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Pollen Visualization of Turkish Flora of Selected Plant Species under Light, Scanning, and Transmission Microscopy

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Abstract: This study aims to examine pollen morphologically via studies of 16 plant taxa from Turkish flora. The pollen structures of the taxa examined, and their microscopic relevance, was discussed and studied with the help of microscopic visualization using scanning and transmission microscopies. Pollen grains were first acetolyzed, and then quantitative and qualitative pollen features were used to evaluate the species. The pollen grains were prolate, spherical, elliptic, tricolporate, 3-colpate, and hexacolpate. Sculpturing patterns of surfaces vary from reticulate, micro-reticulate, and striate regulate. As the findings reveal, palynological data can aid in the taxonomic classification of Turkish floral species. Microscopic implications can be made via micromorphological examination to correctly identify the species. While the pollen morphology of 16 taxa collected from the study area was studied for the first time from this region, the palynological research of some taxa was introduced to the literature for the first time with this study. Pollen morphology and photographic and statistical data of the taxa in our study were determined. This study contributed to bee plant research, melisapalinological studies, and systematic botanical flora studies.

Keywords: Turkish flora; pollen morphology; surface; clustering analysis (CA); principle component analysis (PCA)

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1. Introduction

Palynological features provide ancillary knowledge and indulge the systematic stand of species with its individual family. Pollen morphology can help support the taxonomic approach [1]. The word palynology was first confined by Hyde and William [2]. Since then, the botanical sciences have become an advanced sub-division with distinct applications. The microscopic investigation of spores, pollen, and other imperceptible structures is known as palynology [3]. Previous studies revealed that pollen morphology shows advancement in taxonomic characteristics like pollen type, exine and sexine appearance, position, measurement, number of apertures, type, and ornamentation [4]. Pollen grains are considered advantageous in morphological and phylogenetic studies [5–10]. Angiosperm might possess pollen apertures that are categorized based on their type; position and number vary from simple to composite [11].

Among angiosperm, pollen morphology plays an important role in advanced classification, which is carried out by using the maximum number of possible characteristics [12]. The division of families into subfamily tribes and subtribes is facilitated by palynology [13]. Different families have been divided into subfamilies and tribes on the bases of pollen morphology [14,15]. Some characteristics are specific to the family, which could include the size of pollen, type of sculpturing, aperture type, etc., which become a key to identifying that family. Asteraceae angiosperms show the most diverse types of pollen, showing the maximum number of pollen characteristics compared to any other taxa [7,16–18].

Palynology is also helpful for various uses, such as atmospheric pollen inspection, spore production, and the archaeological excavation of shipwrecks. Primarily, the investigation of morphological diversity in pollen is directly concerned with the classification of plants [18,19]. Palyno-morphological characteristics are fruitful for solving various taxonomic problems at species, genus, and family levels [7,20–22].

The primary objective of this research is to offer detailed insight into micromorphological features via SEM and TEM among selected plants from Turkey. In our study, taxa belonging to Lamiaceae, Boraginaceae, Fabaceae, Amaryllidaceae, Scrophulariaceae, Apiaceae, and Convolvulaceae families were selected and palynologically analyzed. The pollen morphology of 16 taxa collected from the study area was studied for the first time from this region, and palynological research of some taxa was introduced to the literature for the first time with this study. The current study provides a comprehensive analysis of the palyno-morphology for determining the extent to which these microscopic techniques can be utilized to discriminate among the taxa analyzed.

2. Materials and Methods

2.1. Specimen Collection and Plant İdentification

The 16 taxa belonging to different families were collected from Turkey (Figure 1, Table 1). The collection period runs from April 2020 to November 2021, including two seasons. Localities, times, and collector numbers are given in Table 1. After collecting the plants, they were then identified with the help of flora from Turkey. Plant material was identified, and voucher specimens were deposited in a Turkish herbarium (name it and insert its acronym). Flowering specimens were collected. Anthers were removed to create the specimens. Pollen was extracted directly and investigated with microscopic tools.

Figure 1. Map of the collection sites of the study area (Source: Google). The red zone represents the working area.

2.2. Light Microscopy (LM)

Pollen obtained from the samples was prepared according to the methods of Ertman and Wodehouse [23,24]. Some changes were made to the acetolysis procedure. Plant matter was marked by applying a drop of glycerin jelly. Each specimen received 4–5 prepared slides in total. Pollen samples were measured from at least 20 completely evolved grains according to the pattern beneath a Euromex CMEX- 10 PRO light microscope (Papenkamp 20, Holland) (100×). These measurements are reported in Tables 2 and 3 and in Figures 2–7. The terminology used is mainly from Faegri and Iversen, Ertman, and Kılıç et al. [25–27].

Table 1. Plant sampling, vouchering, and endemism.

No	Plant Name	Locality	Endemic Situation	Voucher Specimen
1.	*Ajuga reptans* L.	Bingöl-Tunceli	-	AD-257
2.	*Micromeria cremnophila* Boiss. Et Heldr. subsp. *anatolica* P. H. Davis	Elazig-Harput castle	Endemic	AD-1011
3.	*Onosma bornmuelleri Hausskn.*	Bingol	Endemic	AD-200
4.	*Onosma sericeum* Wild.	Bingol	-	AD-176
5.	*Rindera caespitosa* (A. DC.) Bunge	Bingol	Endemic	AD-78
6.	*Paracaryum* Schreber Boiss. subsp. *cristatum* Schreber Boiss.	Elazig	Endemic	AD-1012
7.	*Paracaryum strictum* (Koch) Boiss.	Elazig	Endemic	AD-1051
8.	*Astragalus aucheri* Boiss.	Elazig	Endemic	AD-1034
9.	*Astragalus aureus* Wild.	Bingol	-	AD-155
10.	*Astragalus dactylocarpus* Boiss.	Bingol	-	AD-230
11.	*Ixiolirion tataricum* Pallas Herbert subsp. *montanum* Labill. Takht.	Bingöl	-	AD-158
12.	*Linaria kurdica* Boiss. Et HOHEN. subsp. *kurdica* Boiss. Et Huet	Elazig	-	AD-1098
13.	*Linaria grandiflora* Desf.	Bingol	-	AD-253
14.	*Foeniculum vulgare* Miller	Bingol	-	AD-543
15.	*Convolvulus carduchorum* Davis	Elazig	Endemic	AD-123
16.	*Convolvulus reticulatus* Choisy subsp. *reticulatus* Choisy	Bingol	-	AD-112

Table 2. Qualitative micromorphological pollen characteristics of selected plants.

		Plant Name	Shape	Type and Number of Aperture	Exine Ornamentation	Colpi Orientation
1.	Lamiaceae	*A. reptans*	Oblate-spheroidal	3-colporate	Tectatae-granulatae	Sunken, angular
2.		*M. cremnophila* ssp. *anatolica*	Sub-oblate	6-colpate	Reticulate	Sunken
3.	Boraginaceae	*O. bornmuelleri*	Sub-prolate	3-colporate	Scabrate-reticulate	Sunken
4.		*O. sericea*	Sub-prolate	3-colporate	Scabrate-reticulate	Sunken
5.		*R. caespitosa*	Sub-prolate	3-colporate	Granulate	Pseudocolpus, margo
6.		*P. cristatum* ssp. *cristatum*	Prolate	6-colporate	Rugulate	Pseudocolpus, margo
7.		*P. strictum*	Prolate	6-colporate	Rugulate	Pseudocolpus, margo

Table 2. *Cont.*

		Plant Name	Shape	Type and Number of Aperture	Exine Ornamentation	Colpi Orientation
8.	Fabaceae	*A. aucheri*	Prolate	3-colporate	Reticulate	Sunken, angular, margins distinct
9.		*A. aureus*	Prolate	3-colporate	Reticulate	Sunken, angular, margins distinct
10.		*A. dactylocarpus*	Prolate	3-colporate	Reticulate	Sunken, angular, margins distinct
11.	Amaryllidaceae	*I. tataricum* subsp. *montanum*	Sub-prolate	3-colporate	Perforate	Sunken, angular, margins distinct
12.	Scrophulariaceae	*L. kurdica* subsp. *kurdica*	Oblate-spheroidal	3-colporate	Reticulate	Sunken
13.		*L. grandiflora*	Oblate-spheroidal	3-colporate	Reticulate	Sunken
14.	Apiaceae	*F. vulgare*	Per-prolate	3-colporate	Scabrate	Sunken
15.	Convolvulaceae	*C. carduchorum*	Oblate-spheroidal	3-colporate	Perforate	Sunken
16.		*C. reticulatus* subsp. *reticulatus*	Oblate-spheroidal	3-colporate	Perforate	Sunken

Table 3. Quantitative pollen features via microscopic measurements.

	Plant Name	Exine Thickness	Polar Diameter	Equatorial Diameter	P/E Ratio	Colpi Length	Colpi Width	Pores Length	Pores Width
Lamiaceae	*A. reptans*	1.1 ± (0.1)	22.4 ± (2.0)	22.7 ± (2.1)	0.98	16.7 ± (1.2)	7.6 ± (1)	-	-
	M. cremnophila ssp. *anatolica*	1.5 ± (1.2)	40 ± (2.3)	46.6 ± (1.3)	0.85	23.5 ± (1.3)	8.3 ± (1.6)	-	-
Boraginaceae	*O. bornmuelleri*	1.0 ± (0.5)	17.3 ± (1.0)	14.7 ± (1)	1.17	13.9 ± (0.3)	-	3 ± (0.5)	3.29 ± (0.5)
	O. sericea	1.0 ± (0.5)	17.8 ± (1.0)	14.9 ± (0.1)	1.19	13.8 ± (0.1)	-	2.83 ± (0.3)	2.29 ± (0.5)
	R. caespitosa	0.5 ± (0.3)	13 ± (1.0)	10 ± (0.1)	1.30	7.1 ± (1.0)	2.5 ± (1.2)	3.8 ± (0.5)	5.3 ± (0.3)
	P. cristatum ssp. *cristatum*	0.24 ±(0.2)	9.1 ± (0.3)	5.6 ± (0.1)	1.62	5.3 ± (1.3)	1.5 ± (1.5)	2.9 ± (0.3)	4.3 ± (0.7)
	P. strictum	0.23 ± (0.1)	10.1 ± (0.5)	5.2 ± (0.2)	1.94	6.5 ± (1.3)	1.9 ± (1.1)	2.8 ± (0.5)	4.7 ± (0.3)
Fabaceae	*A. aucheri*	0.45 ± (0.1)	35.5 ± (1.9)	23.6 ± (1.28)	1.50	29.2 ± (1.3)	4.2 ± (0.3)	6.3 ± (0.2)	4.2 ± (0.5)
	A.aureus	0.5 ± (0.1)	35.8 ± (1.2)	23.5 ± (1.0)	1.52	27.5 ± (0.3)	3.9 ± (0.9)	5.9 ± (0.6)	4.2 ± (0.7)
	A. dactylocarpus	0.5 ± (0.1)	38.7 ± (2.2)	25.3 ± (1.5)	1.52	29.8 ± (1.5)	5.5 ± (1.2)	7.4 ± (1.2)	5.7 ± (1.1)
Amaryllidaceae	*I. tataricum* subsp. *montanum*	1.8 ± (0.7)	51.2 ± (4.3)	41.6 ± (3.3)	1.23	38.9 ± (1.9)	15.0 ± (1.8)	-	-
Scrophulariaceae	*L. kurdica* subsp. *kurdica*	0.9 ± (0.2)	10.9 ± (1.2)	11.2 ± (1.1)	0.97	8.01 ± (2.3)	2.4 ± (2.3)	4.4 ± (1.4)	4.5 ± (1.8)
	L. grandiflora	0.82 ± (0.2)	10.0 ± (1.2)	10.3 ± (1.3)	0.97	7.85 ± (1.2)	3.8 ± (0.9)	4.23 ± (1.2)	3.98 ± (1.5)
Apiaceae	*F. vulgare*	1.8 ± (0.5)	26.8 ± (1.9)	10.0 ± (1.8)	2.68	14.9 ± (2.9)	4.0 ± (2.3)	3.8 ± (0.9)	4.1 ± (0.3)
Convolvulaceae	*C.carduchorum*	2.1 ± (0.4)	42.3 ± (1.3)	43 ± (1)	0.97	18.2 ± (1.9)	13.7 ± (1.1)	-	-
	C. reticulatus subsp. *reticulatus*	1.9 ± (0.3)	41.9 ± (0.9)	43.5 ± (1.2)	0.97	17.3 ± (1.2)	12.4 ± (1.3)	-	-

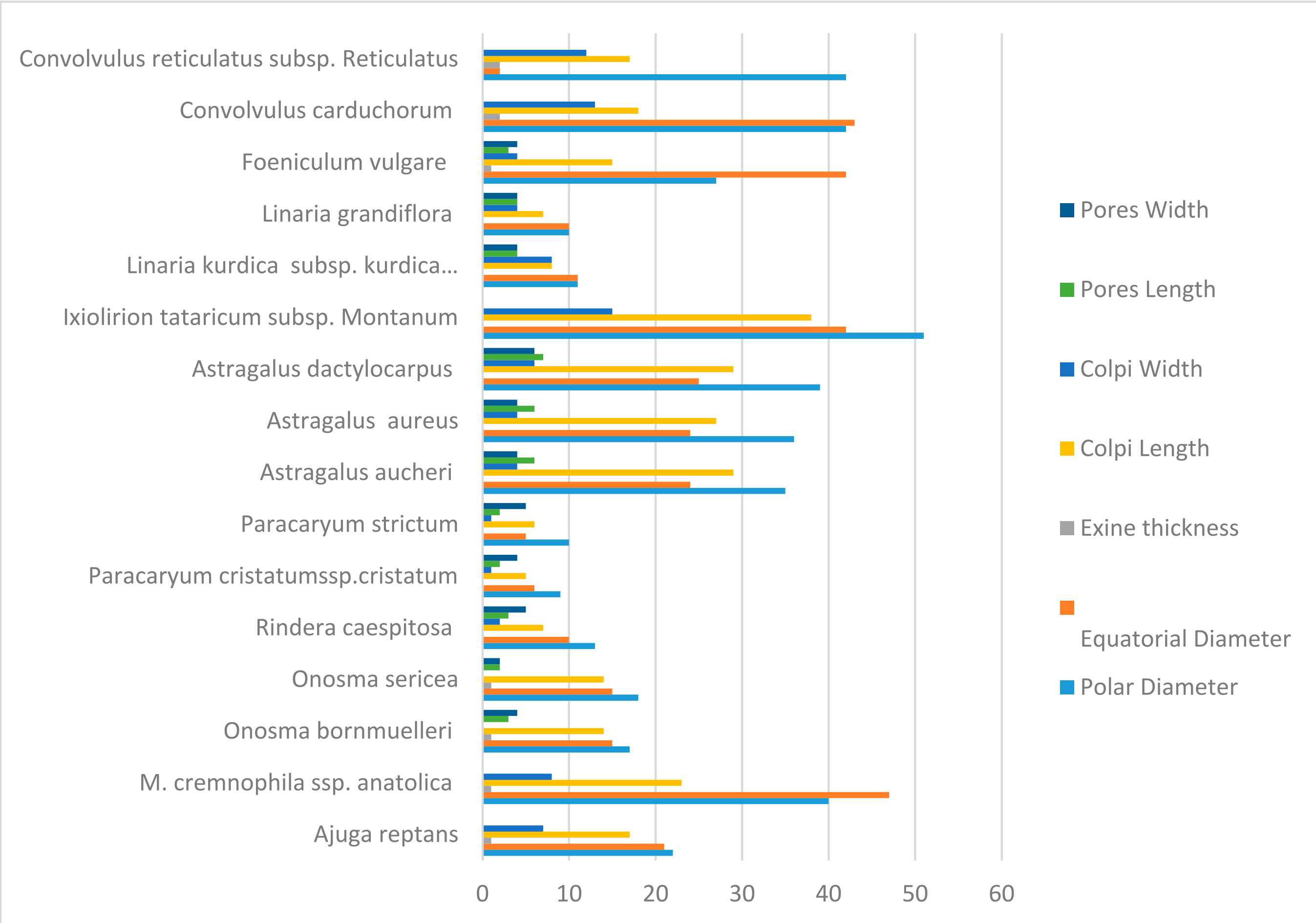

Figure 2. Graph of pollen's morphological measurements of the studied taxa.

2.3. Scanning Electron Microscopy (SEM)

Pollens belonging to each taxon were placed on metal carriers covered with double-sided adhesive tape under a binocular microscope and later coated with gold on a JEOL JSM 6510 (Akishima, Tokyo) (operating voltage range: 500 V–30 kV). The pollen appearances and detailed surface ornamentations of the studied pollen were obtained from Bingöl University Central Laboratory. Microphotographs at 1500×–10,000× magnification were taken for each taxon.

2.4. Transmission Electron Microscopy (TEM)

A transmission electron microscope (Beijing, P.R. China) (TEM, Tecnai G2 F20 S-TWIN, 200 kV) was used to analyze the sculpturing of pollen exine. The pollen sampled for TEM analysis was prepared by placing a drop of pollen on a carbon-coated copper TEM grid and allowed to evaporate (150 × 30 Mx).

2.5. Cluster Analysis and PCA (Principal Component Analysis)

The pollen data were subjected to cluster analysis from numerical taxonomic methods for the 16 taxa. For this analysis, XLSTAT 2022 (Denver, CO, USA) software and the UPGMA statistical method were used. The results of these analyses were illustrated using dendrograms and evaluated in terms of numerical chemotaxonomic relationships. In the resulting cluster analysis tree, the relationships of the species are indicated (Figure 8).

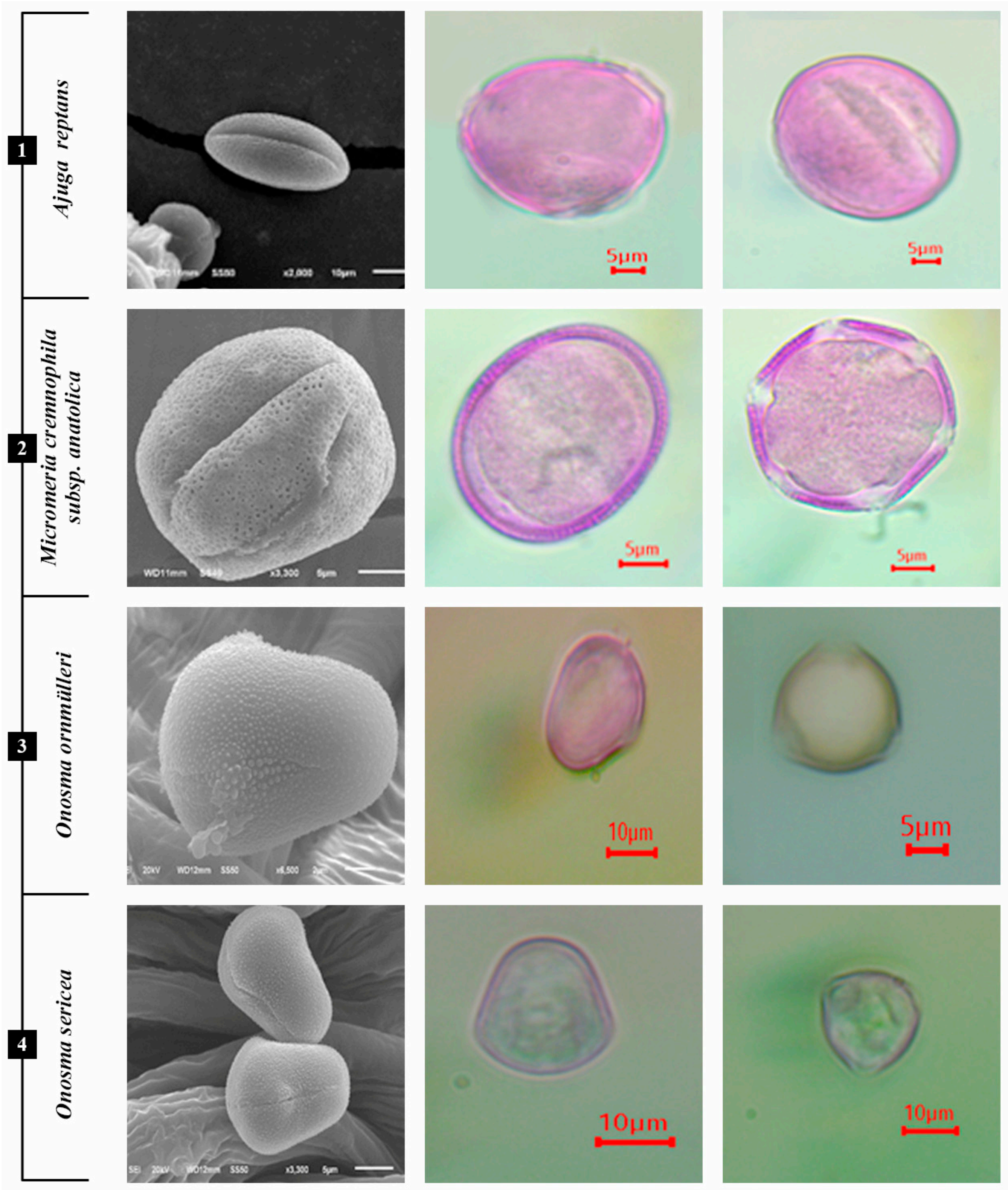

Figure 3. Scanning and light microscopic images of *A. reptans*, *M. cremnophila* subsp. *anatolica*, *O. bornmülleri*, and *O. sericea*.

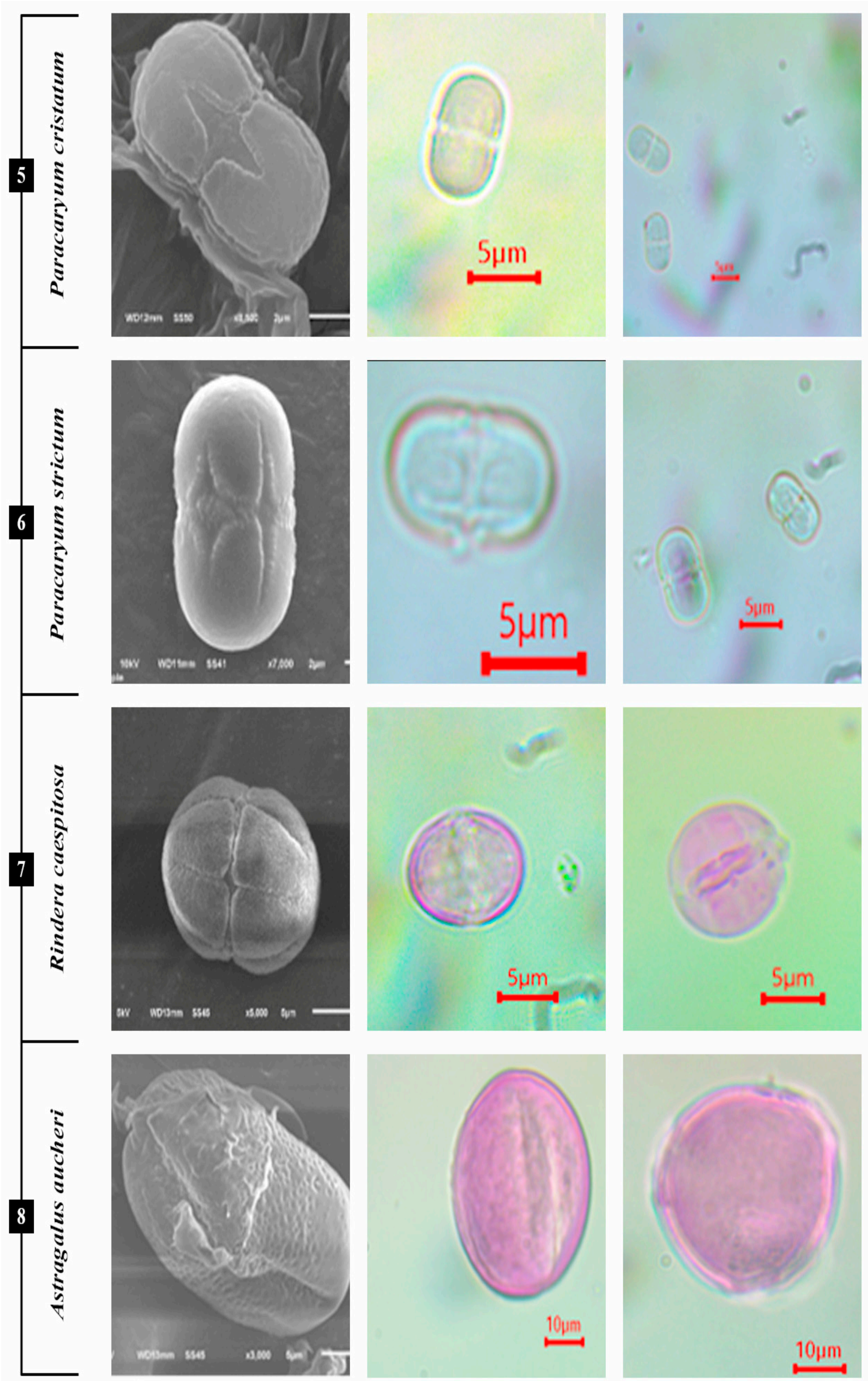

Figure 4. Scanning and light microscopic images of *P. cristatum*, *P. strictum*, *R. caespitosa*, and *A. aucheri*.

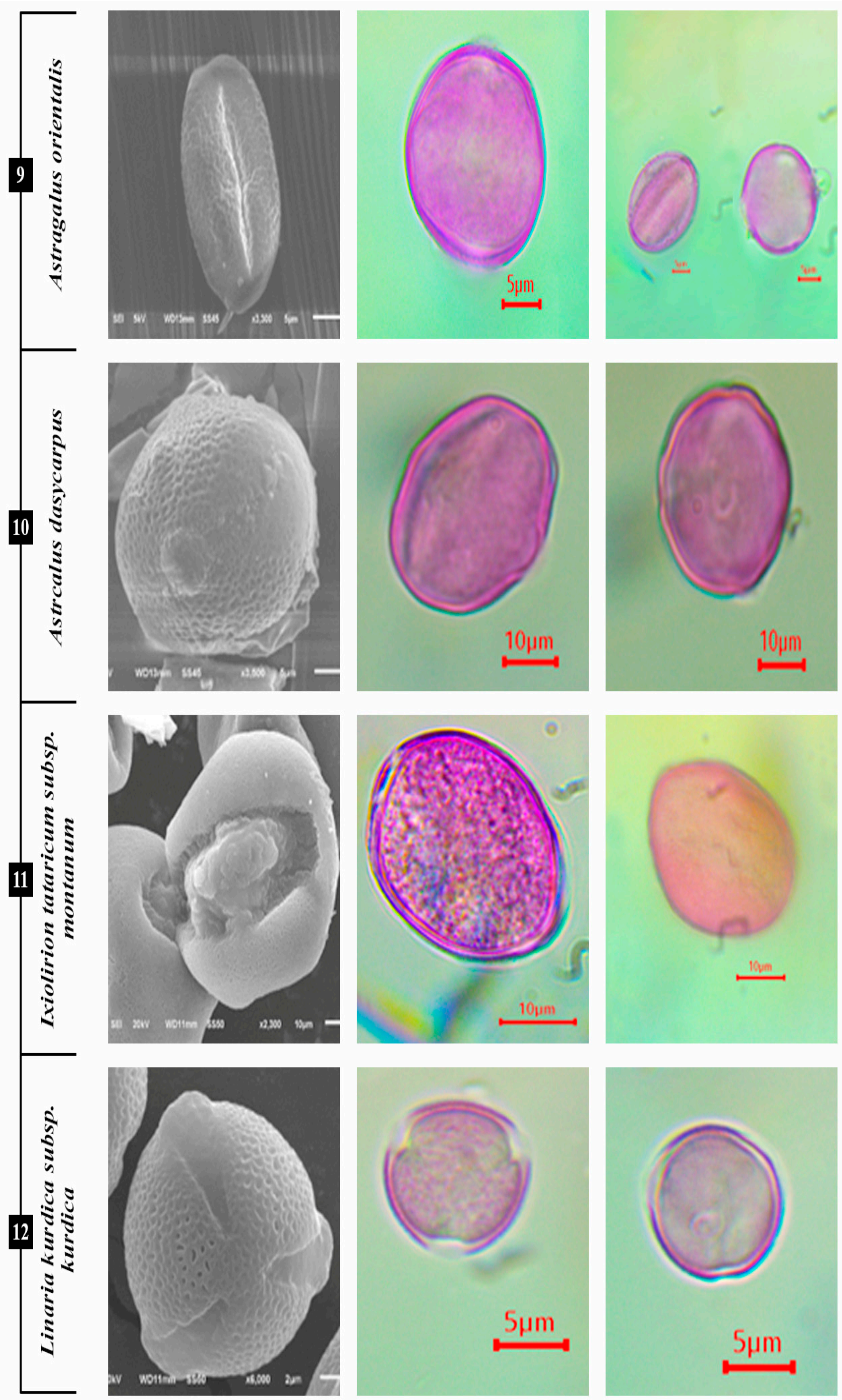

Figure 5. Scanning and light microscopic images of *A. orientalis*, *A. dasycarpus*, *I. tataricum* subsp. *montanum*, and *L. kurdica* subsp. *kurdica*.

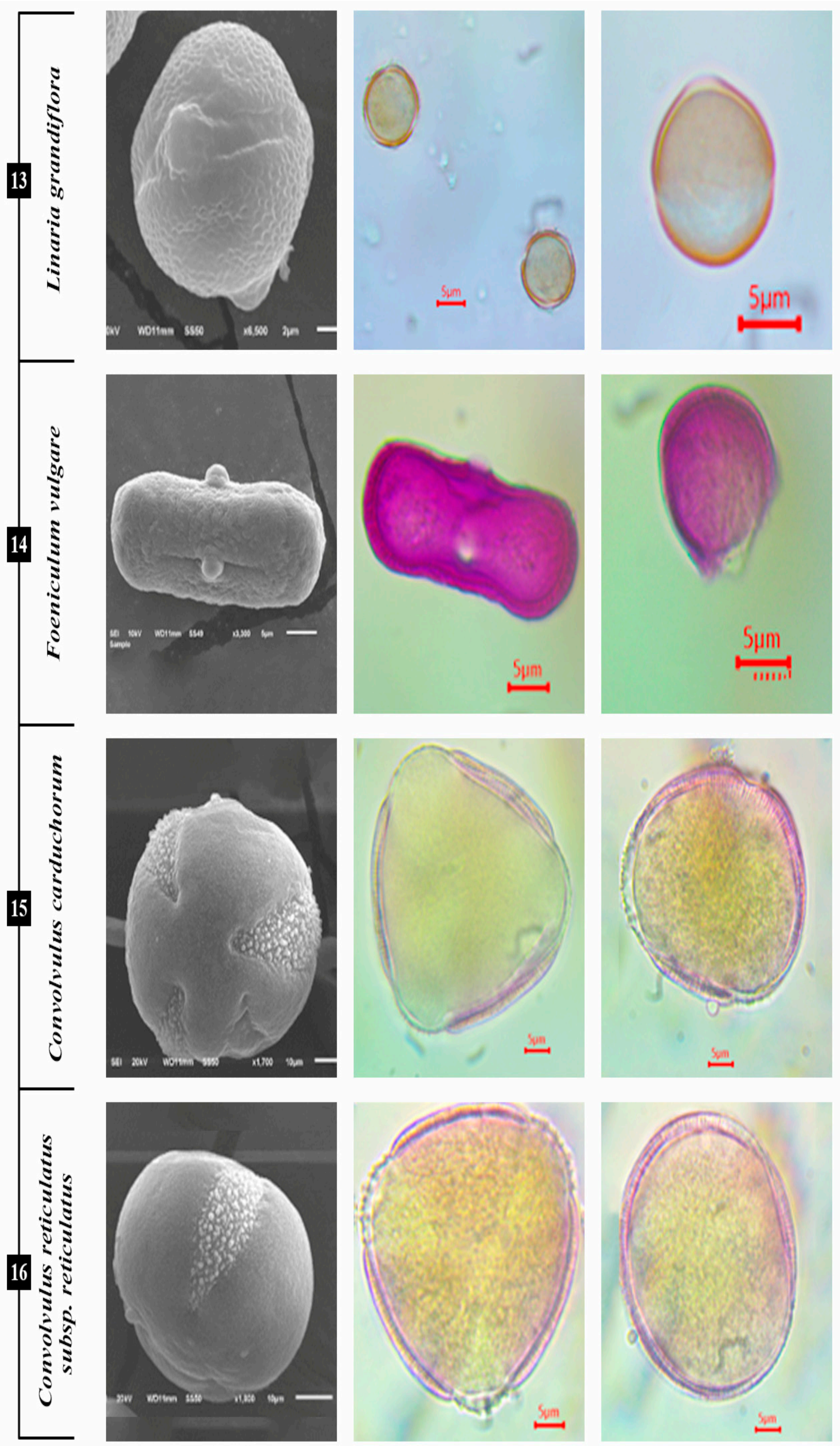

Figure 6. Scanning and light microscopic images of *L. grandiflora*, *F. vulgare*, *C. carduchorum* and *C. reticulatus* subsp. *reticulatus*.

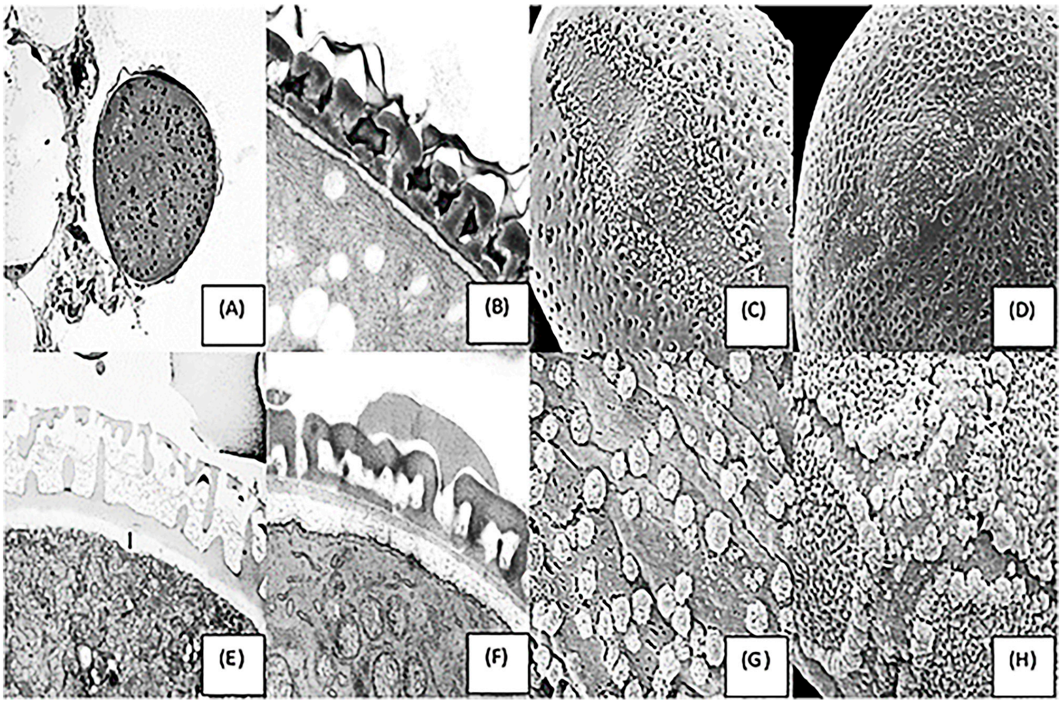

Figure 7. TEM micrographs: (**A**,**B**) *A. reptans*, (**C**) *A. aucheri*, (**D**) *A. orientalis*, (**E**) *C. reticulatus* subsp. *reticulatus*, (**F**) *L. kurdica* subsp. *kurdica*, and (**G**,**H**) *R. caespitosa*.

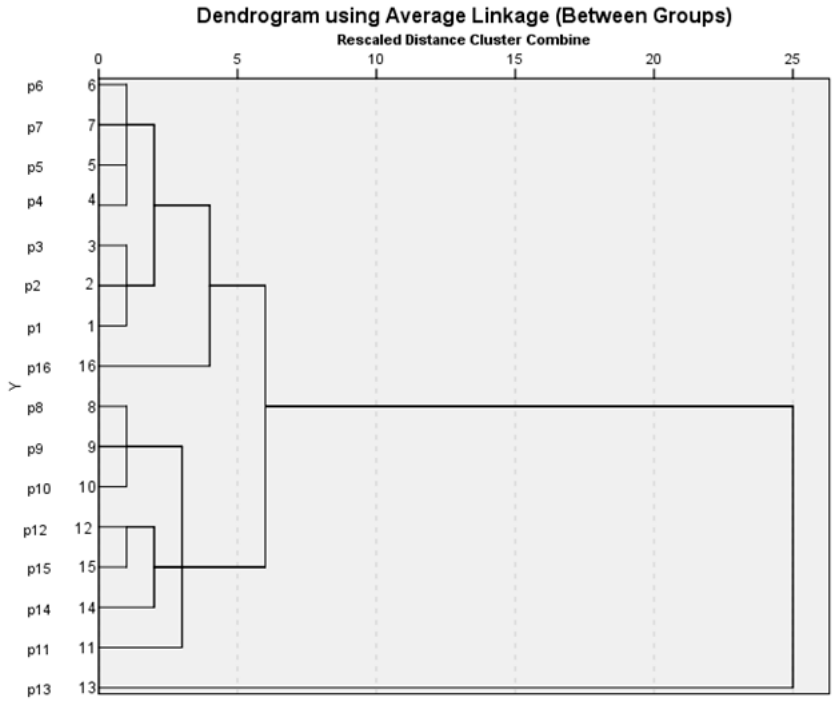

Figure 8. Clustering analysis of palynological data of taxa: Plant name p1—*Ajuga reptans*; p2—*Micromeria cremnophila* ssp. *anatolica*; p3—*Onosma bornmuelleri*; p4—*Onosma sericea*; p5—*Rindera caespitosa*; p6—*Paracaryum cristatum* ssp. *cristatum*; p7—*Paracaryum strictum*; p8—*Astragalus aucheri*; p9—*Astragalus aureus*; p10—*Astragalus dactylocarpus*; p11—*Ixiolirion tataricum* subsp. *montanum*; p12—*Linaria kurdica* subsp. *kurdica*; p13—*Linaria grandiflora*; p14—*Foeniculum vulgare*; p15—*Convolvulus carduchorum*; p16—*Convolvulus reticulatus* subsp. *reticulatus*.

Multivariate analysis was conducted to identify the structure of variability and to measure the distances between groups. These analyses were performed on complete data sets. The UPGMA (unweighted pair-group average linkage) clustering method based on Pearson distances was used to measure the similarities between each measured unit (Figure 9). Principle component analysis (PCA) and cluster analysis (CA) were used to evaluate the pollen morphology data of 16 taxa. The raw data were standardized to the same weight as previously reported.

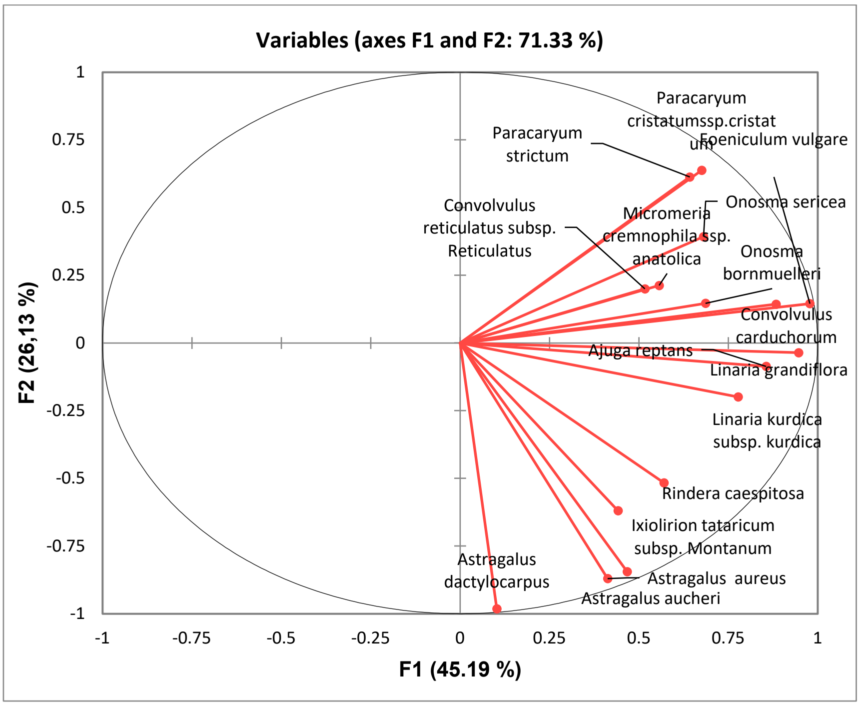

Figure 9. PCA correlation circle from the analysis of the palynological data of taxa.

3. Result

The micromorphological pollen features of selected 16 taxa from Turkey are summarized in Tables 2 and 3. The pollen's ultrastructure characteristic illustrations are shown in microscopic detail in Figures 3–7.

3.1. Pollen Descriptions

3.1.1. *A. reptans*

The results of light microscopy examinations in the pollen of the *Ajuga reptans* taxon revealed the tricolpate structure: polar axis of 22.4 ± (2.0) µm; equatorial axis of 22.7 ± (2.1) µm; pollen shape: oblate-spheroidal, (P/E = 0.98); polar aspect; colpus length of 16.7 ± (1.2) µm in equatorial aspect; and colpus width of 7.6 ± (1) µm. Exine thickness was 1.1 ± (0.1) µm, and their ornamentation showed a singletate granulate ornamentation according to SEM results (Figure 3).

3.1.2. *M. cremnophila* subsp. *anatolica*

Polar axis of 40 ± (2.3) µm, equatorial axis of 46.6 ± (1.3) µm, exine of 1.5–2.4 µm, and P/E: 0.85 sub-oblate. Pollen ornamentation showed reticulated ornamentation according to the SEM results.

3.1.3. *O. bornmuelleri* and *O. sericeum*

O. bornmuelleri have the following characteristics: polar axis 17.3 ± (1.0) µm; equatorial axis 14.7 ± (1) µm; pollen shape subprolate; (P/E = 1.17); polar aspect, colpus length 13.7 ± (0.1) µm in equatorial aspect; and pore width 3 × 3.29 µm. Exine thickness was 1.0 ± (0.5) µm, and their ornamentation showed a scabrad-granulate ornamentation according to SEM results. The polar axis of *O. sericeum* pollen grains was 17.8 ± (1.0) µm, equatorial axis was 14.9 ± (0.1) µm, pollen shape was subprolate, (P/E = 1.19), polar aspect, equatorial aspect, colpus length was 13.8 ± (0.1) µm, and pore width 2.83 µm × 2.29 µm. Exine thickness was 1.0 ± (0.5) µm, and the ornamentation showed a scabrad ornamentation according to the SEM results. Both *Onosma* species are similar in terms of pollen morphology (Figure 3).

3.1.4. *R. caespitosa*

Polar axis 13 ± (1.0) µm, equatorial axis 10 ± (0.1) µm, and pollen shape subprolate (P/E = 1.3). Exine thickness was 0.5 ± (0.3) µm, and ornamentation showed granulate ornamentation according to SEM results.

3.1.5. *P. cristatum* subsp. *cristatum*, *P. strictum*

The pollen grains have a 6-heterocolpate and are hexagonal from the polar view. The aperture margins are strongly thickened. Polar axis was 9.1 ± (0.3) µm, equatorial axis was 5.6 ± (0.1) µm, exine was 0.24 ± (0.2), P/E ratio was 1.62, and the pollen shape was prolate with respect to *P. cristatum* subsp. *cristatum*. The *P. strictum* pollen grains have a 6-heterocolpate and are hexagonal from the polar view. Polar axis was 10.1 ± (0.5) µm, equatorial axis was 5.2 ± (0.2) µm, exine was 0.23 ± (0.1), and P/E ratio was 1.94. The pollen was elliptical from the equatorial view, ±hexagonal from the polar view, and the pollen shape was prolate.

3.1.6. *A. aucheri*, *A. aureus*, *A. dasycarpum*

Polar axis 35.5 ± (1.9) µm, equatorial axis 23.6 ± 1.28 µm, P/E ratio 1.50, and pollen shape prolate in *A. aucheri*. According to the SEM results of the ornamentation, it was determined that they showed micro-reticular ornamentation. The *A. aureus* pollen grains have a polar axis of 35.8 ± 1.2 µm, equatorial axis of 23.5 ± 1.0 µm, P/E ratio of 1.52, and prolate pollen shape. Amb diameter average 20 ± 1.5. Exine thickness subtectate 0.5 ± 0.1 µm. According to the SEM results of the ornamentation, it was determined that they showed micro-reticulate ornamentation (Figure 5). Polar axis 38.7 ± 2.2 µm, equatorial axis 25.3 ± 1.5 µm, P/E ratio 1.52, and pollen shape prolate in *A. dasycarpum*. According to the SEM results of the ornamentation, it was determined that they showed micro-reticular ornamentation (Figures 4 and 5).

3.1.7. *I. tataricum* subsp. *montanum*

Pollen grains heteropolar, monosulcate, and polar axis 51.2 ± (4.3) mm, equatorial axis 41.6 ± (3.3) µm, P/E: 1.23 sub-prolate. Exine 1–1.8 µm, and the ornamentation showed a reticulate ornamentation according to SEM results.

3.1.8. *L. kurdica* subsp. *kurdica*

Pollens with radial symmetry, isopolar, tricolporate; polar axis 10.9 ± (1.2) µm, equatorial axis 11.2 ± (1.1) µm. Exine thickness 0.9 ± 0.2 µm. P/E: 0.97. Exine is reticulate. Colpus length 8.01 ± (2.3) µm; colpus width 2.4 ± (2.3) µm.

3.1.9. *L. grandiflora*

Pollens with radial symmetry, iso polar, tricolporate; polar axis 10.0 ± (1.2) µm, equatorial axis 10.3 ± (1.3) µm. Exine thickness 0.82 ± 0.2 µm. P/E: 0.97. Exine is reticulate. Colpus length 7.85 ± (1.2) µm; colpus width 3.8 ± (0.9) µm.

3.1.10. *F. vulgare*

Polar axis 26.86 ± (1.9) µm, equatorial axis 10.07 ± (1.8) µm, exine 1.8–2.1 µm, pollen shape 2.68 per-prolate, colpus length 14.90 µm, colpus width 4.9 µm, por çapı 3.8 µm. The ornamentation showed a rugulate-striate ornamentation according to the SEM results.

3.1.11. *C. carduchorum* and *C. reticulatus* subsp. *reticulatus*

Convolvulus species have an isopolar pollen shape. Pollen grains are radial and the pollen type is three zonocolpate in *C. carduchorum*. The pollen grains were symmetrical. The polar axis was 42.3 ± (1.3) m, equatorial axis was 43.5 ± (1.2) mm, exine was 2.1 ± (0.4), P/E: 0.97, and pollen shape was oblate-spheroidal in *C. carduchorum*. The polar axis was 42.3 ± (1.3) µm, equatorial axis was 43.5 ± (1.2) µm, exine was 2.1 ± (0.4) µm, P/E: 0.97, and pollen shape was oblate-spheroidal in *C. reticulatus* subsp. *reticulatus*.

Principle component analysis (PCA) and cluster analysis (CA) were performed to identify the palynological data of taxa. The PCA was then performed with Varimax rotation using the matrix correlation configuration. The main components of the principal component analysis were PC1 with 45.19% and with PC2 26.13%, respectively. The total load of PC1 and PC2 was 71.33%. The Kaiser–Meyer–Olkin (KMO) method was conducted to examine the correlation of the variables. KMO was at 0.798, which is considered acceptable. Barlett's test of sphericity also showed a statistical significance at alpha 0.04 for the data set. PCA analysis, which clarified the relationship between the taxa and pollen morphology, was explained with two possible groups (PC1 and PC2). The results are presented in Figure 9.

4. Discussion

The morphological parameters of pollen provide additional information in plant taxonomy and systematics. Pollen analyzed by LM showed high variability in equatorial diameter, polar diameter, width, and length of the colpi and exine. Variations were observed in the pollen morphology of a wide range of species selected from a particular area (Figure 2). This suggests that palynological data play a vital role in evolutionary studies. Pollen size and exine ornamentation are two of the most prominent diagnostic features of plant species in systematic studies. This study differs from previous studies on species collected from the study area in terms of qualitative and quantitative microscopic characteristics.

Kose et al. [28] studied the pollen grains of eight different *Ajuga* species and measured the pollen morphology of *A. reptans* suboblata-subprolata and tricolpatae, with P/E = 0.89 and the orientation is tectatae-granulate; they determined that the exine thickness was 1.46 µm. According to these results, *A. reptans* pollen grains collected from the Bingöl region and the pollen findings studied by Kose et al. show differences, albeit partially. In another study, *Onosma bornmuelleri* pollen grains have an equatorial appearance, a polar axis of 25.08 µm, and an equatorial axis of 21.47 µm [29]. Ornamentation in the polar region was determined as granulate. The exine is 1.05 µm thick [29]. In our study, the polar axis was 22.4 ± (2.0) µm and the equatorial axis was 22.7 ± (2.1) µm, while in the other study, the polar axis was 19.02 ± 2.15 µm, and the equatorial axis was 21.32 ± 2.1 µm. This situation caused different pollen shape results.

Bigazzi et al. [30] reported that *Paracaryum cristatum* pollen grains were small and had a polar axis of 10.7–14.4 µm, equatorial axis of 10.2–13.4 µm, were elliptic in the equatorial view and hexagonal in the polar view, and ranged from prolate-spheroidal to subprolate in shape (P/E: 1.06–1.25), with (3-)6-hetero-colpate and ectocingulate. The composed apertures are spindle-shaped and rarely rhombic, with margins not thickened or only slightly thickened; the endoapertures are elongate, about 1.5 × 3 mm, situated at the equator, and exhibited a granular membrane; the simple colpi are narrow and generally shorter than colporate apertures. According to the results of this study, the pollen's shape was found to be prolate with respect to *P. cristatum* subsp. *cristatum* and *P. strictum*. The pollen morphologies of *P. cristatum* subsp. *cristatum* found in this study were close to each other. *Rindera caespitosa*'s pollen shape was subprolate and ornamentation showed granulate ornamentation according to this study's SEM results. Our results show similarities with the studies conducted.

In a study of two sections of this genus, with respect to *Astragalus falcatus*, *A. odoratus*, *A. ornithopodioides*, *A. stevenianus* var. *stevenianus*, *A. stevenianus* var. *kochianus*, and

A. jodostachys species with a polar axis (P) of 24.53–40.76 µm and equatorial axis (E) of 21.82–31.48 µm, pollen shapes were determined as prolate-spheroidal, prolate, or subprolate. Ekici et al. studied the revision of the Hololeuce Bunge (*Astragalus*) section of Turkey. They found that the pollens of all species were tricolporate types and had reticulate ornamentation. As a result of pollen measurements, they stated that the pollen shapes showed slight differences, but pollen characteristics did not show systematically significant differences between species [31]. As a result of light and electron microscopy studies of the specimens in the Onobrychoidei section of the genus *Astragalus* L., they reported that the pollen was generally tricolporate, prolate, subprolate, or prolate-spheroidal. The polar axis of the pollen varied between 23.4 and 42.6 µm, and the equatorial axis varied between 14.3 and 36.4 µm; their external appearance was elliptic or flattened oval in the meridional optical region and trilobat and sometimes tetrabulate in the polar optical region [32]. The shape of pollen belonging to *A. aucheri* and *A. orientalis* species that we determined in this study is prolate. In this respect, our results show parallelism with the studies carried out. Akan et al. (2005) studied the pollen morphology of taxa belonging to the Alopecias Bunge section of the genus *Astragalus* in Turkey, employing light and electron microscopy. They found that the pollen shape of the taxa was subprolate or prolate-spheroidal, the pollen type was tricolpate, and ornamentation was microreticulate [33]. The ornamentation of the *Astragalus* species in our study was determined as reticulate. Thus, pollen, especially polar ornamentation in the region, differed from each other.

Oybak Dönmez and Işık [34] determined that the *Ixiolirion* sp.'s pollen ornamentation was micro-reticulate, and it has a polar axis of 50 $\pm$ (1.34), equatorial axis of 43 $\pm$ (1.26), and exine of 1–1.5 µm. In another study of *Linaria kurdica* subsp. *kurdica*, the pollen was tricolporate and prolate-spheroid; the polar axis was 16.95–15.37 µm, equatorial axis was 14.88–15.86 µm, and exine thickness was 0.94 µm (exine embellishment reticulate). The intin was 0.61 µm thick, the colpus length was 12.5 µm, and colpus width was determined to be 3.4 µm [35]. According to the results of this study, *L. kurdica* subsp. *kurdica* pollens are isopolar and tricolporate. However, the polar and equatorial lengths were found to be smaller in this taxon from Turkey.

The pollen morphology studies of the *Convolvulus* genus taxa in this study were presented for the first time in this study. Qualitative and quantitative observations of the pollen of *Convolvulus carduchorum* and *C. reticulatus* subsp. *reticulatus* were observed to be compatible within the genus [36].

Beekeeping is an agricultural activity in which honey bee colonies are used for the purpose of maximizing the worker bee population during the periods when the nectar flow is most abundant and for the production of honey, pollen, royal jelly, and pollination of plants [37]. Baydar and Gürel found that Fabaceae-type pollens are among the most preferred for honey bees and that pollen has an important role in the Fabaceae pollen of species belonging to the family compared to species from both families because they are much richer in protein and minerals [38]. Pollen quality is also important in pollen preferences. Fabaceae is the most important species in the flora. In our study, there are three specimens from the Fabaceae family. However, all pollens analyzed in this study were important species for beekeeping.

Qualitative and quantitative observations obtained as a result of palynological data were grouped by cluster analysis (Figure 8). According to this result, the taxa at the family and genus levels gave results that were consistent with their morphological and systematics. Taxa in the same genus were also found in close clades. Pollen morphology data were examined from all aspects with both statistical methods and data visualization techniques. All methods supported each other by making the study more comprehensive. Pollen morphologies studied for the first time have made a great contribution to the science of systematic taxonomy.

5. Conclusions

The analyzed plant taxa have 3-colporate, zono-colporate, mono-sulcate, and 6-colpate pollen with reticulate and micro-reticulate exine-sculpturing patterns. The pollen grains' qualitative data were utilized to evaluate pollen types and differentiate species at the genus level. It was concluded that individuals within the genus, which are morphologically similar to each other, can be easily distinguished by using the data obtained as a result of palynological studies. For example, the genus *Onosma*, *Paracaryum* in the family Boraginaceae in this study can easily be distinguished from each other thanks to the data provided by this and other studies in the study of pollen analysis in honey and beekeeping. The type and number of apertures, exine ornamentation, and pollen shapes of the species in these genera are different from each other. Furthermore, quantitative data concerning pollen are important tools for finding the similarities and dissimilarities between taxa. The morphological features of pollen that are provided in this study might be useful for systematic and phylogenetic analysis.

In this study, the comprehensive palynological significance of the reported species was examined. The available information emphasizes that pollen's morphology characteristics of the studied species can be used to accurately identify the species. Significant variation among palynological characteristics was studied during both quantitative and non-quantitative analyses. The traits examined in this study were reviewed and compared with previous literature. When comparing the current results of the reported species with previous results, a degree of similarity emerged. Taxonomic data based on qualitative traits were established for the investigated species for precise recognition and identification. Pollen morphology, as well as photographic and statistical data of the taxa, were determined in this study. While the pollen morphology of 16 taxa collected from the study area was studied for the first, the pollen morphologies of some taxa were also introduced to the literature for the first time in this study. The species analyzed with TEM were visualized in taxa collected from this region for the first time. This study contributed to bee plant research, melissopalynological, and systematic botanical flora studies.

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