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POLLEN MORPHOLOGY OF ONAGRACEAE IN IRAN

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This study aims to assess the pollen characteristics of Iranian Onagraceae based on SEM techniques to evaluate the possibility of giving them a role in systematic analysis. Pollen grains of the studied taxa included radially symmetrical, tetrad tetra-hederal, isopolar, triangular to triangular-spheroidal, 3-zonoporate, with viscin threads, rugulate-granulate, granulate, verrucate and striate sculpturing. Polar axis ranged between 30 and 80 µm. Equatorial axis showed variation from 40 to 90 µm. Moreover, arm diameter ranged between 8.85 to 18.83 µm, pore diameter 8.72 to 21.69 µm. PCOA and Cluster analysis of pollen characteristics did not clearly show sectional divisions based on Flora Iranica as well as Flora of Iran. There are not significant differences in size, shape, pore structure and arm characteristics to delimit all species in Onagraceae. Moreover, Pollen grains are not efficient for generic delimitation in the studied taxa in Iran. Therefore, even though that the examined taxa cannot be distinguished exclusively by pollen, it may be a useful tool for delimitation some taxa in Onagraceae along with other evidence.

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Key words: Palynology; Onagraceae; *Epilobium*; *Circaea*; *Oenothera*; Iran

ریخت شناسی گرده در خانواده گل مغربی در ایران

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این مطالعه با هدف ارزیابی صفات گرده شناسی مبتنی بر تصاویر فناوری میکروسکپ الکترونی نگاره و ارزیابی نقش آنها در تحلیل سیستماتیکی خانواده گل مغربی به انجام رسیده است. دانه‌های گرده تاکسون‌های مطالعه شده با تقارن شعاعی، چهار تایی چهار وجهی، جورقطبی، سه گوش تا سه گوش-کروی، سه منفذی با رشته‌های ویسکین، با تزئینات میله‌ای، دانه دانه تا مخطط می باشند. محور قطبی بین ۳۰-۸۰ میکرومتر، محور استوایی بین ۴۰-۹۰ میکرومتر می‌باشد. قطر بازوها بین ۸/۸۵ تا ۱۸/۸۳ میکرومتر و قطر منافذ ۸/۷۲ تا ۲۱/۶۹ میکرومتر بوده است. آنالیز تجزیه به مولفه‌های اصلی و آنالیز تحلیل خوشه‌ای تمایز بخشه‌ها را بر مبنای فلور ایرانیکا و ایران نمایان نمی‌سازد. به علاوه تفاوت معناداری در ابعاد، ساختار منافذ و ویژگی‌های بازوها برای تعیین محدوده گونه‌ها درخانواده گل مغربی وجود ندارد. به علاوه دانه‌های گرده برای تمایز تاکسون‌ها مورد مطالعه در سطح جنس چندان کارآمد نیستند. با این وجود حتی اگر تاکسون‌ها به تنهایی بر اساس صفات دانه گرده قابل تفکیک نباشند، این صفت در کنار سایر صفات (صفت کمکی) در تمایز تاکسون‌های می‌تواند موثر واقع شود.

INTRODUCTION

The Onagraceae includes 655 species of herbs, shrubs, and trees belonging to 17 genera (Ford & Gotlieb 2007). The Onagraceae family in Iran is comprised of 23 species belonging to 4 genera including *Epilobium* L. (19), *Oenothera* L. (2), *Circaea* L. (1), and *Ludwigia* L. (1)(Asssadi et al. 2005, Naghinezhad & Sharafi 2009). It has broad distribution, especially in the New World (USA), mainly north western America (Levin & al. 2004). There has been much hybridization and creation of new forms (Stebbins 1950) as well as a lack of sharp diagnostic characters. So, these taxa confront several problems related to diagnostic keys as well as taxonomical complexities. Therefore, assessment of complementary evidence such as pollen characteristics is useful for analysis of complexity. Beer (1906) notes that pollen characteristics of Onagraceae attracted the attention of 19^{th} Century plant taxonomists (Mohl 1834; Scleiden 1846; Tschistakoff 1876). Besides, several studies have suggested that pollen characteristics have taxonomic as well phylogenetic importance including pollen morphology of Onagraceae (Ting 1966; Brown 1967; Makbul & al. 2008), palynology of *Circaea* (Skvarla & al. 2005), pollen taxonomy of *Epilobium* (Makbul & al. 2008), pollen evolution in Onagraceae (Harder & Johnson 2008), evolution and diversity of pollen tetrads in Onagraceae (Liao 1962; John & al. 1975; Skvarla & al. 1975, 1978; Albert & al. 2010; Hesse 2009), diversity of fossil pollen (Grímsson & al. 2012), sculpturing of Onagraceae (Keri & Zetter 1992; Praglowski & al. 1994) along with *Epilobium* (Hesse 1981, 1984; Patel & al. 1984; Rowley & Calugher 1996), pollen development in Onagraceae (Takahashi & Skvarla 1990; Rowley & Skvarla 2006), and finally evolution, reproductive ecology and pollination of Onagraceae (Montgomery & al. 2001; Erbar 2003; Roberstson & al. 2008; Albert & al. 2010; Krakos 2011). However, little attention has been paid to pollen characteristics of Onagraceae in Iran. There are several challenges in taxonomy of Onagraceae such that SEM techniques have been used to study the evidence related to Iranian Onagraceae. This study aims to assess the pollen characteristics of Iranian Onagraceae based on SEM techniques to evaluate the possibility of giving them a role in systematics analysis.

MATERIALS AND METHODS

Plant specimens were collected from natural habitats and from Iranian herbaria HSBU, IRAN HSBU along with the private herbarium of Dr. Akhani (herbarium abbreviation according to Thiers 2008). Vouchers of the specimens are presented in table 1. Pollen grains were extracted from mature anthers and mounted on aluminum stubs. They were then coated with gold in an Emitech EMK 550 sputter. Studies were made with Cam Scan Hitachi SU3500 Electron Microscope. Image tool Ver.30 was used to assess quantitative pollen characteristics. Pollen assessment was performed on 20-30 pollen grains including the shape, size of polar and equatorial axis, viscin threads, and diameter of pore and arms. Quantitative characteristics were coded as multistate characters and used for cluster and PCOA analyses. PAST software was used for all numerical analyses. Terminology has been based on to Punt & al. (2006), Halbritter & al. (2008), and Hesse & al. (2009).

RESULTS

Pollen grains of the studied taxa included radially symmetrical, tetrad tetrahederal as well isopolar. Besides, their pollen are shed as tetrad, occasionally monad**.** In addition, the shape of pollen grains in equatorial view are triangular (*E. roseum, E. palustre*) to triangular-spheroidal (*E. dodonaei, E. algidum, E. angustifolium*) as well elliptic-spheroidal (*E. tetragonum, E. frigidum, E. parviflorum*) to spheroidal (*E. dodonaei, E. algidum, E. angustifolium*) in polar view. Moreover, pollen grains are commonly 3-zonoporate, including sporadically viscin threads with flat superficial connected to the exine surface near the apertures.

Table 1. Voucher specimens of the studied taxa.

Species	**collection data**
Epilobium frigidum Hausskn.	Alborz, Gachsar, Rahimi & Mohhebi sadr, HSBU 87
E. frigidum Hausskn.	Mazandaran, Kelardasht, Rahimi & Mohhebi sadr, HSBU 88
E. montanum L.	Mazandaran, Larijan, Rahimi & Mohhebi sadr, HSBU 89
E. montanum L.	Mazandaran, Polur, Rahimi & Mohhebi sadr, HSBU 90
E. tetragonum L.	Alborz, Karaj, Rahimi & Mohhebi sadr, HSBU 91
E. tetragonum L.	Mazandaran,Siyahbisheh, Rahimi & Mohhebi sadr, HSBU 92
E. tetragonum L.	Alborz,Gachsar, Rahimi & Mohhebi sadr, HSBU 93
E. anatolicum Hausskn.	Alborz,Gachsar, Rahimi & Mohhebi sadr, HSBU 94
E. parviflorum Schreb.	Alborz,Gachsar, Rahimi & Mohhebi sadr, HSBU 95
E. parviflorum Schreb.	Mazandaran, Kelardasht, Rahimi & Mohhebi sadr, HSBU 96
E. parviflorum Schreb.	Tuskacheshmeh, Rahimi & Mohhebi sadr, HSBU 97
E. parviflorum Schreb.	Azerbaijan eastern, Kaleybar, Rahimi & Mohhebi sadr, HSBU 98
E. algidum M. B.	Azerbaijan eastern,Kaleybar, Rahimi & Mohhebi sadr, HSBU 2782
E. angustifolium L.	Azerbaijan eastern, Kaleybar, Rahimi & Mohhebi sadr, HSBU 2779
E. confusum Hausskn.	Karaj, IRAN 43148
E. roseum Schreb.	Ardebil, Amini rad & Eskandari, IRAN 65980
E. stevenii Boiss.	Azerbaijan, marand, IRAN 31120/8
E. gemmascens C. A. Mey.	Mazandaran, chaloos, IRAN 43104/2
E. hirsutum L.1	Alborz, Gachsar, Rahimi & Mohhebi sadr, HSBU 2784
E. hirsutum L. 2	Mazandaran, Kelardasht, Rahimi & Mohhebi sadr, HSBU 2781
E. lanceolatum Sebast. & Maur.	Azerbaijan, astara, HSBU 2787, IRAN 53936/2
E. minutiflorum Hausskn.	Ardebil, Rahimi & Mohhebi sadr, HSBU 2783
E. palustre L.	Kermanshah, IRAN 43143
E. ponticum Hausskn.	Azerbaijan, meshkin shahr, IRAN 31117
E. rechingeri Raven	Mazandaran, AKHANI 11879
E. dodonaei Vill.	Mazandaran, Anzali, HSBU 2780
Circaea lutetiana L.	Mazandaran, Mehrabian & Rahimi & Mohhebi sadr, HSBU 2785
Oenothera biennis L.	Gilan, Rahimi & Mohhebi sadr, HSBU 2786

The exine showing a heterogeneous sculpturing composed of rugulate (*E. confusum, E. tetragonum*), rugulate-granulate (*Oenothera biennis*, *E. minutiflorum*), granulate (*E. palustre*), verrucate (*E. hirsutum* 1) and striate (*E. montanum*) (fig. 1). In addition, polar axis ranged between 30 (*E. algidum*) to 80 µm (*E. hirsutum* 1, *Oenothera biennis*) and Equatorial axis showed variation from 40 (*Circaea lutetiana*) to 90 µm (*Oenothera biennis)*. Also, Polar/equatorial axes were 0.84 (*E. confusum*) to 1.15µm (*E. hirsutum* 1). Moreover, arm diameter ranged between 8.85 (*E. algidum*) and 18.83 µm (*Oenothera biennis*); pore diameter 8.72 (*E. minutiflorum*) to 21.69 µm (*Oenothera biennis*). Length of bulges ranged from 0.28 (*E. confusum*) to 1.489 µm (*E. tetragonum*); width of bulges ranged from 0.21 (*E. confusum*) to 0.43 µm (*E. tetragonum*). An extended palynological data on studied taxa can be seen in tables 3 and 4. The WARD trees of pollen characteristics (fig. 2) showed a relatively high cophenetic correlation (r >0.80). In general, three main clusters were observed. PCA analysis on the basis of 11 palynological variables revealed three components (polar length, equatorial length as well bugles length) accounting for about 72 % of the total variance (fig. 3). These characteristics had the highest correlation value (>0.7) with this axis.

Based on the pollen shape, two types of pollen were identified in the studied taxa.

Type I: elliptic-spheroidal in polar view, triangular in equatorial view, polar axis 40-70 µm, equatorial axis 40-75.31 µm, polar/equatorial 0.84-1.15 µm, pore length 8.72-19.5 µm, arm diameter 11-23 µm, sculpturing rugulate-granulate, rugulate, granulate-verrucate, striate, length of bulges 0.28-1.48 µm as well width of bulges 0.21-0.41 µm and their proportions (length/ width) 1.33-2.08 µm (fig. 22).

Type II: spheroidal in polar view, triangular-spheroidal in equatorial view,, polar axis 47.79-60 µm, equatorial axis 51.63- 70 µm, polar/equatorial 0.86-0.92 µm, pore length 9-19 µm, arm diameter 8.85-16.5 µm, sculpturing striate, granulate, verrucate, length of bulges 0.32-0.52 µm, width of bulges 0.23-0.26 µm, as well their proportions (length/ width) 1.40-1.88 µm (fig. 52).

DISCUSSION

The results proved that Iranian Onagraceae mainly shed their pollen in tetrad tetra-hederal as well in monad albeit in lower frequency. Accordingly, these results confirmed those of Mitrotu (1963) as well as Brown (1967). However, our results were contrary to those reported in Punt & al. (2003) and Makbul & al. (2008). Regardless, the pollen of fossil specimens of *Epilobium* was reported in the monad shape by Grimsson & Zetter Qin Leng (2012). However, Brown (1967) observed presence of monad and tetrad tetra-hederal shapes together in *Epilobium* as well as in other taxa of Onagraceae. Moreover, research reported in Skvarla & al. (1978) showed tetrad tetra-hederal dominantly visible in *Epilobieae*. Recent results confirm that both types, monad and tetrad tetra-hederal, can be seen in diverse geographical conditions. Furthermore, Punt & al. (2003) noted that there is only a tenuous connection between pollen grains and in several cases normally after acetolysis, where pollen is shed in a monad pattern. On the other hand, Erdtman (1960) believed that the monad form is a result of processes such as acetolysis. Moreover, our results do not confirm these opinions and in this study the monad form was considered as a natural pattern as well as a diagnostic characteristic for differentiation. Harder & Johnson (2008) proved that tetrad tetra-hederal improved the efficiency of pollen transfer. In addition, strong inbreeding depression has been reported in Onagraceae (Husband & Schemske 1996), where this response is a recent mechanism. Moreover, the species of *Ludwigia* (Brown 1966) *Circaea* as well as *Oenothera* shed their pollen in monad in fossil as well current samples (Grimsson & al. 2012) along with current taxa (Brown 1967), in line with our results.

The pollen shape manifests relative homogeneity in each genus, while represents diverse types in different studied genera including *Epilobium*, as well as *Oenothera*. So, *Oenothera* shows a wrinkled structure in polar surfaces. Further, this study proved presence of two types in *Epilobium* including triangular and triangular-spheroidal in polar view that confirmed the results of Brown (1967). Apart from this, *Epilobium luteum* Pursh indicates a unique irregular spheroidal shape, which is clearly distinguishable from other Onagraceae (Skvarla & al. 2008). Moreover, our distinguished pollen types are not in accordance with pervious morphological as well molecular based classifications.The studied taxa indicated a relatively diverse sculpturing (e.g. granulate, verrucate, striate, granulate-verrucate and etc.), though it does not appear as clear evidence to support delimitation of closet taxa. Apart from this, Makbul & al. (2008) classified them as bacculate while Rowley & Claugher (1996) did it by rod-shaped sculpturing. In addition, rugulate structures are considered ancestral properties in Angiosperms (Doyle 2009). Skavarla & al. (1975) has named an exine network of Onagraceae as paracrystaline-beaded. Some authors (Rowley 1983, 1996; Hesse 1984) have insisted on presence of a highly complex exine pattern as well as absence of distinct columellae.

An extended overlap was observed in quantitative characteristics. Nevertheless, other characteristics including length of bugles, along with the length and width of pores are considered more important than other evidence in terms of delimitation of these taxa. Generally, the mentioned evidence can be useful in delimitation of taxa when accompanied with other evidence.

Three apertures were seen across all studied species as a well-known characteristic in Onagraceae (Albert & al. 2010). Regardless of this, pollen with more and less than three apertures has also been seen in the mentioned family (Ting 1966; Brown 1967; Skvarla & al. 1978; Praglowski & al. 1994; Punt & al. 2003). Besides, Skvarla & al. (2008) stated that *E. luteum* Pursh. showingthree to four apertures, and sometimes it lacks aperture.

Table 2. Pollen characters of studied taxa.

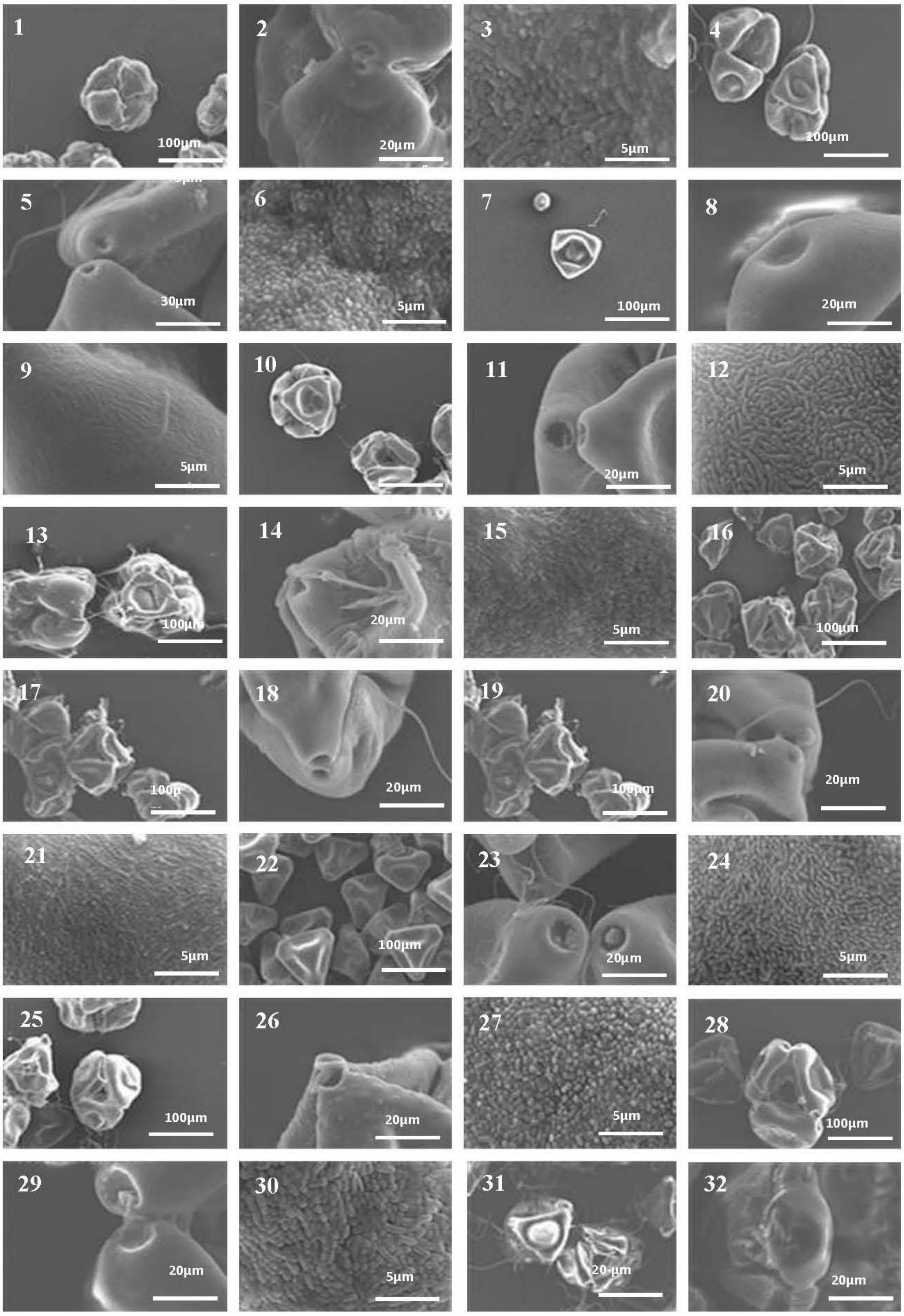
Species	Polar (µm)			Equtorial(µm)			P/E		length pore(µm)			Arm diameter(µm)			Ornamentation length(µm)			Ornamentation width(µm)			p/w		shape	Pollen unit	sculpture
	Var (µm)	SD (µm)	M (µm)	Var (µm)	SD (µm)	M (µm)	Var (µm)	M (µm)	Var (µm)	SD (µm)	M (µm)	Var (µm)	SD (µm)	M (µm)	Var (µm)	SD (µm)	M (µm)	Var (µm)	SD (µm)	M (µm)	Var (µm)	M (µm)			
Circaea. lutetiana	30-50	10	40	30-50	10	40	0.75-1.33	1	10-30	6.3	19	10-12	0.38	11.2	0.44-1	0.14	0.76	0.3-0.57	0.07	0.41	0.93-2.31	1.7	ta	monad	rugulae-granulate
Epilobium algidum	31.09-64.92	7.78	47.79	34.33-67.26	9.21	51.63	0.67-1.33	0.92	5.79-11.57	2.38	9	6.91-11.86	1.29	8.85	0.24-0.56	0.07	0.35	0.16-0.32	0.04	0.23	0.96-2.22	1.5	ta-sp	tetrad	granulate
E. anatolicum	50-80	10	60	50-80	10	70	0.71-1.16	0.86	10-20	5.1	1٥	20-30	4	22	0.3-1.35	0.28	0.72	0.19-0.3	0.03	0.25	1.11-7.10	2.88	ta	tetrad	rugulae-granulate
E. angustifolium	50-80	10	60	60-80	10	70	0.71-1.33	0.86	10-20	3.1	19	10-20	4.7	13	0.21-0.47	0.09	0.32	0.17-0.28	0.03	0.23	0.87-2.23	1.4	ta-sp	monad	rugulae-granulate
E. confusum	40-70	10	50	50-70	10	60	0.66-1.4	0.84	10-20	3.7	18.5	10-20	4.7	1٧	0.19-0.40	0.06	0.28	0.15-0.26	0.03	0.21	0.88-2.10	1.33	ta	tetrad	rugulae
E. dodonaei	40-70	10	60	50-80	10	70	0.66-1.4	0.86	10-20	4.1	18	10-20	4.9	16.5	0.30-0.91	0.15	0.52	0.17-0.31	0.03	0.25	1.18-3.79	2.08	ta-sp	monad	verrucate
E. stevenii	60-80	10	70	60-90	10	70	0.75-1.16	1	10-20	4.7	13	10-20	5.7	17	0.35-0.63	0.08	0.49	0.19-0.32	0.03	0.26	1.4-2.73	1.88	ta	monad	rugulae-granulate
E. frigidum	40-70	10	60	50-70	10	60	0.71-1.4	1	10-20	4.1	12	10-20	4.5	12.5	0.32-0.99	0.19	0.50	0.17-0.30	0.04	0.24	1.22-4.71	2.08	ta	tetrad	0verrucate-granulate
E. gemmascens	60-80	10	70	60-90	10	70	0.75-1.16	1	10-20	5.5	19	20-30	5	22	0.36-1	0.17	0.59	0.20-0.34	0.03	0.28	1.12-3.39	2.1	ta	tetrad	rugulae-granulate
E. hirsutum 1	60-90	10	80	50-90	10	70	0.75-2	1.15	10-20	5.5	19	10-30	5.7	17	0.62-1.49	0.24	0.93	0.33-0.49	0.05	0.40	1.63-4.13	2.22	ta	tetrad	verrucate
E. hirsutum 2	53.42-81.01	7.43	68.64	64.22-85.03	5.07	75.31	0.69-1.12	0.91	7.84-16.91	1.97	11.77	11.37-19.02	2.36	15.54	0.27-0.48	0.06	0.37	0.19-0.31	0.03	0.24	1.19-2.05	1.54	ta	tetrad	rugulae
E. lanceolatum	40-70	10	60	50-80	10	60	0.50-1.4	1	10-20	6	15	10-30	3.1	11	0.32-0.93	0.13	0.55	0.20-0.33	0.04	0.27	1.28-3.08	2.03	ta	tetrad	rugulae-granulate
E. minutiflorum	40.22-58.84	5.12	52.39	37.48-64.46	5.95	54.78	0.75-1.44	0.95	3.72-15	3.52	8.72	10.5-22.5	3.29	16	0.36-0.62	0.07	0.44	0.20-0.29	0.02	0.24	1.34-2.65	1.83	ta	tetrad	rugulae-granulate
E. montanum	60-70	10	70	60-80	10	70	0.75-1.16	1	10-20	4.7	13	10-30	4.7	13	0.31-0.88	0.11	0.51	0.23-0.33	0.03	0.27	1.14-3.82	1.88	ta	tetrad	striate
E. palustre	40-80	10	60	50-80	10	60	0.57-1.16	1	10-30	4.4	11	10-30	6.4	15	0.29-0.77	0.14	0.42	0.18-0.30	0.03	0.23	1.3-3.20	1.82	ta	tetrad	granulate
E. parviflorum	40-80	10	60	50-80	10	70	0.50-1.4	0.85	10-20	4.5	19	10-30	3.7	11.5	0.22-0.69	0.12	0.42	0.17-0.28	0.03	0.23	1.24-3	1.82	ta	tetrad	rugulae-granulate
E. ponticum	50-80	10	70	60-80	10	70	0.83-1.16	1	10-20	5.5	18.4	10-30	4.4	12.5	0.25-0.44	0.05	0.35	0.16-0.26	0.03	0.22	1.11-2.27	1.6	ta	tetrad	rugulae
E. rechingeri	53.26-73.98	6.16	64.63	57.81-71.15	4.11	64.07	0.75-1.22	1	6.3-10.81	1.37	8.93	9.41-22.32	2.7	17.04	0.31-0.76	0.12	0.50	0.20-0.29	0.03	0.24	1.29-3. 8	2.08	ta	tetrad	rugulae-granulate
E. roseum	40-90	10	60	50-70	10	60	0.57-1.5	1	10-20	3.2	14	10-30	3.8	18.3	0.21-0.62	0.09	0.38	0.16-0.29	0.03	0.23	1.03-3.43	1.65	ta	tetrad	rugulae-granulate
E. tetragonum	47.05-80.46	7.96	63.49	45.03-67.52	6.30	55.74	0.90-1.36	1.14	6.87-15.56	2.41	10.9	7.32-18.07	2.81	11.42	1.01-2.22	0.28	1.48	.27-.58	0.07	0.43	1.74-8.22	3.44	ta	tetrad	rugulae
Oenothera. biennis	60-90	10	80	70-90	10	80	0.60-1.28	1	16.06-36	4.87	21.69	13.55-26.91	3.35	18.83	0.27-0.40	0.03	0.34	0.25-0.38	0.04	0.33	0.84-1.37	1.03	ta	monad	rugulae-granulate

Viscin threads are present across all the studied taxa. However, some authors (Liao 1962; Ting 1966; Brown 1967) insisted on existence of viscin threads to determine delimitation of taxa along with identification of species in Onagraceae. These structures have also been reported in fossil pollen of Onagraceae (Zetter & Kerim 1987). Incidence of these threads as an adaptive character is considered a syndrome of zoophilous pollination (Zetter & Hesse 1996). The mentioned threads originated from the proximal pole, which has been confirmed in previous studies (Skvarla & al. 1978). Moreover, our results are in accordance with recent opinion. Regardless in other taxa of family mainly reported near the center of the proximal pole (Patel & al. 1984, Punt & al. 2003). Viscin threads hold monads in dehisced anthers (Schleiden 1846; Tschistiakoff 1866; Muller 1883; Beer 1904; Zander 1935; Lepouse & Romain 1967). Also, it plays an important role in transportation of pollen by pollinators (Piel 1971; Proctor & Yeo 1973).

PCOA and Cluster analysis of pollen characteristics did not clearly show sectional divisions based on Flora Iranica (Raven 1964) as well as Flora of Iran (Azizian 2006). Nevertheless, phylogenetic affinities of the mentioned family at the generic level are now relatively clear (Grimsson & al. 2012). However, some biological events that follow hybridization have appeared in the form of several systematic complexities within Onagraceae. Haussknecht (1884) observed easy hybridization in Sect. *Epilobium.* Moreover, other authors (Brockie 1966, 1970; Raven 1967; Seavy & Raven 1977; Chen & al. 1992; Krakos 2011) have also confirmed these events. Due to these conditions, assessment of all characteristics seems necessary for clarifying affinities and delimitation of species in Onagraceae.

One of the most important achievements of this study was empowerment of the intra-specific variations in *E. hirsutum* senso lato. This problematic structure has led to errors in identification of some populations of *E. hirsutum* senso stricto. However, different populations in several Iranian herbaria have been determined as similar species, regardless to the distinctions in terms of obvious variation in density of leaf trichomes. Pollen characters confirmed this divergence by clear distinctions.

This study is an incomplete but extended research on Onagraceae, as it provides valuable data on pollen morphology of the family in Iran as well assessment of their importance in taxonomy. There are not significant differences in size, shape, pore structure and arm characteristics to delimit all species in Onagraceae. Moreover, Pollen grains are not efficient for generic delimitation in the studied taxa, regardless to noticeable dissimilarities in size, shape, wall thickness as well pore structure which can delimit some species in several genera in Onagraceae (Brown 1967). Therefore, even though the taxa cannot be distinguished exclusively by pollen characters, this characterization can be a useful tool for distinguishing some studied taxa of Onagraceae along with other evidence. Moreover, the present study provides useful basic data in taxonomic studies of the family.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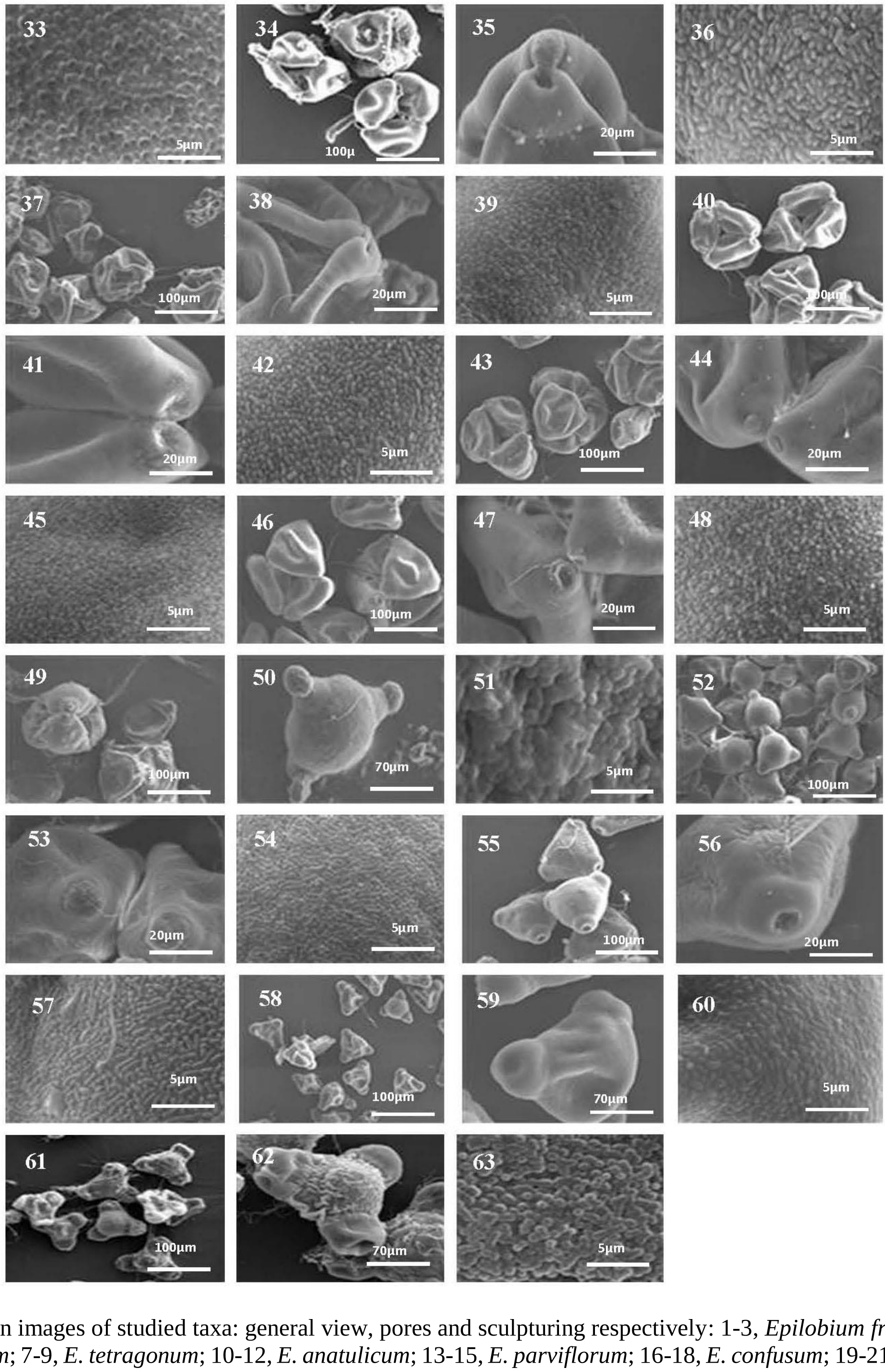

Fig. 1. Pollen images of studied taxa: general view, pores and sculpturing respectively: 1-3, *Epilobium frigidum*; 4-6, *E. montanum*; 7-9, *E. tetragonum*; 10-12, *E. anatulicum*; 13-15, *E. parviflorum*; 16-18, *E. confusum*; 19-21, *E. roseum*; 22-24, *E. stevenii*; 25-27, *E. gemmascens*; 28-30, *E. hirsutum*1; 31-33, *E. hirsutum*2; 34-36, *E. lanceolatum*; 37-39, *E. minutiflorum*; 40-42, *E. palustre*; 43-45, *E. ponticum*; 46-48, *E. rechingeri*; 49-51, *E. algidum*; 52-54, *E. angustifolium*; 55-57, *E. dodonaei*; 58-60, *Circaea lutetiana*; 61-63, *Oenothera biennis*.

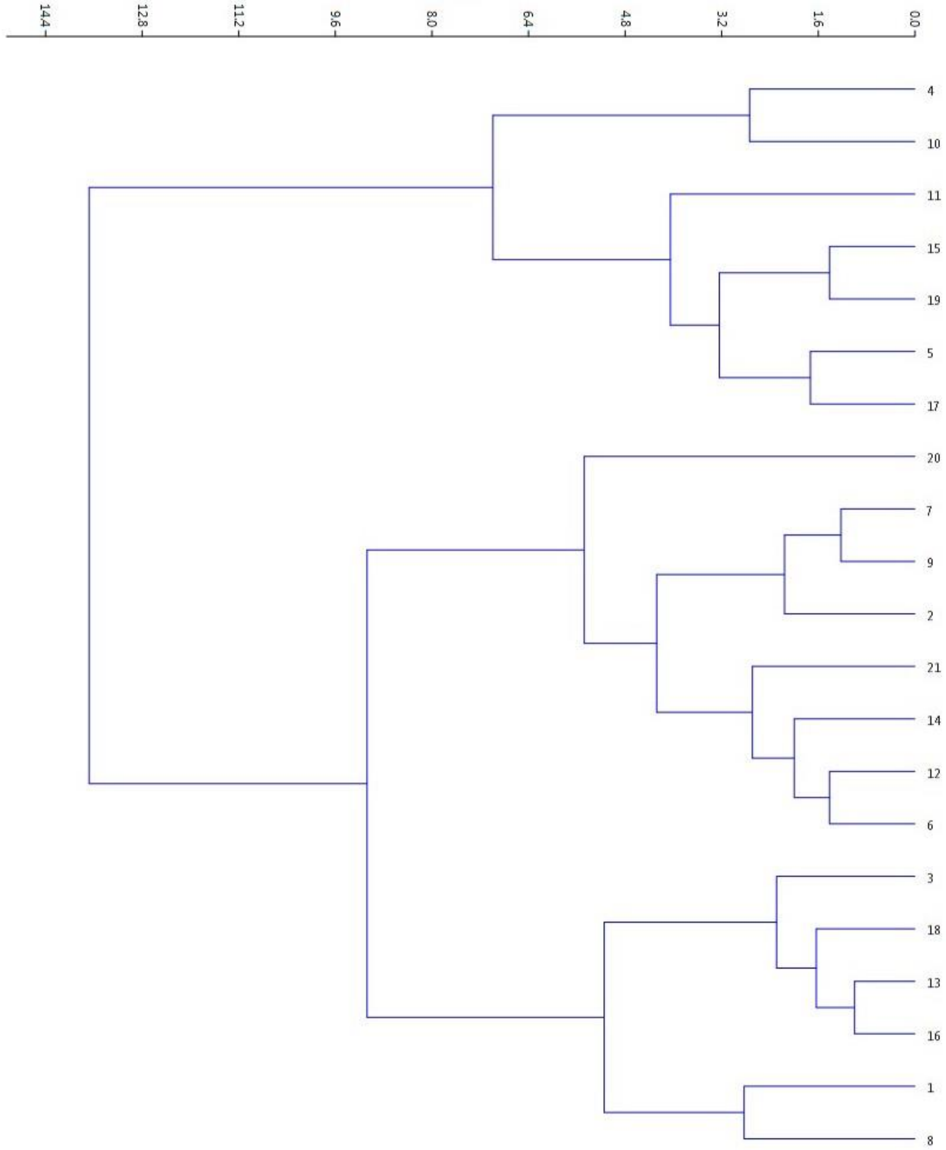

Fig. 2: Ward dendrogram of studied taxa based on pollen evidence: 1, *Epilobium algidum*; 2, *E. anatolicum*; 3, *E. angustifolium*; 4, *E. tetragonum*; 5, *E. confusum*; 6, *E. roseum*; 7, *E. stevenii*; 8, *E. frigidum*; 9, *E. gammascens*; 10, *E. hirsutum*1; 11, *E. hirsutum*2; 12, *E. lanceolatum*; 13, *E. minutiflorum*; 14, *E. montanum*; 15, *E. palustre*; 16, *E. parviflorum*; 17, *E. ponticum*; 18, *E. rechingeri*; 19, *E. dodonaei*; 20, *Circaea lutetiana*; 21, *Oenothera biennis.*

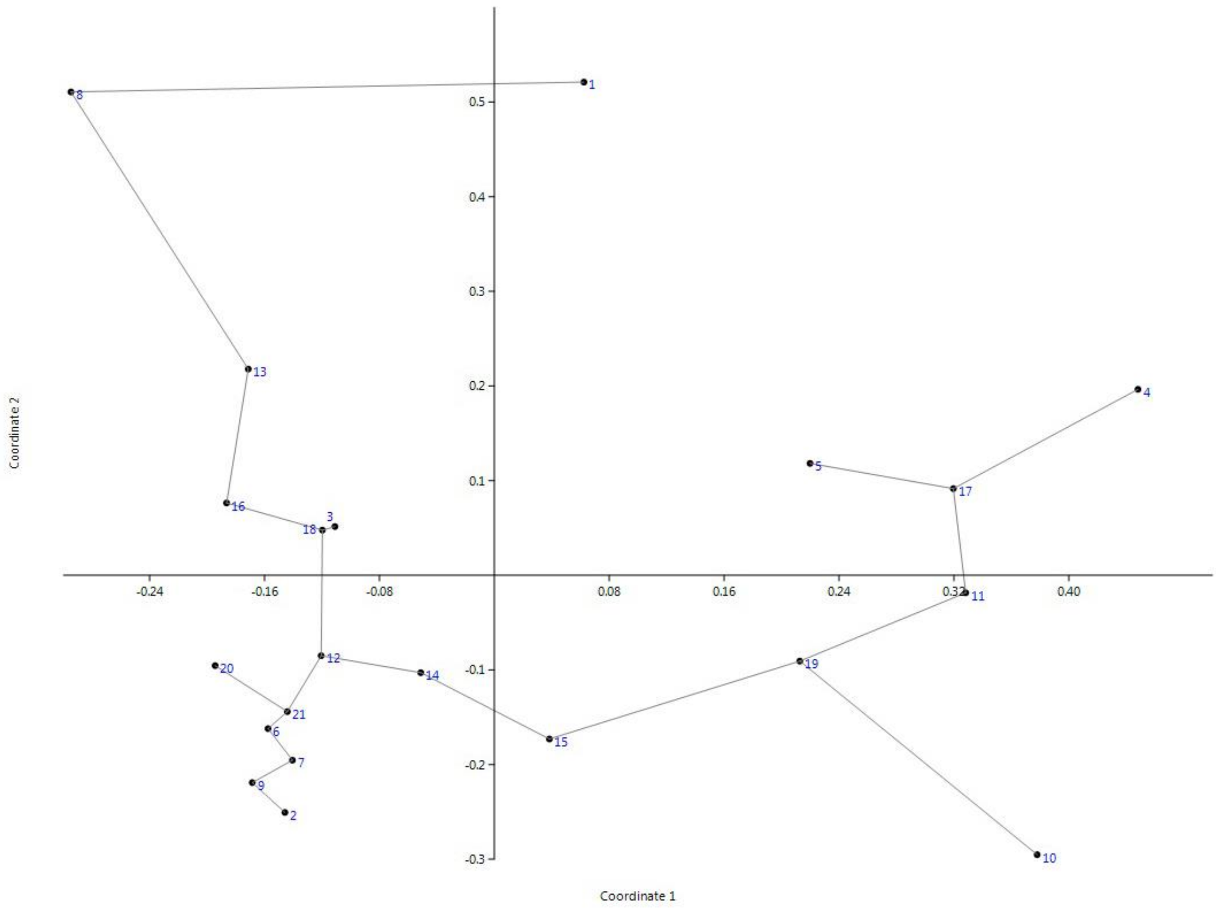

Fig. 3. PCA plot of studied taxa based on pollen evidence: 1, *Epilobium algidum*; 2, *E. anatolicum*; 3, *E. angustifolium*; 4, *E. tetragonum*; 5, *E. confusum*; 6, *E. roseum*; 7, *E. stevenii*; 8, *E. frigidum*; 9, *E. gammascens*; 10, *E. hirsutum*1; 11, *E. hirsutum*2; 12, *E. lanceolatum*; 13, *E. minutiflorum*; 14, *E. montanum*; 15, *E. palustre*; 16, *E. parviflorum*; 17, *E. ponticum*; 18, *E. rechingeri*; 19, *E. dodonaei*; 20, *Circaea lutetiana*; 21, *Oenothera biennis.*

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