COMPARISON OF MORPHOLOGICAL AND MICROMORPHOLOGICAL STUDIES IN THE GENUS PAPAVER SECT. OXYTONA (PAPAVERACEAE) AND INTERSPECIFIC HYBRIDS

Z. Tavakkoli & M. Assadi

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The leaf epidermal structure of seven taxa from the genus *Papaver* L. sect. *Oxytona* are investigated. All taxa have the type of anomocytic stomata on abaxial surface and no stomata is present on the adaxial surface. Within the section, the species and interspecific hybrids could be identified by the following characters: stomata size, the shape of the epidermal cells and anticlinal walls on leaf abaxial surface. Seed characteristics of the species of this section were studied using scanning electron microscopy and stereomicroscope. Seeds are reniform or sometimes falcate; the epidermal cells shape varies from polygonal and rectangular to irregular polygonal and rectangular; anticlinal walls are nearly straight in *P. bracteatum* and slightly sinuate in *P. orientale* and *P. setiferum*. The sculpturing pattern of testa is granulate-perforate in all taxa. Pollen grains size of seven taxa and the exine structure of *P. bracteatum* and *P. lasiothrix* were studied by SEM. *P. bracteatum* has the smallest pollen grains and *P. setiferum* × *P. bracteatum* the largest pollen. The basic shape of the pollen grains is prolate-spheroidal. The aperture is tri-to tetracolpate. Moreover, taxonomically important characters of taxa were observed and measured. A positive correlation is found between stomata and pollen grains size and chromosome number. Our results revealed that seed morphological characteristics have not taxonomic value in separation of taxa from each other. Also, *P. lasiothrix* could be reduced as synonymy of *P. bracteatum*. Interspecific hybrid of *P. setiferum* × *P. bracteatum* is recorded here for the first time from NW Iran. For these taxa, taxonomical characteristics, localities and geographical distributions are presented.

Zahra Tavakkoli (correspondence <zatavakkoli6@gmail.com>), Dept. of Plant Biology, Faculty of Biological Sciences, Kharazmi Univ. Tehran - Iran. -Mostafa Assadi <assadi@rifr-ac.ir>, Research Institute of Forests and Rangelands, P. O. Box 13185-116, Tehran, Iran.

Key words. *Papaver*, leaf epidermis, seed, pollen grains, morphology, Iran.

مقایسه مطالعات مورفولوژی و میکرومورفولوژی بخشه sect. Oxytona از جنس Papaver و دورگه‌های بین گونه‌ای آنها

زهرا توکلی، عضوهیئت علمی دانشکده علوم زیستی دانشگاه خوارزمی.

مصطفی اسدی، استاد پژوهشی مؤسسه تحقیقات جنگلها و مراتع کشور.

در این مطالعه ویژگیهای مورفولوژیکی و میکرومورفولوژیکی 7 تاکسون از بخشه *Papaver* sect. *Oxytona* با استفاده از استریومیکروسکوپ و میکروسکوپ نوری و الکترونی شرح داده شده و با یکدیگر مقایسه شدند. بر اساس صفات میکرومورفولوژی: اندازه سلولهای روزنه، شکل سلولهای اپیدرمی و دیواره جداکننده سلولها در سطح زیرین برگ، تا حدود زیادی تایید کننده تقسیم‌بندی مورفولوژیک گونه‌ها و دورگه‌های بین گونه‌ای است؛ اندازه دانه‌های گرده می‌تواند به عنوان صفات متمایز کننده گونه‌ای به شمار رود؛ ویژگیهای دانه برای تفکیک گونه‌ها مفید نمی‌باشد. همچنین شرح کوتاهی از ویژگیهای مورفولوژیکی تاکسونهای این بخشه همراه با کلید شناسایی آنها ارایه شده است. بررسیهای انجام شده برای دو گونه *P. bracteatum* و *P. lasiothrix* نشان می‌دهد که این دو گونه تا حدود زیادی مشابه هستند. بنابراین مطابق با نظر Goldblatt گونه *P. lasiothrix* مترادف *گونه P. bracteatum* معرفی می‌گردد. در این مقاله دورگه دو والدی *P. setiferum* × *P. bracteatum* برای اولین بار از ایران گزارش می‌گردد.

INTRODUCTION

Members of *Papaver* section *oxytona* Bernh. are characterized by their perennial habit, poricidal capsule dehisence and dark filament and anthers (Carolan et al. 2006). The section comprises three species of *P. bracteatum* Lindl. , *P. lasiothrix* Fedde and *P. orientale* L. in Iran (Cullen 1966) while Goldblatt (1974) listed three species including *P. bracteatum*: diploid (2n=14); *P. orientale*: tetraploid (2n=28) and *P. pseudo-orientale* (Fedde) Medw.: hexaploid (2n=42) that are found predominatly in the Caucasus mountains, eastern Turkey and north western Iran. Also, *P. lasiothrix* was reduced to the synonymy of *P. bracteatum* (Goldblatt 1974). In addition, two biparental hybrids were reported in Iran by Goldblatt (1974): 1) *P. orientale* × *P. bracteatum* in the Shuran area, 2) *P. oreintale* × *P. pseudo-orientale* in the Siah Cheshmeh area, near the Turkish border where their parents occur together. Morphologically, the species and two interspecific hybrids in this section were described by Goldblatt. The species of the section are particularly important due to their medical alkaloid contents such as morphine, codein and thebaine (Parmaksize & Ozean 2010). Also, they studied the thebaine alkaloid content in dry capsules of the species belonging to sect. *Oxytona* and revealed the majority of *P. bracteatum* accessions contained high thebaine from 0.6% to 2.5% and none of the *P. orientale* accessions contained any thebaine. While the thebaine content of the *P. pseudo-orientale* accessions ranged from none to 1.5%. Goldblatt (1974) reported the alkaloids found in the sect. *Oxytona* which were studied by Lalezari et al. (1973); Shafiee et al. (1975) and coworkers in Tehran University as following: Dominant alkaloid in *P. bracteatum* is thebaine with a little alpinigenine. *P. orientale* has oripavine as a dominant alkaloid with trace amounts of isothebaine and *P. pseudo-orientale* contains high isothebaine with trace amounts of thebaine and oripavine and other trace alkaloids. Also, thebaine and oripavine were identified in *P. bracteatum* × *P. orientale* and isothebaine with oripavine in *P. orientale* × *P. pseudo-orientale*.

Ojala et al. (1990) investigated the species of *P. bracteatum*, *P. orientale* and *P. pseudo-orientale* and F_1 hybrids between these species. They presented morphological, Cytological and chemical findings of these taxa.

Levy and Milo (1991) studied the alkaloids and chromosome number of *P. bracteatum*, *P. pseudo-orientale*, the generations of F_1, F_2, back cross of F_1 with both parents and their results were consistent with those reported in previous studies (Shaffiee et al. 1975; Ojala et al. 1990; Goldblatt 1974 and Milo et al. 1986). Also, they indicated that the alkaloid contents of the capsules and roots of the plants are determined by regulatory genes and introduced a genetic model for the inheritance of these alkaloids.

The stomata length and pollen diameter of *P. bracteatum*, *P. orientale* and *P. pseudo-orientale* were reported by Goldblatt as follows: the mean of stomata length and pollen diameter is respectively 26 and 25.5 in *P. bracteatum*; 36 and 27.4 in *P. orientale*; 50 and 28.8 in *P. pseudo-orientale*. On the other hand, the name of *P. setiferum* Goldblatt was proposed for *P. pseudo-orientale* (Goldblatt 2011) which is a homonym for the hybrid taxon that determined by Camus (*P. orientale* × *P. lateritium*).

This study aims to: 1- Survey the pollen, seed and leaf epidermal characters of taxa of the section and discuss their taxonomical value. 2- Describe a new interspecific hybrid of the section for Iran. 3- Investigate the synonymy of *P. lasiothrix* and *P. bracteatum* micromorphologically

MATERIAL AND METHODS

Our study included four species and three hybrids of sect. *Oxytona*. The voucher specimens are listed in Table 1. In this study, seeds, leaves and stamens were collected from their natural habitats and herbarium specimens of TARI and FAR. Also, diagnostic characters were considered for each taxon and compared with the studies of Goldblatt (1974) and Ojala et al. (1990) (Table 2). In light microscopy (LM) studies: pollen grains were prepared using acetolysis method of Erdtman (1989), then mounted on slides in glycerin jelley. The polar (P) and equatorial (E) axes of 30 pollen grains for each taxon were measured by Jenus microscope at a ×100 magnification. The matured leaves of each taxon were selected and boild in water being macerated in 20% Naocl solution for about 6 hours. Leaf epidermis samples were stained in a solution of 0.1% fushin and methylblue 1% for 2-3 min., then fixed in glycerin. Also, the epidermal samples were taken from the mid-lamina regions of adaxial and abaxial surface, which is considered to be least variable (Wilkinson 1974; Chen et al. 2008). Mostly two or more leaves were collected for each species and at least 3-5 slides were prepared from each taxon. The type of stomata complex, shape of epidermal cells and pattern of anticlinal walls, length and breadth of stomata and stomata density per square millimeter of the leaf surface were recorded using Ziess microscope (Table 3).

30 mature seeds belonging to four specimens were measured and studied for shape, color, length, width, epidermal cell shape and anticlinal walls by Zies stereomicroscope, Stemi Sv6 (Table 5).

In scanning electron microscopy (SEM), small

Table 1. List of examined *Papaver* taxa in micromorphological studies.

Taxa	Collection Data
P. bracteatum	Azerbaijan: Urumieh, Targevar, 1750 m, 15. Jun. 1972, 1750 m, Pabot 5722-A-TARI.-Tehran: E. of Tehran, polour, 2350 m, 16. 6. 1973, Babakhanlou & Amin 14721-TARI; E. of Tehran, 3 km from Lar protected station, 2500 m, 18. 7. 1972, Dini & Arazm 14722-TARI; E. of Tehran, Polour, 2280 m, 25. 6. 2009, Tavakkoli 34306-FAR.
P. lasiothrix	Tehran: E. of Tehran, Polour, 2280 m, 25. 6. 2009, Tavakkoli 26930-FAR; Between Karaj and Chalus, Kandowan, 2500 m, 16. 6. 1974, Amin & Bazargan 19329-TARI.
P. orientale	Gilan: the road of Assalem to Khakhal, 1900 m. 23. 6. 2011, Tavakkoli –FAR.-Azerbaijan: Ardebil, Meshkinshahr, Mooeil village, yeylagh-e Rezvan, 17. 8. 2012, Taghizadeh 54245-FAR; Ardebil, Lahrood, Sabalan, Shabil, 2700-2750 m, 16. 7. 2012, Salimi 59905-FAR; Lisar protected area, 2200 m, Bidarlord 1747, 1748-FAR
P. setiferum	Azerbaijan: Arasbaran protected area, Doughroon mt. 2300 m. 23. 6. 76, Runemark & Assadi 21861-TARI; Ardebil, Khalkhal, the road of Khalkhal to Assalem, Taghizadeh 70378-FAR.
P. setiferum × *P. bracteatum*	Azerbaijan: 20 km from Razi to Germi, Histi kuh, East of Seyed Lar village, 1600-2000 m, 21. 6. 1980, Mozaffarian & Nowrozi 34717-TARI; 14 km from Keshmesh-Tappeh to Khoy on road of Kelisa Kandi, 1400 m, 18. 6. 1978, Assadi & Mozaffarian 30219-TARI.
P. setiferum × *P. orientale*	Azerbaijan: 6. Km from Germi to Ani, W. of Easemar village, 900-1200 m. 23. 6. 1980, Mozaffarian & Nowrozi 34945-TARI; Urumieh, Targevar, 1750 m, 15. Jun. 1972, Foroughi 5722-TARI.
P. orientale × *P. bracteatum*	Gilan: the road of Assalem to Khalkhal, 1900 m, 23. 6. 2011, Tavakkoli 6364-FAR.-Azerbaijan: 30 km after Makou on road to Boralan, 28. 6. 1982, 1600 m, Akbarzadeh& Salari 41147-TARI.

portions of leaf material of all taxa, mature seeds of *P. bracteatum* and *P. lasiothrix*, *P. orientale* and *P. setiferum* and pollen grains of *P. bracteatum* and *P. lasiothrix* were mounted on aluminum stubs and sputter coated with gold. At least 2-3 materials from each taxon were scanned and their micro-and macromorphological characters were studied (Figs. 15-28; 44-55; 56-61).

The terminology of seed coat and pollen grains sculpturing and stomata type follows Barthlott (1981) & Koul et al. (2000); Hesse et al. (2009) and Van Cotthem (1970) respectively.

RESULTS

The morphological characters of specimens and interspecific hybrids of *Papaver* sect. *Oxytona* are given in Table 2 and compared with the results of Goldblatt and Ojala et al. (Golblatt 1974; Ojala et al. 1990). Leaf epidermis characteristics of the taxa under light and scanning electron microscopy are summarized in Table 3.

Micromorphological characters of the seeds and pollen grains of the species of *P. bracteatum*, *P. lasiothrix*, *P. orientale* and *P. setiferum* under stereomicroscope, light and electron scanning microscopy are presented in Tables 4, 5.

Epidermal characters

The results are summarized in Table 3 and Figs. 1-28.

Shapes of the epidermal cells

Shape of epidermal cells on the adaxial surface are polygonal and polygonal-nearly irregular in hybrids (Figs. 9,11,13) whereas on the abaxial surface are polygonal-nearly irregular (Figs. 2, 4, 6, 15, 16, 17, 18, 19, 20) with the exception of four taxa with irregular epidermal cells: *P. setiferum* (Figs. 8, 21, 22), *P. setiferum* × *P. bracteatum* (Figs. 10, 23, 24), *P. setiferum* × *P. orientale* (Figs. 12, 25, 26) and *P. orientale* × *P. bracteatum* (Figs. 14, 27, 28).

Anticlinal walls of the epidermal cells

The patterns of the anticlinal walls of the epidermal cells are divided into four types:

- Straight-curved type:

In the section, the straight-curved type of the anticlinal walls is observed in *P. bracteatum* (Ad.; Fig. 1), *P. lasiothrix* (Ad.; Fig. 3) and *P. orientale* (Ad.; Fig. 5).

- Curved-nearly sinuate (or slightly sinuate) type:
This type occurs in *P. setiferum* (Ad.; Fig. 7), *P. setiferum* × *P. bracteatum* (Ad.; Fig. 9), *P. setiferum* × *P. orientale* (Ad.; Fig. 11), *P. bracteatum* (Ab.; Figs. 2, 15, 16) and *P. lasiothrix* (Ab.; Figs. 4, 17, 18).
- Nearly sinuate-sinuate type:
In section *Oxytona*, this type occurs in *P. orientale* (Ab.; Figs. 6, 19, 20) and *P. orientale* × *P. bracteatum* (Ab.; Figs. 14, 27, 28).
- Strongly sinuate type:
Strongly sinuate type occurs in *P. setiferum* (Ab.; Figs. 8, 21, 22), *P. setiferum*× *P. bracteatum* (Ab.; Figs. 10, 23, 24) and *P. setiferum*× *P. orientale* (Ab.; Figs. 12, 25, 26).

Stomata apparatus
All taxa of this section have anomocytic stomatal type on the abaxial surface and do not have any stomata on the adaxial surface.

Stomata size
The size of the stomata is 35.1-49.72 × 29.66-36.4 µm. Among the species, *P. setiferum* has longer stomata than *P. orientale* and the stomata size of *P. orientale* is longer than *P. bracteatum* and *P. lasiothrix* (Table 3).

The hybrids have longer stomata than three species of *P. bracteatum*, *P. lasiothrix* and *P. orientale* (Table 3).

Stomata density
The stomata density varies amongst the different taxa, ranging from 63.18 stomata per square millimeter in *P. setiferum* (sparse) to 291.81 in *P. lasiothrix* (dense) (Table 3).

Epicuticular secretions
The cuticle of leaf abaxial surface of all the taxa examined is covered with dense crystalloid threads and sparse flakes (Figs. 15-28).

Seed characters
According to the Table 5 and Figs. 44-55. seeds are brown to dark brown, slightly lustrous, reniform, sometimes falcate, convex at the dorsal side and concave at the ventral side with reticulate sculpturing. The average of seed length in the species examined is 0.93-1.08 mm and the average of width is 0.63-0.74 mm. The seed coat cells are oblong, elongated polygonal, rectangular, polygonal, irregular polygonal or rectangular. Anticlinal walls are nearly straight in *P. bracteatum* and *P. lasiothrix* and nearly sinuate in *P. orientale* and *P. setiferum*. The testa surface ornamentation in the species studied is granulate-perforate.

Pollen grains characters
Characters of pollen grains are shown in Table 4 and Figs. 29-43 & 56-61. The pollen grains are monad, tricolpate or sometimes tetracolpate, radially symmetrical and isopolar. The mean of polar axis (Table 4) is 25.16µm in *P. bracteatum* to 35.31µm in *P. setiferum* × *P. orientale*, but the mean of equatorial axis is 24.64µm in *P. bracteatum* to 31.25µm in *P. setiferum* × *P. orientale*. The shape of the pollen grains according to Erthman(1989) is prolate-spheroidal and oblate-spheroidal.

The exine sculpturing in *P. bracteatum* and *P. lasiothrix* is microechinate (Table 4; Figs. 58, 61). The mean of spinuli length varies from 288.06 nm in *P. bracteatum* to 303.3 nm in *P. lasiothrix*, but spinuli base ranges from 470.92 nm in *P. bracteatum* to 512.66 nm in *P. lasiothrix*.

Morphological characters
Papaver sect. **Oxytona** Bernh., Linnaea 8: 463 (1833).
Syn.: *Papaver* sect. *Macrantha* Elkan, Tent. Monog. *Papaver*. 13. 1839; *Calomecon* Spach, Hist. Nat. 7: 9. 1939.
Setose perennial herbs. Petals pale orange to dark red, usually four and sometimes six, with or without black to purple spot. Filaments dilated. Capsules to ovoid or obovoid; stigmatic disc as broad as the capsule, with 8-20 rays.

Key to the species of the sect. *Oxytona*
1. Flowers with 3-8 bracts. Pedicels 3-28 cm long, with adpressed or patent bristles. Buds erect; calyx with adpressed bristles. Petals dark red with basal black spot **1. P. bracteatum**
- Flowers without bract or with 1-2 bracts. Pedicels long or short. Calyx with patent or nearly patent bristles. Petals with or without spot 2
2. Flowers without bract. Pedicels 17-39 cm. Buds pendulous; calyx with patent bristles. Petals pale orange, mostly without spot. **2. P. orientale**
- Flowers without bract or with 1-2 bracts. Pedicels 1.5-20 cm. Buds erect; calyx with nearly patent bristles. Petals orange red, occasionally with rectangular black spot near the base. **3. P. setiferum**

1. **Papaver bracteatum** Lindl., Coll. Bot. tab. 23 (1821).
Figs. 62, 63
Syn.: *P. orientale* var. *bracteatum* (Lindl.) Ledebour,

Table 2. Comparison of taxonomic treatments of the genus *Papaver* sect. *Oxytona*.

Taxa	Authors	Stem high (cm)	Pedicel length/stem	Bracts number	Bud	Calyx bristles	Petal color	Stigmatic rays number	Stem leaves number
P. bracteatum 2n=14	This study	54.5 (40-85)	0.42 (0.14-0.65)	4.91 (3-7)	Erect	Adpressed	Dark red with basal black spot and length more than width	15 (13-19)	4.33 (3-6)
	Ojala et al.	92.5 (66-115)	0.27 (0.11-0.36)	4.7 (3-7)	Erect	Adpressed	Dark red	-	-
	Goldbaltt	Robust habit up to 1 m	One third of stem length	3-8	Erect	Adpressed	Dark red with dark marking usually longer than width and running to base	12-24	5-7
P. lasiothrix 2n=14	This study	40 (26-52)	0.38 (0.37-0.39)	4.3 (4-7)	Erect	Adpressed	Dark red with basal black spot and with length more than width	15 (13-19)	4
	Ojala et al.	-	-	-	-	-	-	-	-
	Goldbaltt	-	-	-	-	-	-	-	-
P. orientale 2n=28	This study	55.69 (38-90)	0.53 (0.29-0.82)	0.0 (-)	Pendulous	Patent	Pale orange	12.38 (8-19)	3.4 (2-5)
	Ojala et al.	84.5 (59-98)	0.53 (0.37-0.85)	0.0 (-)	Pendulous	Nearly patent	Pale orange	-	-
	Goldbaltt	Slender, 30-70 cm, occasionally to 1 m	-	-	Pendulous	Patent	Pale orange with pale blue or white rectangular marking above the base	8-15	2-3
P. setiferum 2n=42	This study	52 (33-84)	0.34 (0.21-0.65)	0.6 (0-2)	Erect	Nearly patent	Orange red with rectangular black spot near the base	13 (10-19)	3.6 (2-4)
	Ojala et al.	82.1 (30-119)	0.27 (0.04-0.47)	1.0 (0-4)	Usually erect	Nearly patent	Red	-	-
	Goldbaltt	40-60, rarely 80 cm	Upper most leaf on the upper third of the stalk	Mostly bracteate	Erect	Nearly patent	Deep orange with heavy black marking near the base	9-19	5-6
P. setiferum × P. bracteatum 2n=28	This study	60 (48-73)	0.18 (0.09-0.35)	3.5 (1-6)	-	-	Orange; some petals with rectangular black spot and with length more than width to small spots	12.6 (11-15)	5.5 (3-7)
	Ojala et al.	47.8 (46-50)	0.31 (0.12-0.42)	2.5 (1-4)	Usually erect	Nearly patent	Red	-	-
	Goldbaltt	-	-	-	-	-	-	-	-
P. setiferum × P. orientale 2n=35	This study	46.14 (32-64)	0.25 (0.11-0.4)	0.0 (-)	-	-	Pale orange; some petals with rectangular spot near the base	11 (9-13)	5.8 (4-7)
	Ojala et al.	61.8 (33-78)	0.51 (0.36-0.64)	0.0 (-)	Usually erect	Nearly patent	Orange	-	-
	Goldbaltt	-	-	-	-	-	Intermediate P. setiferum and P. orientale; without marking	-	-
P. orientale × P. bracteatum 2n=21	This study	53.2 (43-72)	0.43 (0.2-0.94)	1.25 (0-2) or with peduncular leaves near the flower	Pendulous	Patent	Pale orange; without spot	13 (9-15)	3.6 (2-6)
	Ojala et al.	-	-	-	-	-	-	-	-
	Goldbaltt	-	-	0.0; Peduncular leaves near the flower	-	-	Intermediate between P. bracteatum and P. orientale; with pale marking	-	-

Table 3. Characters of the leaf epidermis in *Papaver* taxa (sect. *Oxytona*) viewed with light microscopy.

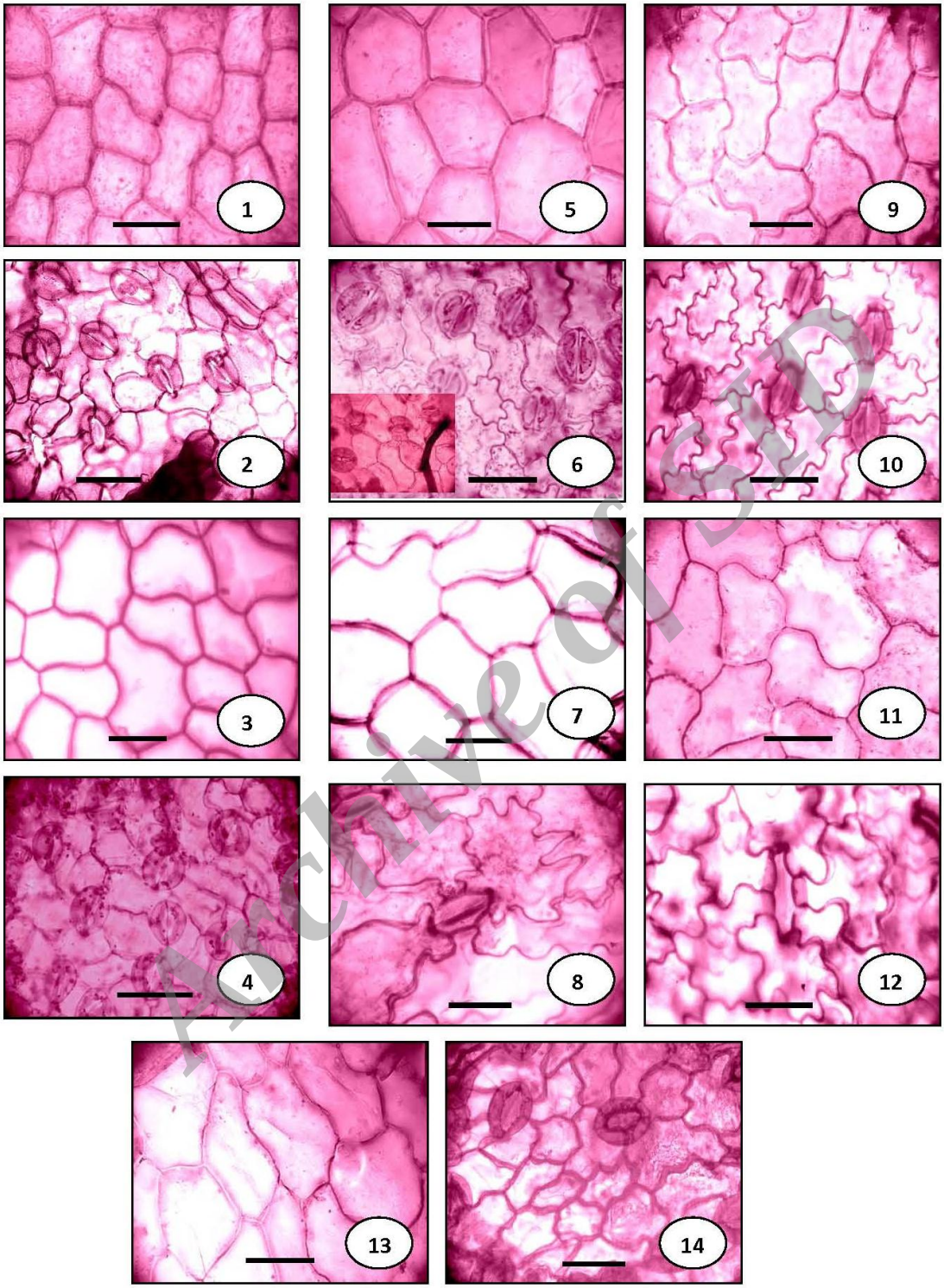
Taxa	Adaxial epidermis			Abaxial epidermis						
	Shape of cells	Pattern of anticlinal wall	Stomata in adaxial surface	Size of stomata (µm)		Shape of cells	Pattern of anticlinal wall	Stomata density (mm^2)	Stomata type	Figures
				Stomata length	Stomata width					
P. bracteatum	Polygonal-slightly irregular	Straight-curved; sometimes slightly sinuate	Absent	26.49 (36.29) 44.08	24.89 (30.71) 38.56	Polygonal-nearly irregular	Curved-slightly sinuate	274.38	Anomocytic	1,2
P. lasiothrix	Polygonal-slightly irregular	Straight-curved; sometimes slightly sinuate	Absent	30.51 (34.62) 46.13	20 (29.66) 39	Polygonal-nearly irregular	Curved-nearly sinuate	291.8	Anomocytic	3,4
P. orientale	Polygonal-slightly irregular	Straight-curved; sometimes slightly sinuate	Absent	35 (42.9) 50	24 (32.26) 40	Irregular	Nearly sinuate-sinuate	140.38	Anomocytic	5,6
P. setiferum	Polygonal-slightly irregular	Curved-slightly sinuate	Absent	40 (47) 65	30 (34.4) 39	Strongly irregular	Strongly sinuate	68.18	Anomocytic	7,8
P. setiferum × *P. bracteatum*	Polygonal-nearly irregular	Curved-nearly sinuate	Absent	30 (43.58) 80	25 (35.68) 45	Strongly irregular	Strongly sinuate	158.73	Anomocytic	9,10
P. setiferum × *P. orientale*	Polygonal-slightly irregular	Curved-nearly sinuate	Absent	36 (49.86) 70	25 (32.77) 39	Strongly irregular	Strongly sinuate	161.87	Anomocytic	11,12
P. orientale × *P. bracteatum*	Polygonal-slightly irregular	Curved-slightly sinuate	Absent	39 (49.72) 55	32 (36.4) 48	Irregular	Nearly sinuate - sinuate	142.86	Anomocytic	13,14

Table 4. Pollen morphological data of the studied taxa of the genus *Papaver* sect. *Oxytona* (P= polar axis, E= equatorial axis).

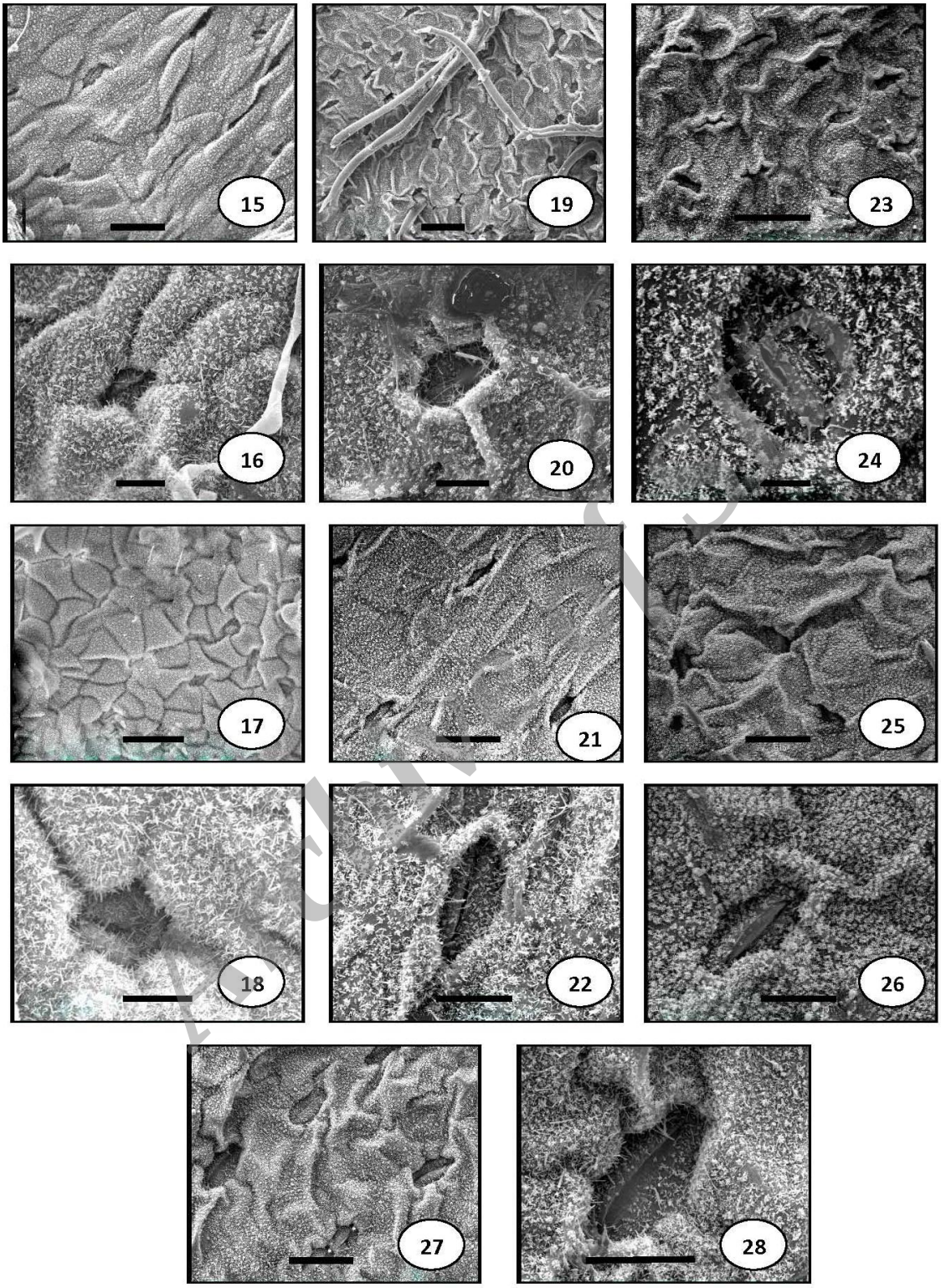
Taxa	Polar axis µm	Equatorial axis µm	P/E	Shape	Sculpturing	Spinule length (nm)	Spinule base (nm)	Distance between spinuli (nm)
P. bracteatum	22.5 (25.16) 30	20 (24.64) 27.5	1.2	Prolate-spheroidal	Microechinate	195.65 (288.06) 431.03	136.36 (187) 250.94	347.82 (470.92) 627.85
P. lasiothrix	21 (25.36) 29	23 (25.03) 27	1.01	Prolate-spheroidal	Microechinate	239.13 (303.31) 399.66	174.25 (200.83) 663.24	353.3 (512.66) 663.24)
P. orientale	20 (27.14) 35	25 (28.72) 36	0.94	Oblate-spheroidal	-	-	-	-
P. setiferum	22 (32.09) 40	26 (31.09) 40	1.03	Prolate-spheroidal	-	-	-	-
P. setiferum × *P. bracteatum*	20 (35.31) 44	25 (31.5) 40	1.12	Prolate-spheroidal	-	-	-	-
P. setiferum × *P. orientale*	21 (31.42) 40	22 (31.25) 40	1.005	Prolate-spheroidal	-	-	-	-
P. orientale × *P. bracteatum*	20.3 (28.7) 35.8	19.6 (25.32) 31	1.13	Prolate-spheroidal	-	-	-	-

Table 5. Seed morphological data of the examined species of the genus *Papaver* sect. *Oxytona.*

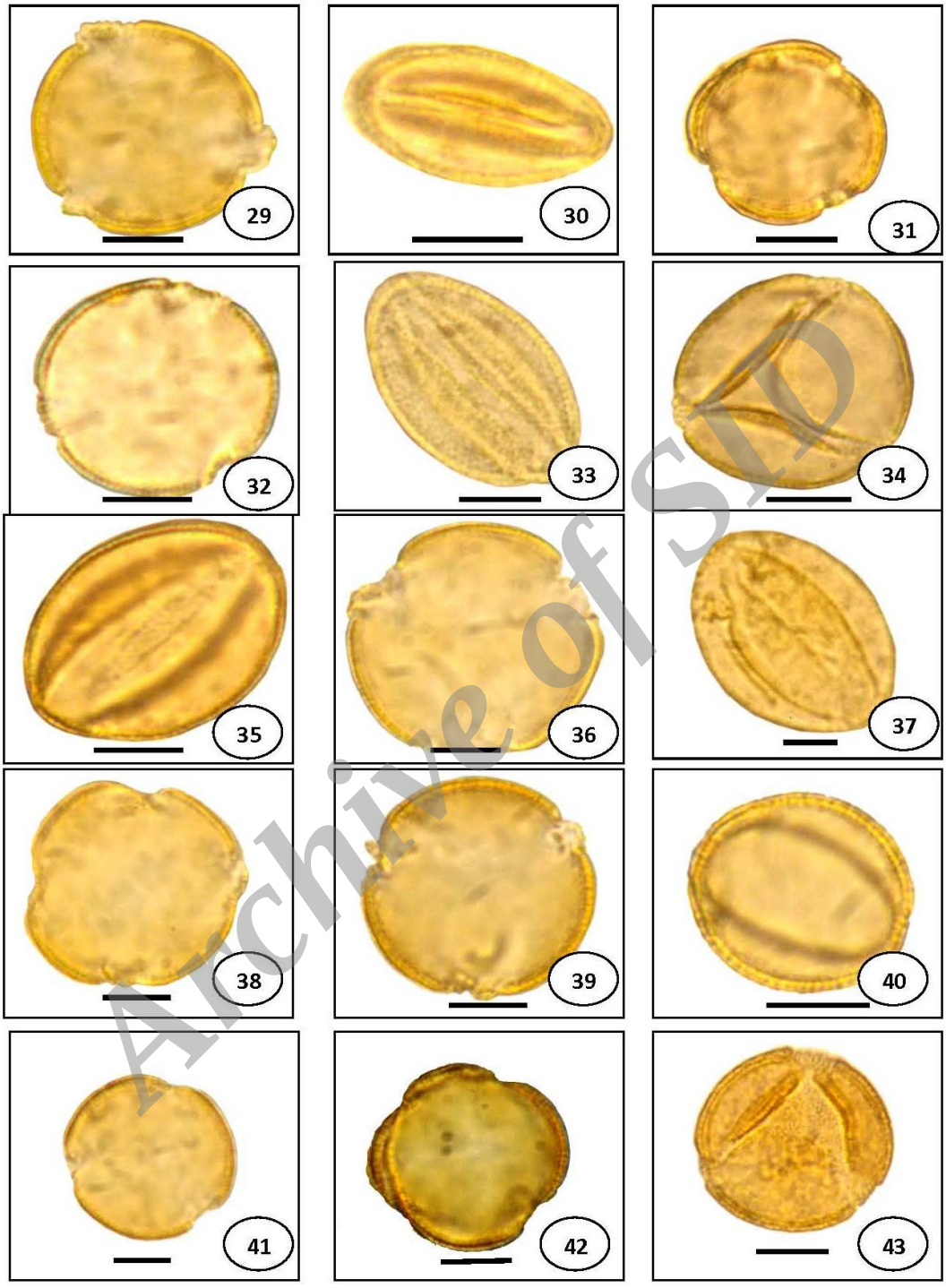
Taxa	Seed length	Seed width	Pattern of anticlinal wall	Seed sculpturing	Testa sculpturing	Epidermal cells shape	Seed shape	Seed color	Figures
P. bracteatum	0.8 (0.99)1.4	0.5 (0.74) 0.9	Nearly straight	Reticulate	Granulate-perforate	Oblong-elongated polygonal in upper part; polygonal in lower part	Reniform; dorsal side convex; ventral side concave	Brown-dark brown; slight lustrous	44, 45, 46
P. lasiothrix	0.7 (0.93) 1	0.4 (0.68) 0.8	Nearly straight	Reticulate	Granulate-perforate	Oblong-elongated polygonal in upper part; polygonal in lower part	Reniform; dorsal side convex; ventral side concave	Brown-dark brown; slight lustrous	47, 48, 49
P. orientale	0.6 (0.84) 1.2	0.44(0.69) 0.9	Sligthtly sinuate	Reticulate	Granulate-perforate	Oblong-elongated polygonal in upper part; rectangular-polygonal in lower part	Reniform-almost falcate; dorsal side convex; ventral side concave	Brown-dark brown; slight lustrous	50, 51, 52
P. setiferum	0.74(0.97) 1.2	0.37 (0.58) 0.9	Sligthtly sinuate	Reticulate	Granulate-perforate	Rectangular, polygonal or elongated polygonal in upper part; elongated polygonal, irregular polygonal-rectangular in lower part	Reniform, occasionally falcate; dorsal side convex; ventral side concave	Brown-dark brown; slight lustrous	53, 54, 55



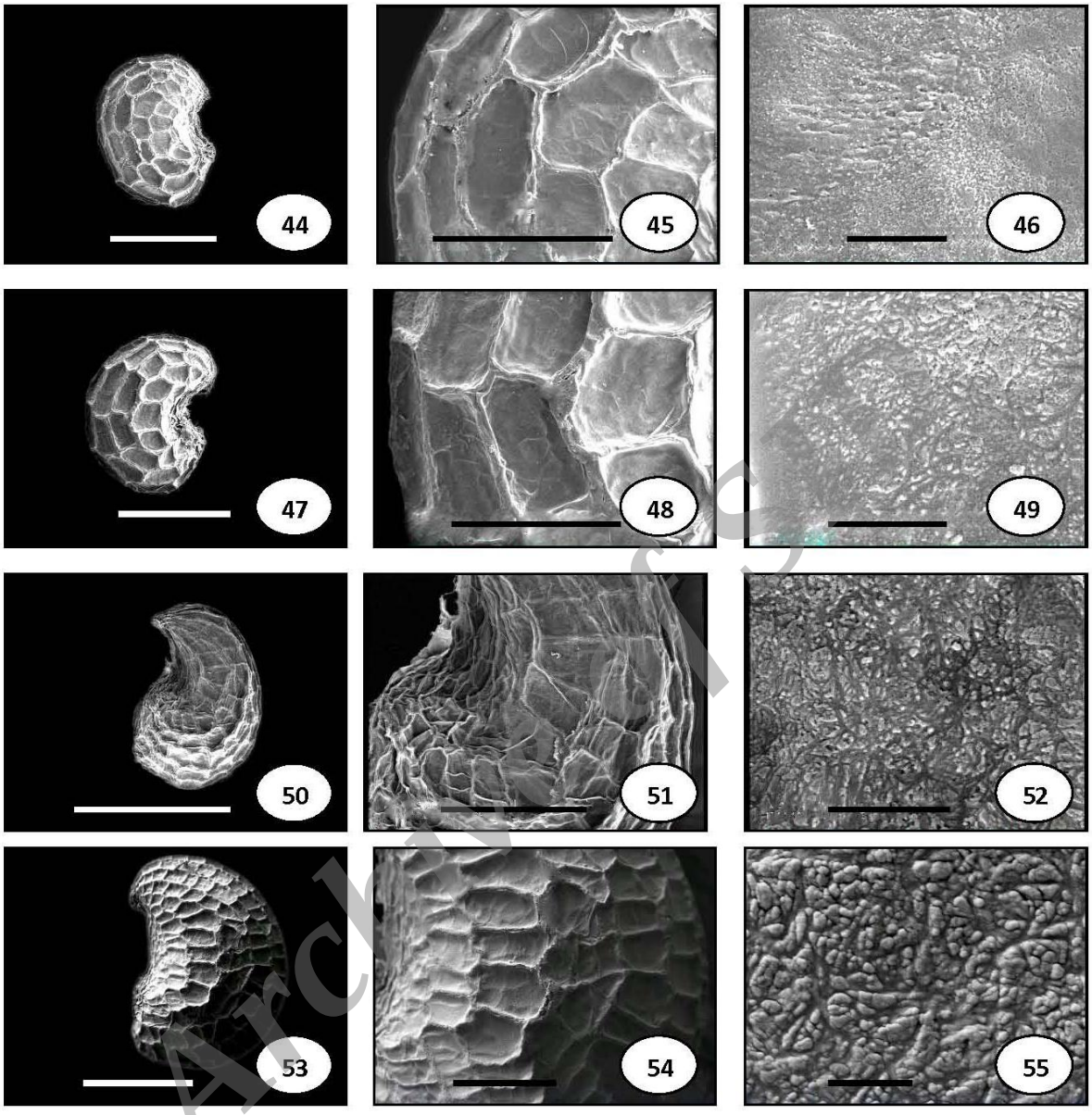
Figs. 1-14: LM micrographs of leaf epidermis in *Papaver* sect. *Oxytona*. 1-2) *P. bracteatum*, 1) Ad. 2) Ab.; 3-4) *P. lasiothrix*, 3) Ad. 4) Ab.; 5-6) *P. orientale*, 5) Ad. 6) Ab.; 7-8) *P. setiferum*, 7) Ad. 8) Ab.; 9-10) *P. setiferum* × *P. bracteatum*, 9) Ad., 10) Ab.; 11-12) *P. setiferum* × *P. orientale*, 11) Ad. 12) Ab.; 13-14) *P. orientale* × *P. bracteatum*; scale bars=50 µm.



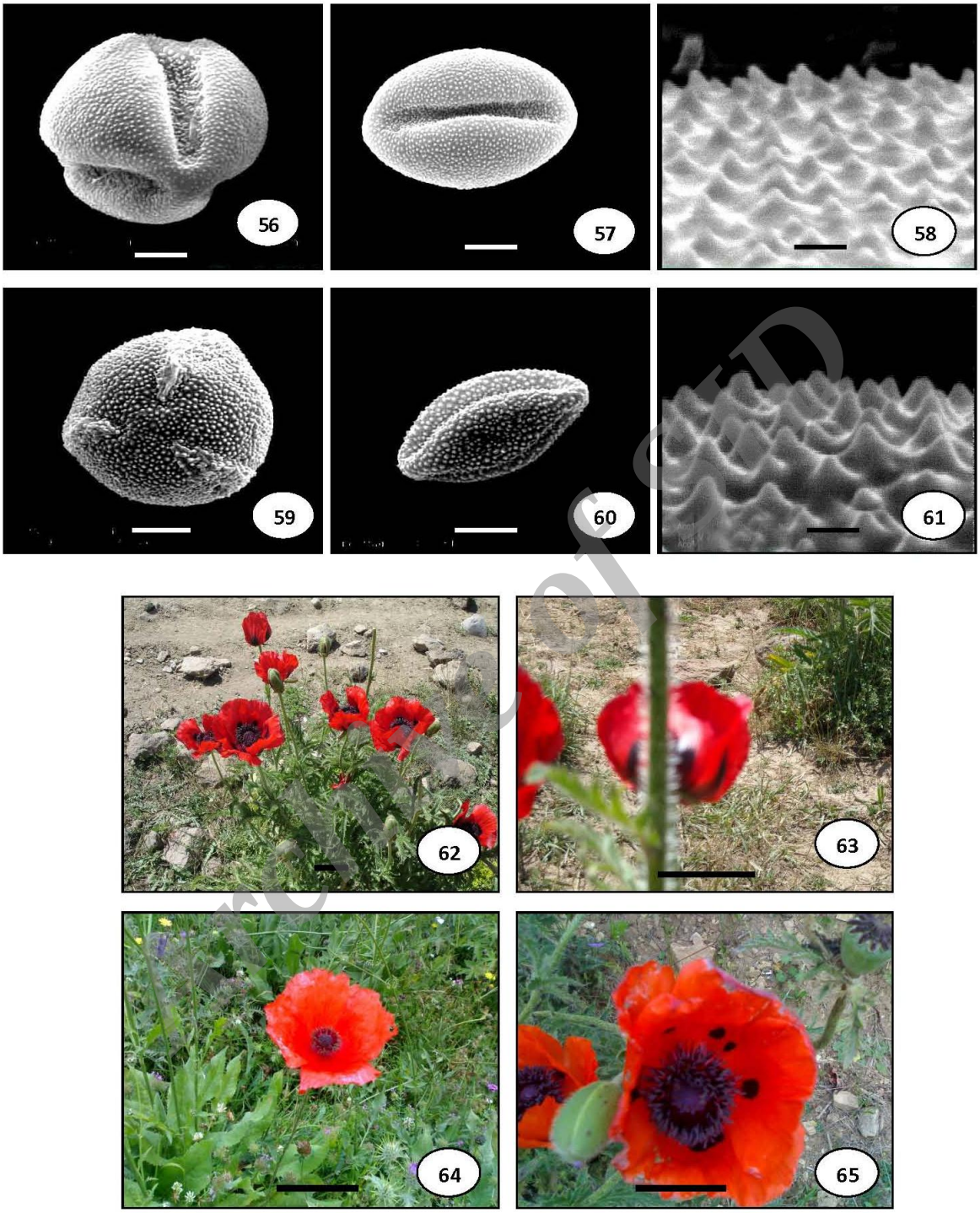
Figs. 15-28: SEM micrographs of leaf abaxial surface in *Papaver* sect. *Oxytona*. 15-16) *P. bracteatum*; 17-18) *P. lasiothrix*; 19-20) *P. orientale*; 21-22) *P. setiferum*; 23-24) *P. setiferum*× *P. bracteatum*; 25-26) *P. setiferum* × *P. orientale*; 27-28) *P. orientale* × *P. bracteatum*; scale bars=50 µm.



Figs. 29-43: LM micrographs of pollen grains in *Papaver* sect. *Oxytona*. 29-31) *P. bracteatum*, 29,31) polar view, 30) equatorial view; 32-33*) P. lasiothrix*, 32) polar view, 33) equatorial view; 34-35) *P. orientale*, 34) polar view, 35) equatorial view; 36-38) *P. setiferum*, 36,38) polar view, 37) equatorial view; 39-40) *P. setiferum* × *P. bracteatum*, 39) polar view, 40) equatorial view; 41-42) *P. setiferum* × *P. orientale*: polar view; 43) *P. orientale* × *P. bracteatum*: polar view. Scale bars= 10 µm.



Figs 44-55: SEM micrographs of seeds in *Papaver* sect. *Oxytona*: 44-46) *P. bracteatum*; 47-49) *P. lasiothrix*; 50-52) *P. orientale*; 53-55) *P. setiferum*. (44, 47, 50, 53) scale bars=0.5 mm; (45, 48, 51, 54) scale bars= 200 µm; (46, 49, 52, 55) scale bars=5 µm.



Figs. 56-61: SEM micrographs of pollen grains in *Papaver* sect. *Oxytona*: 56-58) *P. bracteatum*; 59-61) *P. lasiothrix*. (56, 57, 59, 60) scale bars=5 µm; (58, 61) scale bars=500 nm.

Figs. 62-68: Photographs of taxa of the genus *Papaver* sect. *Oxytona*: 62) *P. bracteatum*; 63) *P. lasiothrix*; 64) *P. orientale*; 65) *P. setiferum*; 66) *P. setiferum* × *P. bracteatum*; 67) *P. setiferum* × *P. orientale*; 68) *P. orientale* × *P. bracteatum*. Scale bars= 5 cm.

Fl. Ross., 1: 91. 1842; Trautvetter, Act. Hort. Peter. 11:495. 1873; *Calomecon bracteatum* (Lindl.) Spach, Hist. Nat. Veg. Phan. 7: 9. 1839; *P. lasiothrix* Fedde in Engl. Pflanzenr. 4: 366. 1909; *P. pulcherrimum* Fisch. ex Steud., Nom., ed. 201: 266. 1840, nom. nud.

Perennial herbs, 40-85 cm high. Cauline leaves 3-6 in number. Pedicels 3-28 cm long, with adpressed to patent bristles. Buds erect, with adpressed bristles on the calyx. Flowers with 3-8 floral bracts; petals dark red with a basal black spot and with length more than width.

Typus. Cultivated specimen in London.

Distribution in Iran. N., NW, W. and C. of Iran.

General distribution. Turkey, Iran and Caucasus.

Selected specimens. **Mazandaran**: Siah bisheh. Chalus valley, 2100 m, 18 Jun 1971, Sabeti 1784**.-Azerbaijan**: Arasbaran protected area, Doghrun mountain, 2500 m, 15. 7. 1977, Assadi & Sardabi 23872; Urumieh: Targevar, 2200 m, 28. Jul. 1972, Sabeti 7685**.-Tehran**: 8.5 km NE of Tehran, Polour protected station, 2600 m, 16. 7. 1972, Babakhanlou 14834.

2. **Papaver orientale** L., Spec. Plant. 508 (1753).
Fig. 64

Syn.: *P. pollakii* A. Kerner, Wiener III. Gartenzeitg. 272 (1888); *Calomecon orientale* (L.) Spach., Hist. Nat. Veg. 7: 9. 1839; *P. grandiflorum* Moench, Meth. 247. 1794, nom. illeg., Superfl. pro *P. oriental* L.; *P. spectabile* Salisb., Prod. 373. 1796, nom. illeg., Supefl. pro *P. orientale* L.; *P. orientale* var. *paucifoliatum* Trautv., Act. Hort. Petr. 4: 345. 1876; *P. paucifoliatum* (Trautv.) Fedde in Engl., Pflanzenr. 4: 366. 1909; *P. orientale* var. *parviflorum* Busch, Fl. Cauc. Crit. 3: 42-43. 1904, nom. illeg., Superfl. pro *P. orientale* var. *paucifoliatum* Trautv.

Perennial herbs, 38-90 cm high. Cauline leaves 2-5 in number. Pedicels 17-39 cm long, with adpressed bristles. Buds pendulous, with patent bristles on the calyx. Flowers without floral bracts. Petals pale orange and without spot or sometimes with basal white spot.

Typus. Turkey.

Distribution in Iran. N. and NW of Iran.

General distribution. Turkey, Iran, Caucasus and Afghanestan.

Selected specimens. **Gilan**: Asalem to Khalkhal, 1900 m, 29. 5. 1978, Wendelbo & Assadi 27771; Manjil, Sardegah, 2150 m, 18. Jun. 1973, Makouii 1007; Mountain above Damesh, East of Rudbar, 2000-2100 m, 22. 6.1975, Wendelbo & Ann Ala 18216. - **Azerbaijan:** Arasbaran protected area, 2400-2700 m, 15. 7. 1977, Assadi & Sardabi 24315; Kuh-e Sabalan, 2690 m, 28. Jul. 1972, Foroughi 7687; Ardebil: Shahbil, 2700 m, 18. 7. 2002, Jamzad & Azimi 82624; 50 km. W. of Khoy, near the Turkish border, between the village Aland and Razi, 2450 m, 25. 7. 1990, Assadi & Olfat 68881.

3**. P. setiferum** Goldblatt, Novon 21: 182 (2011).
Fig. 65.

Syn.: *P. pseudo-orientale* (Fedde) Medw.,Isv. Kavkazsk. Muz. [Bull. Mus. Cauc.] 11. 207. 1918. nom. illeg., non *P.* × *pseudo-orientale* E. G. Camus, J. Bot. (Morot) 12 (10): 156. 1898; *P. bracteatum* var. *pseudo-orientale* Fedde in Engl., Pflanzenr. 4. 365: *P. intermedium* DC. Mem. Soc. Phys. Geneva 7. 301.

1836, nom. illeg.
Perennial herbs, 30-85 cm high. Cauline leaves 2-4 in number. Pedicels up to 20 cm long, with adpressed bristles. Buds erect with nearly patent bristles on the calyx. Flowers with 1-2 to without floral bracts. Petals orange red with rectangular black spot and with width more than length.
Typus. Caucasus.
Distribution in Iran. N. NW and C. of Iran.
General distribution. Turkey, Iran and Transcaucasus.
Selected specimens. Gilan: Manjil, Sardegah, 2150 m, 18. Jun. 1973, Makouii 1007. -**Azerbaijan**: 40 km from Razi to Germi, 1700 m, 22. 6. 1980, Mozaffarian and Nowrozi 34794; Arasbaran protected area, 1300 m, 1975, Savabi s.n. -**Tehran**: Fasham, 23. Jul. 1994, 1960 m, Parnia 32454-FAR.

Hybrids

Papaver setiferum × *P. bracteatum*
Fig. 66
Perennial herbs, 48-72.5 cm high. Cauline leaves 3-7 in number. Pedicels up to 26 cm long with adpressed bristles. Flowers with 1-6 floral bracts. Petals orange to orange red; some petals with black to deep purple rectangular spot; spots with width more than length and relatively small.

This hybrid differs from *P. setiferum* in having several floral bracts and similar to it by having petals with rectangular spot near the base, also differs from *P. bracteatum* in having 1-2 floral bracts (similar to *P. setiferum*) and purple to black rectangular spot on some petals.

The hybrid is found in Arasbaran area where *P. setiferum* and *P. bracteatum* occur together.
Specimens seen. **Azerbaijan**: Arasbaran protected area, Savabi & Raofi s.n.; 20 km from Razi to Germi, Histi kuh, East of Seyed Lar village, 1600-2000 m, 21. 6. 1980, Mozaffarian & Nowrozi 34717.

The hybrid *P. setiferum* × *P. bracteatum* is recorded ere for the first time in Iran.

Papaver setiferum × *P. orientale*
Fig. 67
Perennial herbs, 33-64 cm high. Cauline leaves 4-7 in number. Pedicels 3-18 cm long, with adpressed bristles. Peduncular leaves 1-2 in number and near the flower. Petals orange or orange red, without spot or with purple rectagngular spot on some petals.

This hybrid differs from *P. orientale* by having purple rectangular spot with width more than length and near the base on some petals and short peduncle; also differs from *P. setiferum* by having petals with or without spot in a flower.

Specimens seen. **Gilan**: Manjil, 24 km from Rostamabad to Dogaheh, 1700 m, 15. 6. 1980, Mozzafarian & Nowrozi 33967**.-Azerbaijan**: Salavat, Goli Daragh, 24. 6. 1980, Mozaffarian & Nowrozi 35005; 6 km from Germi to Ani, W. of Easemar village, 900-1200 m, 23. 6. 1980, Mozaffarian & Nowrozi 34945.

P. setiferum × *P. orientale* occurs in N. and NW of Iran where the parent species could be found growing in close vicinity making hybridization.

Papaver orientale × *P. bracteatum*
Fig. 68
Perennial herbs, 46-72 cm high. Cauline leaves 2-6 in number. Pedicels 11-38 cm long, with adpressed to nearly adpressed bristles. Petals pale orange, without spot, with 1-2 large bracts under or near the flower.

This hybrid differs from *P. orientale* in having floral bracts and from *P. bracteatum* in having pale orange petals and without spot.
Specimens seen. **Azerbaijan**: between Maku and Khoy, Arab-Dizechi, 2150 m, 29. 6. 1978, Assadi & Mozaffarian 30250; Urumieh, Naghadeh, 1700 m, 27. Jun. 1972, Sabeti 5725.

NW of Iran is locality where *P. orientale* and *P. bracteatum* could be found in close vicinity.

DISCUSSION

The morphological characteristics of perennial species and their interspecific hybrids with different ploidy levels are in accordance with the description given by Goldblatt (1974), Ojala & Rousi (1986) and Ojala et al. (1990). The most variable taxonomic characters of the section are presence or absence of petal spot; shape, color, size and position of petal spot; peduncle length and floral bracts number.

P. bracteatum is identified by numerous floral bracts; dark red petals with a black spot and running to base. *P. lasiothrix* differs from *P. bracteatum* by having patent bristles on the peduncle (Cullen 1966). On the other hand, the members were observed with nearly patent bristles on the peduncle or with adpressed bristles on the upper part and spreading on the lower part of peduncle. Therefore, the separation of this species from *P. bracteatum* is difficult. Our morphological observations confirm Goldblatt hypothesis in which *P. lasiothrix* reduced as the synonymy of *P. bracteatum*.

In *P. orientale* flowers are pale orange and the spot and floral bracts are absent. The buds are pendulous with spreading bristles on the calyx. Also, the peduncle is relatively long.

P. setiferum is recognized by deep orange petals

with a black rectangular spot and broader than length near the base. The buds are erect with nearly patent bristles on the calyx. The flowers are without or with 1-2 floral bracts.

In the hybrid *P. setiferum* × *P. bracteatum*, petals are pale red with rectangular spot above the base and width more than length (similar to *P. setiferum*) or reduced to a small spot or some petals lacked spot. The number of floral bracts is greater and more than *P. setiferum* but smaller and less than *P. bracteatum*. Generally, this hybrid more closely resembles *P. setiferum*.

Our observations on *P. setiferum* × *P. orientale* combination indicate that mostly many morphological features of *P. setiferum* predominate in the hybrid. The individuals of this hybrid have petals with rectangular spot near the base (similar to *P. setiferum*) or lacked spot (similar to *P. orientale*). The peduncle is almost short. Also flowers have 1-2 floral leaves on the peduncle.

The hybrid plants with the parents of *P. orientale* and *P. bracteatum* have long peduncle. Petals are orange or orange red and without spot. The flowers are with 1-2 large floral bracts or with peduncular leaves near the flower. The general appearance of the hybrid is very similar to *P. orientale*.

All of the taxa investigated of sect. *Oxytona* lacked any stomata on adaxial surface. Also, the upper epidermis cells are relatively similar in shape and outlines of anticlinal walls. Therefore, epidermal characters of upper surface do not support the separation of taxa from each other. Although in abaxial surface, the anomocytic stomatal type and epicuticular wax secretions are similar in all of the taxa but the features such as stomata size, epidermal cells and anticlinal walls shape have taxonomic value in distinguishing of taxa.

P. bracteatum and *P. lasiothrix* show similar epidermal characters (Table 3). Therefore our study confirms the systematic treatments of Goldblatt (1974) in which *P. lasiothrix* reduced to the synonymy of *P. bracteatum*.

Among the species studied, the stomata size of *P. bracteatum* in average (35.45 × 30.18) is obviously smaller than those of other species (Table 3). In *P. setiferum*, epidermal cells shape of abaxial surface is strongly irregular and anticlinal walls are strongly sinuate. In contrast, *P. orientale* has irregular epidermal cells and nearly sinuate-sinuate anticlinal walls. These are obviously different from those of *P. bracteatum* which have polygonal-nearly sinuate epidermal cells and curved-slightly sinuate anticlinal walls. On the other hand, the distinction of the three species of *P. bracteatum*, *P. orientale* and *P. setiferum* based on leaf epidermal characters is in agreement with their separation on the basis of morphological and cytological data and their alkaloid compositions (Goldblatt 1974; Ojala et al. 190; Milo et al. 1986).

In the hybrids of *P. setiferum* × *P. bracteatum* and *P. orientale* × *P. bracteatum*, epidermal characters of poylploid parents are dominant which are consistent with morphological data and chemical spectra reported in the hybrids and interspecific hybrids between *P. somniferum* and the section *Oxytona* species (Ojala and Rousi 1986). Also, morphological and epidermal characteristics of *P. setiferum* × *P. orientale* correspond to alkaloid content of hexaploid parent.

From the palynological view, two species of *P. bracteatum* and *P. lasiothrix* are completely similar in pollen grains size and shape. Both of species have microechinate tectum. The length, the base and the distance of spinuli are similar together. Thus palynological characters support the systematic treatment of Goldblatt, and *P. lasiothrix* should be treated as a synonymy of *P. bracteatum.*

In the species examined, *P. bracteatum* has the smallest pollen grains in average (25.26 × 25.08) and *P. setiferum* the largest grains in average (32.09 × 31.09). The shape of pollen grains in the taxa studied is prolate-spheroidal but oblate-spheroidal shape is found in the *P. orientale*. Also the pollen grains of all of the taxa studied are usually tricolpate but tetracolpate grains are found in most of them. Therefore, the pollen grains size has taxonomically significance between the taxa investigated in sect. *Oxytona*. Similar to morphological and epidermal characteristics, the hybrids inherit pollen grains size of hexaploid parent (*P. setiferum* × *bracteatum* & *P. setiferum* × *oreintale*) and tetraploid parent (*P. orientale* × *bracteatum*) (Table 4).

The seeds ultra-structural studies are revealed that their surface sculpturing is reticulate and granulate-prforate. This type of testa surface sculpturing is common among the species studied. The general shape of the seeds is very similar being reniform or occasionally falcate. The species investigated in the section have seeds with relatively similar size and color. Seed morphological data in *P. bracteatum* and *P. lasiothrix* are completely similar (Table 5).

Thus in agreement with the findings of Goldblatt, *P. lasiothrix* can be introduced as a synonymy of *P. bracteatum*. Three species of *P. bracteaum*, *P. orientale* and *P. setiferum* have slightly differences in testa cells shape and anticlinal walls (Table 5). Hence seed morphological characters do not support the separation of the species from each other.

CONCLUSION

1. Our morphological and micromorphplogical observations show high similarties between the two species *P. bracteatum* and *P. lasiothrix* which are in agreement with Goldblatt hypothesis in which *P. lasiothrix* reduced as a synonymy of *P. bracteatum*.
2. Within the genus *Papaver* sect. *Oxytona*, the species investigated are relatively similar in epidermal characters of leaf adaxial surface and seed morphology.
3. Among studied micromorphological characters, the differences in stomata and pollen grains size, epidermal cells and anticlinal walls shape of taxa are proportional to their differences in chromosome number and alkaloid compositions.
4. It seems, morphological and micromorphological characteristics of the hybrids between diploid and polyploidy species are inherited from polyploidy parents.

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