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MICROMORPHOLOGICAL STUDIES OF POLLENS OF THE GENUS *PSYCHROGETON* BOISS. FROM PAKISTAN AND KASHMIR

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Abstract

This genus has not been examined yet either by Light Microscopy or by Scanning Electron Microscopy, not only in Pakistan but all over the world. This is the first time that its micromorphological study of the pollens has been carried out. Systematic studies of this genus Psychrogeton Boiss. have been done and six species are found in Pakistan & Kashmir (Qaiser et al, 2021). Psychrogeton has been placed in the family Asteraceae, Tribe Astereae, Sub-tribe Asteranae, Erigeron-Conyza group, (Bremer, 1994). In this paper we are discussing about the palynological attributes of five species of Psychrogeton and their systemic implication in their identification and circumscription of species. P.alexeenkoi is reported from Pakistan but we couldn't find any herbarium material for the micromorphological studies.Pollens are Sub-oblate, Oblate-spheroidal, radially symmetrical, isopolar, tricolporate. Tectum is perforate/non-perforate and echinate. Spines apices are rounded blunt – acuminate with variable interspinal distance. On the basis of pollen shape, twsix distinct pollen types are recognized. P.aucheri type and P.andryaloides-type.

Introduction:

The genus *Psychrogeton* Boiss. is placed in Erigeron –Conyza group of Sub-tribe Asteranae, Tribe Astereae, Family Asteraceae (Bremer, 1994). *Psychrogeton* Boiss. natively belongs to Anatolia, Central Asia, Iran, Afghanistan and Northern Areas of Pakistan, it is also reported from N. India and China. The genus is studied systematically in Pakistan and six species are circumscribed from Pakistan and Kashmir (Qaiser et al, 2021); *Psychrogeton alexeenkoi* Krasch., *P.amorphoglossus* (Boiss.) Novopokr., *P.andryaloides* (DC) Novopokr. ex Krasch., *P.aucheri* (DC.) Grierson, *P.chitralicus* Grierson and *P.olgae* (Regel & Schmalh.) Novopokr. ex Nevski. We couldn't find any herbarium material of *Psychrogeton alexeenkoi* Krasch for the micromorphological studies, that's why it is not included in this study, yet it is reported from Pakistan and its specimens are lodged in international herbaria. Pollens of *Psychrogeton* Boiss. have now been studied first time all over the world.

Asteraceae is a eurypalynous family (Erdtman, 1952). However, the genus *Psychrogeton* is stenopalynous. Wodehouse (1935) gave a detailed account of pollens of Compositae. He described the characters and coined the terminology also. Other workers such as Erdtmann (1952), Stix (1960), Saad (1961), Jafri (1966), Skvarla & Larson (1965), Skavrla & Turner (1966, 1969), Askerova (1970, 1971, 1973, 1976, 1985, 1992), Robbin *et al* (1979), Sachdeva & Malik, (1986),Faegri & Iverson (1989), Nakajima & Monteiro (1995), Blackmore &Pearson (1996), and Kaya *et al.,* (1996) also worked on this family. Stix (1960) used electron microscopy to reveal the internal morphology of pollen material. In Pakistan, Perveen, Qaiser (1994-2005) and different workers have value-added precious contributions on palynological studies. They have studied the complete pollen flora of Pakistan.

Pollen morphology of Compositae (Asteraceae) was contributed by Perveen (1997); Vilatersana (2001), the Carthamus Complex and the genus *Aegialophila in Centaurinae,* Asteraceae), Abid et *al.,* (2002), (Palynology of Inula), Celik et *al* (2005) discussed Morphology, Anatomy, Ecology and Palynology of two *Centaurea* species from Turkey. Meo & Khan (2006) worked on pollens of genus *Calendula* from Pakistan. Tzvelev (2007) described new taxa and new combinations in Asteraceae from Central Asia. Wortley et *al* (2008) worked on palynology and discussed the evolution of Arctotidae–Compositae.

Taxonomic Study of Gochnatieae and on its basis evolutionary sequences were discussed by Blackmore et *al.,* (2009). They also discussed evolution of pollens in Asteraceae. Wang et *al.,* (2009) worked on Pollen Morphology of Cichorieae –Asteraceae and also discussed its taxonomic significance. The genus *Tibestoseris* and the new genus *Pseudoyoungia* of Cichorieae were described from Eastern Himalya (Maity et *al* 2010). In Iran, Shabestari (2013) worked on pollens of *Centaurea* (Asteraceae). Pollen Morphology and its taxonomic significance were discussed by Telleria et *al* (2013) in the tribe Gochnatieae which supported a recent concept of "Basal Compositae". Another new aspect of Forensic palynology was studied by Alotaibi et al (2020). Souza et al studied pollens of Piptolepis (Asteraceae) from Brazil (2022). There are no records of palynological study of *Psychrogeton* from Pakistan & Kashmir.

All these studies conclude that micro-morphological studies are significant to delimit the taxa not only at generic level but also on specific level. The present data elaborates the micromorphology of cypsela and Pollens of *Psychrogeton* Boiss. from Pakistan & Kashmir.

Material & Methods:

Pollens of four species of genus *Psychrogeton* were taken from herbarium material lodged at KUH, NHI (RAW) and E (Holmgren, 1990). Pollen grains were observed under a Scanning Electron Microscope. For SEM, the acetolyzed material was transferred with a fine pipette on the metal stub, on which double sided adhesive cello tape was attached previously. Then the stubs were coated with gold in the sputtering chamber (Ion-Sputter JFC-1100). The finer descriptive details were carried out on a Joel Microscope (JSM-2). Pollens of one species *P.aucheri* were coated by Auto Fine Coater: JEC-3000 FC. Coating was done on 20 m A current for 30 seconds, then studied under Scanning Electron Microscope Jeol (Japan) model JSM-IT 100.

Mostly up to 5 pollens / species were studied, in the case where the material was not sufficient readings were reduced. The measurements are based on 4-5 readings from each specimen. The following characters were studied. Polar axis (*P*), Equatorial diameter (*E*), aperture size, apocolpium, mesocolpium and

exine thickness were measured (Table: 1). In the description of pollen, the terminology used is in accordance with Erdtman (1969), Kremp (1965), Faegri & Iverson (1989) and Hesse et *al*., (2009).

Results:

General Pollen Characters in the genus *Psychrogeton*:

Pollen grains radially symmetrical, isopolar, polar length ranges from 10-20 µm, equatorial length ranges 15-23 µm; sub-oblate to oblate /oblate spheroidal, sometimes triangular in polar view or circular, Tectum echinate, smooth or layered, some species perforate; aperture tricolporate, zonoaperturate. Tricolporate pollen is mostly basal colporate (pore is towards the basal end of colpi), sometimes apical colporate (pore near the apical region of colpi).

Key to Pollen Types

1 + Pollen Oblate-suboblate..... *P.aucheri* type

– Pollen Spheroidal..... *P. andryaloides* type

Description of Pollen types:

***Pollen type - I*: *P.aucheri* type** ***Pollen Class:* Tricolporate, zonocolporate.**

***P/E Ratio:* 0.80-0.87** ***Shape:* Oblate-suboblate**

Ornamentation: Tectum echinate, Spine smooth-rough; Perforate/Non-Perforate, Spine 2.4-2.6 µm, apex acute-acuminate.

Aperture: Colpi ovate-broad ovate.

***Measurements*:** Polar Axis (12.00-) 15.00 (-18.00) µm and Equatorial Diameter (15.00-) 18.50 (-22.00) µm. P/E Ratio: 0.80-0.87 ; Colpi ovate, Colpi length (7.00-) 9.00 (-11.00) µm. Apocolpium (4.00-) 5.00 (-6.00) µm; Mesocolpium (4.00-) 5.00 (-6.00) µm. Tectum echinate; Spine length 2.5-2.6 µm; interspinal distance 0.50-2.00 µm; Spine ovate- broad ovate, bent- straight; apex acute- sub-acuminate; Exine thickness 3.12- (4.37) -5.00 µm, surface smooth-rough /layered.

Species Included: P.aucheri, P.olgae, P.amorphoglossus.

1	**+**	**Tectum perforate.....**	***P.aucheri***
	-	**Tectum non-perforate.....**	**3**
2	**+**	**Tectum smooth, colpi broad ovate; P/E ratio < 0.85.....**	
		Tectum rough/layered, colpi narrow ovate; P/E ratio > 0.85	***P.olgae***
	-	**upto 0.87.....**	***P.amorphoglossus***

***Pollen Type II*: *P.andryaloides type.* *Pollen Class*: Tricolporate, zonocolporate.**

***P/E Ratio:* 0.88-0.90** ***Shape:* Oblate-Spheroidal**

Ornamentation:Tectum echinate, spine smooth; non-perforate *Aperture:* Tricolporate, Colpi long**.**

***Measurements*:** Polar Axis P (14.00-) 17.00 (-20.00) µm and Equatorial Diameter E (16.00-) 19.00 (-22.00) µm; Colpi length (9.00-) 9.50 (-10.00) µm; Apocolpium (4.00-) 5.00 (-6.00) µm; Mesocolpium (7.00-) 8.00 (-9.00) µm; Tectum echinate; Spine length 2.0 µm; interspinal distance 1.0-1.5 µm; Spine ovate, mostly straight; apex acute; Exine thickness 3.25 (4.25) 5.25 µm; surface smooth-rough/layered.

Species included*: *P.andryaloides, P.chitralicus.

1	**+**	**Tectum rough,spine apices straight.....**	***P.chitralicus***
	-	**Tectum smooth, spine apices slightly bent.....**	***P.andryaloides***

Discussion:

There is no report on palynology of the genus *Psychrogeton*. Its pollen morphology has been studied first time, hence taxonomic treatment has been given to the genus by different workers of this region but no work is done in Pakistan.

Psychrogeton is the member of Asteraceae, which is a eurypalynous family with echinate exine, while the genus understudy is a stenopalynous taxa.

Pollen Study:

Plant has following distinguishing pollen characters i.e Pollen grains radially symmetrical, isopolar, sub-oblate to oblate or oblate spheroidal, sometimes triangular or circular in polar view; aperture tricolporate, zonocolporate with echinate tectum. Tricolporate pollen is mostly basal colporate (pore is towards the basal end of colpi), sometimes apical colporate (pore near the apical region of colpi). Although *P.alexeenkoi* is reported from Pakistan. But we have not found its any specimen to study that's why its micromorphological studies are not included in this paper.

Based on morphology of pollen 2 types of pollens are recognized i.e, Pollen type-I: *P. aucheri*, Pollen type-II: *P.andryaloides* type.

These types are based on the shape of pollens. The shape of the pollens ranges from sub-oblate to spheroidal, P/E ratio lies between 0.80-0.90. Type–I includes 3 species, *P.amorphoglossus, P.aucheri* & *P.olgae* with P/E ratio upto 0.87. This group furthers bifurcates on the basis of smooth or perforate pollen surface. *P.aucheri* is the species with perforations on surface, and rest of the two species, tectum is non-perforate. Second group, has non-perforate tectum. It includes 2 species i.e *P.andryaloides* and *P.chitralicus;* both of them have the thick caespitose body with basal leaves and varied type of indumentums, but also have some differences morphologically but their pollens are almost similar. There are minute differences among the micromorphological details of pollens of all these species so they are identifiable. *P.chitralicus* has echinate tectum with straight spine having rough and layered surface, while *P.andryaloides* has spines with slightly bent apices and smooth tectum.

Appendix I: Voucher specimens for Pollen Morphology

1. *P.amorphoglossus* (Boiss.) Novopokr.:(i) Deosai Plain, Jan Alam and Ghulam Nabi, 1497, 20-08-2002, (KUH).

(ii) Chitral: Khot Garhi, Torikhoo (Turkoh), (72.38'N, 36.32'E), 14-08-2005, *Haider Ali* 2732, (KUH).

2. *P.andryaloides* (DC) Novopokr. ex Krasch. *var. andryaloides*: Base Camp, Hushu Valley, Baltistan, E.Nasir & G.L. Wehtsu, 5970, 5-7-1955, (RAW).

3. *P.andryaloides var. poncinsii*: Chaupursan Upper Hunza, Jan Alam & Abdul Hameed 1233, Late July-2002, (KUH).

4. *P.aucheri* (DC.) Grierson: Lutkhoo, Gobor Shasadin, 10-08-2005, *Haider Ali* 2583, (KUH).

5. *P.olgae* (Regel & Schmalh.) Novopokr. ex Nevski.: Peer rout Village Borophow, Astore, Ali Noor 1322, 14-05-2008, (KUH).

6. *P.chitralicus* Grierson: Above Batain Distt. Ushu, Stewart R.R & Rahman A., 25309, 28-7-1953, (RAW).

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Table No. 1: Measurements of Pollens of *Psychrogeton* species.

Species	Polar Axis (µm)	Equatorial Axis (µm)	P/E	Shape (Equatorial View)	Colpi Length (µm)	Apo-colpium (µm)	Meso-colpium (µm)	Pollen Surface	Spine Length (µm)	Interspinal distance (µm)	Apex of Spine
P. olgae	12(13.5)15	15(16.5)18	0.81	Suboblate	10	4	6	Echinate	±2.6	1.0-2.0	Ovate, acuminate, bent, smooth
P. aucheri	17(18)19	20(21)22	0.85	Suboblate	±7	5-6	5-6	Echinate Perforate	±2.50	0.5-1	Broad ovate, acute, bent, layered
P. amorphoglossus	17(17.5)18	19(20)21	0.87	Suboblate	8-10	4	6-7	Echinate	±2.50	0.5-1.0	Ovate, acuminate, bent, layered
P. chitralicus	14(16)18	16(18)20	0.88	Oblate-spheroidal	9	4	6	Echinate	±2.94	1.0-1.5	Ovate, acute, straight, rough and layered
P. andryaloides	15(17)19	17(19)21	0.89	Oblate-Spheroidal	10	5-6	7-9	Echinate	±2.0	1.0-1.5	Ovate, Acute Straight, almost smooth

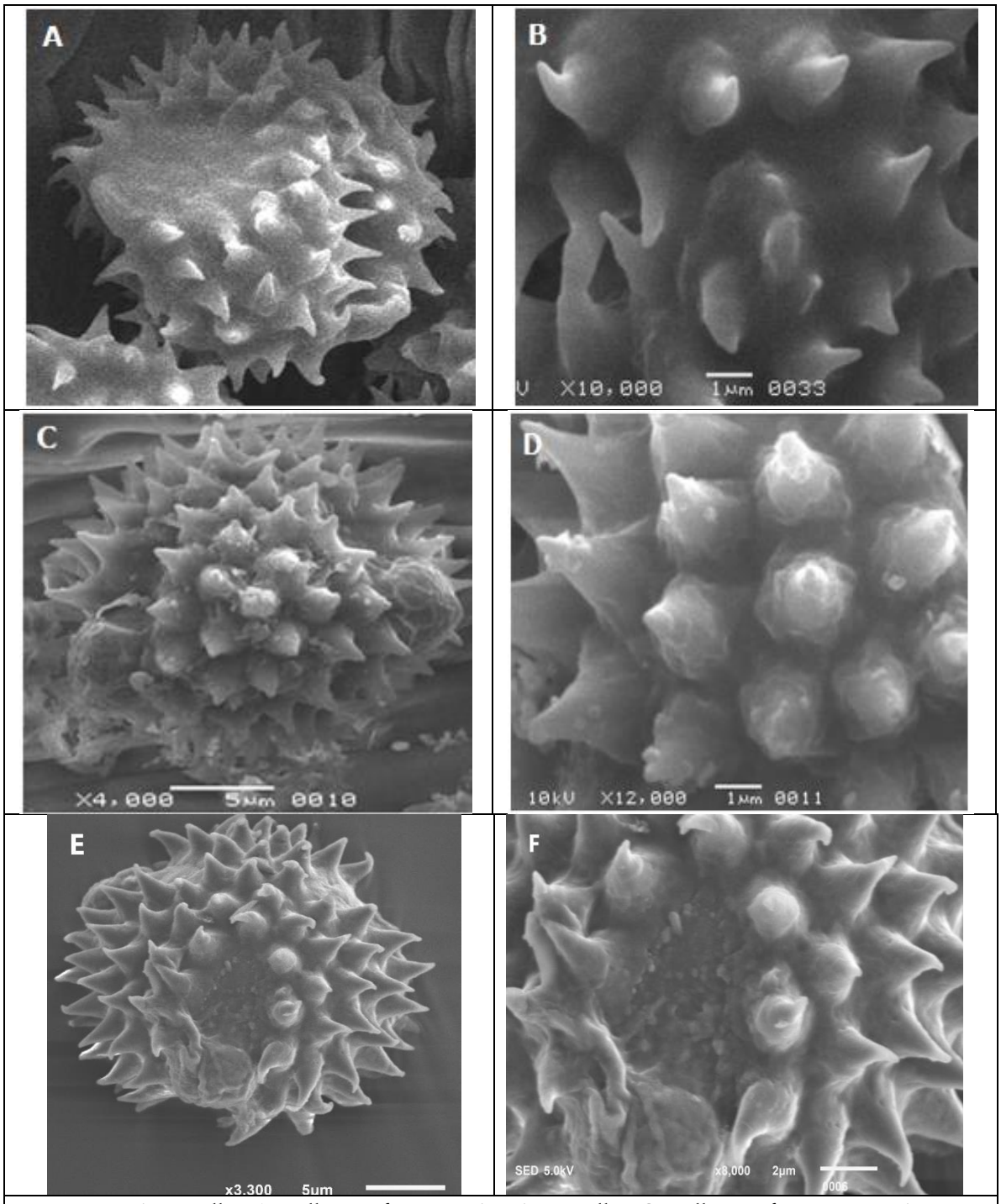


Figure 1: *P.olgae*: Pollen A, Pollen surface B; *P.chitralicus*: Pollen C, Pollen surface D, *P. aucheri* : Pollen E, Pollen surface F.

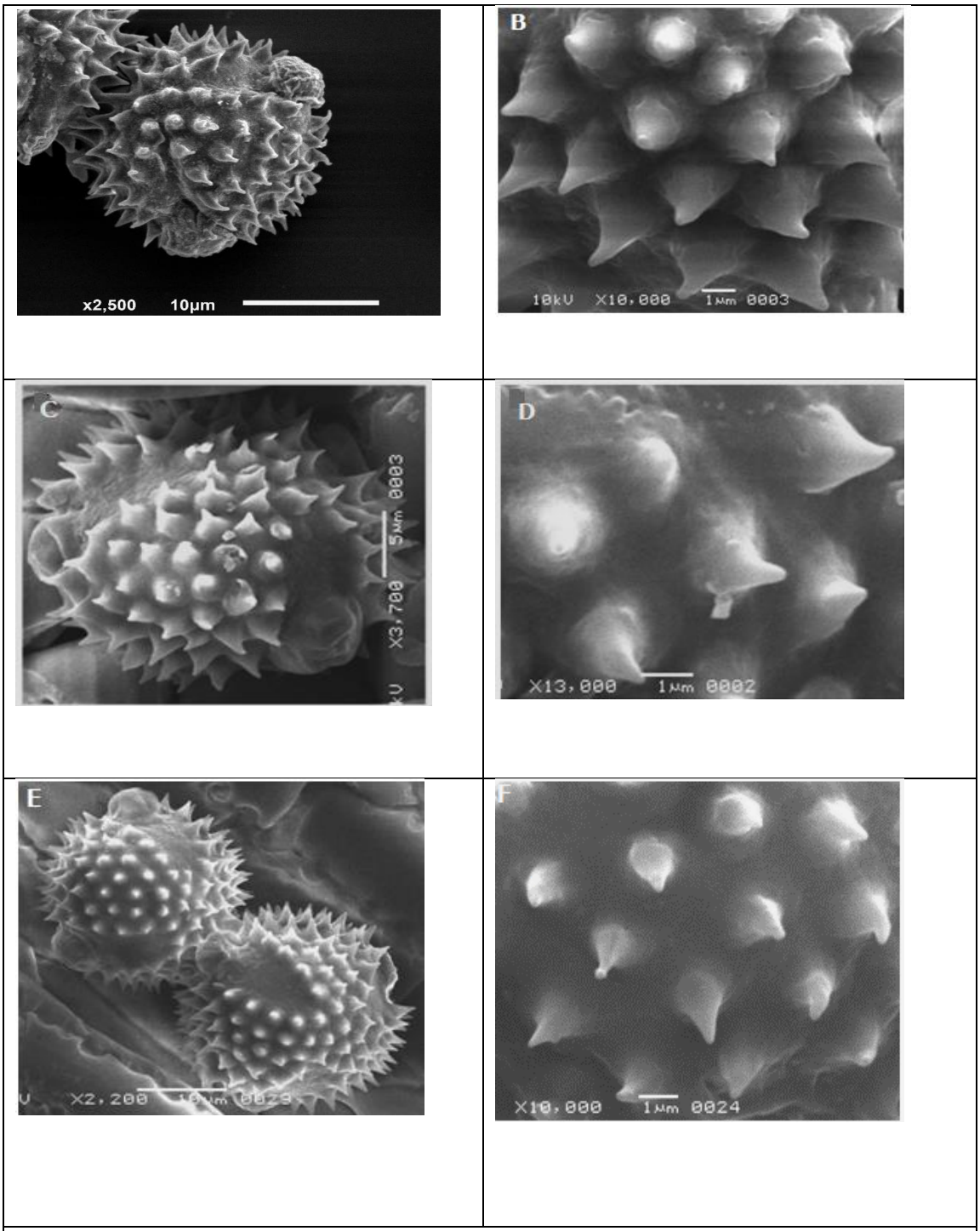


Figure 2: *P.amorphoglossus* : Pollen A, Pollen surface B; *P.andryaloides var. poncinsii*: Pollen C, Pollen surface D, *P.andryaloides var. andryaloides*: Pollen E, Pollen surface F.

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