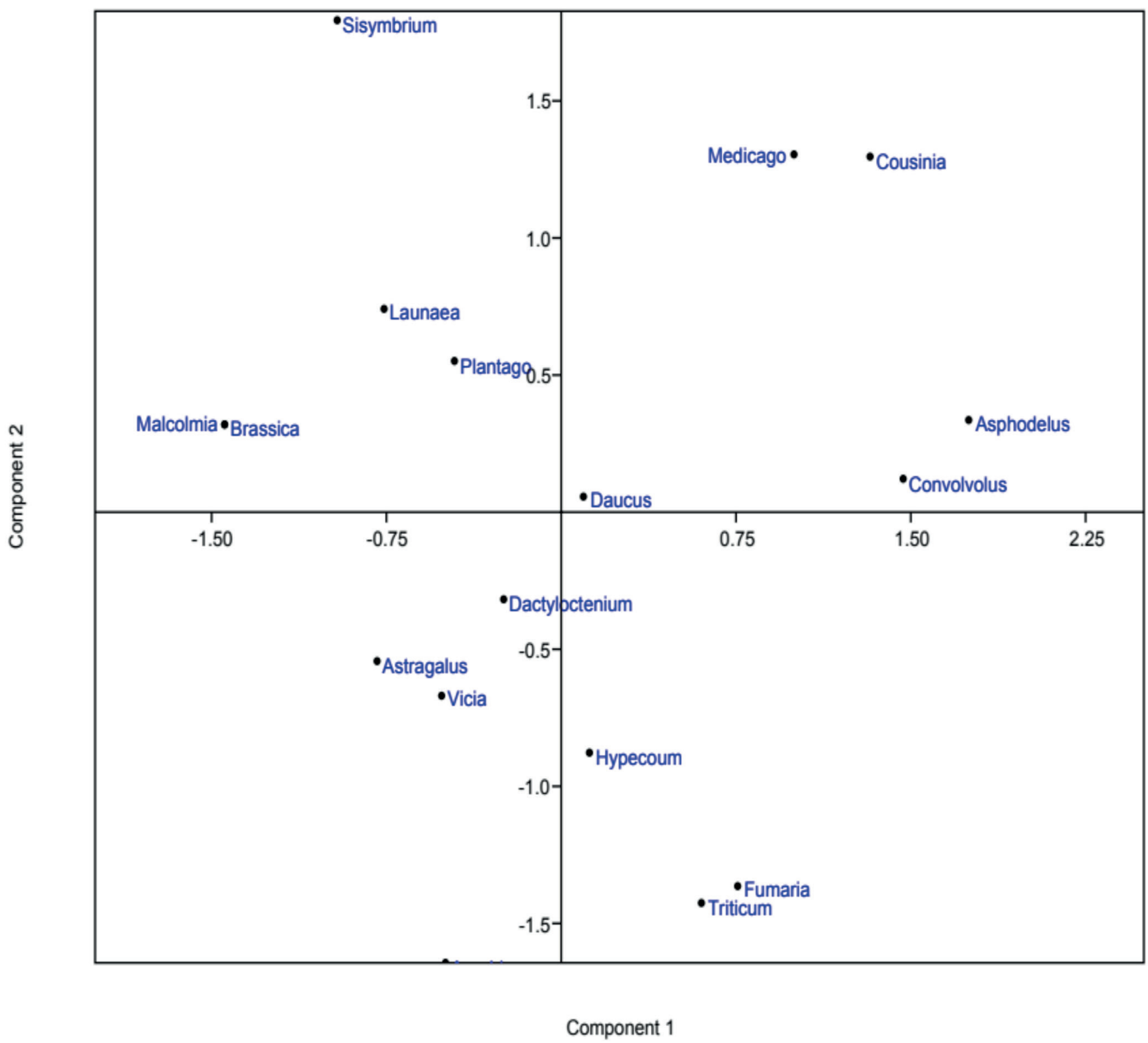

Figure 7. PCA based on the coded pollen grain morphological characters.

Table 2. Micromorphological qualitative features of pollen.

S.no	Species name	Pollen size	Pollen type	Pollen shapes	Colpi/Pore	Spines	Exine sculpturing
1	*Daucus carota* L	Small	Tricolporate	Oblate-Spheroidal	Present	Absent	-
2	*Launaea procumbens* (Roxb) Ramayya and Rajagopal	Small	Tricolporate	Oblate-Spheroidal	Present	Present	Echinate
3	*Plantago amplexicauli*s Cav	Small	Pantaporate	Subprolate	Absent	Absent	Psilate
4	*Medicago monantha* (C.A.Mey) Trautv	Large	Tricolporate	Prolate-Spheroidal	Present	Absent	Psilate
5	*Cousinia minuta* Boiss	Large	Tricolporate	Prolate-Spheroidal	Present	Absent	-
6	*Convolvolus arvensi*s L.	Medium	Tricolporate	Oblate-Spheroidal	Present	Absent	-
7	*Dactyloctenium aegyptium* (L.) Willd	Small	Monoporate	Prolate-Spheroidal	Present	Absent	Psilate
8	*Vicia sativa* L	Small	Tricolporate	Oblate-Spheroidal	Present	Absent	Psilate
9	*Fumaria indic*a (Hausskn.) Pugsley	Medium	Tricolpate	Oblate-Spheroidal	Present	Absent	-
10	*Sisymbrium irio* L.	Small	Tricolpate	Prolate-Spheroidal	Present	Absent	Reticulate
11	*Malcolmia cabulica* (Boiss.) Hook. f. and Thomson	Small	Tricolpate	Oblate-Spheroidal	Present	Absent	Reticulate
12	*Arnebia hispidissima* (Lehm.) A. DC.	Small	Tricolpate	Oblate	Present	Absent	-
13	*Astragalus hamosus* L	Small	Tricolporate	Oblate-Spheroidal	Present	Absent	Psilate
14	*Hypecoum pendulu*m L.	Small	Tricolpate	Oblate-Spheroidal	Present	Absent	-
15	*Brassica campestris* L	Small	Tricolpate	Oblate-Spheroidal	Present	Absent	Reticulate
16	*Asphodelus tenuifolius* Cav	Medium	Monocolpate	Prolate-Spheroidal	Present	Absent	-
17	*Triticum aestivum* L.	Medium	Monoporate	Oblate-Spheroidal	Present	Absent	Psilate

Table 3. Micromorphological quantitative features of pollen.

S.no	Taxon	P/E ratio	Exine thickness Mean (Min-Max) SE (um)	Polar diameter Mean (Min-Max) SE (um)	Equatorial diameter Mean (Min-Max) SE (um)	Length of colpi Mean (Min-Max) SE (um)	Width of colpi Mean (Min-Max) SE (um)	Length of spine Mean (Min-Max) SE (um)	Width of spine Mean (Min-Max) SE (um)
1	*Daucus carota* L	0.96	1.74 (1.35-2.10) ±0.14	19.77 (17.70-21.00) ±0.57	20.55 (17.40-23.70) ±1.25	7.77 (6.30-9.15) ±0.50	9.42 (7.50-11.25) ±0.79	A	A
2	*Launaea procumbens* (Roxb) Ramayya and Rajagopal	0.95	2.01 (0.90-3.30) ±0.41	22.44 (21.00-25.20) ±0.74	23.40 (18.60-25.80) ±1.34	10.89 (9.45-15.00) ±1.05	3.00 (2.25-3.90) ±0.29	1.89 (1.20-2.55) ±0.24	1.71 (1.05-3.15) ±0.37
3	*Plantago amplexicaulis* Cav	1.18	1.26 (0.60-1.95) ±0.27	22.38 (19.50-26.40) ±1.31	18.96 (17.70-20.40) ±0.50	3.78 (2.25-6.45) ±0.73	3.93 (3.00-6.00) ±0.54	A	A
4	*Medicago monantha* (C.A.Mey) Trautv	1.11	1.41 (0.90-1.80) ±0.16	27.42 (26.10-28.95) ±0.49	24.54 (22.50-25.65) ±0.54	12.87 (7.05-18.75) ±1.91	8.79 (6.75-10.20) ±0.72	A	A
5	*Cousinia minuta* Boiss	1.04	2.70 (1.95-3.90) ±0.36	28.62 (27.45-30.30) ±0.50	27.45 (23.70-32.40) ±1.62	8.64 (4.20-15.30) ±2.15	7.20 (3.75-12.75) ±7.20	A	A
6	*Convolvolus arvensis* L.	1.16	1.83 (1.20-3.00) ±0.31	51.72 (44.40-59.25) ±2.95	44.43 (34.05-54.90) ±3.78	20.25 (10.95-36.45) ±4.39	21.33 (14.55-29.10) ±2.40	A	A
7	*Dactyloctenium aegyptium* (L.) Willd	1.07	0.99 (0.60-1.50) ±0.16	22.68 (19.80-22.85) ±1.29	21.06 (19.35-23.40) ±0.87	A	A	A	A
8	*Vicia sativa* L	0.99	1.23 (0.60-1.95) ±1.23	22.68 (16.50-28.50) ±2.10	22.74 (18.45-30.00) ±2.45	7.11 (3.75-10.35) ±1.29	5.64 (2.10-8.70) ±1.32	A	A
9	*Fumaria indica* (Hausskn.) Pugsley	0.89	1.02 (0.45-1.65) ±0.20	31.77 (28.80-35.55) ±1.25	35.64 (24.45-42.45) ±3.13	4.44 (2.85-7.80) ±0.89	5.37 (4.50-6.00) ±0.30	A	A
10	*Sisymbrium irio* L.	1.03	1.68 (1.35-2.10) ±0.13	18.24 (15.75-21.00) ±0.90	17.55 (15.60-18.75) ±0.62	4.05 (2.55-6.45) ±0.67	4.35 (2.70-5.25) ±0.47	A	A
11	*Malcolmia cabulica* (Boiss.) Hook. f. and Thomson	0.91	1.35 (0.90-1.80) ±0.15	12.69 (11.40-13.50) ±0.40	13.86 (12.15-15.30) ±0.68	6.60 (3.45-9.15) ±0.95	3.75 (1.50-7.50) ±1.14	A	A
12	*Arnebia hispidissima* (Lehm.) A. DC.	0.71	1.08 (0.60-1.80) ±0.22	13.65 (12.90-14.70) ±0.30	19.20 (12.30-28.50) ±3.41	6.18 (2.10-10.95) ±1.45	5.61 (1.80-10.35) ±1.44	A	A
13	*Astragalus hamosus* L	0.98	0.75 (0.45-1.05) ±0.11	18.69 (17.70-19.95) ±0.44	18.90 (18.30-19.35) ±18.69	8.94 (6.60-11.40) ±0.88	3.63 (2.10-6.00) ±0.66	A	A
14	*Hypecoum pendulum* L.	0.94	0.81 (0.45-1.35) ±0.15	23.01 (22.20-23.85) ±0.33	24.24 (22.50-25.65) ±0.55	12.24 (8.70-15.30) ±1.29	6.63 (4.35-10.05) ±0.99	A	A
15	*Brassica campestris* L	0.98	1.44 (0.90-1.95) ±0.18	18.93 (17.40-21.00) ±0.67	19.26 (15.00-22.80) ±1.45	7.02 (3.45-11.70) ±1.367	3.06 (1.20-9.00) ±1.49	A	A
16	*Asphodelus tenuifolius* Cav	1.06	1.53 (1.20-1.95) ±0.13	47.01 (44.10-50.25) ±1.08	44.19 (32.85-56.25) ±3.73	34.26 (22.50-43.35) ±3.59	17.28 (6.90-32.10) ±4.60	A	A
17	*Triticum aestivum* L.	1.18	1.08 (0.75-1.50) ±0.13	38.85 (37.20-40.50) ±0.60	32.67 (27.00-38.25) ±2.33	A	A	A	A

Table 4. Pollen fertility percentage for spring melliferous flora from Southern Khyber Pakhtunkhwa, Pakistan.

S.no	Species name	Fertile pollen	Sterile pollen	Fertility (%)
1	*Daucus carota* L	198	25	89
2	*Launaea procumbens* (Roxb) Ramayya and Rajagopal	58	11	84
3	*Plantago amplexicaulis* Cav	36	10	78
4	*Medicago monantha* (C.A.Mey) Trautv	50	11	82
5	*Cousinia minuta* Boiss	85	12	88
6	*Convolvolus arvensis* L.	55	17	76
7	*Dactyloctenium aegyptium* (L.) Willd	52	9	85
8	*Vicia sativa* L	68	16	81
9	*Fumaria indica* (Hausskn.) Pugsley	27	9	75
10	*Sisymbrium irio* L.	176	24	88
11	*Malcolmia cabulica* (Boiss.) Hook. f. and Thomson	113	25	82
12	*Arnebia hispidissima* (Lehm.) A. DC.	63	17	79
13	*Astragalus hamosus* L	244	27	89
14	*Hypecoum pendulum* L.	148	19	89
15	*Brassica campestris* L	478	38	93
16	*Asphodelus tenuifolius* Cav	48	7	87
17	*Triticum aestivum* L.	34	17	67

Acknowledgments

The authors extend their appreciation to Taif university, Saudi Arabia, for supporting this work through project number (TU-DSPP-2024-254).

Funding

This research was funded by Taif University, Taif, Saudi Arabia (TU-DSPP-2024-254).

Conflict of interest

The authors of this manuscript have no conflicts of interest to declare it.

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