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A Palynological Review for Some Species of Family Boraginaceae Juss. From the Egyptian Flora

ABSTRACT

The pollen grains of 22 species covering 11 genera are investigated. These selected species are represented by the two subfamilies *Heliotropioideae* and *Boraginoideae* with 7 tribes. Light and scanning electron microscopic techniques are followed. This work aims to introduce a well-developed identifying constant and valuable features of pollen grains. General pollen morphological characters are examined: the size, shape, polarity, apertures types & features, colpi characters, endoapertures and tectum characters. Two patterns of grains are observed; hetero- and homoaperturate. Results are significantly variant; grains are monads, mostly minute- or small-sized and prolate or subprolate. Endoapertures shape is lo- or lalongate. Eight pollen subtypes and 2 subtypes are derived from the apertures types & features, and tectum characters. These results are consumed to describe different taxa. A palynological studying of the Boraginaceae is, however, of great taxonomic significance.

Keywords: Europalynous; heteroaperturate; diorate; endoapertures; endocingulus; bicolpate.

1. INTRODUCTION

Boraginaceae Juss. [1] is a large cosmopolitan family with nearly 2700 species in c. 150 genera, according to [2] Boraginaceae is represented by 19 genera and 57 species. Boraginaceae members are widely distributed in the temperate and tropical regions of the Old and New world [3]. In the last two decades, taxonomic limits of genera, tribes, and even subfamilies have dramatically changed according to the macro-morphological characters and molecular data [4].

Currently, *Boraginaceae* s.l. is classified into six subfamilies: *Codonideae*, *Wellstedioideae*, *Cordioideae*, *Ehretioideae*, *Heliotropioideae*, and *Boraginoideae*. This classification is based on distribution, habit, as well as floral and fruit characters. Moreover, the subfamily *Boraginoideae* has recently been divided into nine tribes basically on the complex characters of the fruit. These are *Trigonotideae*, *Eritricheae*, *Cynoglosseae*, *Trichodesmeae*, *Lithospermeae*, *Borageneae*, *Echieae*, *Echiochileae*, and *Myosotideae* [3].

Today, we have seen that the study of palynology is a distinct and important botanical discipline. Pollen grains present several features of taxonomist interest [5]. Pollen morphology could provide an additional tool to estimate the tribe limits and the systematic relationships occurring among genera and infrageneric taxonomic units [6].

Members of *Boraginaceae* are europalynous, with a great diversity in morphological types, classified under either colporate or heterocolpate main types, and in which many genera or even species can be recognized by their pollen characters [7, 8, 9, 10, 11].

Pollen morphology has been examined and described in regional pollen floras [7, 12,13, 14, 15]; while others have studied certain tribes [10, 6] or even some taxa within a single genus [9, 11, 16, 17, 18]. For Egypt, little attention has been focused on the pollen morphology; it's only represented by [19].

The aim of this investigation is to give an analysis of pollen morphological characters of 22 species most common in the Egyptian flora. To assess the variability of palynology as a good taxonomic criterion at different systematic ranks; species or even tribes. Looking for an evaluation of the little palynological studies in Egypt about the Boraginaceae species, and for expecting some valuable features that can provide better identification for some confusing taxa.

2. MATERIAL AND METHODS

2.1. Plant specimens:

The pollen grains of 22 taxa covering 11 genera and include two subfamilies Heliotropioideae and Boraginoideae with seven tribes (involve 2 subspecies, 4 varieties); these specimens were obtained from herbarium that deposited at which found in Cairo University Herbarium (CAI) and South Valley University-Qena Herbarium (QNA: a proposed acronym. The selected specimens are arranged corresponding to their placement in [3].

The specimens have been determined by the 2nd author (El Hadidy) and updated according to the contribution sources of [20, 2, 21], as well as, [22].

Voucher specimens and permanent slides are deposited in South Valley University-Qena Herbarium (QNA). Details of collecting the investigated taxa are reported in Tab. 1.

2.2. Investigating methods:

The survey is based on light and scanning electron microscopy to give a clear view of the pollen general morphological criteria: the size, shape, P/E ratio, polarity, apertures types & features, colpi characters, endoapertures and tectum characters.

2.2.1. Light microscopy (LM):

All samples were taken from mature anthers of the flowering buds. Material was boiled in H_2O for a few minutes, rehydrated in a 10% aqueous solution of KOH on a clean slide, stained with 10% safranin, mounted in glycerin jelly, and sealed with paraffin for LM observations [23]. Permanent slides were examined by light microscopy (Labomed-Labo America, Inc.) at magnification 100X. Photographs were obtained by the author using Leica-DM1000 microscope equipped with Leica-EC digital camera. Measurements of pollen grains were based on 10-15 pollen grains for each sample.

2.2.2. Scanning electron microscopy (SEM):

Un-acetolyzed pollen grains were mounted onto metallic holders previously coated with double-sided self-adhesive tape [23]. Each sample was sputter-coated with gold in JEOL JFC 1100 E ion sputtering device, and examined with JEOL JSM 5400 LV Scanning Electron Microscopy, accelerated by a voltage of 10-15 KV at SEM Unit, Assiut University, Egypt.

2.3. Pollen properties:

In this study, the terminology is followed the [24]. Colpus length is described as “terminal” when the colpus length represents 0.8-0.9 times of the total polar axis length; “subterminal” when it represents 0.7-0.8 times of the polar axis; “short” when it represents 0.5-0.7 times of the polar axis length; “very short” when it represents less than 0.5 times of the polar axis length. Simple ectocolpi correspond to the pseudoapertures (without endoapertures); while compound ectocolpi correspond to the true apertures (with endoapertures).

Table 1: Details of the collecting information of species studied

Subfamily: Heliotropioideae Gürke [**Heliotropium** L., Sp. Pl., ed. 1: 130 (1753)].				
Plant samples		**Specimens examined**		
Section	**Taxon**	**Collection origin, collector**	**Geo-region/ Date**	**Voucher**
Chametropium Griseb.	**1.** *Heliotropium supinum* L.	Esna city, Qena Governate: south Egypt, *A. Osman*, s.n.	Nv/2003	QNA
Odnotropium Griseb.	**2.** *H. arbianense* Fresen.	South Sinai, Sant Catherine, Gebel Catherine, *A. Osman*, s.n.	S/2004-05	QNA
Pterotropium (A. DC.) Bunge.	**3.** *H. bacciferum* Forssk. subsp. *bacciferum* var. *bacciferum* Forssk.	Gebel Elba, Wadi Habib, *A. El-Hadidy*, s.n.	Ge/1983	CAI
		Cairo-Ismailia desert-road, 46 Km NE of Cairo, *El-Bakry*, 426.	Dg/ 29-4-1980	CAI
		Suez-road, Km 23, *EL-Hadidi*, s.n.	Dg/ 4-4-1952	CAI
		Gebel Elba: E. of Hamra Dam, Wadi Ibib, *J. Osborn & I. Helmy* s.n.	Ge/ 6-3-1967	CAI
	4. *H. bacciferum* subsp. *bacciferum* var. erosum (Lehm.) El Hadidy	Giza: N. of Giza Pyramids, as a weed among *Ricinus* plants (in deserts), *G. Täckholm*, s.n.	Nv/ 5-11-1926	CAI
	5. *H. ramosissimum* Lehm. (Sieb. ex A. DC.)	Naqada city, Qena Gov. south Egypt, *A. Osman*, s.n.	Nv/1998	QNA
		Kharga oasis: near the town, *G. Täckholm*, s.n.	On/ 16-1-1928	CAI
	6. *H. pterocarpum* (A.DC.) Hochst. & Steud. ex Bunge	Gebel Elba, Wadi Habib, *A. Osman*, s.n.	Ge/2004	QNA
		Expedition to Gebel Elba district: Sand dune of Mersa Abu Naam, *Täckholm et al* 601	Ge/ 25-1-1962	CAI
		Expedition to Gebel Elba district: Wadi Yahameib, *Täckholm et al* 244C	Ge/ 22-1-1962	CAI
		Expedition to Gebel Elba district: Delta of Wadi Serimtai, *Täckholm et al* 1592	Ge/ 5-2-1962	CAI
Subfamily: Boraginoideae Gürke				
Plant samples		**Specimens examined**		
Tribe	**Taxon**	**Collection origin, collector**	**Geo-region/ Date**	**Voucher**
Eritrichieae Benth. & Hook. f., Gen. Pl. 2(2): 836 (1876).	**7.** *Asperugo procumbens* L.	South Sinai, Sant Catherine, Gebel Catherine, *A. El-Hadidy*, s.n.	S/ 1983	CAI
	8. *Lappula spinocarpos* (Forssk.) Asch. ex Kuntze	Wadi Hashim, 48 km before Mersa Matrouh, *I. El-Garf,* s.n.	Mm/1999	CAI
Cynoglosseae DC. in Meisn., Pl. Vasc. Gen., "Comm." 2: 187 (1840).	**9.** *Paracaryum intermedium* (Fresen.) Lipsky	South Sinai, Sant Catherine, Gebel Catherine, *A. Osman*, s.n.	S/ 2004-05	QNA
		Gebl Mousa, Sinai, *Täckholm et al*, s.n.	S/ 22-4-1961	CAI

		10. *Paracaryum rugulosum* (DC.) Boiss.	South Sinai, Sant Catherine, Gebel Catherine, *A. Osman*, s.n.	S/ 2004-05	QNA
Trichodesmeae Zak. in Serawsch. 4: 5 (1941).		**11.** *Trichodesma africanum* (L.) R. Br. var. *africanum*	Pharmacy faculty farms, south valley university, Qena governorate, *N. Hussein*, s.n.	Nv/2011	QNA
		12. *Trichodesma africanum* L. R. Br. var. *heterotrichum* Bornm. & Kneucker	East of Cairo, Wadi Dowiaqa, *A. El- Hadidy*, s.n.	Nv/1983	CAI
			East of Cairo, Wadi Dowiaqa, *G. Täckholm* s.n.	Nv/ 3-12-1926	CAI
			Wadi Hammat near Wadi Hôf, *V. Täckholm* s.n.	Nv/ 12-2-1952	CAI
		13. *Trichodesma ehrenbergii* Schweinf.	Expedition to Gebel Elba district: Wadi El Shallal, Bir area, *Täckholm et al*, 519.	Ge/ 24-1-1962	CAI
Lithospermeae (DC.) Gürke in Engl. & Prantl, Nat. Pflanzenfam. 4(3a): 118 (1894).		**14.** *Alkanna tinctoria* Tausch	Alexandria: Mandara, *Hassib*, s.n.	Mm/ 13-8-1929	CAI
		15. *Alkanna orientalis* (L.) Boiss.	Sinai: Sinai Mountain region, *Hassib*, s.n.	S/ April 1940	CAI
		16. *Arnebia decumbens* (Vent.) Coss. & Kralik	Mariut: Ras el Hekma, *Botany Dept. Excursion*, s.n.	Mm/ 2-5-1955	CAI
		17. *Arnebia hispidissima* (Lehm.) DC.	Gebel Elba, Wadi Habib, *A. Osman*, s.n.	Ge/2004	QNA
Boragineae Riedl in Rech., Fl., Iran. 48: 231 (1967).	**Genus: Anchusa** L., Sp. Pl., ed.1, 133 (1753).	**Subgenus: Anchusa** Feinbrun, Fl. Palaest., 3: 82 (1978); D. F. Chamb. in Davis, P. H. [ed.]. Fl. Turkey 6: 390 (1978). **18.** *A. hybrida* Ten.	On the road Alexandria- Burg El-Arab at kilo 17, *M. Imam*, s.n.	Mm/ 22-3-1956	CAI
		Subgenus: Buglossoides (Reichenb.) Guşul., Bul. Fac. Şti. Cernăuţi 1: 280 (1927); Riedl in Rech., Fl. Iran. 48: 236 (1967); Feinbrun, Fl. Palaest. 3: 84 (1978). **19.** *A. aegyptiaca* (L.) A. DC.	Mediterranean coast, Wadi Hashim, 25 km West Matrouh, *A. Osman*, s.n.	Mm/ 2006	QNA
		20. *A. milleri* Sperng.	South Sinai, Sant Catherine, Gebel Catherine, *A. Osman*, s.n.	S/2004-05	QNA
			Sinai: Wadi el Ereiq, *Hassib*, s.n.	S/7-4-1940	CAI
		Subgenus: Hormuzakia (Guşul.) Chamb., Notes Roy. Bot. Gard. Edinb. 35(3): 298 (1977); Chamb. In P. H. Davis, Fl. Turkey 6: 401 (1978). **21.** *A. aggregata* Lehm.	Alexandria province, Alexandria, *A. Amer*, 9740.	Mm/ 1987	CAI

	Genus: Gastrocotyle Bunge	**22.** *G. hispida* (Forssk.) Bunge	Mediterranean coast, Wadi Hashim, 25 km West Matrouh, *A. Osman*, s.n.	Mm/ 2006	QNA
			3 km north of Al- Arish, *L. Boulos*, s.n.	Di/ 1955	CAI
		23. *Echium rauwolfii* Delile	Pharmacy faculty farms, south valley university, Qena governorate, *A. Osman*, s.n.	Nv/2005	QNA
		24. *Echiochilon fruticosum* Desf.	Burg El- Arab, *Botany Dept. Excursion*, s.n.	Mm/ 2016	CAI

3. RESULTS AND DISCUSSION

3.1. General description of the pollen grains of Boraginaceae Juss.

Pollen grains are dispersed as monads, minute or small sized, occasionally medium; radially symmetrical, isopolar or subisopolar, and rarely bilaterally symmetrical or heteropolar; mostly prolate or subprolate, sometimes prolate-spheroidal or perprolate. The equatorial view is rectangular to rectangular-elliptic or elliptic with hemispherical (rounded) or flattened ends, and with convex or parallel faces, with or without medial constriction at the equator, rarely ovate-triangular or triangular and circular-lobate to polygonal or triangular in the polar view. Apertures are colporate, colpate or heterocolpate. Apertures are characteristics to the subfamilies and tribes. In Heliotropioideae (Heliotropium), the pollen grains are 6-8-heterocolpate; while 3-9(-10) are colporate in Boraginoideae as (Boragineae, Echieae, Lithospermeae, and Trichodesmeae). In Eritricheae and Cynoglosseae are tricolporate alternate with three pseudocolpi. The length and width of simple (pseudocolpi) and compound colpi are variable, but simple colpi are commonly narrower and longer. Some genera in Eritricheae and Cynoglosseae possess slight to strong medial constriction as (*Asperugo*, *Lappula*, *Paracaryum*); rarely with endocingulus such as in (*Paracaryum*). Endoapertures lolongate or lalongate, rarely circular; sometimes operculate as (*Heliotropium*) or protruded as (*Echium*); commonly monorate, rarely diorate as in (*Arnebia*, *Lappula*). The sculpture of the pollen grains either extends over the entire grain or only over a part of it (i.e. sculpture at the apocolpia differs than that on the mesocolpia). Tectum psilate, perforate, rugulate, fossulate, rarely micro-echinate or reticulate to micro-reticulate.

The main pollen grains morphological characters:

1. Size: the size of pollen grains greatly variable ranges from minute to medium-sized pollen grains; polar axis diameter ranges from 6.0-36.0 µm and the equatorial diameter ranges from 4.0-28.0 µm; much smaller pollen grains are found in *Paracaryum intermedium* (P= 6.0-10.0 µm, E= 4.0-5.5 µm), then in *Asperugo procumbens* whereas (P= 8.0-10.0 µm, E= 5.0-6.0 µm). The largest pollen size is found in *Anchusa undulata* subsp. *hybrida* (P= 32.0-36.0 µm, E= 23.0-25.0 µm), and in *Heliotropium supinum* in which (P= 30.0-34.0 µm, E= 18.0-25.0 µm).

2. Shape & polarity:

2.1. Shape: a considerable variation in pollen grains shapes results. The original shapes prolate and spheroidal are detected and their sub-forms; which are integrated into the term sub-spheroidal including: prolate spheroidal, sub-prolate, perprolate and oblate spheroidal. The lowest P/E ratio average is 0.93 (oblate-spheroidal to spheroidal) in *Trichodesma eherenbergii* (Fig. 2.1, 2.2 & 2.5), the highest P/E ratio average is 2.2 (perprolate) in *Lappula spinocarpos* (Fig. 1.2, Fig. 1.5) (Tab. 3).

2.2. Polarity: major pollen grains of Boraginaceae are isopolar, heteropolar, and subisopolar grains occasionally appear. Heteropolar grains in *Echium rauwolfii*, *Alkanna tuberculata* and *A. orientalis* due to that one pole is narrower than the other. Some species of *Echium* also are found to be heteropolar by [25]. Subisopolar grains present in three taxa of *Heliotropium*: *H. bacciferum* subsp. *bacciferum*; var. bacciferum and var. erosum, and *H. ramosissimun* due to a connecting bridge appears between some colpi margins. Otherwise, taxa are isopolar and radially symmetric, and appreciably that only *Echichilon fruticosum* is isopolar and bilaterally symmetric.

3. Apertures types & characters: pollen grains of Boraginaceae mainly found to be either homoaperturate grains or heteroaperturate ones. It is characterized also by large variation of aperture morphology [26]. Homoaperturate pollen grains includes the tribes: Boragineae, Echieae, Echiochileae, Lithospermeae and Trichodesmeae of the subfamily Boraginoideae; heteroaperturate pollen grains includes the subfamily: Heliotropioideae and two tribes of the Boraginoideae; Cynoglosseae and Eritrichieae. Homoaperturate colpate grain is only in *Echichilon fruticosum* (bicolpate). Those of the colporate aperture type vary by the different number of apertures ranging from 3 to poly-colporate, Echieae, Trichodesmeae and *Alkanna sp*. (Lithospermeae) are 3-colporate, Boragineae and *Arnebia sp*. (Lithospermeae) are 4 to poly (8-9)-colporate. The tribes Cynoglosseae and Eritrichieae and the subfamily Heliotropioideae are heteroaperturate grains; all taxa found in this group are 6-heteroaperturate except in *H.* ramosissimum; 8-heteroaperturate and in *H. bacciferum* subsp. *bacciferum* are 6-8-heteroaperturate. Here we agree with [27] who observed the presence of heteroapertures in Boraginaceae (Tab. 2-4).

3.1. Colpi characters: different shapes of colpi are observed mostly linear and fusiform, rhombic and oblong are also presented. Colpi diameters range from 0.1-5.0 µm. the narrower colpus is linear, with acute ends, 0.1-0.2 µm in diam. as in *Lappula spinocarpos*. The widest one is oblong, with obtuse ends, 3.0-5.0 µm in diam. in *Arnebia decumbens*. The length of the colpi is compared relative to the P-axis length. This feature is revealed by describing the colpi as very short, short, subterminal and terminal. This character is somewhat also indicating the colpi length values. Colpi margins appear not thickened or thickened either granulated, perforated or echinated (Tab. 2-4).

3.2. Endoapertures characters: endoaperture shapes mostly found elliptic, rarely circular. Lolongate endoapertures are common, hereafter lalongate ones. A high variation in their sizes, the largest one is lolongate, elliptic and 6.0-9.0 x 3.0-4.0 µm in *Arnebia decumbens*; the smallest one is lalongate, c. 1.0 x 2.0 µm in *Paracaryum intermedium*. Endoapertures present singly for each colpus (monorate) of all taxa studied except in *Lappula spinocarpos* and two species of *Arnebia*: *A. decumbens* and *A. hispidissima* where as two endoapertures for each colpus (diorate) (Tab. 2-4).

4. Wall characters: tectum sculpture ornamentations are remarkably different, most of them are psilate perforate. An integration with other shapes presents such as reticulate, granulate, rugulate, and echinate. The simple pattern of sculpture elements occurs in *Alkanna sp*. (psilate), and in *Echium rauwolfii* (micro-reticulate). Otherwise, taxa are characterized by the compound pattern of more than one type of tectal elements. Most taxa studied showed an obvious differentiation between apocolpia and mesocolpia regions' sculpture elements and the reset tectum all over the wall surface. An equatorial band

mostly with a distinct sculptured surface is characteristic for taxa of the tribe Boragineae (Tab. 2-4).

A synopsis for the pollen grains types (these types are derived according to the apertures types and numbers, and the tectum sculpturing):

- **Subfamily Heliotropioideae (Heliotropium)**

TYPE 1: 6-8-Heterocolpate (Fig. 5)

Subtype a. tri- to tetra-colporate alternate with tri- to tetra-pseudocolpi- perforate.

Species included: *H. bacciferum*, *H. pterocarpum*, *H. ramosissimum* & *H. arbainense*.

Subtype b. tricolporate alternate with tri-pseudocolpi- fossulate.

Species included: *H. supinum*

- **Subfamily Boraginoideae**

TYPE 2: 6-Heterocolpate-Perforate (Fig. 1)

Tricolporate alternate with tri-pseudocolpi.

Tribes included: Eritricheae (e.g. *Asperugo*, *Lappula*) & Cynoglosseae (e.g. *Paracaryum*).

TYPE 3: Bicolpate-perforate (Fig. 4)

Species included: *Echiochilon fruticosum* (Tr. Echiochileae).

TYPE 4: 3-Colporate-Micro-reticulate (Fig. 1)

Species included: *Echium rauwolfii* (Tr. Echieae)

TYPE 5: 3-Colporate-Micro-echinate (Fig. 1)

Tribe included: Trichodesmeae (e.g. *Trichodesma*)

TYPE 6: 3-Colporate-Psilate (Fig. 2)

Species included: *Alkanna* (Tr. Lithospermeae)

TYPE 7: 4-9-Colporate-Perforate (Fig. 4)

Tribes included: Boragineae (e.g. *Anchusa*, *Gastrocotyle*) & Lithospermeae (e.g. *Arnebia decumens*).

TYPE 8: 6-7-Colporate-Micro-echinate (Fig. 3)

Species included: *Arnebia hispidissima* (Tr. Lithospermeae).

CONCLUSION

The Boraginaceae pollen grains morphological characters are variable. All grains are monads, commonly minute to small, mostly isopolar, sometimes heteropolar. Subisopolar are found in *Heliotropium*. That genus which is heteroaperturate and have great variation within its pollen characters as well as its species; meeting the findings of [28]. Apertures are colporate, colpate or heterocolpate; and characteristic to subfamilies and tribes. Compound colpi are mostly monorate, diorate are distinct in *Arnebia*. Varying from very

short to long; either terminal or subterminal. Endoapertures are equatorial or subequatorial; even la- or lolongate, and mostly elliptic. Tectum psilate, perforate, rugulate, fossulate, rarely micro-echinate or reticulate to micro-reticulate. The results of this distinctive aspect ensured the taxonomic implications of palynology. And correlate to those of [19]. Assessed the europalynous nature of Boraginaceae and its pollen morphology diversity; adopting [29]. This work introduces here a well-developed identifying constant and valuable features for the pollen grains of Boraginaceae in Egypt.

Ethical approval and consent are not applicable.

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Table 2: Pollen grains characters of the studied species of *Heliotropium* L. (Subfamily: Heliotropioideae):

Sections/Taxa / Characters	Sect. Chamotropium	Sect. Odnotropium	Sect. Pterotropium			
	H. supinum	*H. arbianense*	*H. bacciferum* subsp. *bacciferum*		*H. ramosissimum*	*H. pterocarpum*
			var. *bacciferum*	var. *erosum*		
P- axis (µm)	30 (31) 34	28 (29) 30	23 (25) 29	16.7 (18.1) 20	21(22)23	23.08 (26.86) 31.25
E-axis (µm)	18 (22) 25	21 (23) 24	20 (24) 28 µm	14.2 (15.5) 16.7	16(18)20	15.38 (16.55) 17.5
P/E (mean)	1.4 (1.5) 1.7	1.2 (1.3) 1.4	1.03 (1.04) 1.2	1.1 (1.2) 1.3	1.1 (1.2) 1.3	1.5 (1.6) 1.9
Pollen shape	Prolate	Subprolate to prolate	Prolate-spheroidal to subprolate	Subprolate or less prolate-spheroidal	Subprolate or prolate-spheroidal	Prolate
Med. Constriction	-	-	-	-	-	-
Equatorial band	-	-	-	-	-	-
Apocolpium diam. (µm)	8-11	7-11	9-12	7-9	3-5	8-10
Aperture type	6-heterocolpate	6-heterocolpate	6(-8)-heterocolpate	6(-8)-heterocolpate	8-heterocolpate	6-heterocolpate
Colpus characters:						
1. Shape	S: Linear C: Linear	S: Linear C: Linear	S: Linear C: Linear	S: linear C: linear	S: Linear C: Linear	S: Linear C: Linear
2. Length (µm)	S: 23-27 C: 23-27	S: 19-24 C: 19-24	S: 19-20 C: 18-19	S: 15-17 C: 13-16	S: 18-20 C: 15-18	S: 20-29 C: 20-25
3. Relative to P-axis (? X)	S: Terminal, c. 0.8x C: Terminal, c. 0.8x	S: Subterminal, c. 0.7x C: Subterminal, c. 0.7x	S: Terminal, 0.8-0.9x C: Terminal, c. 0.8x	S: Terminal, 0.8-0.9x C: Subterminal	S: Terminal, 0.8-0.9x C: Subterminal,	S: Terminal, 0.8-0.9 x C:Subterminal, 0.7-0.8x

				, 0.7-0.8x	0.7-0.8x	
4. Diameter (µm)	S: 0.5-1.0 C: 0.5-1.0	S: 0.5-0.75 C: 0.5-0.75	S: 0.5-0.7 C: 1.4-1.6	S: 0.4-0.7 C: 1.0-1.75	S: 0.3-0.5 C: 0.5-1.0	S: 0.3-0.5 C: 0.3-0.6
5. Margin	Not thickened	Not thickened	Not thickened	Not thickened	Not thickened	Not thickened
Endoaperture characters:						
1. Shape	Lalongate (elliptic)	Lalongate (elliptic)	Lolongate (elliptic)	Lolongate (elliptic)	Lolongate (elliptic)	Lolongate, (elliptic)
2. Diameter (µm)	c. 3 x 4	c. 6 x 7	3-4 x 2-2.5	c. 3 x 2	c. 3 x 2	2-3 x 1-2
Wall characters:						
1. Exine thickness (µm)	0.9-1.2	1.3-1.5	1-1.4	0.9-1.4	1-1.4	0.9-1.6
2. Apo-Mesocolpia differentiation	+	-	+	+	+	+
3. Tectum	Perforate-rugulate or fossulate	Psilate-perforate	Perforate-rugulate	Perforate-rugulate	Perforate-rugulate	Perforate-rugulate

Table 3: Pollen grains characters of the studied species in Eritricheae, Cynoglosseae & Trichodesmeae (Subfamily: Boraginoideae).

Tribe/taxa / **Characters**	**Tribe: Eritricheae**		**Tribe: Cynoglosseae**		**Tr. Trichodesmeae**		
	Asperugo procumbens	**Lappula spinocarpos**	**Paracaryum intermedium**	**P. rugulosum**	**Trichodesma africanum**		**T. eherenbergii**
					var. africanum	**var. heterotrichum**	
P- axis (µm)	8 (9) 10	13 (14) 16	6 (8.3) 10	9.78 (10.81) 12.44	20 (22) 23	14 (15) 16	13 (13) 14.2
E-axis (µm)	5 (5.5) 6	5 (6.5) 8	4 (4.6) 5.5	7.78 (8.37) 9.22	17 (18) 18	12 (13) 15	14 (14) 14.4
P/E (mean)	1.5 (1.6) 1.8	2.0 (2.2) 2.6	1.5 (1.8) 1.82	1.26 (1.29) 1.35	1.20 (1.22) 1.24	1.1 (1.2) 1.3	0.92 (0.93) 1
Pollen shape	Prolate	Perprolate	Prolate	Subprolate to prolate	Subprolate	Prolate-spheroidal	Oblate-spheroidal to spheroidal

Median Constriction	Slight	+	Slight	-	-	-	-
Equatorial band	-	-	-	-	-	-	-
Apocolpium diameter (µm)	3-6	6-7	3-5	5-6	4-5	4-5	8-10
Aperture type	6-heterocolpate	6-heterocolpate	6-heterocolpate	6-heterocolpate	3-colpotare	3-colpotare	3-colpotare
Colpus characters:							
1. shape	S: Linear C: Rhombic	S: Linear C: Linear	S: Nearly linear C: Broadly rhombic	S: Rhombic C: Rhombic	Fusiform	Fusiform	Fusiform
2. length (µm)	S: 5-8 C: 3-5	S: 10-13 C: 9-12	S: 4-6 C: 3-4	S: 6-8 C: 4-5	18 (19) 20	11(12) 13	10 (11) 12
3. relative to P-axis (? X)	S: subterminal, c. 0.8x C: very short, c. 0.4x	S: subterminal, c. 0.8x C: subterminal, c. 0.7x	S: short, c.0.6x C: very short, c. 0.4x	S: short, 0.6-0.7x C: very short, c. 0.4x	Terminal, 0.8-0.9x	Terminal, c. 0.8x	Terminal, c. 0.8x
4. diameter (µm)	S: 0.3-0.5 C: 1-2	S: 0.1-0.2 C: 0.3-0.5	S: 0.2-4 C: 0.6-1	S: 1-1.5 C: 2-2.5	1-2	1-2	0.5-1
5. margin	Thickened	Thickened	Thickened	Thickened	Thickened	Thickened	Thickened
Endoaperture characters:							
1. shape	Lalongate (elliptic)	Lalongate (elliptic)	Lalongate (elliptic)	Lalongate (elliptic)	Lolongate (elliptic)	Lolongate (elliptic)	Lolongate (elliptic), rarely circular
2. diameter (µm)	c. 2 x 3	1-2 x 2-2.5	c. 1 x 2	1-1.5 x 2-3	6-7 x 3-4	5-7 x 3-4	4-5 x 3-5
Wall characters:							
1. Exine thickness (µm)	0.3-0.4	0.4-0.5	0.6-0.7	0.64-0.72	1-1.2	0.98-1	1-1.6

2. Apo-mesocolpia differentiation	**+**	**+**	-	**-**	-	-	-
3. Tectum	Psilate-perforate	Psilate-perforate	Psilate-Perforate	Psilate-Perforate	Micro-echinate-perforate	Micro-echinate-perforate	Micro-echinate-perforate

Table 4: Pollen grain characters of Tribe: Boragineae:

Taxa / Characters	Tr. Boragineae				
	Anchusa				Gastrocotyle
	Subg. Anchusa	Subg. Buglossoides		Subg. Hormuzakia	
	A.undulata subsp. *hybrida*	*A. aegyptiaca*	*A.milleri*	*A.aggregata*	*G. hispida*
P- axis (µm)	32 (34) 36	26 (27.30) 28.33	24 (26) 27	23 (26) 29	20 (22) 23
E-axis (µm)	23 (24) 25	16 (18.25) 20	17 (19) 21	18 (22) 24	19 (22) 25
P/E (mean)	1.39 (1.42) 1.44	1.33 (1.5) 1.70	1.28 (1.36) 1.4	1.1 (1.2) 1.3	0.92 (1) 1.1
Pollen shape	Prolate	Prolate	Prolate to subprolatre	Prolate-spheroidal to subprolate	Spheroidal or Prolate-spheroidal
Med. Constriction	-	-	-	-	-
Equatorial band	+	+	+	-	+
Apocolpium diam. (µm)	15-20	15-17	13-16	11-14	4-5
Aperture type	4-colporate	4(-5)-colporate	4-colporate	6-7-colporate	8-9-colporate
Colpus characters:					
1. shape	Fusiform	Fusiform	Fusiform	Fusiform	Rhombic
2. length (µm)	13 (14) 15	10 (12) 13	10 (10.5) 11	9 (12) 14	10 (12) 14
3. relative to P-axis (? X)	Very short 0.4x	Very short 0.3-0.4x	Very short 0.4-0.5x	Very short 0.4x	Short 0.5-0.6x
4. diameter (µm)	0.3-0.7	0.3-0.7	1-2	0.5-1.4	1-1.5
5. margin	Thickened, granulated	Thickened, finely granulated	Not thickened	Thickened, granulated	Thickened, granulated
Endoaperture characters:					
1. shape	Lalongate (elliptic)	Lalongate (elliptic)	Lalongate (elliptic)	Lalongate (elliptic)	Lalongate (elliptic)
2. diameter (µm)	3-5 x 5-8	2-3 x 3-4	2-3 x 4-7	1-2x3-4	c. 3 x 4
Wall characters:					
1. Exine thickness (µm)	0.52-0.89	1-1.61	1.28-1.56	0.75-0.96	0.8-1.0
2. Apo-mesocolpia differentiation	+	+	+	-	+ (reduced)
3. Tectum	Psilate-Perforate with micro-reticulate equatorial band	Psilate-Perforate with reticulate equatorial band	Psilate-Perforate with micro-reticulate equatorial band	Psilate-Perforate with micro-reticulate	Psilate-Perforate with granulated equatorial band

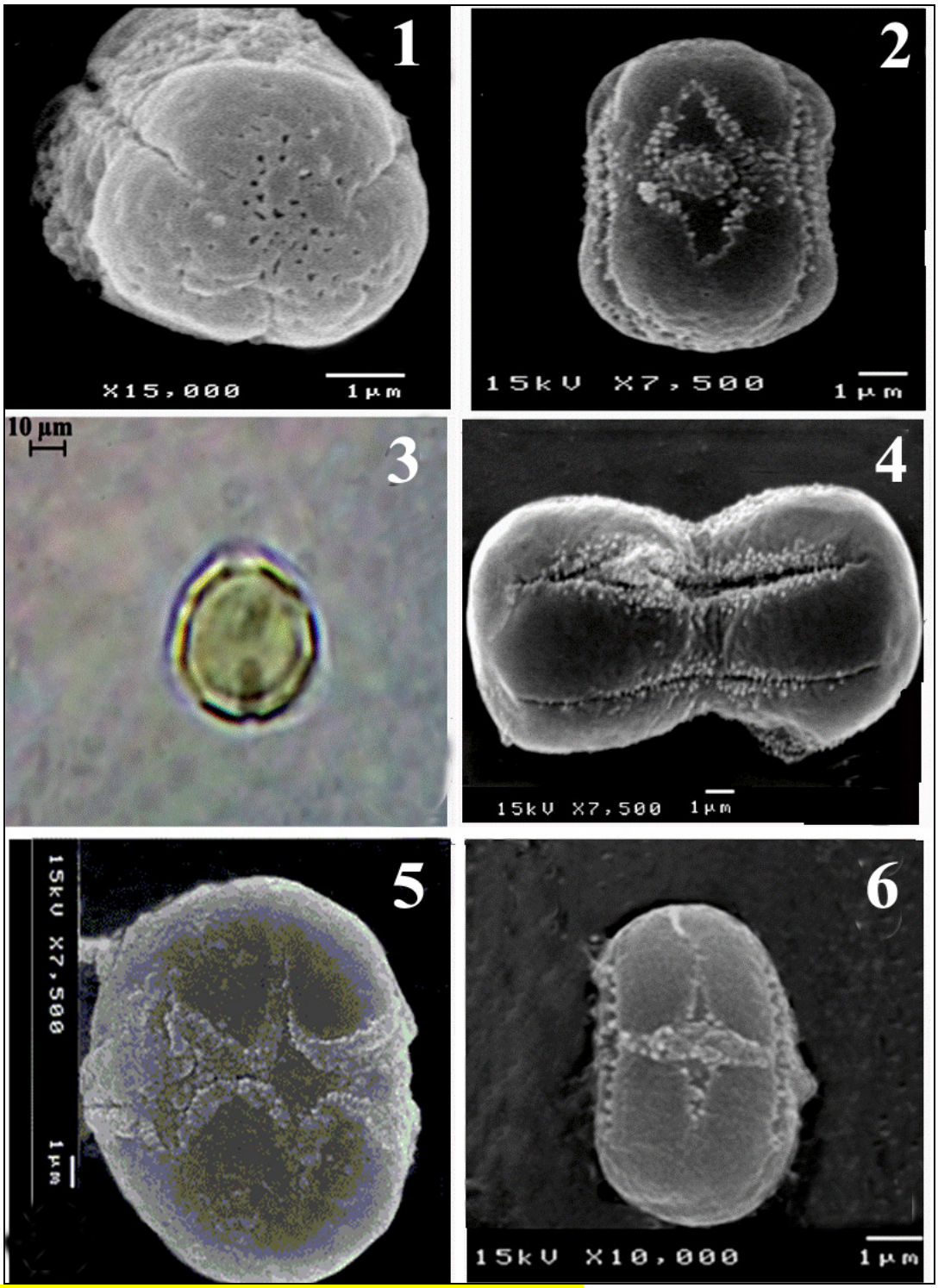

Figure 1. Pollen TYPE 2 (6-heterocolpate-Perforate):

1.1. SEM-photos of pollen shape in polar view in *Asperugo* showing the apocolpium region with perforations. And **1.2**. E-view of *Asperugo* grains shows the simple & compound colpi, lalongate endoaperture and granulated colpi margins.

1.3. LM-image for the P-view in *P. rugulosum* showing the number of apertures.

1.4. SEM-photo in *L. spinocarpos* shows the subequatorial, lalongate endoaperture and densely granulated colpi margins.

1.5. E-view in *P. rugulosum* shows the equatorial ectocingulus and the thickened margins**.**

1.6. E-view in *P. intermedium* shows the simple and compound colpi with the elliptic lalongate endoaperture.

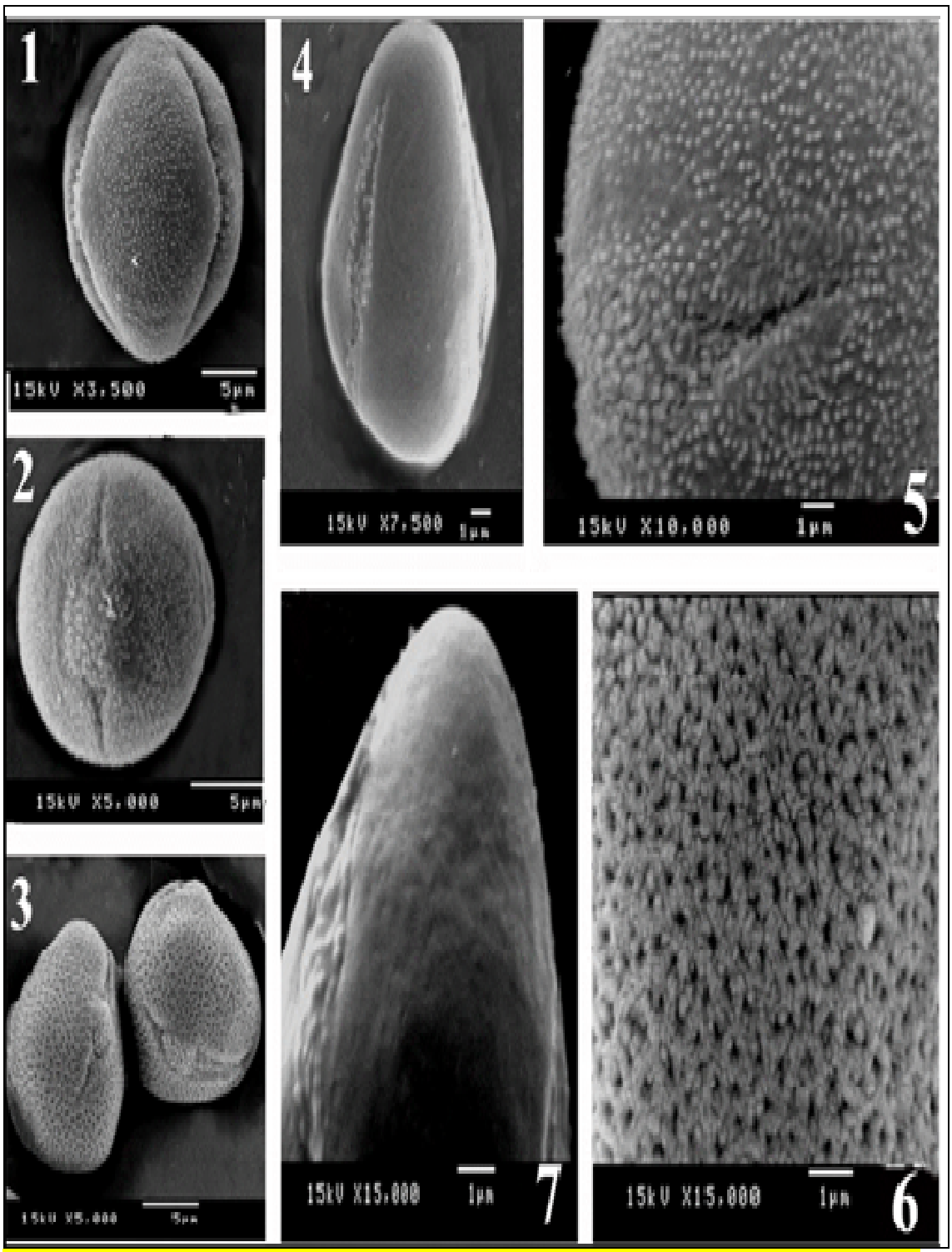

Figure 2. SEM-photos of pollen TYPES 4, 5 & 6 in *Echieae*, *Trichodesmeae* & *Alkanna sp*. respectively.

TYPE 4: 3-colporate-Micro-reticulate in *Echium ggregate*; **2.3.** the E-view shows the fusiform colpi and the endoaperture & **2.6.** Tectum Micro-reticulate.

TYPE 5: 3-colporate-Micro-echinate in *Trichodesmeae*; **2.1.** E-view of *T. africanum*, **2.2.** the lolongate endoaperture of *T. ehrenbergii* & **2.5.** Tectum Micro-echinate.

TYPE 6: 3-colporate-Psilate in *Alkanna* sp.; **2.4.** pollen grains polarity and the colpi margin & **2.7.** Tectum Psilate.

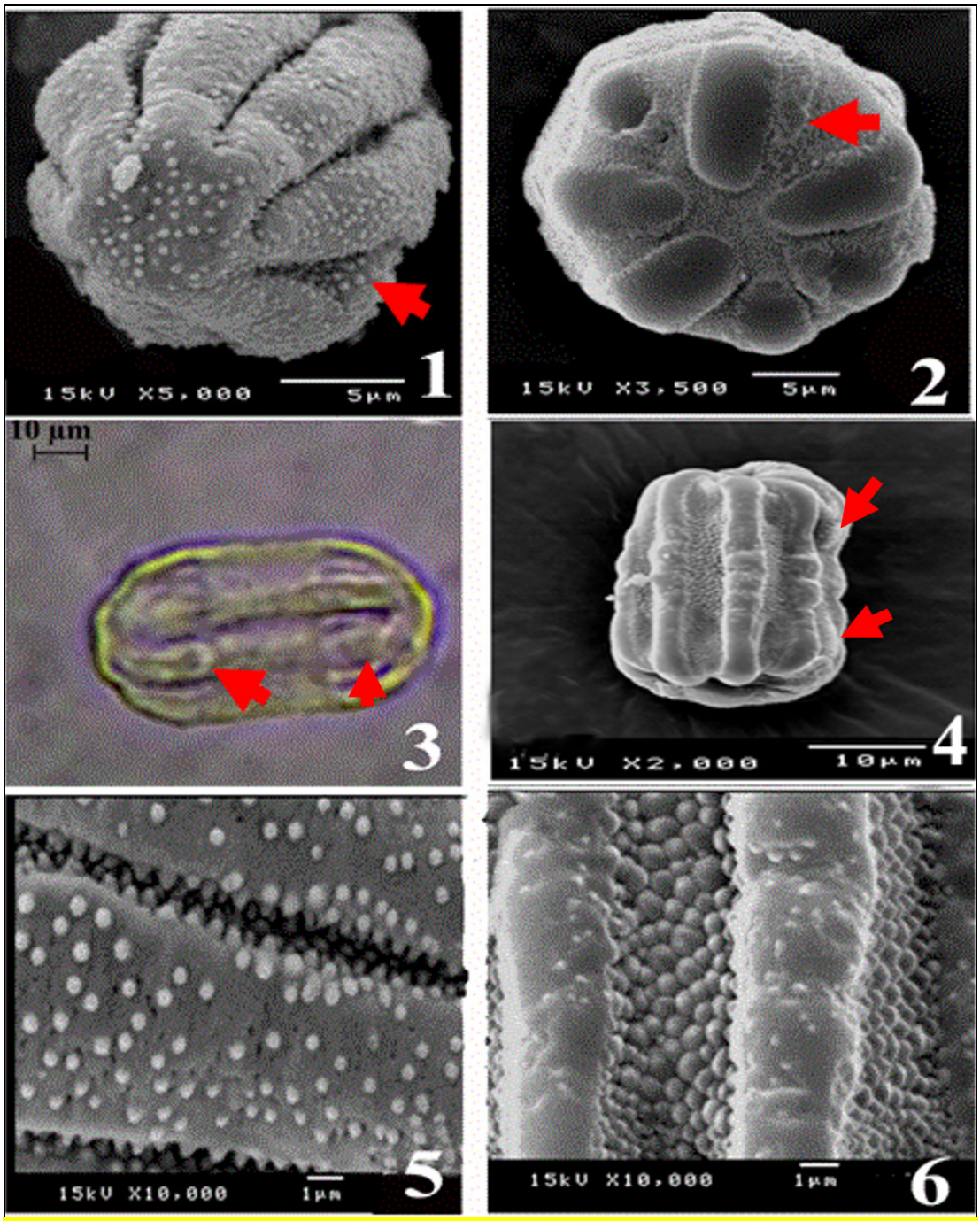

Figure 3. Pollen **TYPES 7 & 8** in *Arnebia sp.*; arrows indicate the subequatorial endoapertures.

3.1. SEM-photo of the P-view in *Arn. hispidissima* showing the terminal, linear colpi; the obvious apocolpium surface & the endoaperture. **3.3.** the E-view in LM-image shows the diorate colpi. **3.5.** the tectum surface and colpi margin of pollen **TYPE 8** (6-7-Colporate-Micro-echinate-perforate) in *Arn. hispidissima*.

3.2. SEM-photo of the P-view in *Arn. decumbens* showing the terminal, oblong colpi; the obscure apocolpium area & the endoaperture. **3.4.** SEM-photo in the E-view shows the diorate colpi (ora are arrowed). **3.6.** the tectum surface and colpi margin of Pollen **TYPE 7** (4-9-Colporate-Perforate) in *Arn. decumbens*.

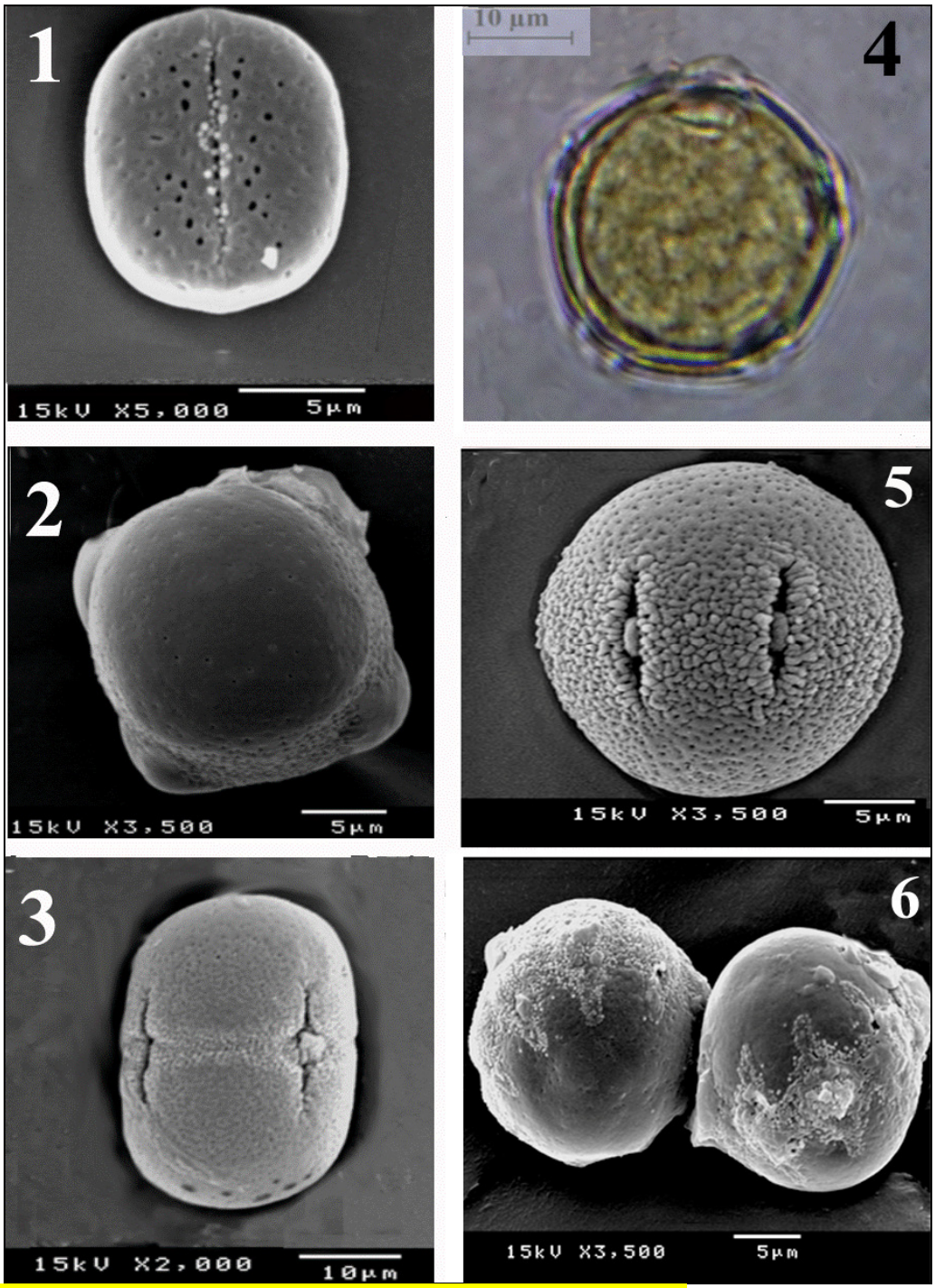

Figure 4. Pollen Echiochilon-**TYPE 3 & TYPE 7** in Boragineae**.**

5.1. E-view of the Bicolpate-Perforate pollen grains in *E. fruticosum*.

Boragineae-**TYPE 7** "4-9-colporate-Perforate" in **4.2.** P-view of the 4-colporate pollens of *Anchusa milleri* shows the apocolpium perforations and **4.3. E-view** in *A. undulata* subsp. *hybrida* shows the micro-reticulate equatorial band.

4.4. LM-image of the P-view shows the 5-colporate pollen grain in *A. aegyptiaca* & **4.5. E-view of grains in** *A. ggregate* show the apo-mesocolpia differentiation and the lolongate endoapertures.

4.6. *Gastrocotyle hispida* of 9-colporate apertures showing the thickened colpi margins and the granulated equatorial band.

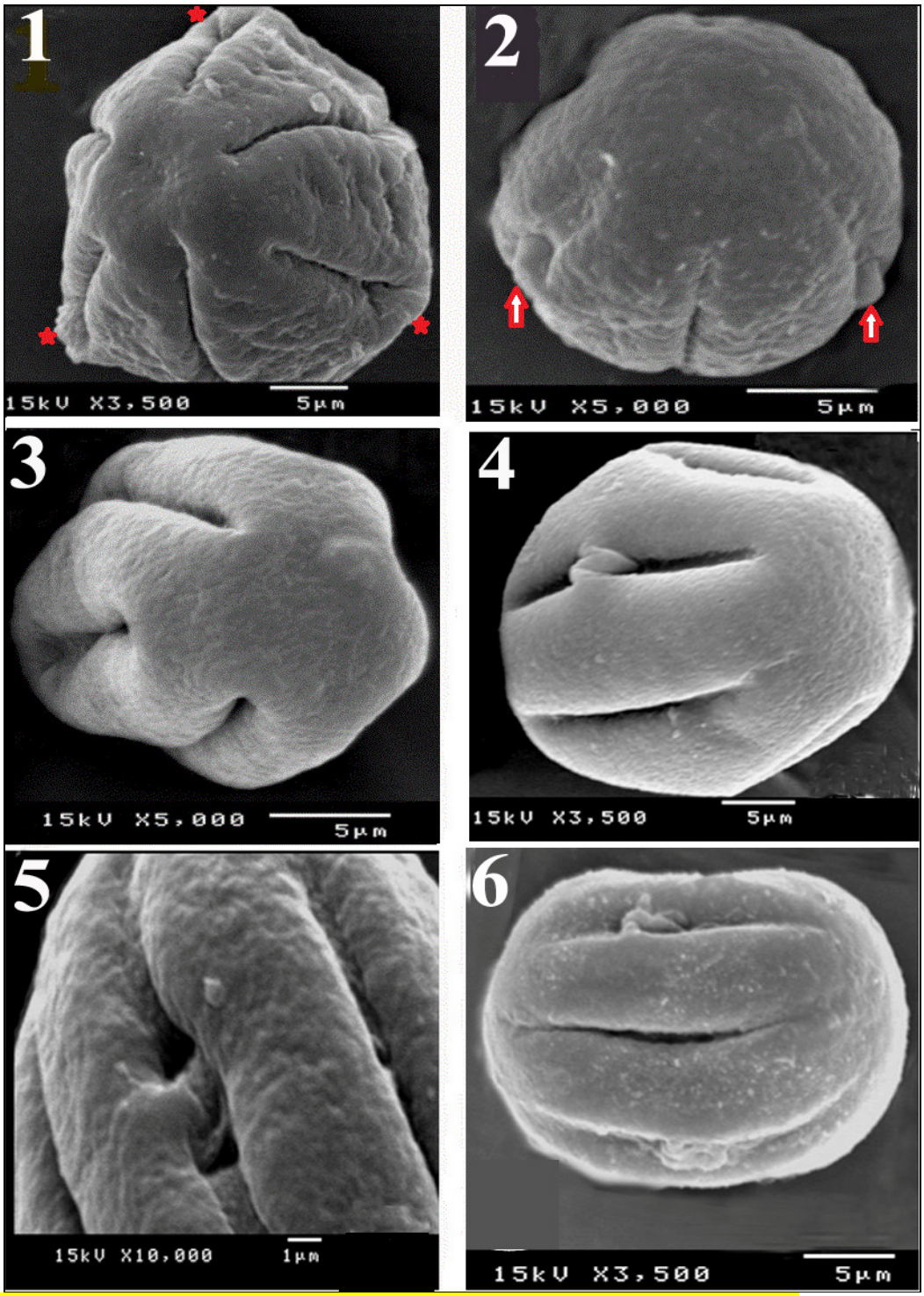

Figure 5. SEM-photos of Heliotropium pollen-**TYPE 1** (**6-8-heterocolpate**):
Subtype a. 6-heterocolpate: 5.1. P-view in *H. supinum* shows the no. of apertures, endoapertures at three compound colpi are asterred.
5.2. oblique P-view in *H. bacciferum* subsp. *bacciferum* var. erosum, endoapertures are arrowed. **5.3.** oblique P-view in *H. pterocarpum* & **5.4.** *H. kassasii* (in comparing).
Subtype b. 8-heterocolpate: 5.5. the sexinous fusion over the endoaperture, colpi margin and the tectum in *H. bacciferum* subsp. *bacciferum* var. bacciferum.
5.6. E-view in *H. ramosissimum* shows two compound colpi and a simple one.