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Pollen morphology of the genus *Lathyrus* L. (Fabaceae) with emphasis on its systematic implications

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Abstract

The pollen morphology of 21 taxa of the genus *Lathyrus*, representing 9 sections *Orobus*, *Lathyrostylis* (=*Platystylis*), *Lathyrus* (syn. *Cicercula*), *Orobon*, *Pratensis*, *Aphaca*, *Viciopsis*, *Linearicarpus* and *Nissolia* from Central Anatolia, Turkey was investigated using light microscopy (LM) and scanning electron microscopy (SEM). The pollen grains of the studied taxa are 3-zonocolporate, spheroidal to prolate and medium to large (P = 28.67-54.32 μ m, E = 20.17-52.58 μ m). Their outline in the equatorial view is quadratic-obtuse-emarginate in *L. tukhtensis*, *L. cilicicus* and *L. inconspicuus* var. *inconspicuus* or elliptical to rectangular-obtuse-convex in the other taxa, and in the polar view it is circular, triangular or quinquangular-obtuse-convex. The surface ornamentation of the mesocolpium varies from obscurely reticulate-perforate in *L. incurvus*, *L. brachypterus*, *L. armeus* and *L. hirsutus*; perforate in *L. tukhtensis* and *L. cilicicus*; reticulate-granulate in *L. cicera*; perforate-granulate in *L. inconspicuus* var. *inconspicuus* to finely reticulate-perforate in the rest of the taxa. The apocolpium and colpus area are psilate or perforate in all taxa except *L. chloranthus*, which exhibits the finely reticulate-perforate pattern. The findings of the study reveal that pollen morphological characters such as the surface sculpturing type, pollen size, shape and outline in equatorial view are mostly useful in distinguishing the studied taxa, but they do not provide strong evidence for the infrageneric delimitation of the genus.

Keywords: Pollen morphology, Lathyrus, Fabaceae, Central Anatolia, Turkey.

Introduction

Lathyrus L. is the largest genus in the economically important tribe Fabeae (syn. Vicieae) of the subfamily Papilionoideae (or Faboideae), with about 170 annual and perennial species in the world (Lewis et al., 2005; ILDIS, 2010). Lathyrus is an almost exclusively temperate genus and its species are distributed throughout the northern hemisphere, with a disjunction in South America (Kenicer, 2008). The eastern Mediterranean region is the main center of diversity for the genus, which is less diversified in North and South America (Kupicha, 1983; Simola, 1986). A few species also reach tropical and the highlands of East Africa (Senn, 1938; Davis, 1970; Kenicer et al., 2005). Endemic species are distributed in all continents, except Australia and Antarctica (Kupicha, 1981). Many species of Lathyrus are mesophytes with erect, climbing or sprawling habit and preferring open woodlands, forest margins, roadside verges, littoral, alpine and drought habitats (Kenicer et al., 2005). Lathyrus species are economically important plants, consisting of food and fodder crops, ornamentals, soil nitrifiers, dune stabilizers, significant agricultural weeds, and model organisms for genetic and ecological research (Chittenden, 1951; Kenicer et al., 2005). L. cicera L., L. latifolius L., L. odoratus L., L. sativus L. and L. tuberosus L. have commercial importance, especially for their ornamental value or as forages and feed. Moreover, L. aphaca L., L.

clymenum L., L. hirsutus L. and L. pratensis L. are very well as weedy aliens and can also be found almost anywhere (Kenicer, 2008). Most researchers separate Lathyrus into 12 or 13 sections in the world (Czefranova, 1971; Kupicha, 1983; Asmussen and Liston, 1998; ILDIS, 2002; Leht, 2009). Kupicha's (1983) infrageneric classification of the genus is the most comprehensive of recent accounts. She classified it into 13 sections on the basis of morphological traits: Orobus, Lathyrostylis, Lathyrus, Orobon, Pratensis, Aphaca, Clymenum, Orobastrum, Viciopsis, Linearicarpus, Nissolia, Neurolobus and Notolathyrus. Since Kupicha's (1983) monograph, Doğan et al. (1992) performed a phenetic analysis of vegetative and floral characters of 54 Lathyrus species in Turkey. Their results showed that these species were clustered into 9 sections, and confirmed sections Orobus, Lathyrostylis, and Clymenum proposed by Kupicha (1983), however disagreed on the circumscription of the remaining sections. Davis (1970) also recognized 58 Lathyrus species from Turkey under 10 sections. Kupicha's (1983) classification system has been generally supported by many recent molecular phylogenetic studies (e.g. Croft et al., 1999; Kenicer et al., 2005). However, cpDNA (Croft et al., 1999) and AFLP analyses (Badr et al., 2002) of Lathyrus species propose that reclassification of some species to different sections may be required. In Turkey, the genus is

represented by 76 taxa (66 species) (Davis, 1970; Davis et al., 1988; Güneş and Özhatay, 2000; Genç and Şahin, 2011), belonging to eleven sections according to the infrageneric system of Kupicha (1983): Orobus, Lathyrostylis (=Platystylis), Lathyrus (syn. Cicercula), Orobon, Pratensis, Aphaca, Clymenum, Orobastrum, Viciopsis, Linearicarpus and Nissolia. Turkey is a major centre of diversity for the genus in Euroasia with a high number of species compared to neighbouring countries: 54 species in Flora Europaea (Tutin and Heywood, 1981), 50 in Flora of the USSR (Fedchenko, 1948), 25 in Flora Iranica (Rechinger, 1979), 18 in Flora of Iraq (Townsend and Guest, 1977) and 11 in Flora of Cyprus (Meikle, 1977). The pollen morphology of some taxa of Lathyrus has been previously studied (Faegri, 1956; Gapotchka and Chamara, 1972; Gapotchka, 1974; Clarke and Kupicha, 1976; Fergusson and Skvarla, 1981; Faegri and Iversen, 1989; Moore et al., 1991; Perveen and Qaiser, 1998; Halbritter, 2000; Mantar et al., 2001, 2003; Beug, 2004; Tosheva and Tonkov, 2005, 2007; Güneş and Aytuğ, 2010; Günes and Cirpici, 2010; Günes, 2011a, b). Moore et al. (1991) classified Lathyrus pollen grains as Vicia cracca type and Lathyrus type. In Bulgaria, Tosheva and Tonkov (2007) found a new pollen type, the Lathyrus filiformis type, indicated by L. filiformis, L. pallescens and L. pancicii from sect. Lathyrostylis. Since 2009, the second author has undertaken a revision study of Lathyrus in Central Anatolia and collected a large number of specimens during his extensive field studies (Çildir, 2011). The present study aims to provide a detailed account of the pollen morphology of 21 taxa of Lathyrus, belonging to 9 sections Orobus, Lathyrostylis, Lathyrus, Orobon, Pratensis, Aphaca, Viciopsis, Linearicarpus and Nissolia distributed in Central Anatolia, using LM and SEM, and to evaluate the usefulness of the palynological data in the classification within the genus.

Results

Pollen grains of 21 *Lathyrus* taxa, placed in 9 sections *Orobus, Lathyrostylis, Lathyrus, Orobon, Pratensis, Aphaca, Viciopsis, Linearicarpus* and *Nissolia* from Central Anatolia ,Turkey show variation in size, shape and exine ornamentation. The main pollen characters of the taxa are summarized in Table 2. SEM micrographs of representative pollen grains studied are illustrated in Figs. 1-5.

Pollen size and shape

The pollen grains of the taxa examined vary in size: polar axis (P) ranges from 28.67 µm, in L. inconspicuus var. inconspicuus, to 54.32 µm, in L. tukhtensis and equatorial axis (E) ranges from 20.17 µm, in L. aphaca var. biflorus, to 52.58 µm, in L. tukhtensis (Table 2). Pollen shape varies from spheroidal to prolate (P/E = 1.00-1.57), mostly frequent subprolate in L. aureus, L. brachypterus, L. digitatus, L. tuberosus, L. chloranthus, L. roseus subsp. roseus, L. pratensis, L. laxiflorus subsp. laxiflorus, L. czeczottianus, L. saxatilis, and L. sphaericus) (Figs. 1a; 2a, g; 3a, g, m; 4a, d, g; 5a, d). Sometimes grain forms coexist between subprolate and prolate in L. czeczottianus or between spheroidal and subprolate in L. incurvus and L. tuberosus (Table 2). According to the equatorial view, the pollen grains can be divided into two groups based on their equatorial view: 1. Quadratic-obtuse-emarginate pollen group in L. tukhtensis, L. cilicicus and L. inconspicuus var. inconspicuus (Table 2; Figs. 2j, m; 5g); 2. Elliptical to rectangular-obtuse-convex pollen group in the rest of the taxa (Table 2; Figs. 1a, d; 2a, d,

g; 3a, d, g, i, m; 4a, d, g, j, m; 5a, d, j). Moreover, the pollen grains in polar view vary from circular as in *L. incurvus*, *L. chloranthus*, *L. cicera*, *L. laxiflorus* subsp. *laxiflorus*, *L. sphaericus* (Figs. 1e; 3h, j; 4e; 5e); triangular as in *L. hirsutus*, *L. pratensis*, *L. inconspicuus* var. *inconspicuous* (Figs. 3e; 4b; 5h) to penta/hexangular-obtuse-convex as in *L. aureus*, *L. brachypterus*, *L. armenus*, *L. digitatus*, *L. tukhtensis*, *L. cilicicus*, *L. tuberosus and L. nissolia* Figs. 1b; 2b, e, h, k, n; 3b; 5k). They sometimes forms coexist triangular to circular in several taxa, such as *L. aphaca* var. *biflorus* and *L. aphaca* var. *affinis* (Fig. 4k, n).

Apertures

The pollen grains are 3-zonocolporate. In the ectoaperture, the colpi are usually long, narrow, sunken (widely sunken in *L. inconspicuus* var. *inconspicuus*), nearly reaching at the poles, straight, widened from mesocolpium to poles, with acute ends. The length of colpus (Clg) ranges from 22.87 μ m, in *L. laxiflorus* subsp. *laxiflorus*, to 42.15 μ m, in *L. brachypterus* (Table 2). In the endoaperture, the porus is large, circular or elliptic, protruding in mesocolpium or not. The length of porus (Plg) ranges from 4.02 μ m, in *L. hirsutus*, to 19.67 μ m, in *L. cilicicus*. The width of porus (Plt) varies from 5.67 μ m, in *L. tuberosus*, to 18.98 μ m, in *L. cilicicus* (Table 2).

Exine ornamentation

Five different types of surface ornamentation are distinguished at the mesocopium: (1) perforate, (2) finely reticulate-perforate (the most common with 15 taxa), (3) obscurely reticulate-perforate, (4) reticulate-granulate and (5) granulate-perforate (Table 2).

Type 1. Perforate

Observed in L. tukhtensis and L. cilicicus (Fig. 2l, o).

Type 2. Finely reticulate-perforate

Observed in L. aureus, L. digitatus, L. tuberosus, L. chloranthus, L. roseus subsp. roseus, L. pratensis, L. laxiflorus subsp. laxiflorus, L. czeczottianus, L. aphaca var. biflorus, L. aphaca var. affinis, L. saxatilis, L. sphaericus and L. nissolia (Figs. 1c; 2i; 3c, o; 4c, f, i, 1, o; 5b, c, f, 1).

Type 3. Obscurely reticulate-perforate

Observed in L. incurvus, L. brachypterus, L. armeus and L. hirsutus (Figs. 1f, 2c, f; 3f).

Type 4. Reticulate-granulate

Observed only in L. cicera (Fig. 3k, l).

Type 5. perforate-granulate

Observed in L. inconspicuus var. inconspicuus (Fig. 5i).

The surface of apocolpium and colpus are psilate or perforate in all taxa except *L. chloranthus* exhibits the finely reticulateperforate pattern (Fig. 3h). While a sunken apocolpium is recognized only in *L. tukhtensis*, *L. cilicicus* and *L. inconspicuus* var. *inconspicuous*, a widely sunken colpus is observed only in *L. inconspicuous* var. *inconspicuous*.

Table 1. Collection data of *Lathyrus* specimens examined here from pollen morphological point of view. Taxa endemic to Turkey are indicated by an asterix (*).

| Таха | Collection data |
|-----------------------------------|---|
| L. aphaca var. biflorus | B4 Konya: Sultanhani to Karapinar, 37°44′467″N–33°33′353″E, 994 m, 22.5.2010, Çildir 123. |
| L. aphaca var. affinis | A4 Ankara: Kalecik, Kızılırmak riverbank, 40°05′018″N–33°27′088″ E, 638 m, 20.6.2009, <i>Çildir 118</i> . |
| L. armenus | A6 Sivas: Zara to İmranlı, 39°52'985"N-38°03'701"E, 1581 m, 22.6.2010, Cildir 168; near Cumhuriyet |
| | University, 1247 m, 18.7.2009, Güneş 2492. |
| L. aureus | A4 Ankara: Çubuk, Karagöl, 40°24'775"N-32°54'706"E, 1512 m, 30.5.2010, <i>Çildir 132</i> . |
| L. brachypterus* | A4 Ankara: Beypazarı to Eğriova, 40°17'960'/N-31°58'724''E, 1534 m, 28.6.2009, Cildir 112. B5 |
| | Aksaray: Hasan Mountain-Helvadere, 1360 m, 9.7.2008, Güneş 1961. |
| L. chloranthus | C4 Konya: Susuz plain, Beyşehir Sultanpınarı, 1097 m. 11. 6.2009, Güneş 2320. |
| L. cicera | B5 Aksaray: Hasan Mountain, 38°10′600″N–34°10′920″E, 1510 m, 23.5.2010, <i>Cildir 125</i> . C4 Konya: |
| | Hadim-Taşkent road 3 km, 1161 m. 11.6.2009, Güneş 2316. |
| L. cilicicus* | C4 Karaman: above Ermenek, 36°37′59.76″N-32°54′33.90″E, 1350 m, 27.6.2009, Celep 1714. |
| L. czeczottianus* | A4 Kastamonu : Ilgaz-Kastamonu ex-road 42. km, 1450 m, 17.7.2009, Güneş 2475. B5 Kayseri: Erciyes |
| | Mountain, 38°36′907″N–35°30′840″E, 1780 m, 19.6.2010, <i>Çildir 157</i> . |
| L. digitatus | B3 Eskişehir: Mihalıççık, 39°53′588″N–31°27′731″E, 1435 m, 16.6.2010, Çildir 146. |
| L. hirsutus | A4 Ankara: METU, near Biology department, 39º53'461"N-32º46'488"E, 850 m, 6.5.2010, Çildir |
| | <i>133a.</i> |
| L. inconspicuus var. inconspicuus | B5 Yozgat: Sorgun to Alaca, 39°54′247″N-34°56′220″E, 1322 m, 24.6.2010, <i>Çildir 177</i> . |
| L. incurvus | A4 Ankara: near Kalecik, 40°05′018″N–33°27′088″E, 638 m, 20.6.2009, <i>Çildir 118</i> . |
| L. laxiflorus subsp. laxiflorus | B3 Eskişehir: Yukarı Kalabak village, 39°32'617"N-30°17'614"E, 1127 m, 27.6.2009, Çildir 108. C4 |
| | Konya: Hadim-Taşkent road 3 km, 1161 m. 11.6.2009, <i>Güneş 2313</i> . |
| L. nissolia | A4 Ankara: Kıcılcahamam, Soğuksu National Park, 40°26'442"N-32°37'272"E, 1392 m, 5.6.2010, |
| | <i>Çildir 141</i> . C4 Konya: Doğanhisar, Güvenlik village, 1456 m. 12.6.2009, <i>Güneş 2324</i> . |
| L. pratensis | B5 Yozgat: Akdağmadeni to Şarkışla, 39°36'980''N–35°56'425''E, 1586 m, 23.6.2010, Çildir 173. |
| L. roseus subsp. roseus | B6 Sivas: Zara to Şerefiye, 39°58'281''N-37°43'434''E, 1639 m, 22.6.2010, Çildir 162a. |
| L. saxatilis | B4 Ankara: near Lalahan, 39°58'043"N-33°07'256"E, 1084 m, 6.5.2009, <i>Çildir 121</i> . |
| L. sphaericus | C4 Konya: Akşehir, Çakıllı village, 1262 m, 12.6.2009, Güneş 2332. |
| L. tuberosus | A4 Ankara: Çubuk, Karagöl, 40°24′775″N–32°54′706″E, 1512 m, 30.5.2010, <i>Çildir 132a</i> . C4 Konya: |
| | Susuz plain, Beyşehir Sultanpınarı, 1097 m. 11.6.2009, Güneş 2321. |
| L. tukhtensis* | A4 Çankırı: Çankırı to Eldivan, 40°29'883"N-32°30'248"E, 1216 m, 27.6.2010, Çildir 179a; Yapraklı |
| | Mountain, 1626 m, 17.7.2009, Güneş 2487. |
| | |



Fig 1. SEM micrographs of pollen grains of the section *Orobus. L. aureus* (**a**) equatorial view, (**b**) polar view, (**c**) ornamentation; *L. incurvus* (**d**) equatorial view, (**e**) polar view, (**f**) ornamentation. Scale bars: (c) $1 \mu m$; (f) $10 \mu m$.

Discussion

Tosheva and Tonkov (2005, 2007), Güneş and Aytuğ (2010), Güneş and Çırpıcı (2010) and Güneş (2011a, b) pointed out that pollen morphological characters were valuable for distinguishing the species in *Lathyrus*. The present study is in agreement with this point of view. The results indicate that the surface sculpturing type of the pollen grains, their size, shape and outline in equatorial view can be of taxanomic value in the separation of the studied taxa. However, these characters are shown to be useless in sectional delimitation of the genus.

Systematic usefulness of pollen size

The largest pollen grains (P = $48.08\pm1.81 \ \mu m \ x \ E = 46.44\pm1.89 \ \mu m$) are found in *L. tukhtensis* from sect. *Lathyrostylis* while the smallest pollen grains (P = $32.98\pm2.15 \ \mu m \ x \ E = 24.18\pm2.02 \ \mu m$) occur in *L. nissolia* from sect. *Nissolia* (Table 2). Most of the taxa examined by us had more or less similar pollen morphologies to those examined by a few researchers (Tosheva and Tonkov, 2005, 2007; Güneş and Aytuğ, 2010; Güneş and Çırpıcı, 2010; Güneş, 2011a, b), except for the differences in size and ratio of the polar axis to equatorial diameter. The values are a little different from those given in this our study.

| Sections | Taxa | Р | Е | P/E | Ex | Clg | Plg | Plt | Outline in | in Ornamentation | |
|---|----------------------|--------------------|--------------------|--------------|-------------------|--------------------|--------------------|--------------------|-----------------|---------------------|--------------|
| | | | | | | 8 | 8 | | equatorial view | in the mesocolpium | |
| Orobus | L. aureus | (47.28-41.23) | (40.48-36.20) | 1.19 | (1.09-0.82) | (32.89-28.43) | (8.33-5.21) | (7.89-6.08) | Eroc | Finely | reticulate- |
| | | (44.77 ± 2.29) | (37.78 ± 1.41) | Sp | (0.92 ± 0.17) | (30.69 ± 1.33) | (7.72 ± 0.63) | (6.96 ± 0.98) | | perforate | |
| | L. incurvus | (40.33-30.55) | (34.26-29.91) | 1.10 | (1.35-0.89) | (31.02-26.80) | (9.92-7.12) | (9.11-6.14) | Eroc | Obscurely | reticulate- |
| | | (35.12 ± 1.16) | (31.90 ± 1.13) | $S(\pm Sp)$ | (1.12 ± 0.23) | (29.48 ± 2.02) | (8.65 ± 0.9) | (8.38 ± 1.02) | | perforate | |
| Lathvrostylis | L. brachypterus | (48.05-40.29) | (38.66-32.70) | 1.18 | (1.25-0.98) | (42.15-38.16) | (13.18-8.77) | (18.12-13.78) | Eroc | Obscurely | reticulate- |
| Landyroolyns | 21 or denyprer do | (43.63 ± 2.23) | (36.97 ± 2.61) | Sp | (1.05 ± 0.16) | (40.24 ± 1.46) | (12.76 ± 2.01) | (16.88 ± 1.21) | 2100 | perforate | Terretailate |
| | | (1010012120) | (001)/==101) | SP | (1100=0110) | (1012121110) | (120022001) | (1010021121) | | periorate | |
| | L. armenus | (39.42-34.54) | (36.31-31.17) | 1.11 | (1.29-0.86) | (29.78 - 24.45) | (13.80-9.19) | (16.23-12.56) | Eroc | Obscurely | reticulate- |
| | | (36.76±2.05) | (33.17±2.24) | S | (0.98 ± 0.13) | (27.65±2.33) | (10.05 ± 1.23) | (14.57 ± 1.26) | | perforate | |
| | L. digitatus | (45.56-40.45) | (33.45-26.89) | 1.32 | (1.03-0.87) | (30.67-25.35) | (9.28-5.67) | (12.65-8.78) | Eroc | Finely | reticulate- |
| | 8 | (42.54 ± 1.13) | (32.23 ± 1.43) | Sp | (0.93 ± 0.14) | (27.24 ± 2.63) | (7.89 ± 1.76) | (10.09 ± 1.31) | | perforate | |
| | L. tukhtensis | (54.32-46.15) | (52.58-44.12) | 1.04 | (1.43-0.85) | (34.65-28.48) | (14.32 - 10.43) | (18.01-13.99) | Rom | Perforate | |
| | | (48.08 ± 1.81) | (46.44 ± 1.89) | S | (1.01 ± 0.14) | (31.19 ± 2.25) | (12.45 ± 2.16) | (16.71 ± 1.45) | | | |
| | L. cilicicus | (44.89-40.67) | (45.98-39.67) | 1.01 | (1.78-1.02) | (41.67-39.98) | (19.67-15.45) | (18.98-14.16) | Rom | Perforate | |
| | | (42.19 ± 1.69) | (41.89±1.46) | S | (1.12 ± 0.16) | (40.19±1.35) | (17.47±1.81) | (16.33±1.31) | | | |
| Lathvrus | L. tuberosus | (48.86-40.15) | (37.95-30.21) | 1.14 | (1.04-0.87) | (35.65-30.46) | (8.67-4.56) | (9.74-5.67) | Eroc | Finelv | reticulate- |
| | | (42.08 ± 2.49) | (36.89±1.80) | S(±Sp) | (0.98 ± 0.2) | (33.16±2.78) | (6.78±1.08) | (7.06±1.08) | | perforate | |
| | L. hirsutus | (45.36-38.35) | (30.52-23.09) | 1.39 | (1.32-0.79) | (40.12-36.52) | (9.49-4.02) | (10.09-6.21) | Eroc | Obscurely | reticulate- |
| | | (40.87 ± 2.56) | (29.34 ± 2.19) | Pr | (0.95 ± 0.2) | (38.28 ± 2.87) | (7.45 ± 1.43) | (7.96 ± 1.21) | | perforate | |
| | L. chloranthus | (50.12-44.21) | (41.90-36.01) | 1.26 | (1.11-0.79) | (37.48-32.82) | (11.04-7.87) | (12.29-9.21) | Eroc | Finely | reticulate- |
| | | (46.82 ± 1.96) | (37.16 ± 2.41) | Sp | (0.94 ± 0.15) | (34.76 ± 1.98) | (9.81 ± 1.46) | (10.86±0.99) | | perforate | |
| | L. cicera | (48.75-46.20) | (36.92-25.91) | 1.38 | (1.81-0.98) | (40.55 - 34.11) | (11.95-9.61) | (11.73-8.54) | Eroc | Reticulate-g | anulate |
| | | (47.32 ± 1.19) | (34.36 ± 2.03) | Pr | (1.21 ± 0.15) | (37.91 ± 3.27) | (10.86 ± 1.24) | (9.19 ± 1.31) | | 0 | |
| Orobon | L. roseus subsp. | (46.76-41.90) | (35.16-31.25) | 1.30 | (1.46-0.98) | (37.41-33.24) | (13.68-9.87) | (12.14-8.41) | Eroc | Finelv | reticulate- |
| | roseus | (43.67 ± 2.14) | (33.64 ± 1.73) | Sp | (1.05 ± 0.17) | (35.55 ± 1.18) | (10.08 ± 1.32) | (9.61 ± 1.21) | | perforate | |
| Pratensis | L. pratensis | (40.67-34.56) | (32.67-26.45) | 1.23 | (1.34-0.78) | (30.65-24.56) | (9.74-6.06) | (12.34-8.89) | Eroc | Finely | reticulate- |
| | 1 | (37.13 ± 2.19) | (30.28 ± 1.66) | Sp | (1.01 ± 0.18) | (27.29 ± 2.79) | (8.91 ± 0.64) | (9.01 ± 0.99) | | perforate | |
| | L. laxiflorus | (42.89-36.24) | (33.76-27.34) | 1.25 | (1.46-0.78) | (27.72-22.87) | (10.34 - 7.89) | (12.43-8.02) | Eroc | Finely | reticulate- |
| | subsp. laxiflorus | (38.25 ± 1.78) | (30.58 ± 2.13) | Sp | (0.94 ± 0.14) | (23.14 ± 2.34) | (8.34 ± 1.12) | (9.21 ± 0.89) | | perforate | |
| | L. czeczottianus | (45.67-38.98) | (34.27-29.54) | 1.33 | (1.05-0.78) | (30.67-25.88) | (11.68-7.89) | (12.19-7.03) | Eroc | Finely | reticulate- |
| | | (42.79±1.87) | (32.23 ± 2.09) | $Pr(\pm Sp)$ | (0.89 ± 0.15) | (27.23 ± 2.13) | (9.04±1.65) | (8.58±0.92) | | perforate | |
| Aphaca | L. aphaca | (54.06-44.42) | (33.05-20.17) | 1.57 | (1.01-0.78) | (37.86-34.15) | (9.67-4.89) | (10.31-7.67) | Eroc | Finely | reticulate- |
| 1 | var. biflorus | (46.44±3.67) | (29.54 ± 3.12) | Pr | (0.92 ± 0.19) | (35.78±1.93) | (6.05 ± 1.54) | (9.56±1.35) | | perforate | |
| | L. aphaca | (53.15-45.32) | (34.15-28.67) | 1.51 | (1.02-0.82) | (35.98-30.31) | (8.92±5.43) | (11.73-8.89) | Eroc | Finely | reticulate- |
| | var. affinis | (46.64±3.89) | (30.81±3.08) | Pr | (0.95 ± 0.21) | (32.37±2.01) | (6.16±1.31) | (9.98 ± 1.42) | | perforate | |
| Viciopsis | L. saxatilis | (40.67-37.21) | (33.01-25.45) | 1.24 | (1.15-0.98) | (36.34-29.45) | (9.98-4.56) | (10.86-8.35) | Eroc | Finely | reticulate- |
| , i i i i i i i i i i i i i i i i i i i | | (38.89 ± 1.57) | (31.50 ± 1.42) | Sp | (1.02 ± 0.3) | (34.61 ± 2.12) | (6.45 ± 1.76) | (7.76 ± 1.05) | | perforate | |
| Linearicarpus | L. sphaericus | (47.77-43.27) | (42.32-35.46) | 1.16 | (0.98 - 0.67) | (35.78-29.32) | (10.23-6.12) | (13.02-8.31) | Eroc | Finely | reticulate- |
| I I I I I I I I I I I I I I I I I I I | I III | (44.81 ± 2.07) | (38.49 ± 2.47) | Sp | (0.91 ± 0.1) | (33.21 ± 2.32) | (8.32 ± 2.65) | (10.11 ± 1.45) | | perforate | |
| | L. inconspicuus var. | (34.28-28.67) | (35.79-30.78) | 1.00 | (1.34-0.93) | (28.54 - 26.58) | (9.27 - 7.23) | (11.84 - 8.09) | Rom | Perforate-granulate | |
| | inconspicuus | (33.57 ± 2.02) | (33.48 ± 1.93) | S | (1.07 ± 0.14) | (27.78 ± 0.96) | (8.25±0.76) | (9.02 ± 1.18) | | Bri | |
| Nissolia | L. nissolia | (36.81-30.32) | (21.14 - 28.97) | 1.36 | (1.11-0.78) | (31.67-25.87) | (11.32-5.34) | (13.07-8.26) | Eroc | Finelv | reticulate- |
| | | (32.98 ± 2.15) | (24.18 ± 2.02) | Pr | (0.96 ± 0.2) | (27.89 ± 2.42) | (8.91 ± 1.14) | (10.18 ± 1.63) | | perforate | |
| | | (| (| | (| (==) | () | (| | 1 | |

Table 2. Summary of pollen morphological data for the taxa studied of *Lathyrus* according to the infrageneric system of Kupicha (1983). All sizes are in µm. Numbers refer to (maximum-minimum) (mean±standard deviation).

Clg = Colpus length, E = equatorial diameter, Eroc = Elliptical to rectangular-obtuse-convex, Ex = Exine thickness, P = Polar diameter, Plg = Porus length regarding the poles, Plt = Porus width regarding the equatorial diameter, Pr = Prolate, Rom=Rectangular-obtuse-emarginate, S = Spheroidal, Sp = Subprolate, ± = pollen shape is rarely present.



Fig 2. SEM micrographs of pollen grains of the section *Lathyrostylis. L. brachypterus* (a) equatorial view, (b) polar view, (c) ornamentation; *L. armenus* (d) equatorial view, (e) polar view, (f) ornamentation; *L. digitatus* (g) equatorial view, (h) polar view, (i) ornamentation; *L. tukhtensis* (j) equatorial view, (k) polar view, (l) ornamentation; *L. cilicicus* (m) equatorial view, (n) polar view, (o) ornamentation. Scale bars: (a, b, d, e, g, h, j, k, m, n) 10 μ m; (c, f, i) 1 μ m; (l, o) 5 μ m.

These minor differences may result from variations in preparation treatments. In the pollen size variability is major and, consequently, the parameters for polar axes and equatorial diameters have a diagnostic value between some of the taxa within the same section, for example, between L. aureus and L. incurvus, between L. digitatus and L. tukhtensis, between L. armenus and L. cilicicus, and between L. sphaericus and L. inconspicuus var. inconspicuus. Based on pollen size and the other examined pollen characters, however, it is impossible to find any important differences between L. aphaca var. biflorus and L. aphaca var. affinis belonging to sect. Aphaca and between L. pratensis and L. laxiflorus subsp. laxiflorus belonging to sect. Pratensis. The sections Nissolia, Orobon and Viciopsis are monotypic (Kupicha, 1983; Asmussen and Liston, 1998; Kenicer et al., 2005). The section Nissolia can be distinguished by its smallest pollen grains (P = $32.98\pm2.15 \ \mu m \ x \ E = 24.18\pm2.02$ μ m) from the other sections studied here. This agrees with the results obtained from previous studies (Moore et al., 1991; Perveen and Qaiser, 1998; Halbritter, 2000; Mantar et al., 2001, 2003; Tosheva and Tonkov, 2005, 2007; Güneş and Aytuğ, 2010; Güneş and Çırpıcı, 2010; Güneş, 2011a, b). However, there appear to be no important pollen morphological differences in distinguishing the sections *Orobon* and *Viciopsis* from the others.

Systematic usefulness of pollen shape

The shape of the pollen grains varies among some of the taxa, even within the same taxon. However, L. armenus, L. tukhtensis, L. cilicicus and L. inconspicuus var. inconspicuus have always spheroidal pollen grains and L. hirsutus, L. cicera, L. aphaca var. biflorus, L. aphaca var. affinis and L. nissolia have always prolate pollen grains. Moore et al. (1991) described two types for Lathyrus pollen: the Vicia cracca type and the Lathyrus type. Beug (2004) presented general description of the pollen morphological characters of taxa assigned to the Lathyrus type. This pollen type was characterized by being 3-zonocolporate, prolate (P/E=1.23-1.91) and medium in size (30-50 µm), with suprareticulate ornamentation and lumina reaching up to 2.5-3.0 µm in diameter, unlike the Vicia type, where lumina is more than 3.0 µm in diameter. The pollen grains were almost psilate at apocolpium and around the apertures and sometimes the reticulum was not well defined. Each aperture was composed of well-defined circular or elliptic endopori and ectocolpi with heavy thick costae. The exine was 1.0-1.5 µm thick and thinner at apocolpium and the endexine was thicker than the tectum. In L. filiformis, L. pallescens and L. pancicii belong to sect. Lathyrostylis, Tosheva and Tonkov (2007) described the Lathyrus filiformis type as a third pollen type. In this type, the pollen had a quadratic (rectangular) outline in its equatorial view. Of the Lathyrus taxa examined to date (Faegri, 1956; Gapotchka and Chamara, 1972; Gapotchka, 1974; Clarke and Kupicha, 1976; Fergusson and Skvarla, 1981; Faegri and Iversen, 1989; Moore et al., 1991; Perveen and Qaiser, 1998; Halbritter, 2000; Mantar et al., 2001, 2003; Beug, 2004; Tosheva and Tonkov, 2005, 2007; Güneş and Aytuğ, 2010; Güneş and Çırpıcı, 2010; Güneş, 2011a, b), the Lathyrus filiformis pollen type was found only in sect. Lathyrostylis. Confirming with previous findings our study also shows that this pollen type is found in the pollen grains of L. tukhtensis and L. cilicicus. Additionally, the Lathyrus *filiformis* type is first identified in the pollen grains of L. inconspicuus var. inconspicuus from sect. Linearicarpus. The other studied taxa have elliptical to rectangular-obtuseconvex pollen grains.

Systematic usefulness of exine ornamentation

The exine ornamentation at the mesocopium proved to be a valuable taxonomic character in *Lathyrus*. It varies from perforate, finely reticulate-perforate, obscurely reticulate-perforate, reticulate-granulate to granulate-perforate. The type of the exine ornamentation is variable within the sections *Orobus*, *Lathyrostylis*, *Lathyrus* and *Linearicarpus*. However, it is constant within the other sections. The finely reticulate-perforate ornamentation is the most frequent type among the examined taxa. The perforate ornamentation is found only in *L. tukhtensis* and *L. cilicicus* (sect. *Lathyrostylis*) whereas the obscurely reticulate-perforate ornamentation is present in *L. incurvus* (sect. Orobus), *L. brachypterus*, *L. armeus* (sect. *Lathyrostylis*) and *L. hirsutus* (sect. *Lathyrus*). *L. cicera* and *L. inconspicuus* var.



Fig 3. SEM micrographs of pollen grains of the sections *Lathyrus* and *Orobon*. Sect. *Lathyrus*: *L. tuberosus* (a) equatorial view, (b) polar view, (c) ornamentation; *L. hirsutus* (d) equatorial view, (e) polar view, (f) ornamentation; *L. chloranthus* (g) equatorial view, (h) polar view; *L. cicera* (i) equatorial view, (j) polar view, (k-I) ornamentation. Sect. *Orobon: L. roseus* subsp. *roseus* (m) equatorial view, (n) polar view, (o) ornamentation. Scale bars: (a, b, d, e, g, h-j, m, n) 10 μ m; (c, k, o) 1 μ m; (f, l) 5 μ m.

inconspicuus each exhibit distinct pollen ornamentation types which could not be confused with the pollen of the other taxa studied. *L. cicera* and *L. inconspicuus* var. *inconspicuus* are well characterized by the reticulate-granulate and granulate-perforate ornamentation, respectively. The surface of the apocolpium and colpus area are psilate or perforate in all taxa except *L. chloranthus* show a distinct pollen ornamentation, differing from all other taxa investigated by its the finely reticulate-perforate pattern at the apocolpium. In conclusion, our study on the *Lathyrus* taxa distributed in Central Anatolia indicates that several pollen morphological characters can be utilized as an important tool to distinguish the taxa studied.



Fig 4. SEM micrographs of pollen grains of the sections *Pratensis* and *Aphaca*. Sect. *Pratensis*: *L. pratensis* (a) equatorial view, (b) polar view, (c) ornamentation; *L. laxiflorus* subsp. *laxiflorus* (d) equatorial view, (e) polar view, (f) ornamentation; *L. czeczottianus* (g) equatorial view, (h) polar view, (i) ornamentation. Sect. *Aphaca: L. aphaca* var. *biflorus* (j) equatorial view, (k) polar view, (l) ornamentation. Scale bars: (a, b, d, e, g, h, j, k, m, n) 10 μ m; (c, f, i, l, o) 5 μ m.

though pollen morphology do not provide strong evidence for comparisons at infrageneric level in *Lathyrus*

Material and methods

Plant material

21 taxa of the genus *Lathyrus* were collected from their natural habitats in Central Anatolia, Turkey. The collected specimens were deposited in the herbarium of Department of Biological Sciences, Middle East Technical University (METU), Ankara and Department of Biology, Kafkas University, Kars. The voucher specimens are listed in Table 1.



Fig 5. SEM micrographs of pollen grains of *L. saxatilis* in the sections *Viciopsis*, *Linearicarpus* and *Nissolia*. Sect. *Viciopsis*: *L. saxatilis* (a) equatorial view, (b-c) ornamentation. Sect. *Linearicarpus*: *L. sphaericus* (d) equatorial view, (e) polar view, (f) ornamentation; *L. inconspicuus* var. *inconspicuus* (g) equatorial view, (h) polar view, (i) ornamentation. Sect. *Nissolia*: *L. nissolia* (j) equatorial view, (k) polar view, (l) ornamentation Scale bars: (a, d-h, j, k) 10 µm; (b, i) 5 µm; (c, l) 1 µm.

Light microscopy (LM)

Pollen grains were first treated with 70% alcohol to remove oily substances and then embedded in glycerine jelly stained with basic fuchsin following the method of Wodehouse (1935). Polar axis (P), equatorial axis (E), colpus length (Clg), exine thickness (Ex), porus length (Plg) and porus width (Plt) were measured from 30 fully developed grains per sample under a Leica DMLB2 microscope (1000×). Results are provided as minimum, maximum and mean±standard deviations. P/E ratios were also calculated.

Scanning electron microscopy (SEM)

Pollen grains were transferred directly to stubs with doublesided adhesive tape and micrographs were obtained using Jeol JSM-6400 SEM and Pei Quanta 400F SEM to determine their ornamentation (Doğan, 1988). The terminology follows that of Punt et al. (2007) and Özler et al. (2011).

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References

- Asmussen C, Liston A (1998) Chloroplast DNA characters, phylogeny and classification of *Lathyrus* (Fabaceae). Am J Bot 85: 387-401.
- Badr A, EL Shazly H, El Rabey H, Watson LE (2002) Systematic relationships in *Lathyrus* sect. *Lathyrus* (Fabaceae) based on amplified fragment length polymorphism (AFLP) data. Can J Bot 80: 962-969.
- Beug H (2004) Leitfaden der Pollenbestimmung fur Mitteleuropa und angrenzende Gebiete. Göttingen.
- Chittenden FG (1951) The Royal Horticulural Society Dictionary of Gardening 3: 1132-1134. Oxford: Clarendon Press.
- Clarke G, Kupicha F (1976) The relationships of the genus *Cicer* L. (Leguminosae): the evidence from pollen morphology. Bot J Linn Soc 72: 35-44.
- Croft AM, Pang ECK, Taylor PWJ (1999) Molecular analysis of *Lathyrus sativus* L. (grasspea) and related *Lathyrus* species. Euphytica 107: 167-176.
- Czefranova ZV (1971) Review of the species of the genus *Lens* Mill. Nov Syst Pl Vas 8: 197-204.
- Çildir H (2011) Morphology, anatomy and systematics of the genus *Lathyrus* L. (Leguminosae) in Central Anatolia, Turkey. Ph.D. Thesis. Departmet of Biology, Middle East Technical University, Ankara, Turkey.
- Davis PH (1970) *Lathyrus* L. In: Davis PH (ed.), Flora of Turkey and the East Aegean Islands, vol. 3, 328-369. Edinburgh University Press, Edinburgh.
- Davis PH, Mill RR, Kit T (1988) Flora of Turkey and East Aegean Islands, vol. 10. Edinburgh University Press, Edinburg
- Doğan M (1988) A scanning electron microscope survey of the lemma in *Phleum, Pseudophleum* and *Rhizocephalus* (Gramineae). Notes Roy Bot Gard Edinb 45(1): 117-124.
- Doğan M, Kence A, Tigin C (1992) Numerical taxonomic study on Turkish *Lathyrus* (Leguminosae). Edinb J Bot 49: 333-341.
- Faegri K (1956) Palynological studies in NW European Papilionaceae. Bot. Mus. Bergen.
- Faegri K, Iversen J (1989) Textbook of pollen analysis. Blackwell, Chichester.
- Fedchenko BA (1948) *Lathyrus*. In: Komarov VL, Shishkin BK, Bobrov EG, Flora of the USSR, vol. 13, 363-395. Leguminosae: *Oxytropis*, *Hedysarum*. Leningrad: Izdatel'stvo Akademii Nauk SSSR, Mosqua.
- Fergusson K, Skvarla J (1981) The pollen morphology of the subfamily Papilionoideae (Leguminosae). In: Polhill R, Raven P (eds.), Advances in Legume Systematics, vol. 2, 859-896. Royal Botanic Gardens, Kew.
- Gapotchka GP (1974) On the palynomorphology of the species of the tribe Vicieae from the family Fabaceae. Vest. Mosk. Univ. ser. 6 Biol. 29: 93-98.
- Gapotchka GP, Chamara LP (1972) The development of sporoderma in *Lathyrus niger* (L.). Bernh. Vest. Mosk. Univ. ser. 6 Biol. 27: 50-54.

- Genç H, Şahin A (2011) A new species of *Lathyrus* L. (Fabaceae) from Turkey. J Syst Bot. 49(5): 505-508.
- Güneş F, Özhatay N (2000) Lathyrus L. In: Güner A, Özhatay N, Ekim T, Başer KHC (eds.), Flora of Turkey and the East Aegean Islands, vol. 11, 92-94. Edinburgh University Press, Edinburgh.
- Güneş F, Aytuğ B (2010) Pollen morphology of the genus *Lathyrus* (Fabaceae) section *Pratensis* in Turkey. Int J Agric Bio 12: 96-100.
- Güneş F, Çırpıcı A (2010) Pollen morphology of the genus *Lathyrus* (Fabaceae) section *Cicercula* in Thrace (European Turkey). Acta Bot Croat 69: 83-92.
- Güneş F (2011a) The pollen morphology of Turkey species from *Lathyrus* section *Platystylis* (Sweet) Bässler (Fabaceae). Plant Syst Evol 293: 75-90.
- Güneş F (2011b) The Pollen morphology of some *Lathyrus* L. (Fabaceae) taxa from Turkey. Int J Agric Bio 13(3): 301-308.
- Halbritter H (2000) *Lathyrus latifolius, Lathyrus tuberosus.* In: Buchner R, Weber M (eds.), PalDat–a palynological database: Descriptions, illustrations, identification, and information retrieval.

http://www.paldat.botanik.univie.ac.at/

- ILDIS (2002). International Legume Database and Information Service, World database of legumes, version 6.05. Southampton, UK.
- ILDIS (2010). International Legume Database and Information Service. http://www.ildis.org/
- Kenicer G, Kajita T, Pennington R, Murata J (2005) Systematics and biogeography of *Lathyrus* (Leguminosae) based on internal transcribed spacer and cpDNA sequence data. Am J Bot 92: 1199-1209.
- Kenicer G (2008) An introduction to the genus *Lathyrus* L. Curtis's Bot Mag 25(4): 286-295.
- Kupicha FK (1981) Vicieae. In: Polhill, RM, Raven, PM (eds.), Advances in Legume Systematics, pp. 377-381. Royal Botanic Gardens, Kew.
- Kupicha FK (1983) The infrageneric structure of *Lathyrus*. Notes Roy Bot Gard Edinb 41: 209-244.
- Leht M (2009) Phylogeny of Old World *Lathyrus* L. (Fabaceae) based on morphological data. Feddes Repert 120(1-2): 59-74.
- Lewis GM, Schrire B, Mackinder B, Lock M (2005) Legumes of the world, Kew Press, London.

- Mantar N, Şahin A, Bağcı E, Çobanoğlu D (2001) Lathyrus annuus L. ve L. cicera L. (Fabaceae) nin Anatomisi, Morfolojisi ve Palinolojisi Üzerinde Bulgular. Fırat Üniversitesi Fen ve Müh. Bil. Der. 13(2): 67-78.
- Mantar N, Bağcı E, Şahin A, Gür N (2003) *Lathyrus sativus* L. ve *L. hirsutus* L. (Fabaceae / Leguminosae) Türleri Üzerinde Morfolojik, Palinolojik ve Anatomik Bir Çalışma. Fırat Üniversitesi Fen ve Müh. Bil. Der. 15(3): 303-314.
- Meikle RD (1977) Flora of Cyprus, vol. 1, 566-578. Bentham-Moxon Trust, Royal Botanic Gardens, Kew.
- Moore PD, Webb JA, Collinson M (1991) Pollen Analysis (second edition). Blackwell Sci. Publication, London.
- Özler H, Pehlivan S, Kahraman A, Doğan M, Celep F, Başer B, Yavru A, Bagherpour S (2011) Pollen morphology of the genus *Salvia* L. (Lamiae) in Turkey. Flora 206: 316-327.
- Perveen A, Qaiser M (1998) Pollen flora of Pakistan VIII Leguminosae (subfamily: Papilionoideae). Turk J Bot 22: 73-91.
- Punt W, Hoen PP, Blackmore S, Nilsson S, Le Thomas A (2007) Glossary of pollen and spore terminology. Rev Palaeobot Palynol 143: 1-81.
- Rechinger KH (1979) Lathyrus L. In: Chrtkova-Zertova A, van der Maesen LJG, Rechinger KH (eds.), Flora Iranica, no. 140, Papillionaceae, vol. I. Vicieae. Akademische Druck und Verlagsanstalt, Graz.
- Senn HA (1938) Experimental data for the revision of the genus *Lathyrus* Linn. Am J Bot 25: 67-78.
- Simola LK (1986) Structural and chemical aspects of evolution of evolution of *Lathyrus* species. Proceedings of the International Symposium of Ibeas, Paris, pp. 225-239.
- Tosheva A, Tonkov S (2005) Pollen grains morphology of Bulgarian species from the section *Orobus* (L.) Gren. et Godr. (genus *Lathyrus*, Fabaceae). Acta Bot Croat 64: 275-287.
- Tosheva A, Tonkov S (2007) Pollen grains morphology of the Bulgarian species from section *Lathyrostylis* (genus Lathyrus, Fabaceae). Phytol Balcan 13: 393-400.
- Townsend CC, Guest E (1977) Flora of Iraq, 549-573. Baghdad: Ministry of Agriculture and Agrarian Reform of the Republic of Iraq,
- Tutin TG, Heywood VH (1981) Lathyrus L. Flora Europaea, vol. 2, 136-145. Cambridge: Cambridge University Press.
- Wodehouse RP (1935) Pollen Grains. McGraw Hill, New York.