Pollen morphology of the Bulgarian species from section *Lathyrostylis* (genus *Lathyrus*, *Fabaceae*)

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Abstract. The pollen morphology of five species from the section *Lathyrostylis* (*Lathyrus*, *Fabaceae*) distributed in Bulgaria (*Lathyrus digitatus*, *L. filiformis*, *L. pallescens*, *L. pancicii* and *L. pannonicus*) was studied with LO microscope and SEM. The pollen grains are 3-zonocolporate, of *prolate*, *prolate-sphaeroidal* and *oblate-sphaeroidal* type, medium in size, elliptical or rectangular (quadratic) in equatorial view and circular to slightly triangular-obtuse-convex in polar view. The ornamentation is predominantly tectum perforatum (punctate-foveolate). The rectangular pollen grains in equatorial view (*L. filiformis*, *L. pallescens* and *L. pancicii*) should be differentiated into a new pollen type *Lathyrus filiformis*.

Key words: Bulgaria, *Lathyrostylis*, *Lathyrus*, pollen morphology

Introduction

The genus *Lathyrus* L. (*Fabaceae*) belongs to the tribe *Vicieae* (Adans.) DC., with about 150 species related to 13 sections (Kupicha 1983). The section *Lathyrostylis* (Griseb.) Bässler comprises 20 perennial species distributed in Central and South Europe, Southwest Asia and Northwest Africa. Although fairly large, this section is remarkable for its relative uniformity, its species share several highly evolved and specialized characters (leaves with few narrowly linear-lanceolate leaflets with parallel venation, inflorescence, lax, several-flowered and often with spathulate style at the apex). Most representatives are characteristically disjuncted and about 60% are endemic to Southwest Asia (Anatolia). The limited geographical distribution and narrow range of variability in this section, combined with its large size, imply that many of the species are of comparatively recent origin, and that the group is still actively evolving (Kupicha 1983).

The section *Lathyrostylis* is represented by five species in the Bulgarian flora: *Lathyrus digitatus* (M. Bieb.) Fiori, *L. filiformis* (Lam.) Gay, *L. pallescens* (M. Bieb.) K. Koch, *L. pancicii* (Jurišić) Adamović, and *L. pannonicus* (Jacq.) Garcke (Tosheva 2005). The species *L. pancicii* is endemic to the Balkan Peninsula.

Pollen-morphological data for the genus *Lathyrus* is available in the publications of Faegri (1956), Gapotchka (1974), Clarke & Kupicha (1976), Fergusson & Skvarla (1981), Faegri & Iversen (1989), Moore & al. (1991), Reille (1995, 1998), Halbritter (2000), and Beug (2004). For the species of the section *Lathyrostylis* distributed in Bulgaria, there is information in literature only for *L. pannonicus* (Kuprianova & Alyoshina 1972; Reille 1998; Halbritter 2000; Beug 2004) and for *L. filiformis* (Reille 1992). Earlier pollen-morphological information is not available for the other species in this section.

This paper is the third in a series of contributions related to the pollen-morphological characteristics of the Bulgarian representatives of genus *Lathyrus* studied by LO microscopy and SEM. These papers present and discuss the pollen morphology of the species from sections *Lathyrus* (Tosheva & al. 2003) and *Orobus* (Tosheva & Tonkov 2005).

Material and methods

Pollen material was collected from natural populations of the concerned taxa and from specimens kept in the Herbarium of Sofia University (SO). The voucher specimens are also deposited in this herbarium.

Pollen grains for LO examinations were prepared following the standard procedure of Erdtman (1960). Six pollen-morphological characters were measured with an Amplival (Carl-Zeiss Yena) microscope at magnification of × 800: P (polar diameter), E (equatorial diameter), Lc (colpus length), Lp (porus length = diameter), M (mesocolpium), A (apocolpium), and the P/E ratio. Thirty measurements were made for each character and the mean values and ranges are shown in Table 1. Light microphotographs were taken at magnification of × 3000. For SEM examination, the acetolysed pollen grains were coated with gold as dry specimens with JEOL-JFC-1200 coater. Microphotographs were obtained with JEOL-JSM-5510 SEM at magnification of × 1900–25000 (Plates I–III).

The pollen-morphological descriptions follow the terminology of Punt & al. (1994).

Results and discussion

L. digitatus (M. Bieb.) Fiori var. *puberulus* (Jordanov) Kožuharov (Plate I, Figs 1-4)

Black Sea Coast (*South*): Maslen Nos, (Primorsko), MG-67, 41°59' N, 27°23' E, 12.05.2005, coll. *B. Assyov* & *A. Tosheva* (SO 103948); Thracian plain : Haskovo, LG--84, 41°58' N, 25°39' E, 30.07.2003, coll. *A. Tosheva* & *D. Pavlova* (SO102679).

Pollen class: 3-zonocolporate (seldom 2-zonocolporate).

Pollen group: prolate (P/E = 1.43).

Dimensions: Medium size [PnE = 30.0 (41.6) 50.6 × 22.1 (29.4) 37.9 μm].

Apertures: Ectoapertures – colpi: long, straight, with acute ends, with costae along the margins of the colpi, colpus membrane covered by granules; endoapertures – pori: large, circular to slightly lalongate.

Outlines: Equatorial view rectangular-obtuse-convex to elliptic for the 2-zonocolporate grains; polar view triangular-obtuse-convex to nearly circular.

Ornamentation: Tectum perforatum (punctate-foveolate). Apocolpium and aperture area almost psilate or with scattered punctae.

Exine: The thickness in mesocolpium is 1.00 μm and 0.75 μm in apocolpium. Nexine is well developed, thinner, or as thick as sexine; infratectum as thick as tectum. Structural elements of the infratectum: columellae, occasionally mixed with granules.

Table 1. Measurements of the pollen-morphological characters.

Species	P, μm	E, μm	Lc, μm	Lp, μm	M, μm	A, μm	P/E
L. digitatus	30.0 (41.6) 50.6	22.1 (29.4) 37.9	22.2 (28.6) 36.3	4.0 (5.5) 9.5	12.6 (17.6) 23.7	9.5 (13.1) 17.4	1.20 (1.43) 2.00
L. filiformis	26.9 (32.8) 37.9	30.0 (35.3) 39.5	18.9 (24.2) 28.4	3.2 (6.0) 7.9	15.5 (22.8) 30.0	9.5 (16.2) 20.5	0.83 (0.93) 1.09
L. pallescens	30.0 (43.6) 53.7	31.6 (42.3) 52.7	22.1 (31.0) 42.7	4.7 (6.8) 12.0	20.5 (26.5) 34.8	11.1 (19.6) 34.8	0.90 (1.03) 1.17
L. pancicii	30.0 (35.5) 43.5	28.4 (34.4) 41.1	18.9 (25.9) 30.0	3.9 (6.0) 7.9	14.2 (21.4) 26.9	11.1 (15.8) 20.5	0.95 (1.03) 1.10
L. pannonicus	26.9 (35.8) 45.8	19.0 (25.6) 31.6	20.5 (26.2) 33.2	2.4 (5.0) 7.1	11.1 (16.9) 23.7	6.3 (12.6) 19.0	1.20 (1.39) 1.67

P (polar diameter); **E** (equatorial diameter); **Lc** (colpus length); **Lp** (porus length = diameter); **M** (mesocolpium); **A** (apocolpium); **P/E** ratio.

***L. filiformis* (Lam.) Gay** (Plate II, Figs 1-8)

Rhodopi Mts (*Central*): Mougla, 1400 m, KG-91, 41°37' N, 24°30' E, 26.06.2002, coll. *A. Tosheva* & *D. Pavlova* (SO103053).

Pollen class: 3-zonocolporate.

Pollen group: oblate-sphaeroidal (P/E = 0.93).

Dimensions: Medium size [PxE = 26.9 (32.8) 37.9 × 30.0 (35.3) 39.0 μm].

Apertures: Ectoapertures – colpi: short, wide, with acute ends, with costae along the margins of the colpi, colpus membrane covered by fine granules; endoapertures – pori: large, circular.

Outlines: Equatorial view rectangular (quadratic); polar view circular to slightly triangular-obtuse-convex. A triangular concavity is outlined in the polar area.

Ornamentation: Tectum perforatum (punctate-foveolate); apocolpium and aperture area psilate. The triangular concavity covered by granules.

Exine: The thickness in mesocolpium is 1.85 μm and 1.20 μm in apocolpium. Sexine is well developed, twice as thick as nexine. Infratectum as thick as tectum and columellae are the structural elements.

According to Reille (1992), the pollen grains are 3-zonocolporate.

***L. pallescens* (M. Bieb.) K. Koch** (Plate I, Figs 5-8)

Sofia region: Beledie Han, F-74, 42°53' N, 23°08' E, 28.05.2005, coll. *A. Tosheva* (SO 103951); Mt Sredna Gora (*Western*): Mt Lozenska, GN-91, 42°36' N, 23°29' E, 02.06.2002, coll. *A. Tosheva* (SO 103950).

Pollen class: 3-zonocolporate.

Pollen group: prolate-sphaeroidal (P/E = 1,03).

Dimensions: Medium size [PxE = 30.02 (43.63) 53.72 × 31.6 (42.26) 52.14 μm].

Apertures: Ectoapertures – colpi: short, straight, with acute ends, with costae along the margins of the colpi, colpus membrane covered by granules; endoapertures – pori: large, circular.

Outlines: Equatorial view rectangular (quadratic); polar view circular to slightly triangular-obtuse-convex. A triangular concavity is outlined in the polar area.

Ornamentation: Tectum perforatum (punctate), the largest punctae are observed in mesocolpium. Apocolpium and adjacent area to the aperture are psilate. The triangular concavity is covered by granules.

Exine: The thickness in mesocolpium is 1.00 μm and 0.75 μm in apocolpium. Nexine is well developed, slightly thinner, or as thick as sexine. Infratectum is well developed, as thick as tectum, structural elements – columellae and granules.

***L. pancicii* (Jurišić) Adamović** (Plate III, Figs 1-4)

Znepole region: Mt Lyubasha, FN-43, 42°46'N, 22°46'E, 17.09.2004, coll. *A. Tosheva* & *B. Assyov* (SO 103624).

Pollen class: 3-zonocolporate.

Pollen group: prolate-sphaeroidal (P/E = 1.03).

Dimensions: Medium size [P×E = 30.0 (35.5) 43.4 × 28.4 (34.4) 41.1 μm].

Apertures: Ectoapertures – colpi: straight, with acute ends, with costae along the margins of the colpi, colpus membrane covered by fine granules; endoapertures – pori: large, circular.

Outlines: Equatorial view rectangular (quadratic); polar view circular to slightly triangular-obtuse-convex. A triangular concavity is outlined in the polar area.

Ornamentation: Tectum perforatum (punctate). Apocolpium and aperture area are psilate. The triangular concavity is covered by granules.

Exine: The thickness in mesocolpium is 1.80 μm and 1.50 μm in apocolpium. Sexine is twice as thick as nexine. Infratectum is well developed, with columellae as structural elements.

***L. pannonicus* (Jacq.) Garcke subsp. *varius* (K. Maly) P.W. Ball** (Plate III, Figs 5-8)

Sofia region: Beledie Han, FN-74, 42°53' N, 23°08' E, 28.05.2005, coll. *A. Tosheva* (SO 103952); Mt Sredna Gora (*Western*): Mt Lozenska, FN 91, 42°36' N, 23°25' E, 03.06.2003, coll. *A. Tosheva* (SO 103953).

Pollen class: 3-zonocolporate.

Pollen group: prolate (P/E = 1.39).

Dimensions: Medium size [PxE = 26.86 (35.77) 45.82 × 18.96 (25.64) 31.60 μm].

Apertures: Ectoapertures – colpi: long, straight, with acute ends, with costae along the margins of the colpi, colpus membrane covered by granules; endoapertures – pori: large.

Outlines: Equatorial view – elliptic to slightly rectangular-obtuse-convex; polar view – triangular-obtuse-convex to nearly circular.

Ornamentation: Tectum perforatum (punctate-foveolate), gradually transformed into suprareticulum in the equatorial area. Apocolpium and aperture area are almost psilate or with single punctae.

Exine: The thickness in mesocolpium is 1.25 µm and 0.75 µm in apocolpium. Nexine is thinner (1/3–2/5 as thick as exine) than sexine. Infratectum is well developed, as thick as tectum, with columellae as structural elements.

Our results confirm and supplement the data provided by other authors (Kuprianova & Alyoshina 1972; Reille 1998; Halbritter 2000; Beug 2004). According to Beug (2004), the size of the pollen grains is P = 31.0 (35.0) 39.0 µm. Halbritter (2000) reports for *prolate*-type pollen medium in size and circular in equatorial view. According to Kuprianova & Alyoshina (1972), pollen is elliptical in equatorial view and triangular-obtuse in polar view, PxE = (33.8) 36.4–37.6 × 26.0–28.6 µm. The protuberances on the tectum surface form a slightly distinct reticulum.

The studied palynotaxonomical characters (pollen size, outline in equatorial view and P/E ratio) assign the pollen grains to two pollen groups:

Group I. Pollen grains rectangular (quadratic) in equatorial view, P/E<1.14, medium in size, polar area covered by different in size granules (*L. filiformis, L. pallescens* and *L. pancicii*).

Group II. Pollen grains elliptical in equatorial view, P/E = 1.3–2.00 (*prolate*), medium in size, polar area psilate, occasionally with puncta (*L. digitatus, L. pannonicus*). The pollen grains of *L. digitatus* are slightly larger.

The pollen morphology of the taxa from section *Lathyrostylis* is quite different from the results obtained for sections *Lathyrus* (Tosheva & al. 2003) and *Orobus* (Tosheva & Tonkov 2005). All pollen grains studied by us are 3-zonocolporate, medium to large in size. Differences are observed in the shape in equatorial view and P/E ratio, as the pollen grains of sections *Lathyrus* and *Orobus* appear predominantly elliptical and related to the *sub-prolate* pollen group. Sexine is thicker than nexine in the pollen grains of the species from section *Lathyrostylis* due to a well-developed infratectum, while sexine of the species from sections *Lathyrus* and *Orobus* is generally as thick as nexine.

Conclusions

The present investigation has shown that section *Lathyrostylis* is a heterogeneous group, according to the pollen morphology of the studied taxa.

The pollen grains are 3-zonocolporate, medium in size. The smallest pollen grains belong to *L. filiformis* and the largest to *L. pallescens*. The species are assigned to three pollen types:

1. **Prolate:** *L. digitatus, L. pannonicus*;

2. Oblate-shaeroidal: *L. filiformis*;

3. Prolate-sphaeroidal: *L. pallescens*, *L. pancicii*.

The shape in equatorial view is elliptic to rectangular-obtuse-convex (*L. digitatus*, *L. pannonicus*), or rectangular (*L. filiformis*, *L. pallescens*, *L. pancicii*). In polar view, the pollen grains are circular to triangular-obtuse-convex.

The aperture system is composed of ectoapertures (colpi) and endoapertures (pori). The colpi are straight, varying in length and width, usually with acute ends and costae along the colpus margins. The colpus membrane is covered by granules. The pori are circular, large.

The thickness of the exine is 0.75–1.85 µm; the sexine is thicker than the nexine, with well-developed infratectum, usually as thick as tectum. Columellae are the structural elements of the infratectum,, occasionally mixed with granules (*L. digitatus, L. pallescens*).

The ornamentation is tectum perforatum (punctate or punctate-foveolate), seldom transformed to suprareticulum in the equatorial area (*L. pannonicus*).

Usually the pollen grains of the *Lathyrus* species are referred to *Vicia cracca*-type, or *Vicia*-type (Faegri & Iversen 1989; Moore & al. 1991), owing to the presence of transitional types of sculpture. Our results show that the studied species should be related to the *Lathyrus*-type, as recently suggested by Beug (2004). In our opinion, the pollen grains which are rectangular (quadratic) in equatorial view (*L. filiformis*, *L. pallescens*, *L. pancicii*) should be differentiated into a new pollen type: *Lathyrus filiformis.*

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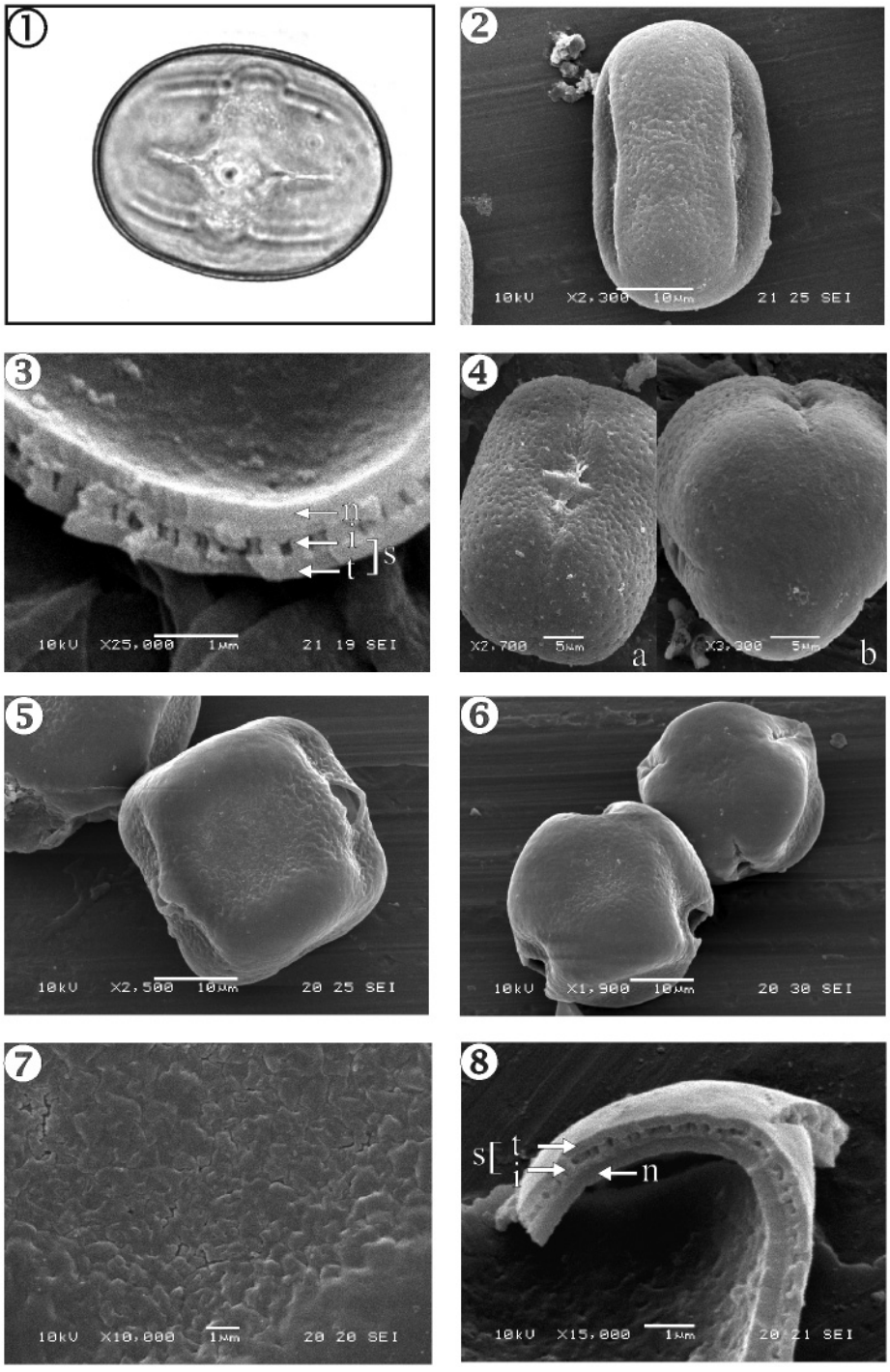
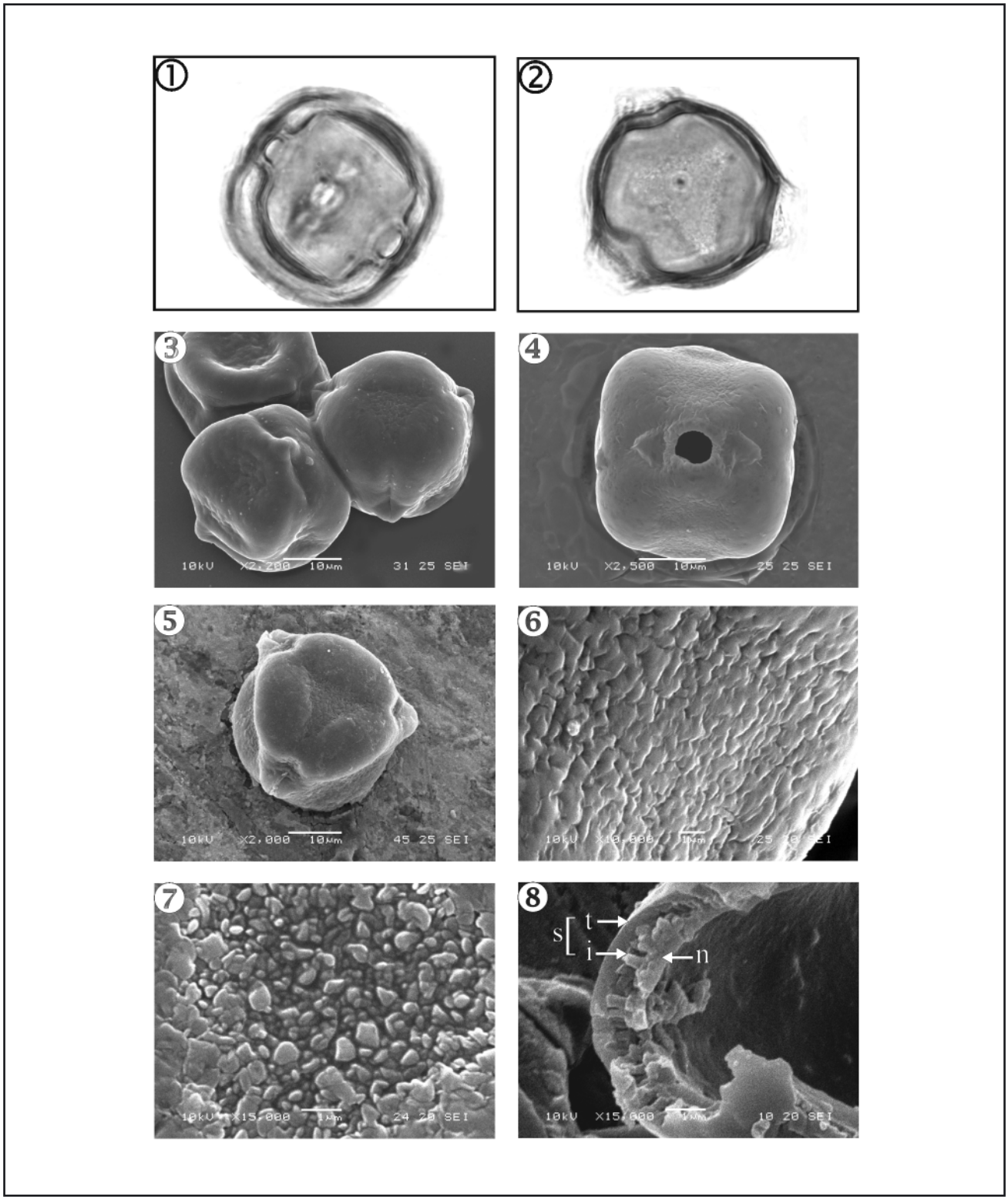
Plate I



Figs 1-4. Pollen grains of *L. digitatus*: **1,** equatorial view, outline, LO, × 3000; **2**, equatorial view, apertures, SEM; **3**, exine structure: **s** – sexine (**t** – tectum, **i** – infratectum), **n** – nexine SEM; **4**, pollen grains: **a** – 2-zonocolporate, **b** – 3-zonocolporate, SEM;

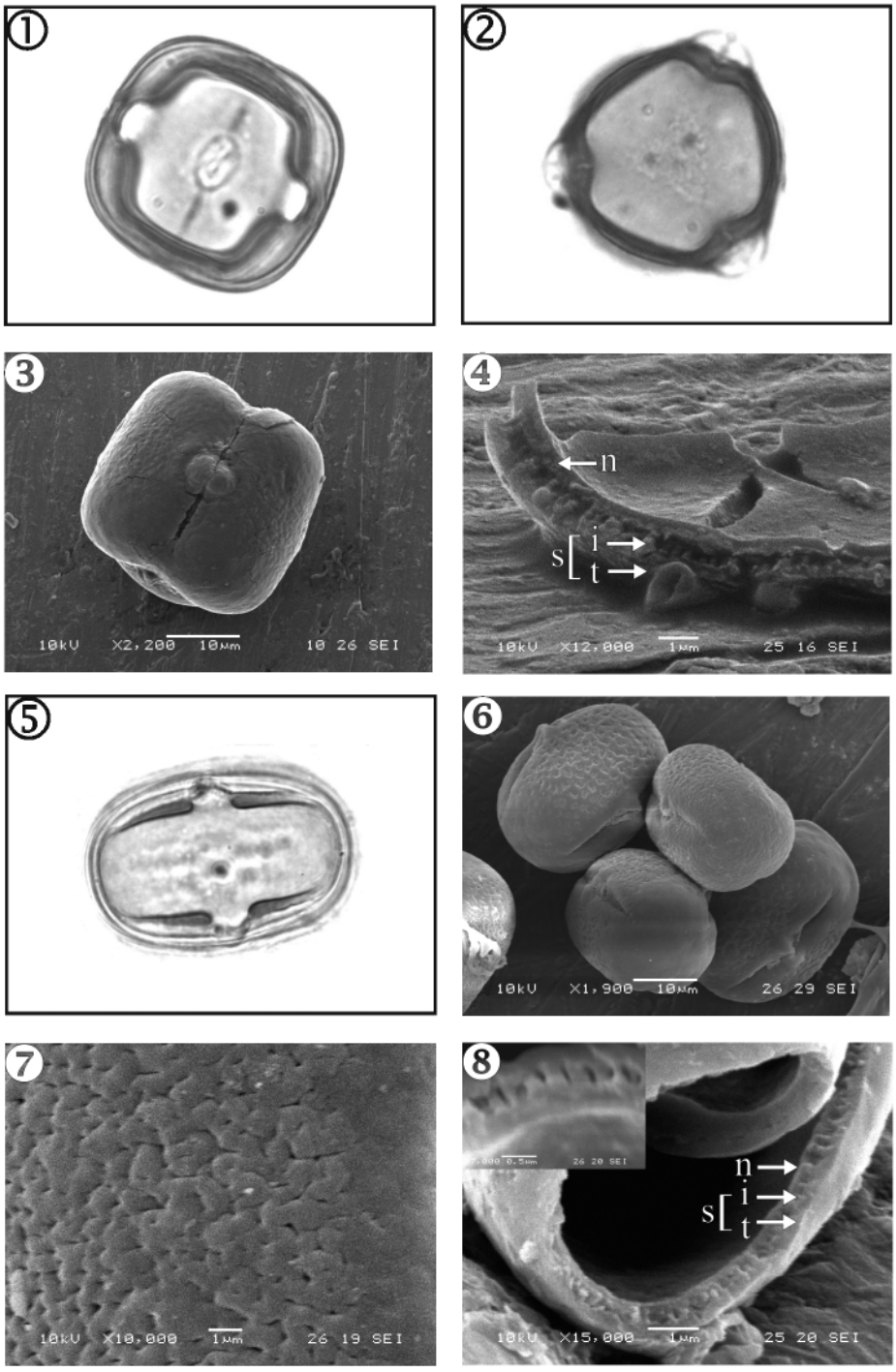
Figs 5-8. *L. pallescens*: **5**, equatorial view, ornamentation, apertures, SEM; **6**, polar view, apertures, SEM; **7**, ornamentation in mesocolpium, SEM; **8**, exine structure, SEM.

Plate II



Figs 1-8. Pollen grains of *L. filiformis*: **1**, equatorial view, outline, LO, × 3000; **2**, polar view, outline, LO, × 3000; **3**, equatorial and polar view, ornamentation, apertures, SEM; **4**, equatorial view, apertures, SEM; **5**, polar view, apertures, SEM; **6**, ornamentation in mesocolpium, SEM; **7**, ornamentation in apocolpium, SEM; **8**, exine structure: **s** – sexine (**t** – tectum, **i** – infratectum), **n** – nexine SEM.

Plate III



Figs 1-4. Pollen grains of *L. pancicii*: **1**, equatorial view, outline, LO, × 3000; **2**, pollar view, outline, LO, × 3000; **3**, equatorial view, apertures; **4**, exine structure: **s** – sexine (**t** – tectum, **i** – infratectum), **n** – nexine SEM;

Figs 5-8. *L. pannonicus*: **5**, equatorial view, outline, LO, × 3000; **6,** equatorial and polar view, ornamentation, apertures, SEM; **7**, ornamentation in mesocolpium, SEM; **8**, exine structure, SEM.

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