

**Pollen micromorphology of some species of *Gagea* in Iran and its taxonomic implication**

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Pollen grains of eight species of *Gagea* were examined using Light Microscope (LM) and Scanning Electron Microscope (SEM). Results showed that, selected quantitative characters of pollen grains (polar axis, equatorial axis, P/E ratio, width of muri, lumina or pore number per 2  $\mu\text{m}^2$  in the exine surface, and lumina or pore size) show significant variation providing potential diagnostic characters in discrimination of taxa in the genus. Among the pollen qualitative characters, pollen shapes varied from oblate to oblate-spheroid, and pollen size can be classified as medium (less than 50  $\mu\text{m}$ ) and large (more than 50  $\mu\text{m}$ ). Exine ornamentation in equatorial view and on sulcus margin region show also variation among the studied taxa. Cluster analysis of selected pollen morphological characters indicated two pollen groups and three types. The palynological types supported taxonomical situation of *G. sect. Didymobulbos*, *G. sect. Plecostigma*, and *G. sect. Platyspermum*. There is no pollen type for studied taxa of *G. sect. Stipitatae*. In addition, palynological issues suggest a close relationship between *G. sect. Didymobulbos* and *G. sect. Stipitatae*.

**Keywords:** Exine ornamentation, *Liliaceae*, pollen group, pollen type, sulcus margin**گرده‌شناسی برخی گونه‌های جنس *Gagea* در ایران و کاربرد تاکسونومیکی آن\***

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ویژگی‌های ریزریخت‌شناسی دانه گرده در هشت گونه از جنس *Gagea* (سوسنیان) به وسیله میکروسکوپ نوری (LM) و میکروسکوپ الکترونی نگاره (SEM) مورد بررسی قرار گرفت. نتایج نشان داد که صفات انتخابی کمی ریزریخت‌شناسی دانه گرده (طول قطبی، طول استوایی، نسبت طول قطبی به استوایی، ضخامت muri و تعداد و اندازه lumina و روزنه‌ها در سطح آگزین) متفاوت بودند. همچنین، در مورد صفات کیفی دانه گرده (شکل و اندازه)، به ترتیب بیضوی پهن تا کروی-بیضوی و متوسط و بزرگ مشاهده شد. تزیینات آگزین در سطح استوایی و کنار شیار در گونه‌های مورد بررسی نیز متفاوت بود. دندروگرام به دست آمده، گونه‌های مورد بررسی را در دو گروه و سه تیپ تقسیم‌بندی نمود. تیپ‌های گرده‌ای از موقعیت تاکسونومیکی *G. sect. Plecostigma*، *G. sect. Didymobulbos* و *G. sect. Platyspermum* حمایت نمودند. از آرایه‌های مورد بررسی در *Gagea sect. Stipitatae* تیپ گرده‌ای تشکیل نشد. همچنین، گروه‌های گرده‌ای تا حدودی ارتباط تاکسونومیکی *G. sect. Didymobulbos* همراه با *G. sect. Stipitatae* نشان دادند.

**واژه‌های کلیدی:** تزیینات آگزین، تزیینات کنار شیار، تیپ گرده، سوسنیان، گروه گرده

## Introduction

*Gagea* Salisb. is one of the largest genera of *Liliaceae* with approximately 300 species in the world, especially Irano-Turanian (IT) floristic region [(central diversity of *Gagea* sections *Didymobulbos*, *Platyspermum*, *Plecostigma*, and *Stipitatae*) (Peterson *et al.* 2008, Zarrei *et al.* 2011, Tison *et al.* 2013, Kayıkçı *et al.* 2014, Peruzzi 2016, Peterson *et al.* 2019)]. Previously, some infrageneric classifications were provided by some taxonomists such as Koch (1849), Boissier (1882), Terracciano (1905a,b), Terracciano (1906), Pascher (1904), Pascher (1907), Stroh (1937), and Uphof (1958–60). Some earlier researches also tried to recommend an appropriate taxonomical classification especially based on the study of Levichev (1990), Peterson *et al.* (2008), and Zarrei *et al.* (2009). The taxonomical situation of *Gagea* sections *Didymobulbos*, *Platyspermum*, and *Stipitatae* showed to be heterogenous (Peterson *et al.* 2008, Zarrei *et al.* 2011, Peruzzi 2012). In this way, number of sections in the taxa of this genus decreased or increased (Peterson *et al.* 2008, Peruzzi *et al.* 2008, Zarrei *et al.* 2009, Zarrei *et al.* 2011, Tison *et al.* 2013, Tekşen & Erkul 2015, Peterson *et al.* 2019). Based on taxonomical research, 13 sections were proposed by Levichev (*l.c.*), Peterson *et al.* (*l.c.*), and Zarrei *et al.* (*l.c.*). The infrageneric of this genus were increased to at least 14 sections based on morphological, anatomical, and ontogenetic characters provided by Peruzzi *et al.* (2008), Peruzzi (2011), Tison *et al.* (2013), and Tekşen & Erkul, (2015). However, based on variations in morphological characters, Zarrei *et al.* (2011) revised the infrageneric classification of this genus and divided it into only seven sections.

In the Flora of Iran, diversity of *Gagea* sections *Didymobulbos*, *Platyspermum*, and *Stipitatae* (Zarrei *et al.* 2011) is appropriately undertaken. An extensive study is also done on the palynological status of *Gagea* by Zarrei & Zarre (2005) to distinguish taxonomical relationship of the genus. This research aimed to update the palynological data from the mentioned problematic sections of *Gagea* in Iran.

## Materials and Methods

The pollen grain samples were obtained from the herbarium of the Ferdowsi University of Mashhad (FUMH), Mashhad (Iran) which are listed in table 1. Pollen grains were acetolysed according to Erdtman's technique (1960) for light microscopy. Accordingly, 30 pollen grains of each studied species were measured by Leitz Light Microscope (HM-LUX3) and the images were then taken by Dino Camera (AM-423). For SEM preparation, the dried pollen grains were transferred to stubs, coated with gold, and examined using a Scanning Electron Microscope (JEOL-JSM-840). The pollen terminologies were explored by Punt *et al.* (2007), Hesse *et al.* (2009), and Halbritter *et al.* (2018). The qualitative pollen grain micromorphological characters were coded (Table 2). MVSP (Multi-Variate Statistical Package) software along with UPGMA (Un Weighted Pair-Group Analysis) method based on Euclidean distances was applied to construct of palynological dendrogram from quantitative and qualitative micromorphological and ultrastructure characters. Principal Component Analysis (PCA) by MVSP software was used both for the studied species (as PCA case scores) as well as the pollen grain characters (as PCA variable loadings) (Kovach 1999).

## Results

### - Light microscopy observations

In this research, pollen quantitative characters such as polar and equatorial axes (minimum, maximum, and average measurements) and P/E ratio were measured and listed in table 2. In addition, two pollen qualitative characters (pollen shape and pollen size type) were detected for all studied species (Table 2). Oblate pollen shapes were seen in all studied taxa except *G. vegeta* which was oblate-spheroid (Table 2, Fig. 1).

Large pollen type or size observed in *G. chomutovae* (more than 50  $\mu\text{m}$ ) and the rest had medium pollen type or size based on the average size of equatorial axis (less than 50  $\mu\text{m}$ ) (Table 2).

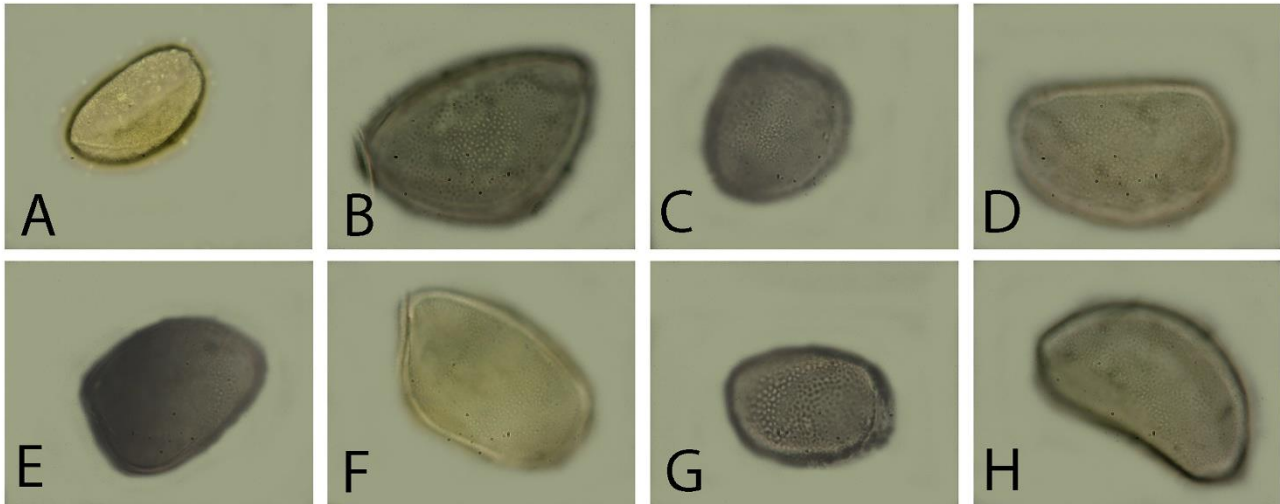
**Table 1.** Information about the studied species in the present research [All specimens are deposited in Herbarium of the Ferdowsi University of Mashhad (FUMH).]

Taxon	Locality along with related data
<i>Gagea afghanica</i> A.Terracc	S. Khorasan prov.: Tabas, 12.3.1995, Joharchi & Zangooei (FUMH 26168)
<i>G. chomutovae</i> (Pascher) Pascher	Razavi Khorasan prov.: Quchan, Garmab, 20.4.1991, Joharchi & Zangooei, (FUMH 19369)
<i>G. dschungarica</i> Regel.	N. Khorasan prov.: Bojnurd, 28.5.2006, Memariani & Zangooei (FUMH 37818)
<i>G. kunawurensis</i> (Royle) Greuter Syn.: <i>G. ova</i> Stapf.	Razavi Khorasan prov.: Chenaran, 21.4.1991, Joharchi & Zangooei (FUMH 19388)
<i>G. reticulata</i> (Pall.) Schult. et Schult.f.	N. Khorasan prov.: Torbat-e Jam, 4.4.1991, Joharchi & Zangooei (FUMH 19217)
<i>G. setifolia</i> Baker	Razavi Khorasan prov.: Gonabad, 9.4.1991, Joharchi & Zangooei (FUMH 19254)
<i>G. tenera</i> Pascher	Razavi Khorasan prov.: Quchan, 15.4.2008, Joharchi & Zangooei (FUMH 39418)
<i>G. vegeta</i> Vved.	N. Khorasan prov.: Bojnurd, 17.4.2008, Joharchi & Zangooei (FUMH 39510)

**Table 2.** Pollen grains quantitative and qualitative characters from LM and SEM observation in the examined taxa

Taxon	Polar axis ( $\mu\text{m}$ ) (Min., Mean $\pm$ SD, Max.)	Equatorial axis ( $\mu\text{m}$ ) (Min., Mean $\pm$ SD, Max.)	P/E	Pollen shape	Pollen size	E <sub>0</sub>	Se <sub>0</sub>	Sa <sub>0</sub>	Wm ( $\mu\text{m}$ ) (Min. & Max.)	N	Lo ( $\mu\text{m}$ ) (Min. & Max.)
<i>Gagea afghanica</i>	20.0–27.9 $\pm$ 2.4–40.0	32.0–39.4 $\pm$ 1.7–49.0	0.70	Oblate	Medium	Pt	P	T	-	3	0.1–0.5
<i>G. chomutovae</i>	25.0–31.8 $\pm$ 3.8–41.0	37.5–51.9 $\pm$ 6.8–89.0	0.61	Oblate	Large	Tp	Tp	T	-	4	0.1–0.3
<i>G. dschungarica</i>	25.0–28.3 $\pm$ 1.3–30.0	37.5–41.4 $\pm$ 4.2–51.0	0.68	Oblate	Medium	Tp	Tp	T	-	3	0.1–0.4
<i>G. kunawurensis</i>	25.0–33.6 $\pm$ 4.7–42.0	37.5–47.8 $\pm$ 5.3–72.0	0.70	Oblate	Medium	Mp	Tp	T	-	1	0.2–0.9
<i>G. reticulata</i>	22.5–27.6 $\pm$ 5.4–41.0	37.5–47.7 $\pm$ 3.6–69.0	0.57	Oblate	Medium	Fs	Sf	T	-	5	0.1–0.3
<i>G. setifolia</i>	22.5–27.9 $\pm$ 2.3–40.0	37.5–44.1 $\pm$ 4.6–65.0	0.63	Oblate	Medium	Mc	F	T	0.3–0.7	2	0.2–0.9
<i>G. tenera</i>	17.5–24.8 $\pm$ 2.8–36.0	35.0–41.0 $\pm$ 2.8–63.0	0.60	Oblate	Medium	Rc	Mc	T	0.4–1.0	1	0.2–1.6
<i>G. vegeta</i>	27.5–32.5 $\pm$ 3.6–37.5	37.5–41.2 $\pm$ 3.1–50.0	0.78	Oblate-spheroid	Medium	Ts	Ts	T	-	-	-

E<sub>0</sub>: Exine ornamentation, Pt: Perforate-tuberculate, Tp: Tuberculate-perforate, Mp: Macrotuberculate-perforate, Fs: Foveolate-striate, Mc: Microreticulate-cristatate, Rc: Reticulate-cristatate, Ts: Tuberculate-striate, Se<sub>0</sub>: Sulcus margin ornamentation, P: Perforate, Sf: Striate-foveolate, F: Foveolate, Sa<sub>0</sub>: Sulcus membrane ornamentation, T: Tuberculate, Wm: Width of muri, N: Lumina or pore number per 2  $\mu\text{m}^2$  in the exine surface, and Lo: Lumina or pore size.

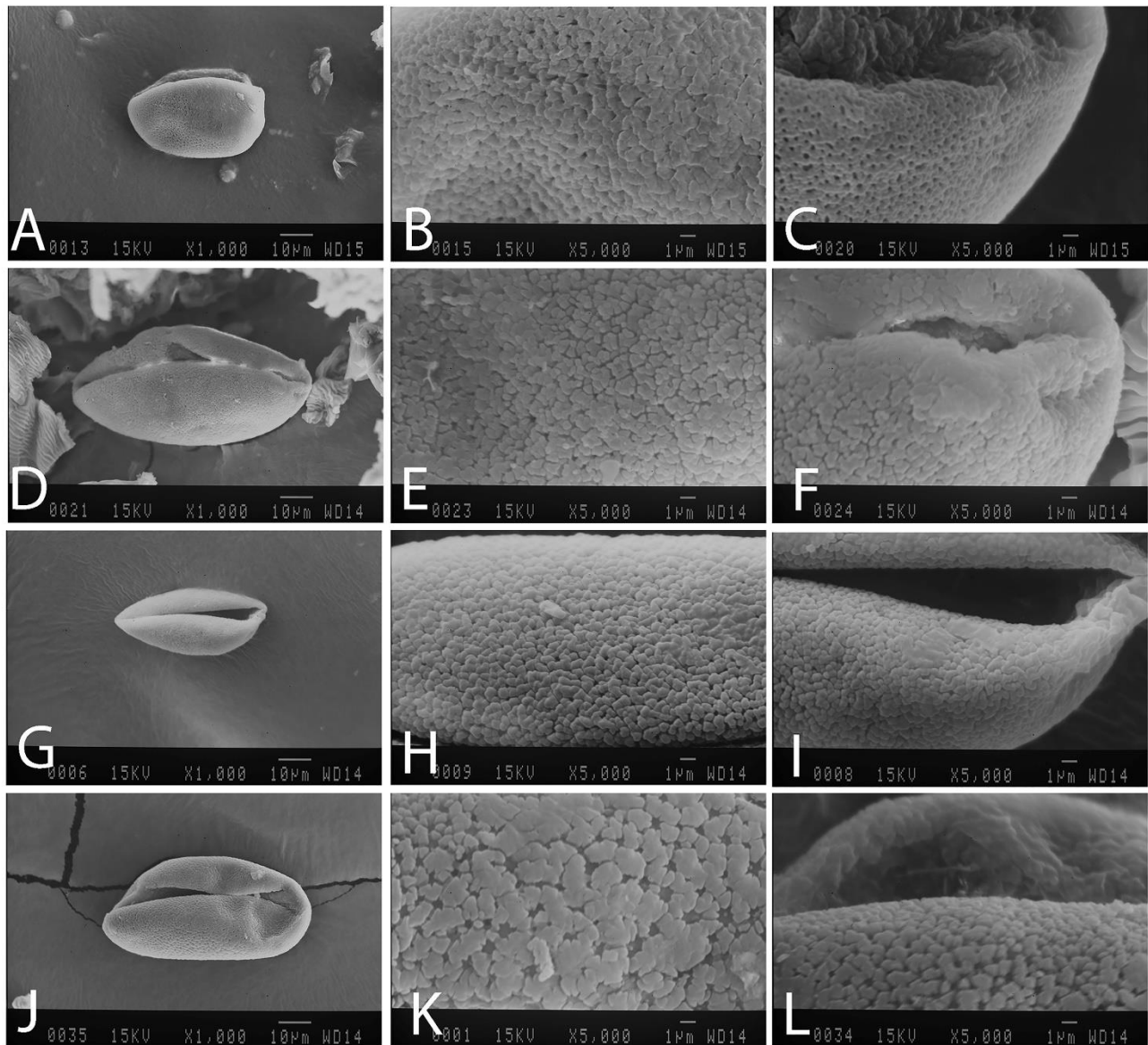


**Fig. 1.** LM micrographs of the pollen grains (Equatorial view): A. *Gagea afghanica*, B. *G. chomutovae*, C. *G. dschungarica*, D. *G. kunawurensis*, E. *G. reticulata*, F. *G. setifolia*, G. *G. tenera*, H. *G. vegeta* (Bars = 10  $\mu\text{m}$ ).

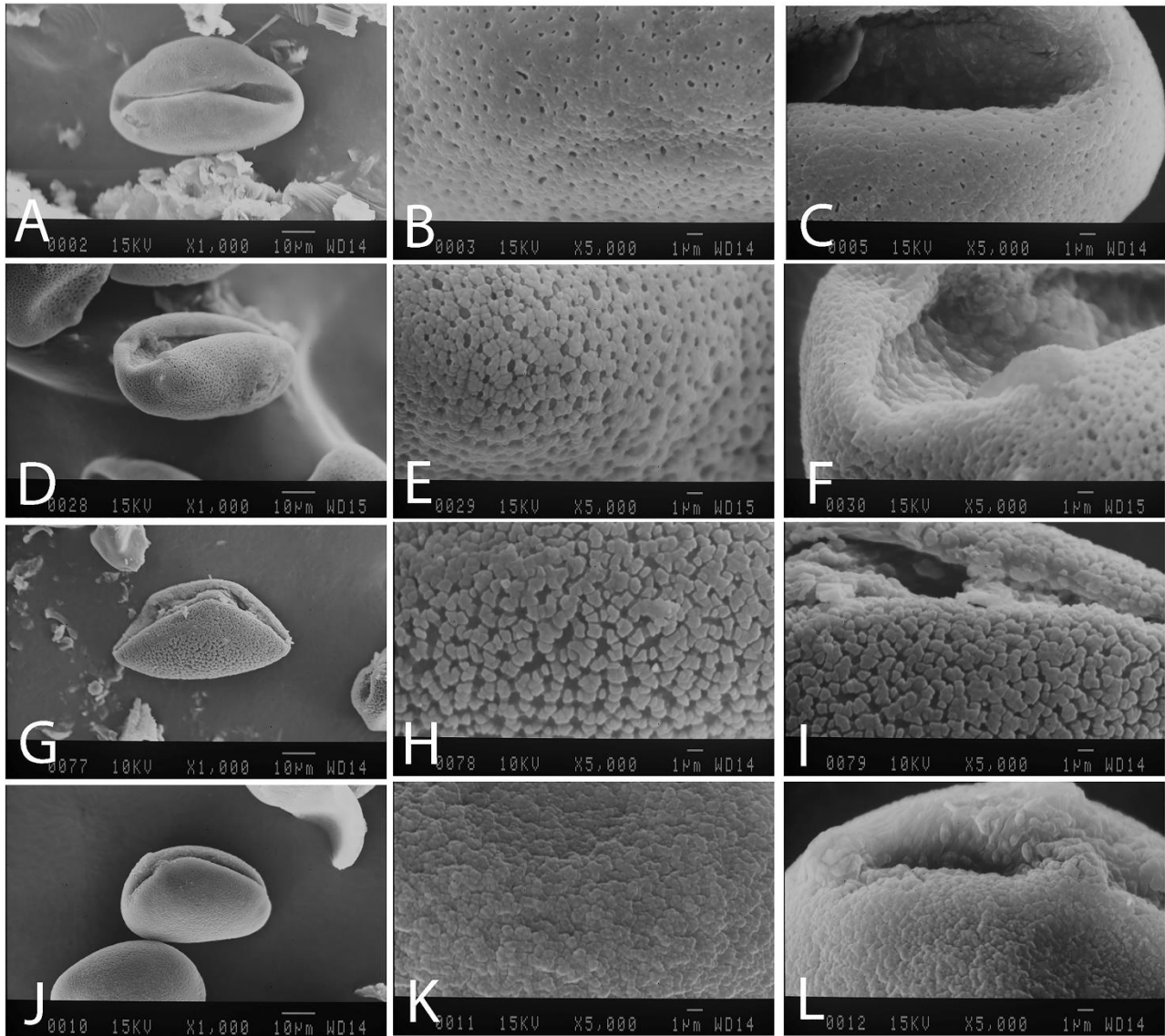
#### - Scanning electron microscopy observations

Four pollen qualitative characters including exine ornamentation of the equatorial surface, sulcus margin ornamentation, and sulcus membrane ornamentation along with lumina per 2  $\mu\text{m}^2$  in the exine surface were examined (Table 2, Figs 1–2). Seven types of pollen equatorial surface exine ornamentation were observed in the studied taxa as follows: perforate-tuberculate (*Gagea afghanica*) (Fig. 2B), tuberculate-perforate (*G. chomutovae* and *G. dschungarica*) (Figs 2E, 2H), macrotuberculate-perforate (*G. kunawurensis*) (Fig. 2K), foveolate-striate (*G. reticulata*) (Fig. 3B), microreticulate-cristate (*G. setifolia*) (Fig. 3E), reticulate-cristate (*G. tenera*) (Fig. 3H), and tuberculate-striate (*G. vegeta*) (Fig. 3K). In addition, sulcus margin ornamentation had six kinds of exine ornamentations (Table 2). *Gagea chomutovae*, *G. dschungarica*, and *G. kunawurensis* had tuberculate-perforate sulcus margin ornamentation (Figs 2F, 2I, 2L). This ornamentation was perforate, striate-foveolate, foveolate, microreticulate-

cristate, and tuberculate-tri-ate in *G. afghanica*, *G. reticulata*, *G. setifolia*, *G. tenera*, and *G. vegeta*, respectively (Figs 2C, 3C, 3F, 3I, 3L). Sulcus membrane ornamentation was similar in all studied taxa (tuberculate) (Table 2, Figs 2C, 2F, 2I, 2L, 3C, 3F, 3I, 3L). The width of muri and lumina size (*G. setifolia* and *G. tenera*) and pore size (the other studied taxa) varied in the exine ornamentation of the equatorial surface (Table 2). There were 1–5 lumina or pores counted per 2  $\mu\text{m}^2$  exine surface in the studied taxa except *G. vegeta* (Table 2). *Gagea kunawurensis* and *G. tenera* which had the fewest lumina or pore on the equatorial exine surface (1 per 2  $\mu\text{m}^2$ ) (Table 2). The number of lumina in the exine surface region of *G. reticulata* (Five per 2  $\mu\text{m}^2$ ) was higher than other species. *Gagea afghanica* and *G. dschungarica* both had three lumina per 2  $\mu\text{m}^2$  in the equatorial exine surface (Table 2). Two lumina and four pores were seen in the equatorial exine surface of pollen in *G. setifolia* and *G. chomutovae* (Table 2).



**Fig. 2.** SEM micrographs of the pollen grains: A–C. *Gagea afghanica* (A. Equatorial view, B. Exine ornamentation, C. Sulcus membrane and sulcus margin ornamentation), D–F. *G. chomutovae* (D. Equatorial view, E. Exine ornamentation, F. Sulcus membrane and sulcus margin ornamentation), G–I. *G. dschungarica* (G. Equatorial view, H. Exine ornamentation, I. Sulcus membrane and sulcus margin ornamentation), J–L. *Gagea kunawurensis* (J. Equatorial view, K. Exine ornamentation, L. Sulcus membrane and sulcus margin ornamentation) (Bars = A, D, G, J: 10  $\mu$ m, B, C, E, F, H, I, K, L: 1  $\mu$ m).



**Fig. 3.** SEM micrographs of the pollen grains: A–C. *Gagea reticulata* (A. Equatorial view, B. Exine ornamentation, C. Sulcus membrane and sulcus margin ornamentation), D–F. *G. setifolia* (D. Equatorial view, E. Exine ornamentation, F. Sulcus membrane and sulcus margin ornamentation), G–I. *G. tenera* (G. Equatorial view, H. Exine ornamentation, I. Sulcus membrane and sulcus margin ornamentation), J–L. *G. vegeta* (J. Equatorial view, K. Exine ornamentation, L. Sulcus membrane and sulcus margin ornamentation) (Bars = A, D, G, J: 10  $\mu$ m, B, C, E, F, H, I, K, L: 1  $\mu$ m).

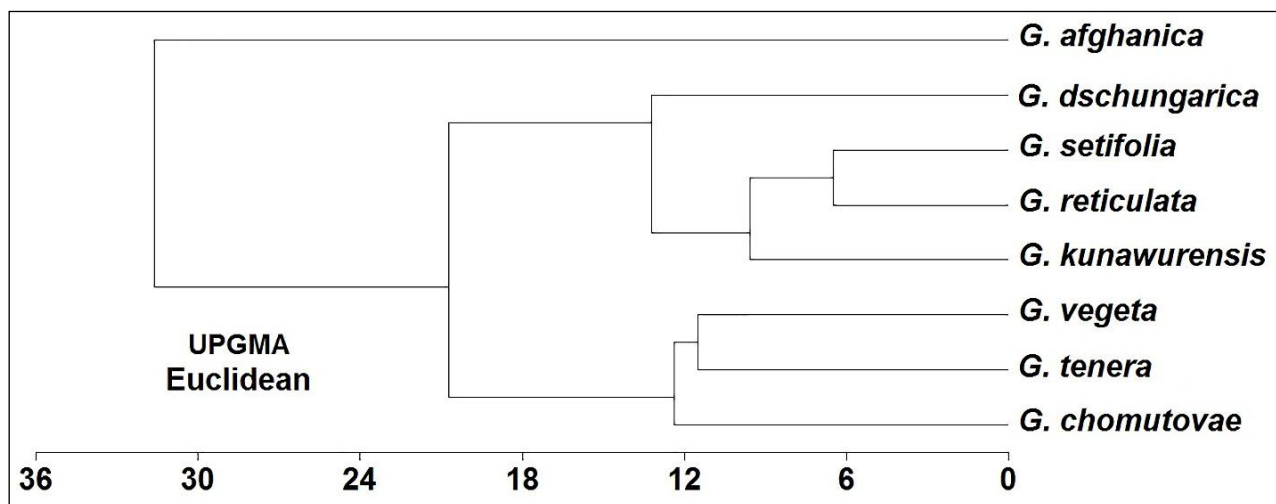
#### - Unweighted Pair-Group Analysis (UPGMA)

According to the UPGMA method based on Euclidean distances from the pollen grain characters under light microscopy and scanning electron microscopy observations, a dendrogram was constructed for eight species (Table 3, Fig. 3). Accordingly, *G. reticulata* and *G. setifolia* placed in node 1 (Table 3, Fig. 3). *Gagea kunawurensis* and node 1 placed in node 2 (Table 3, Fig. 3). *Gagea tenera* and *G. vegeta* constructed node 3 (Table 3, Fig. 3). *Gagea chomutovae* and node 3 constructed node 4 (Table 3, Fig. 3). Node 2 along with *G. dschungarica*

constructed node 5 (Table 3, Fig. 3). Nodes 4 and 5 placed in node 6 based on pollen micromorphological and ultrastructure characters (Table 3, Fig. 3). Node 7 included node 6 and *G. afghanica* (Table 3, Fig. 3). Based on placement of nodes, two pollen groups and three types were obtained as follows: type 1 or type *Plecostigma* (*G. afghanica*); group 1 included type 2 or type *Platyspermum* (*G. reticulata* and *G. setifolia*), *G. dschungarica*, and *G. kunawurensis*; group 2 included type 3 or type *Didymobulbos* (*G. tenera* and *G. vegeta*) and *G. chomutovae* (Fig. 3).

**Table 3.** The pollen grains micromorphological characters (variables) were analyzed in the studied species (cases)

Cluster analysis				
Analyzing 17 variables and 8 cases				
UPGMA				
Node	Group 1	Group 2	Dissimilarity	Object in group
1	<i>Gagea reticulata</i>	<i>Gagea setifolia</i>	6.481	2
2	<i>G. kunawurensis</i>	Node 1	9.565	3
3	<i>G. tenera</i>	<i>G. vegeta</i>	11.505	2
4	<i>G. chomutovae</i>	Node 3	12.377	3
5	Node 2	<i>G. dschungarica</i>	13.227	4
6	Node 4	Node 5	20.714	7
7	Node 6	<i>G. afghanica</i>	31.614	8



**Fig. 4.** Dendrogram of the studied species which was analyzed by MVSP software based on UPGMA method (numbers which located under of dendrogram indicate euclidean distances and nodes, respectively) from pollen data. Two pollen groups and two pollen types were obtained as follows: type 1 or type *Plecostigma* (*Gagea afghanica*); group 1 included type 2 or type *Platyspermum* (*G. reticulata* and *G. setifolia*), *G. dschungarica* and *G. kunawurensis*; group 2 included type 3 or type *Didymobulbos* (*G. tenera* and *G. vegeta*), and *G. chomutovae*.

#### - Principal Components Analysis (PCA)

The results of PCA including eigenvalues, percentages, and cumulative percentages for both axes, PCA case scores (as the studied species), and PCA variable loadings (as pollen grain micromorphological characters) which were obtained from LM and SEM observations are shown in table 4 and figure 4. There were three types and two groups of case scores in both axes: type 1: *Gagea afghanica* (positive in both axes), type 2: *G. reticulata* and *G. setifolia* (positive in axis 1 and negative in axis 2), and type 3: *G. vegeta* and *G. tenera* (negative in axis 1, positive and negative

in axis 2); group 1, type 2, *G. dschungarica* and *G. kunawurensis* (positive and negative in both axes); and group 2, type 3, *G. chomutovae* (negative in axis 1, positive and negative in axis 2) (Table 4, Fig. 4).

Based on the PCA variable loadings, one group (A) and seven characters were made. Characters Pi, Pm, Ei, and Em (minimum and average of polar and equatorial axes) (positive in both axes); character Ea (maximum of equatorial axis) (positive in axis 1 and negative in axis 2), and characters group A, including rest of the pollen characters (in the central part of both axes) (Table 4, Fig. 4).

The results of PCA analysis based on the studied species and the overlapping of pollen grain micromorphological characters showed that, pollen character Ea (minimum of equatorial axes) had major role in situation of types 1 in the palynological biplot (Fig. 4). Pollen characters including Ei and Em (minimum and average of equatorial axes) had major roles in the situation

of some members of group 1 (*G. kunawurensis*) (Fig. 4), although characters Pi and Pm (minimum and average of polar axes) had minor roles in the situation of all studied species. Rest of the studied pollen characters in the central part of the biplot (characters group A) showed major roles (Fig. 4).

**Table 4.** The pollen grain morphological characters analyzed by principal components analysis

	Axis 1	Axis 2
Eigen values	210.425	19.299
Percentage	83.28	7.638
Cumulative percentage	83.28	90.918

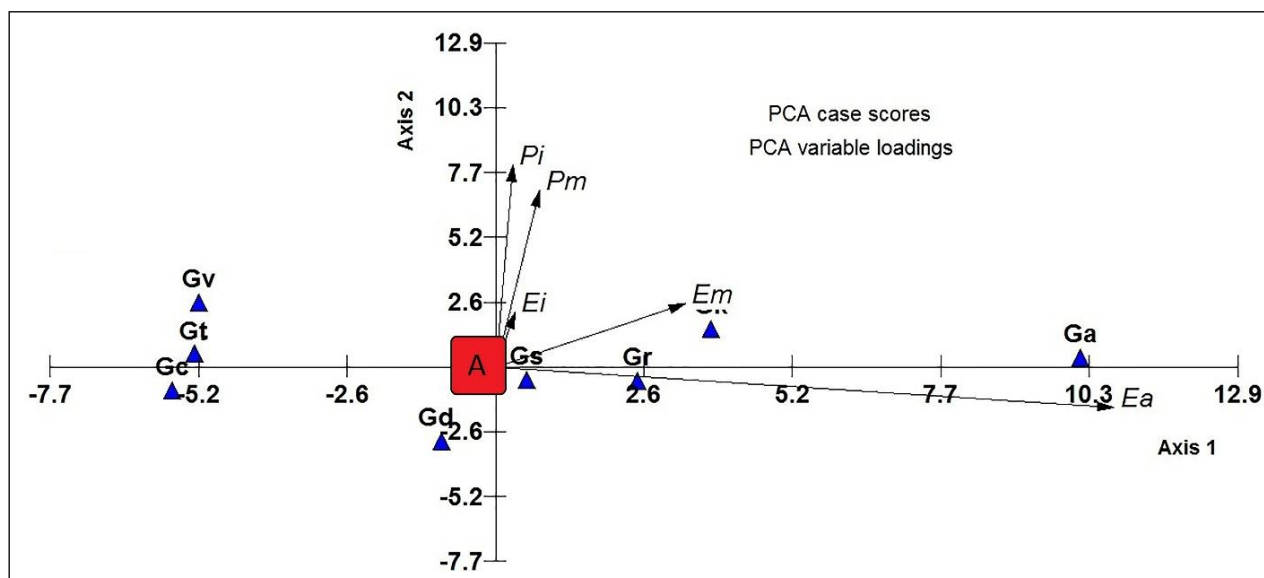
  

Pollen character	Abbreviation	Axis 1	Axis 2
Minimum of polar axis	Pi	0.026	0.702
Average of polar axis	Pm	0.067	0.612
Maximum of polar axis	Pa	0.16	0.061
Minimum of equatorial axis	Ei	0.029	0.19
Average of equatorial axis	Em	0.288	0.221
Maximum of equatorial axis	Ea	0.939	-0.142
P/E: Polar/equatorial ratio	P/E	-0.003	0.011
Pollen shape	Ps	-0.009	0.05
Pollen type	Pz	0.018	0.008
Exine ornamentation	Eo	-0.024	0.014
Sulcus margin ornamentation	Sm	-0.026	0.013
Sulcus membrane ornamentation	Se	0	0
Minimum width of muri	Mi	0	-0.026
Maximum width of muri	Ma	-0.001	-0.065
Minimum of lumina size	Li	0.002	-0.009
Maximum of lumina size	La	0.002	-0.082
Lumina or pore number per 2 $\mu\text{m}^2$	N	0.044	-0.096

Taxon	Abbreviation	Axis 1	Axis 2
<i>Gagea afghanica</i>	Ga	10.181	0.387
<i>G. chomutovae</i>	Gc	-5.618	-0.924
<i>G. dschungarica</i>	Gd	-0.939	-2.959
<i>G. kunawurensis</i>	Gk	3.742	1.489
<i>G. reticulata</i>	Gr	2.475	-0.549
<i>G. selifolia</i>	Gs	0.542	-0.509
<i>G. tenera</i>	Gt	-5.226	0.517
<i>G. vegeta</i>	Gv	-5.157	2.548





**Fig. 5.** Principal components analysis scatterplot obtained from overlapping of species (case scores) and pollen grain characteristics (variable loadings) loadings on both axes. Case scores (species): *Gagea afghanica* (Ga), *G. chomutovae* (Gc), *G. dschungarica* (Gd), *G. kunawurensis* (Gk), *G. reticulata* (Gr), *G. setifolia* (Gs), *G. tenera* (Gt), *G. vejeta* (Gv). Variable loading (pollen grain characters): characters Pi, Pm, Ei, and Em, and Ea (minimum and average of polar and equatorial axes) (positive in both axes), characters Ea (maximum of equatorial axis) (positive in axis 1 and negative in axis 2), and characters group A including rest of the pollen characters (in the central part of both axes).

## Discussion

Morphological features and phylogenetical relationships of taxa *Gagea* sections *Didymobulbos*, *Platyspermum*, and *Stipitatae* have been reported (Peterson et al. 2008, Zarrei et al. 2009, Peterson et al. 2019). The taxa of sect. *Stipitatae* (Pascher) Davlianidze were distributed among the other clades in different positions of phylogram (polytomy) (Peterson et al. 2008). In a later study, this section was included in sect. *Platyspermum* Boiss. (as a subsect.) forming a polytomy with other clades (Zarrei et al. 2009). Zarrei et al. (2011) revised the classification of this genus and placed sections *Platyspermum* and *Stipitatae* in *G.* sect. *Plecostigma* (Turcz.) Pascher.

In the present research, the taxa of *Gagea* sections *Didymobulbos*, *Platyspermum*, and *Stipitatae* were placed in two palynological groups. Furthermore, studied taxon of *G.* sect. *Plecostigma* (*G. afghanica*) segregated from the others as sister group. In the present study, the taxa of *Gagea* sect. *Platyspermum* constructed one palynological type. The studied species of *G.* sect. *Stipitatae* (in palynological group 1 and 2) had

heterogeneous palynological situation. This palynological result corroborated with phylogenetical situation of *Gagea* sect. *Stipitatae* (Peterson et al. 2008).

The palynological research in Iran showed that, exine ornamentation had the potential taxonomical value to the delimitation of studied species from this genus (Zarrei & Zarre 2005). Following four types of pollen grains can be distinguished in the proximal face as reticulate, microreticulate, foveolate, and perforate:

In the reticulate type, three subtypes were distinguished in the distal face of reticulate types as reticulate, microreticulate, and perforate. In the microreticulate type, two exine ornamentation were detected as follows: exine reticulate at distal face and exine perforate at distal face. In addition, in the foveolate type, exine foveolate at the distal face was seen and exine perforate being at the distal face (Zarrei & Zarre 2005).

In general, the pollen typification of Zarrei & Zarre (l.c.) partly supported the taxonomical values in the section levels. These researchers helped the other characters (pollen size, type of muri, and columella and lumina size) along with this pollen typification which

constructed pollen key to segregate the species of this genus.

Morphological evidences (cymose inflorescence and fibrous-papery tunic) supported the taxonomical placement of sect. *Plecostigma* (Zarrei *et al.* 2011). In addition, pollen micromorphological characters verified this situation (Zarrei & Zarre 2005). All taxa had foveolate or perforate pollen type (Zarrei & Zarre *l.c.*). Three species of this section were examined by Zarrei & Zarre (*l.c.*). *Gagea afghanica* as one member of this section had perforate exine ornamentation and pluricolumellate infratectum in the proximal face and eutectate ultrastructure in distal face (Zarrei & Zarre 2005). The perforation in different parts of exine ornamentation was seen during this research. In addition, the sulcus membrane ornamentation and pore number per  $2 \mu\text{m}^2$  in the exine surface was added. In the current study, palynological issues placed this species as a segregated group of the other taxa and overlapped with previous palynological research (Zarrei & Zarre *l.c.*) in the exine ornamentation.

*Gagea chomutovae* had reticulate and perforate exine ornamentation in proximal and distal faces with compound muri and simplicolumellate infratectum (Zarrei & Zarre 2005). The present results showed that, this species had tuberculate-perforate exine ornamentation in the equatorial surface and the sulcus margin. Additionally, a tuberculate ornamentation in the sulcus membrane regions was also found. Although Zarrei & Zarre (*l.c.*) showed that, pollen of *G. kunawurensis* (Syn.: *G. ova*) had perforate exine ornamentation; the present investigation indicated that, exine ornamentation was different from above-mentioned study and had variation in different pollen regions (macrotuberculate-perforate in equatorial surface toward tuberculate-perforate in sulcus region). In addition, there was tuberculate intine ornamentation in the sulcus membrane. The pollen data of *G. dschungarica* were reported in this research from Iran for the first time. Furthermore, in comparison, this species is pollen morphologically similar to *G. chomutovae* and *G. kunawurensis* based on qualitative palynological

issues. Although the palynological dendrogram drawn in the present research, it showed a heterogeneous situation in all the taxa of sect. *Stipitatae*. In general, perforate exine ornamentation and tuberculate intine ornamentation had been considered fundamental pollen qualitative characteristics based on our research and previous study of Zarrei & Zarre (*l.c.*).

In the present study, *G. reticulata* and *G. setifolia* were placed in one palynological type. Reticulation and perforation had major roles in exine ornamentation of *G. reticulata* in the proximal and distal faces, respectively (Zarrei & Zarre 2005). Results derived through the present investigation, differed from results of the exine ornamentation achieved by Zarrei & Zarre (*l.c.*). Therefore, this qualitative palynological character is varied in the taxa of sect. *Platyspermum*. It is interesting to state that, the palynological dendrogram obtained from the current study, has accepted the taxonomical situation of sect. *Platyspermum*.

*Gagea tenera* (sect. *Didymobulbos*) was also examined by Zarrei & Zarre (2005). This species had reticulate and microreticulate exine ornamentation in proximal and distal faces with compound muri and simplicolumellate infratectum (Zarrei & Zarre *l.c.*). In our study, reticulate-cristate and microreticulate-cristate exine ornamentation was observed in this taxon in the equatorial surface and sulcus margin regions, respectively. In addition, there was tuberculate intine ornamentation in the sulcus membrane region. Foveolate exine ornamentation with solid muri and pluricolumellate infratectum were seen in *G. vegeta* (Zarrei & Zarre *l.c.*). Based on the observation made in the present investigation, there was tuberculate-striate exine ornamentation and tuberculate intine ornamentation in different part of pollens from *G. vegeta*. Palynological dendrogram obtained from the present observation, placed the taxa of *Gagea* sect. *Didymobulbos* in one type. Although the taxa of *Gagea* sect. *Didymobulbos* partly corroborated with *Gagea* sect. *Stipitatae* based on our palynological dendrogram, the other studied taxa strongly segregated from *Gagea* sect. *Didymobulbos*. This result

did not support the taxonomical revision of Zarrei *et al.* (2011).

Based on the above-mentioned discussion, it is concluded that, there was variation in the pollen micromorphology in the taxa of *Gagea* sections *Didymobulbos*, *Platyspermum*, and *Stipitatae*. Accordingly, pollen grains micromorphology partly supported integration of sections *Platyspermum* and *Stipitatae* in sect. *Didymobulbos*.

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