

BIOSYSTEMATIC STUDY OF SOME PAPAVER (PAPAVERACEAE) SPECIES FROM IRAN

F. Sharifnia, S. Heydarian & F. Salimpour

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In order to clarify the taxonomic state of some Iranian *Papaver* species, cluster and PCA analysis of data sets resulted from macro- and micromorphological study of 14 species belonging to the genus *Papaver* in Iran were performed. Macromorphological characters of all species included in the study were examined. Pollen grains and seeds belonging to the species were scanned using SEM microscopy. High correlation among the results obtained from analyses of 38 morphological, 11 palynological and 6 seed characters was observed especially at lower levels. Based on our results, *P. gaubae* is reduced as a variety named *P. glaucum* var. *gaubae* and the name *P. piptostigma* is regarded as a synonym of the older *P. macrostomum*. In addition, our results do not confirm opinions indicating synonymy of *P. fugax* with the older name *P. armeniacum*. Also, *P. halophilum* is regarded as a variety of *P. macrostomum*.

Key words. *Papaver*, taxonomy, biosystematic, palynology, phenetic, Micro- & Macromorphology, Iran.

مطالعه بیوسیتماطیکی برخی گونه های جنس خشخاش (*Papaver*) در ایران

دکتر فریبا شریف نیا، دانشیار گروه زیست شناسی دانشگاه آزاد اسلامی واحد تهران شمال.

سوده حیدریان، دانشجوی کارشناسی ارشد گروه زیست شناسی دانشگاه آزاد اسلامی واحد تهران شمال.

دکتر فهیمه سلیم پور، استادیار گروه زیست شناسی، دانشگاه آزاد اسلامی واحد تهران شمال.

برای روشن نمودن وضعیت تاکسونومیکی برخی از گونه های خشخاش ایرانی، آنالیز خوشه ای براساس اطلاعات میکرو- ماکرومورفولوژی ۱۴ گونه متعلق به جنس خشخاش انجام شد. صفات ریخت شناسی مورد سنجش قرار گرفت و با استفاده از میکروسکپ الکترونی تصاویری از گرده ها و دانه های این گونه ها تهیه گردید. نتایج بدست آمده از ۳۸ صفت مورفولوژیک تا حد زیادی با نتایج بدست آمده از ۱۱ و ۶ صفت میکرومورفولوژیک گرده و دانه ها مطابقت داشت. براساس این نتایج گونه *Papaver gaubae* به سطح واریته ای از گونه *P. glaucum* تنزل پیدا کرد. گونه *P. piptostigma* با گونه قدیمی تر *P. macrostomum* مترادف شد. بعلاوه نتایج بدست آمده مترادف بودن دو گونه *Papaver armeniacum* و *Papaver fugax* را تایید نکرد. *P. halophilum* به سطح واریته ای از گونه *P. macrostomum* تنزل پیدا کرد.

Introduction

The family *Papaveraceae* with 50 genera and ca. 830 species is mainly distributed in temperate regions of the northern hemisphere, South Africa and southern America (Singh & Jain 2004). Taxonomy and position of *Papaveraceae* has been changed widely (Hutchinson 1973; Kadereit et al. 1995; Hoot et al. 1997; Takhtajan 1997; Judd et al. 1999; Singh et al. 2005). *Papaver* L. (*Papaveroideae*, *Papavereae*) is characteristic in absence of style and the possession of stigmatic tissue arranged radially on a sessile stigmatic disc which crowns the ovary (Kadereit 1988). The genus has long

been at the centre of attention for many botanists and pharmacognosists for presence of important alkaloids like Papaverin, Codein, Morphin,... (Sariyar 2002). *Papaver* comprises 80 annual, biennial and perennial species in central and south-western Asia, central and southern Europe and northern Africa (Kadereit 1993; Kadereit et al. 1997). Molecular phylogeny of *Papaver* has been investigated using chloroplastic *trnK* data sets and RFLP technique (Kadereit et al. 1997). Cullen (1966) reported 26 *Papaver* species, of them five were endemic from Iran, without considering sectional classification of the genus. Fedde (1909) classified the

Table 1. *Papaver* specimens included in the morphological study and SEM microscopy (indicated by an asterisk *).
Abbr.: IAUNT= Islamic Azad University, North Tehran .

P. argemone L.: Tehran, Damavand, Ab-e Sard, 1920 m, Heydariann 6019* (IAUNT); Tehran, Shemiran, Cullen & Sharif 31922 IRAN; Gilan, Deylaman, 1500-1600 m, Termeh & Esfandiari 31921 IRAN; Tehran, Lashgarak, 1570 m, Heydarian 6017* (IAUNT).

P. pavonium Fish. & C. A. May.: Mazandaran, Chalus, 5 km to Kalar Dasht, 1470 m, Heydarian 6020* (IAUNT); Tehran, Gachsar, 1900 m, Salimpour 6021* (IAUNT); Gorgan, Gonbad, Sharif & Reching 32085/1 IRAN.

P. bracteatum Lindl.: Tehran, Lar, 2520 m, Heydarian 6043* (IAUNT); Tehran, Karaj, Kandovan, 2800 m, Iranshahr 31940 IRAN; Mazandaran, Amol, Yoush, Termeh 31939/1 IRAN.

P. armeniacum (L.) DC.: Tehran, Lar, beside of Lar river, 2420 m, Heydarian 6015* (IAUNT); Tehran, Karaj, Kandovan, 2800 m, Cullen 31928/2 IRAN; Tehran, Gadok, Cullen & Behboodi 31934/1 IRAN.

P. fugax Poir.: Kordestan, Sardasht, 1200 m, Afzalrabi 6016* (IAUNT); Kordestan, Baneh, 2000-2200 m, Iranshahr & Cullen 32019/2 IRAN; Azerbaijan, Marand, 1400 m, Iranshahr & Cullen 32025/1 IRAN.

P. glaucum Boiss. & Hausskn.: Ghazvin: Abyek, Ziaran, 2864 m, Heydarian 6022*(IAUNT); Kermanshah, Sharif & Cullen 32029 IRAN.

P. gaubae Cullen & Rech. f.: Ghazvin: Abyek, Ziaran, 2864 m, Heydarian 6023*(IAUNT); Ghazvin: Karaj, between Khor and Fashand, 1560 m, Mosavi & Esfandiari 32028/3IRAN.

P. dubium L. Tehran: Lashgarak, near Rudehen, 1710 m, Heydarian 6025*(IAUNT); Karaj, Chalus road, 2000 m, Heydarian*6026 (IAUNT); Gorgan, Golestan Forest, 920-1000 m, Termeh & Matin 31992 IRAN.; Tehran, Damavand, Absard, 1920 m, Heydarian*6027(IAUNT).

P. tenuifolium Boiss. & Hohen. ex Boiss.: Hamedan: Alvand mountain, Babae & Cullen 34181 IRAN; Tehran: Evin, Zarghani & Cullen 32104/1 IRAN; Tehran: Lashgarak, near Rudehen, 1650 m, Heydarian*6029 (IAUNT).

P. commutatum Fischer & C. A. Mey.: Ardabil: Namin, 1500 m, Heydarian* 6030 (IAUNT); Mazandaran: 15 km to Kelardasht, 1400 m, Heydarian*6031 (IAUNT).

P. chelidonifolium Boiss. & Buhse: Mazandran: Tonkabon, Sehezar forest, 2200 m, Heydarian*6039 (IAUNT); Mazandaran: Hezarjarib, 2200 m, Heydarian*6034 (IAUNT); Gilan: Asalem, Nav, Abbasi & Esfandiari 31945 IRAN.

P. macrostemum Boiss. & Huet.: Ghazvin, Gauba & Cullen 32063 IRAN; Isfahan: between Shahreza and Broojen, Dehaghan village, 2150 m, Iranshahr & Cullen 32060/2IRAN; Ghazvin: between Ghazvin and Tehran, Soltanabad, 1250 m, Heydarian*6004 (IAUNT); Tehran: Ozgol, 1570 m, Heydarian*6011 (IAUNT); Ardabil: Namin, 1420 m, Heydarian*6002(IAUNT).

P. piptostigma Bienert ex Fedde : Gilan: Amarlo, Damash, 1800 m, Daryadel & Cullen 32089/2IRAN; Tehran: Damavand, Absard, Sabzevri & Cullen 3290 IRAN; Tehran: Damavand, Absard, 1920 m, Heydarian*6010 (IAUNT); Ghazvin: Abyek, Ziaran, 1145 m, Heydarian*6013 (IAUNT); Tehran: Lashgark, Ozgil, 1570 m, Heydarian*6003(IAUNT).

P. halophilum (Fedde) Cullen: Kermanshah: Ghasreshirin, Sharif & Cullen 32031/1IRAN. Tehran, Lashgarak, Ozgol, 1570 m, Heydarian*6041(IAUNT); Kermanshah: Gilangharb, Kasegaran, Iranshahr & Cullen32030/3IRAN.

genus into nine sections. According to Kadereit (1988) the genus *Papaver* is divided into 11 sections, of them six are distributed in Iran. They are *Papaver* sections *Oxytona*, *Rhoeadium*, *Argemonidium*, *Carinatae*, *Papaver* and *Mecononidium* appearing in Iran. According to Cullen (1966), *P.* sect. *Carinatae* has three species in Iran: two endemic species *P. halophilum* and *P. piptostigmata* Bienert, and a widely distributed *P. macrostomum* which is very close to *P. piptostigmata*, differing from it in size and shape of capsule. Of four Iranian species belonging to the section *Mecononidium* reported by Cullen (1966), only two are accepted by Kadereit (1993) naming *P. armeniacum* and *P. persicum*, and two others i.e. *P. fugax* and *P. cylindricum* are reduced as synonym under *P. armeniacum* subsp. *microstigmum*, one of its three subspecies distributed in Iran. Furthermore, two

species of the section *Papaver* described from Iran (Cullen 1966), are regarded as synonym by Kadereit (1986b): *P. gaubae* Cullen & Rech. f. was reduced as synonym under *C. glaucum* Boiss. & Hausskn.

Using light microscopy, Rachele (1974) studied palynological aspects of 11 genera belonging to the *Papaveraceae* including three species *P. somniferum*, *P. rhoeas* and *P. dubium*. Kadereit (1986a) studied pollen morphology and exine surface ornamentation of four species belonging to the section *Argemonidium* using SEM (Scanning Electronic Microscope) microscopy.

There is no integrated biosystematic study on the genus *Papaver* in Iran. This paper aims to evaluate the present taxonomic state of some Iranian *Papaver* using a phenetic approach based on the morphological,

micromorphological of seed and palynological evidences.

Materials and methods

Herbarium specimens belonging to 14 *Papaver* species were collected from their natural habitats (partly listed in Table 1). At least three individuals of each species were subjected to morphological and palynological studies (Table 1). 76 morphological qualitative and quantitative characters were examined (Table 2). Quantitative characters were initially encoded into multi-state characters. 14 OTUs (here 14 species) were subjected to a hierarchical cluster analysis using SPSS ver. 9 with Ward method (Norusis 1999). Principal Component Axes (PCA) were extracted and ordination of taxa was performed on the first two PCA using SPSS ver. 9.

Papaver seeds are very small and therefore could not be easily studied with light microscopy. At least 3 ripen seeds from each species was selected and then coated with a very thin layer of gold in order to be prepared for SEM microscopy. Similar methods as described above were used in order to analyze morphological data obtained from study of seeds.

Pollen grains from at least three individuals from each species were prepared for SEM microscopy with the similar method described for seeds. They were taken from unopened flowers using a needle. Pollen grains as well as seeds were studied using a SEM electronic microscope model Philips XL 30. Different magnifications were used for examining seed and pollen grain surfaces. Our terminology for pollen grains is in accord with that of Moore et al. (1991). Similar methods (described above) were used for the hierarchical cluster analysis and PCA analysis of palynological data.

Results and discussion

Scan of seed and pollen grains are presented in Figs. 1-6. Results obtained from the factor analysis of quantitative morphological characters showed that first two components have the most influence in the analysis. ca. 26% of overall variability belongs to the first component including capsule shape (most important character with a variance of 0.895) and ca. 20% of overall variability belongs to the second component including duration (most important character with a variance of 0.98).

As illustrated in the dendrogram resulted from the hierarchical analysis of morphological characters of 14 species, *P. bracteatum* shows the minimum similarity to other species and has segregated from the others at the linkage distance 25 (Fig. 7). Ordination of taxa

based on PCA analysis of first two components yields similar results (Fig. 8). *P. bracteatum* is located at a remote position relatively far from others. It is distinguishable from other studied *Papaver* species in having characters like presence of tiny spines on the plant, present of bracts just below flowers and buds with three furrows. Other species are divided into two clusters at the linkage distance 16 (Fig. 7). The first cluster is then divided into two subclusters at the linkage distance 10. The first subcluster includes *P. armeniacum* and *P. fugax* both from the section *Meconidium*. These two species have similar capsule shape, stigma morphology, valvate capsule opening, rosette and cauline leaves morphology, biennial duration, and their close positions in the dendrogram seem to be logic.

The second subcluster of cluster 1 comprises two species *P. argemone* and *P. pavonium* (Fig. 7) which have similar setose capsule, stigma morphology, morphology of rosette and cauline leaves, and clavate filaments.

The second cluster is divided into two subclusters at the linkage distance 10.89 (Fig. 7). The first subcluster includes three species *P. macrostomum*, *P. piptostigma* and *P. halophilum* from the section *Carinatae*. They are characterized with their similar capsule shape, carinate stigma and anther morphology especially in having a round appendix at the end of anthers. *P. macrostomum* and *P. piptostigma* have more characters in common. The second subcluster of cluster 2 is divided into two groups: the first group including two species *P. gaubae* and *P. glaucum* both from the section *Papaver* with similar capsule shape, stigma morphology, plant color, shape of cauline leaves, amplexicaule base of cauline leaves, and similar anther morphology with a round appendix at the end of anthers; and the second group formed by four species *P. chelidonifolium*, *P. commutatum*, *P. dubium* and *P. tenuifolium* from *P. sect. Rhoeadium* with similar capsule shape, stigma morphology, and morphology of rosette and cauline leaves.

First two components obtained from the factor analysis of characters of seed morphology, i.e. component 1 with less than 52% of overall variance and component 2 with 21% of overall variance have the most influence in the analysis. In the first component, seed shape and seed surface ornamentations with a variance of 0.956, and in the second component ornamentation of lumina margin with a variance of 0.463 were the most important characters and therefore had the most influence in the analysis.

Table 2. Morphological characters and character states among the studied *Papaver* species.

1. duration (annual 1/ biennial 2/ perennial 3); 2. branching position (branched from the base 1/ branched upward near the base 2/ branched along the stem 3/ unbranched 4); 3. habit (decumbent to erect 1/ erect 2/ decumbent 3); 4. stem indumentum (glabrous 1/ densely hispid-setose 2/ grey villous 3/ pilose 4/ patent setose 5/ setose below 6/ strongly bristly 7); 5. plant color (dark green 1/ light green 2/ glaucous 3); 6. morphology of basal leaves (ovate 1/ ovate-elliptic 2/ lyrate 3/ oblong-lanceolate 4/ linear-oblong 5); 7. margin of basal leaves (serrate 1/ dentate 2/ incised 3/ lacerate 4/ pectinate 5); 8. basal leaf segments (pinnatisect 1/ pinnatisect- bipinnatisect 2/ bipinnatisect-tripinnatisect 3); 9. shape of cauline leaves (ovate 1/ obovate 2/ lyrate 3/ oblong-lanceolate 4/ oblong-linear); 10. margin of cauline leaves (serrate 1/ dentate 2/ incised 3/ lacerate 4/ pectinate 5); 11. cauline leaf segments (pinnatisect-bipinnatifid 1/ bipinnatisect-tripinnatifid 2); 12. petal color (light red 1/ dark red 2/ orange 3/ pink 4); 13. spots on petals (absent 1/ present 2/ both states 3); position of spots on petals (absent 1/ basal 2/ middle 3); 15. shape of spots (without spot 1/ curved 2/ oblong 3/ obovate 4/ radial 5); 16. spot color (without spot 1/ black 2/ dark violet-black 3/ black with white margin 4); 17. filament shape (clavate 1/ filiform 2); 18. filament color (dark violet-black 1/ yellow-brown 2); 19. anther shape (globose 1/ oblong 2); 20. anther appendage (present 1/ absent 2); 21. anther color (dark violet-black 1/ yellow-brown 2); 22. ovary shape (globose to subglobose 1/ cylindrical-abruptly attenuate at base 2/ clavate 3/ oblong-ellipsoid 4/ obovate 5); 23. disc teeth (carinate 1/ not carinate 2); 24. capsule color (light green 1/ streaky light green 2/ glaucous 3/ dark green 4); 25. stigma color (dark brown 1/ dark brown-violet 2/ yellow-light brown 3/ green-yellow 4/ violet 5); 26. capsule surface (with bristle 1/ without bristle 2); 27. apex of buds (with two horns 1/ hornless 2); 28. capsule opening (porate 1/ valvate 2); 29. base of cauline leaves (amplexicaule 1/ not amplexicaule 2); 30. stigmatic disc (scarious 1/ coriaceous 2); 31. stigmatic rays high (very low 1/ elevated 2/ very high 3); 32. disc form (flat to slightly convex 1/ flat-convex to highly convex 2/ pyramidal 3/ carinate 4); 33. stigmatic rays (exceeding the free lobes 1/ not exceeding free lobes 2); 34. capsule shape (oblong with pyramidal disc 1/ obovate with flat-slightly convex disc 2/ globose to subglobose with convex disc 3/ globose to subglobose with flat-slightly convex disc 4/ globose-ellipsoid with flat-slightly convex disc 5/ clavate with carinatae disc 6); 35. hairs (absent 1/ soft 2/ rough 3); 36. rosette radical leaves (present 1/ absent 2); 37. bud lobes (two 1/ three 2); 38. plant height (small to medium 1/ large 2).

Table 3. Seed and pollen grain characters and character states among the studied *Papaver* species.

1. seed shape (reniform 1/ oblong-reniform 2); 2. seed surface ornamentation (reticulate 1/ striate 2); 3. ornamentation depth (superficial reticulate 1/ deep reticulate 2/ superficial seriate 3); 4. lumina surface (smooth 1/ tuberculate 2/ microechinate 3/ faveolate 4/ punctuate 5/ microverrucate 6); 5. cell margin (without reticulum 1/ smooth 2/ sinuate 3/ tuberculate 4/ microechinate 5/ punctuate 6); 6. seed color (yellow 1/ light brown 2/ brown 3/ dark brown 4/ black 5/ orange 6); 7. polar length (P; μm); 8. equatorial diameter (E; μm); 9. P/E ratio; 10. colpus length (L; μm); 11. apocolpium (μm); 12. mesocolpium (μm); 13. spinule diameter (μm); 14. number of spinules in 10 square μm ; 15. pollen aperture (porate 1/ culpate 2); 16. colpus openness (open 1/ close 2/ no colpus 3); 17. pollen shape (spherical 1/ prolate 2).

As shown in the cluster analysis (Fig. 9) and ordination of taxa based on PCA analysis of first two components (Fig. 10) of data obtained from seed morphology of 14 species, *P. argemone* has the minimum similarity to other studied species. All other *Papaver* species studied in this paper have obtuse-reniform seeds with reticulate surface. In contrast, *P. argemone* has seeds with seriate surface (Fig. 4, A-B). Our results are highly congruent with those of Kadereit et al. (1997) in case of *P. argemone*. They found that *P. sect. argemonidium* (including *P. argemone*) forms a separate clade more related to *Roemeria* than to other *Papaver* species. Dendrogram and ordination of taxa resulted from analysis of data obtained from seed morphology (Fig. 9-10) were not congruent with those of plant morphology (Fig. 7-8) at higher levels.

However, at lower levels, more congruency between two data sets can be observed. The main cluster is divided into two subclades at the linkage distance 25 (Fig. 9). Three species *P. macrostomum*, *P. halophilum* and *P. piptostigmata* have very similar seed morphology and form a group (Fig. 1, C-H). Two species *P. armenicum* and *P. fugax* from the section *Meconodium* have similar gemmate lumina surface ornamentations and form a nested subcluster under a larger subcluster including themselves plus three former species with smooth lumina surface ornamentations (Figs. 9; 2, A-D). The second subcluster of cluster 1 includes three species from the section *Rhoeadium* with three different lumina surface type: *P. commutatum* with faveolate (Fig. 3, E-F), *P. chelidoniifolium* with microechinate (Fig. 3, G-H)

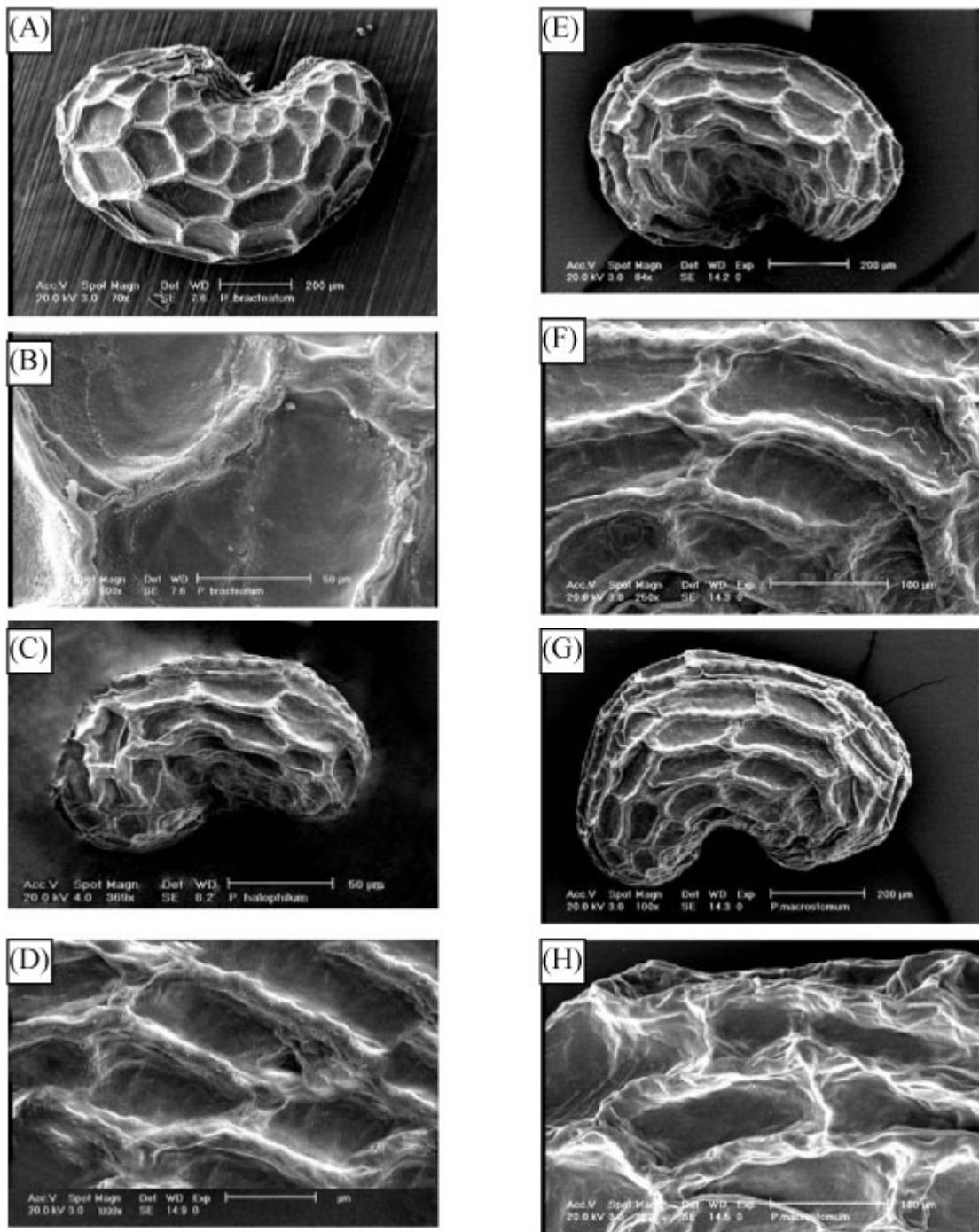


Fig. 1. Seed morphology of *Papaver* species: A (x70) & B (x500): *P. bracteatum*; C (x369) & D (x1000): *P. halophilum*; E (x84) & F (x250): *P. piptostigma*; G (x100) & H (x250): *P. macrostomum*.

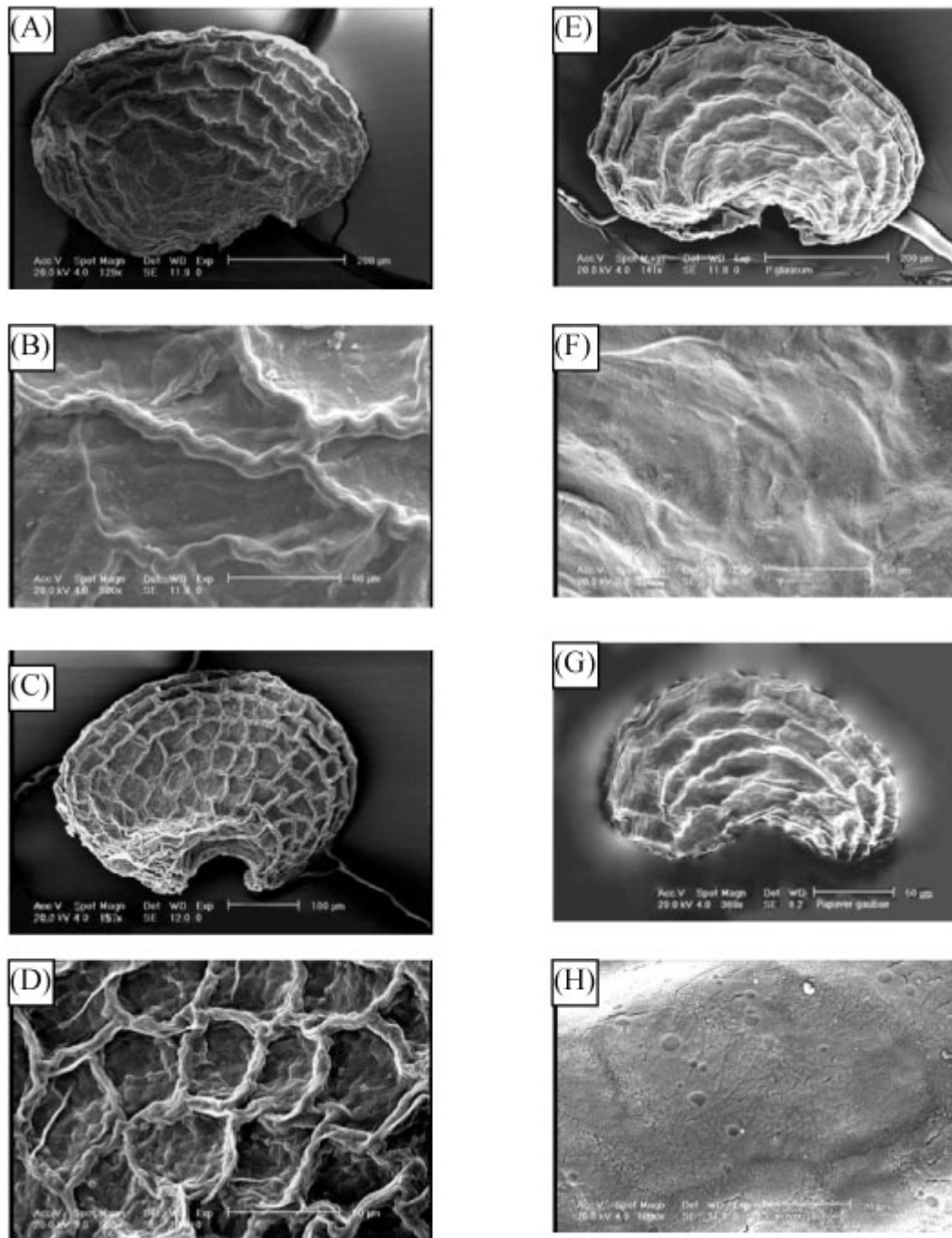


Fig. 2. Seed morphology of *Papaver* species: A (x187) & B (x500): *P. armeniacum*; C (x157) & D (x500): *P. fugax*; E (x141) & F (x1000): *P. glaucum*; G (x369) & H (x1000): *P. gaubae*.

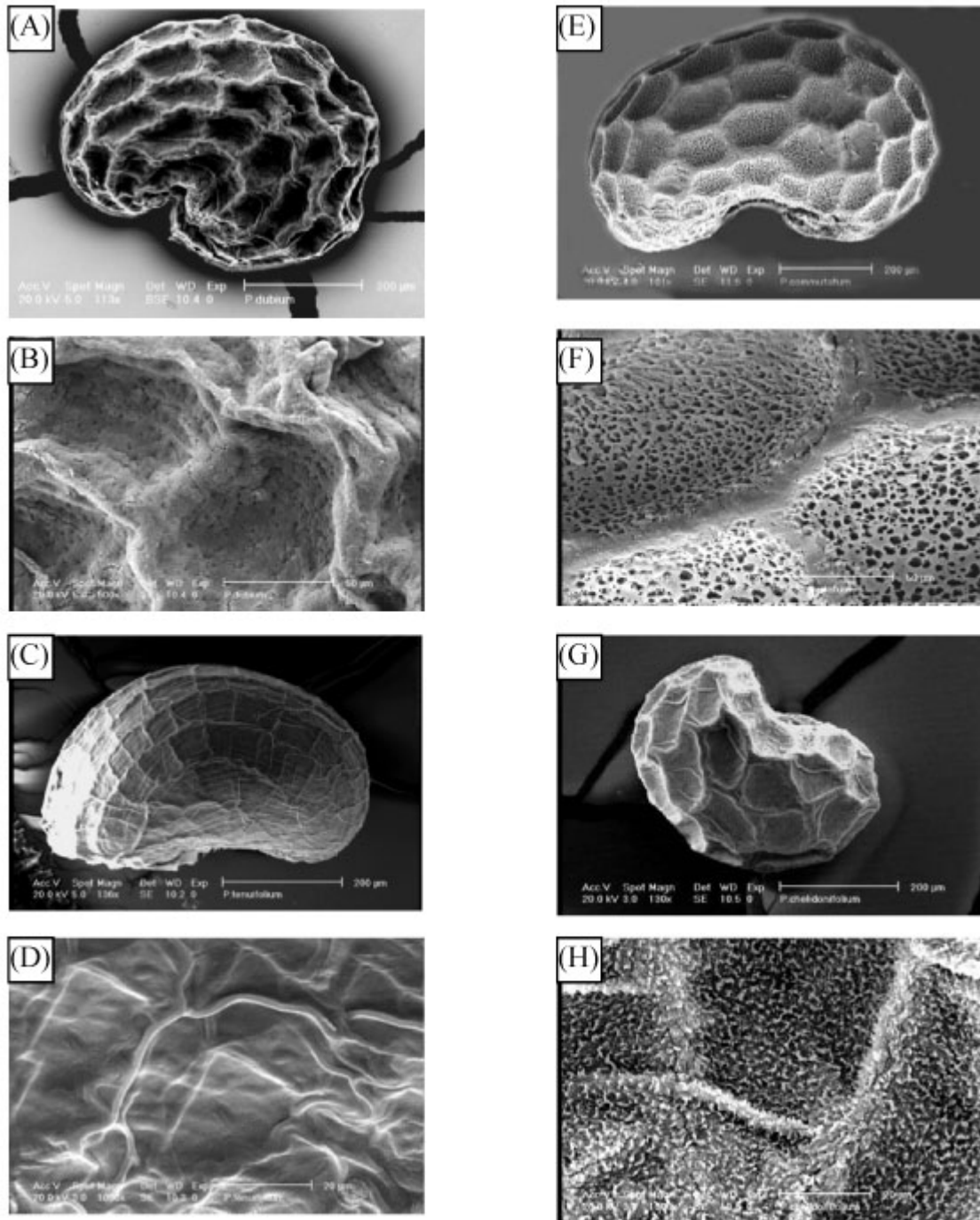


Fig. 3. Seed morphology of *Papaver* species: A (x113) & B (x500): *P. dubium*; C (x136) & D (x500): *P. tenuifolium*; E (x141) & F (x1000): *P. commutaum*; G (x369) & H (x1000): *P. chelidoniifolium*.

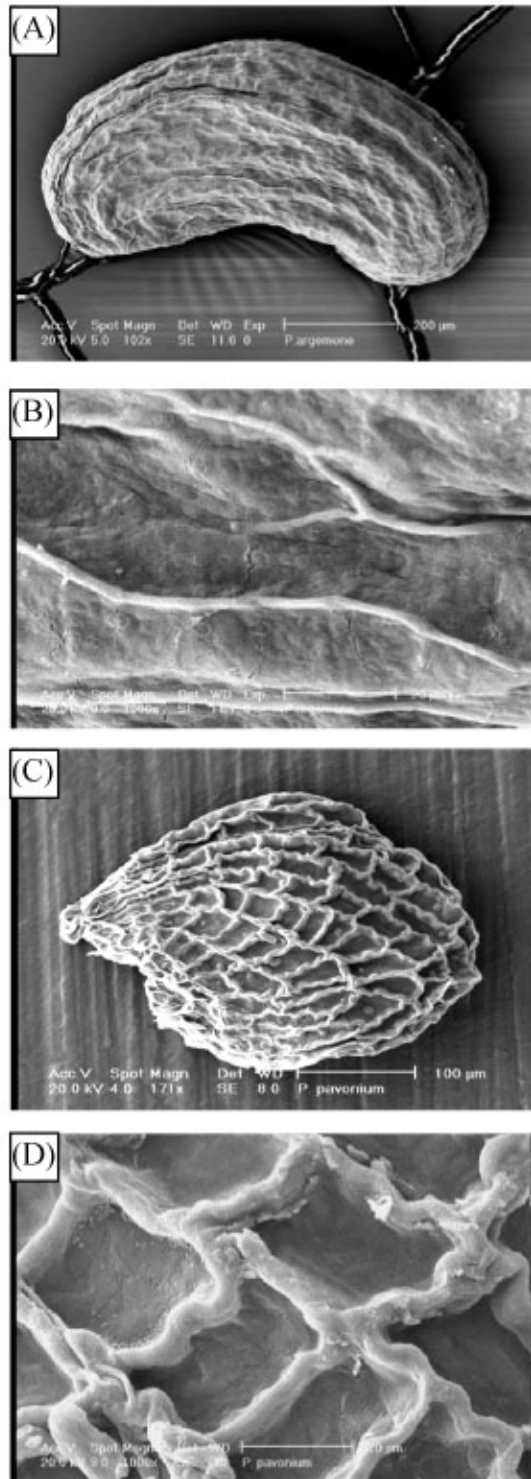


Fig. 4. Seed morphology of *Papaver* species: A (x102) & B (x1000): *P. argemoneae*; C (x171) & D (x1000): *P. pavonium*.

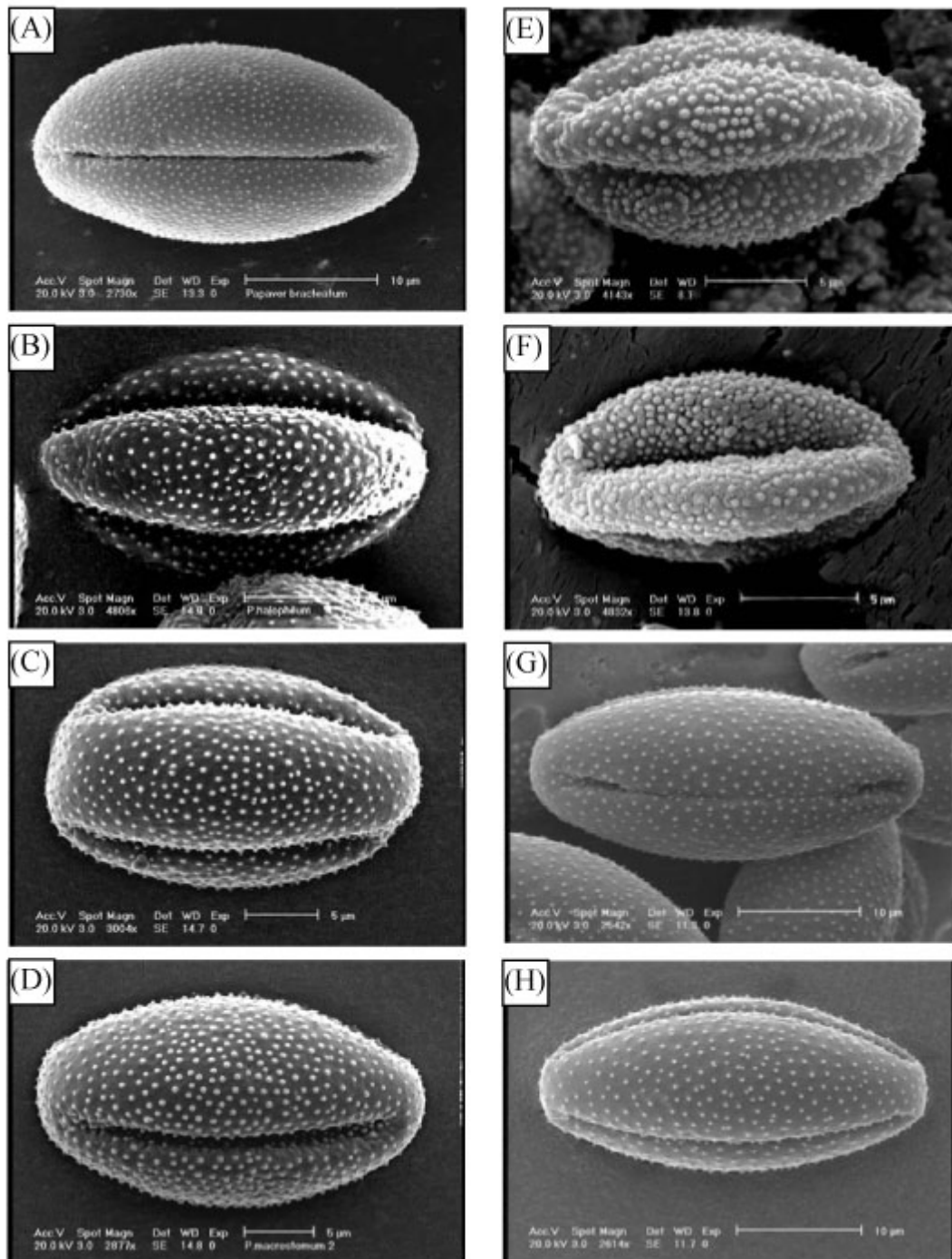


Fig. 5. Equatorial view of pollen grains of A: *P. bracteatum*; B: *P. halophilum*; C: *P. piptostigma*; D: *P. macrostemum*; E: *P. armeniacum*; F: *P. fugax*; G: *P. gaubae*; H: *P. glaucum*.

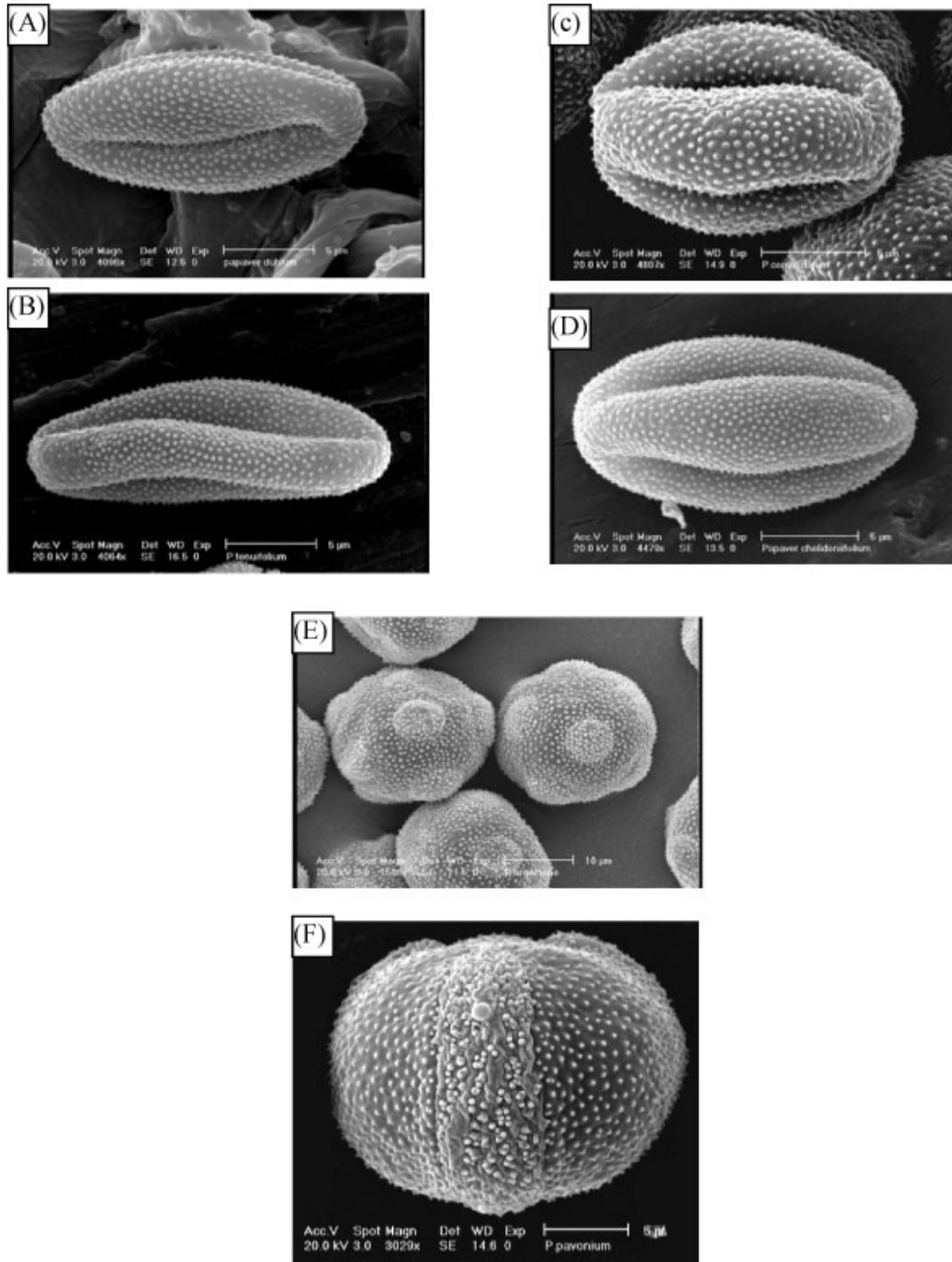


Fig. 6. Equatorial view of pollen grains of A: *P. dubium*; B: *P. tenuifolium*; C: *P. commutatum*; D: *P. chelidoniifolium*; E: *P. argemone*; F: *P. pavonium*.

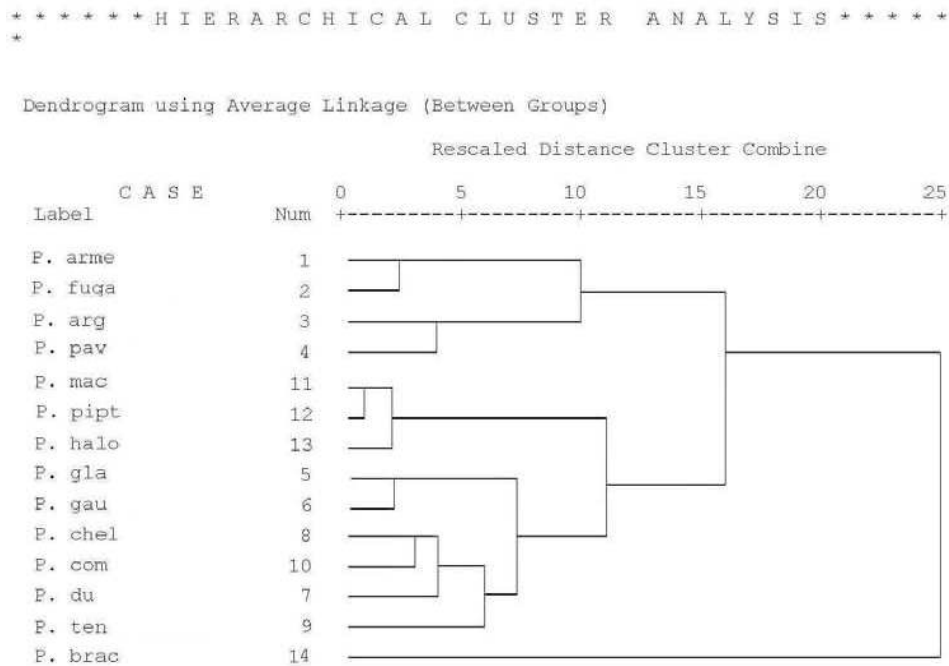


Fig. 7. Cluster analysis of some *Papaver* species resulted from analysis of morphological data using Average Linkage with Ward method. Abbreviations (arme=armeniicum; fuga=fugax; arg=argemone; pav=pavonium; mac=macrostemum; pipt=piptostigma; halo=halophilum; gla=glaucum; gau=gaubae; chel=chelidonifolium; com=commutatatum; du=dubium; ten.=tenuifolium; brac=brateatum).

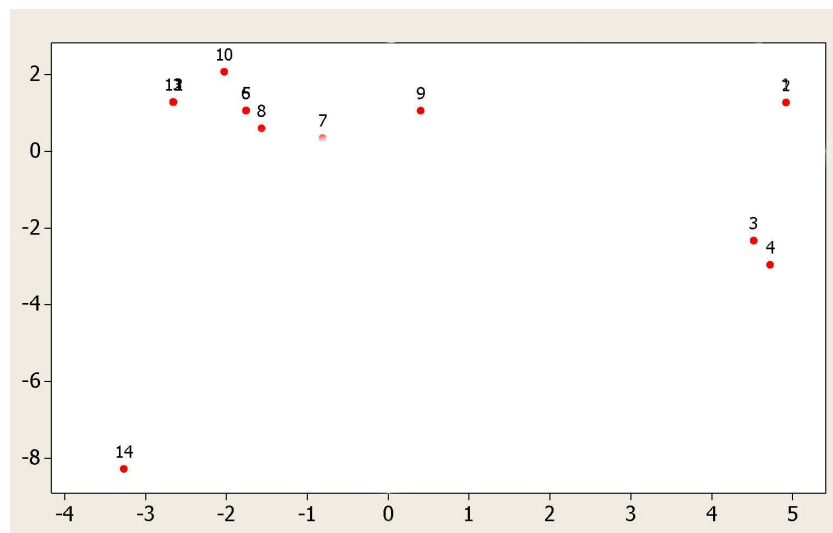


Fig. 8. Ordination of some *Papaver* species based on first two principal components using morphological data (numbers as in Fig. 7).

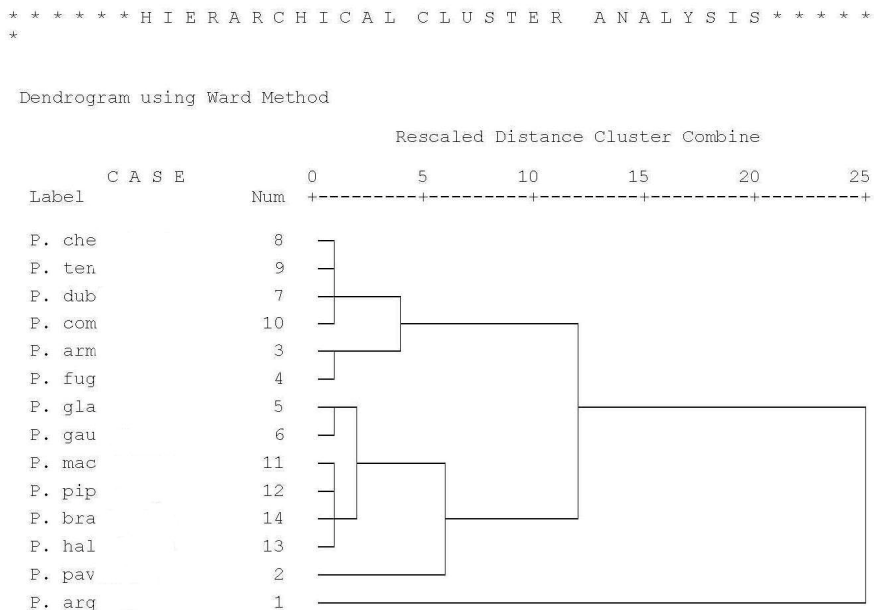


Fig. 11. Cluster analysis of some *Papaver* species resulted from analysis of palynological data using Average Linkage with Ward method (abbreviations as in Fig. 7).

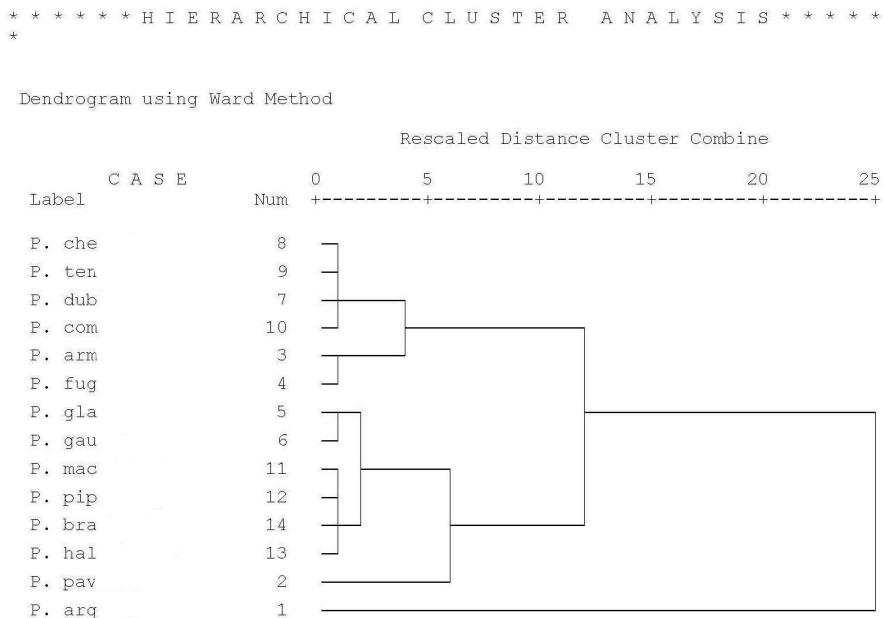


Fig. 12. Ordination of some *Papaver* species based on first two principal component using palynological data (numbers as in Fig. 11).

and *P. dubium* with fossulate lumina surface ornamentation (Fig. 3, A-B). The second cluster comprises five species, of them three species *P. glaucum*, *P. gaubae* and *P. tenuifolium* have the maximum similarities (Fig. 9). *P. glaucum* and *P. gaubae* are also very close to each other in analysis based on the morphological data (Fig. 7).

Factor analysis of palynological characters showed that first two components had the most influence in the analysis: first component with ca. 60% of overall variability including colpi position, aperture condition, apocolpium and mesocolpium length (most important characters with variance of 0.995, 0.994, 0.989 and 0.987 respectively), and the second component with ca. 19% of overall variability including polar axis length (most important character with a variance of 0.919).

Dendrogram (Fig. 11) and ordination of taxa (Fig. 12) based on the palynological data are more congruent to those of seed morphology (Fig. 9-10), with *P. argemone* with spherical and porate pollen grains (Fig. 6, D) and therefore showing the minimum similarity to others. Rest of species with different pollen morphology (Fig. 5-6) are grouped into two clusters at the linkage distance 25 (Fig. 11). The first cluster is divided into two subclusters. In the second subcluster of cluster 1, two closely related species *P. armeniacum* and *P. fugax* are grouped at very low distance linkage. In all our analyses, these two species show a high similarity to each other. *P. fugax* was reduced as a synonym for *P. armeniacum* subsp. *microstigma* by Kadereit (1988). Regarding the presence of many intermediate forms between *P. fugax* and *P. armeniacum* (Cullen 1966), and high similarity in micro- and macromorphology observed in present study, we here agree with Kadereit (1988) to assume *P. fugax* representing an interspecific variation within *P. armeniacum*.

In the second cluster (Fig. 11) two small groups of species are grouped in nested subclusters at very low distance linkages. The first nested subcluster of cluster 2 includes *P. glaucum* and *P. gaubae*, two species that are also grouped in dendrogram resulted from analysis of morphological characters (Fig. 7). These two species plus *P. tenuifolium* are grouped as a nested subcluster in dendrogram obtained from the analysis of seed morphology (Fig. 9). Results obtained from analysis of both macro- and micromorphological data sets stress high similarities between *P. glaucum* and *P. gaubae*. *P. gaubae* was only known from the type locality (Cullen 1966). Based on the study of type material, Kadereit (1988) suggested that *P. gaubae* represents only a depauperate form of *P. glaucum*, because he found the turbinate form of capsule and the bidentate free lobes of the stigmatic disc, i.e. the key diagnostic characters

of *P. gaubae*, in only one of the specimens of type gathering. Our study here is based on the study of more material collected from area near type locality, plus material deposited in herbaria IRAN and IAUN match the description of *P. gaubae*. Both materials of *P. glaucum* and *P. gaubae* show differences in capsule shape and stigma morphology, however they are very close in our analyses. Therefore, we assume *P. gaubae* as a distinct variety under *P. glaucum*:

Papaver glaucum Boiss. & Hausskn. var. **gaubae** (Cullen & Rech. f.) Sharifnia & Heydarian, **stat. & comb. nov.**

Syn.: *P. gaubae* Cullen & Rech. f., Fl. Iranica 34: 16 (1966).

The second nested subcluster of cluster 2 includes four species *P. macrostomum*, *P. piptostigma*, *P. bracteatum* and *P. halophilum* (Fig. 11). *P. bracteatum* is a very distinct species with different morphological characters (see paragraph 2 of results & discussion). Other three species are grouped together in all three analyses presented here (macromorphology, seed morphology and palynology). As noted before, *P. macrostomum* and *P. piptostigma* are much related to each other, differ only in size and shape of capsule. In study of herbarium material at herbaria IRAN and IAUNT, we observed some intermediate forms, so that in our opinion they could not be circumscribed as separated species (Sharifnia & et al 2008) In addition, according to Flora of USSR (Popov 1937), Flora Orientalis (Boissier 1867), Flora of Iraq (Townsend & Guest 1966) and Flora of Turkey (Davis 1965), *P. macrostomum* shows high degree of interspecific morphological diversity. We here reduce the newer name *P. piptostigma* (Fedde in Engler, Pflanzenr. 40: 336 (1909)) as synonym for the older *P. macrostomum* (Boiss., Fl. Orient. 1: 115 (1867)).

In study of herbarium material deposited at herbaria IRAN and IAUNT, and herbarium material collected by us from wild, we found that despite many similarities between two species *P. macrostomum* and *P. halophilum*, they could be distinguished from each other mainly based on their clear difference in size of plants. *P. macrostomum* has 20 – 50 cm long stems, while *P. halophilum* has 10-20 cm long stems. Therefore, we here agree with Fedde (1909) in assuming *P. halophilum* as a variety of *P. macrostomum*:

P. macrostomum Boiss. & Huet var. **halophilum** Fedde in Engler, Pflanzenr. 40: 336 (1909).

Syn.: *P. halophilum* (Fedde) Cullen, Fl. Iranica 34: 17 (1966).

Identification key for two varieties of *P. macrostomum*.

1. Plant 30-50 cm high. Stem branched from base. Petals often with spots. Capsule about 20-25mm long

var. *macrostomum*

-Plant 10-20 cm high. Stem without branches. Petals often without spot. Capsule about 7-9 mm long

var. *halophilum***Acknowledgments**

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References

- Boissier, E. 1867: *Papaver* in *Flora Orientalis* vol: 1: 105-118. -Basileae & Genevae.
- Cullen, J. 1966: *Papaver* in K. H. Rechinger(ed.) *Flora Iranica* no.34: 10-27. -Akademische druck- verlags ansalt, Graz-Austria.
- Fedde F. 1909: *Papaver* in A. Engler *Das Pflanzenreich* IV. 104: 288-386. -Leipzig.
- Hoot, S. B., Kadereit, J. W., Blattner, F. R., Jork, K. B., Schwarzbach, A. & Crane, P. R. 1997: Data congruence and the phylogeny of the *Papaveraceae* s. l. based on four data sets: *atpB* and *rbcL* sequences, *trnK* restriction site variation, and morphological characters. -*Syst. Bot.* 22: 575 - 590.
- Hutchinson, J. 1974: *The Families of Flowering plants*. -Oxford. Univ. Press.
- Judd, W. S., Campbell, C. S., Kellogg, E. A., Stevens, P. F. 1999: *Plant Systematic A phylogenetic approach*. - Sinaur Associates , Inc, USA.
- Kadereit J. W. 1986a.: A revision of *Papaver* section *Argemonidium*. -Notes from the Royal Botanic Garden Edinburgh 44: 25-43.
- Kadereit J. W. 1986b: A revision of *Papaver* L. sect *Papaver* (*Papaveraceae*). -*Botanische Jahrbücher für Systematik, Pflanzengeschichte und Pflanzengeographie* 108: 1-16.
- Kadereit, J. W. 1988: Sectional affinities and geographical distribution in the genus *Papaver* L. (*Papaveraceae*). - *Beitr. Biol. Pflanzen* 63: 139-156.
- Kadereit, J. W. 1993: A revision of *Papaver* L. sect. *Meconidium* Spach. - *Edinburgh Journal of Botany* 50: 125-148.
- Kadereit, J. W., Blattner, F.R., Jork, K.B. & Schwarzbach, A. 1995: The Phylogeny of *Papaveraceae* s. l.; morphological, geographical and ecological implication in Jensen, U. & Kadereit, J. W. (ed) *Systematic and evolution of the Ranunculiflorae*. -*Pl. Syst. Evol. Suppl.* 9. 133-145.
- Kadereit, J. W., Jork, K. B. & Schwarzbach, A. 1997: The phylogeny of *Papaver* s. l.: Polyphyly or monophyly? - *Pl. Syst. Evol.* 204: 75 - 98.
- Norusis, M. J. 1999: *SPSS 9.0 , guide to data analysis*. - Amazoon, Chicago.
- Popov, M. G. 1937: *Papaveraceae* in V. L. Komarov *Flora of the USSR* 7: 573-717 (English translation). -Moskva & Leningrad.
- Rachele, D. R. 1974: Pollen morphology of the *Papaveraceae* of the Northern United States and Canada, pp.152-159.
- Sariyar , G. 2002. Biodiversity in the alkaloids of Turkish *Papaver* Species. -*Pure Appl. Chem.*, vol. 74, No. 4, pp. 557-574.
- Sharifnia, F., Heydarian, S. & Salimpour, F. 2008: Revision of *Papaver macrostomum* Boiss. and *P. piptostigma* Bienert ex Fedde in Iran. -*Journal of Plant Sciences Researches Islamic Azad University, Gorgan Branch* vol. 1, no 10: 32-41.
- Takhtajan, A. L. 1997: *Diversity and classification of flowering plants* .-Columbia University Press. New York. 642pp.
- Singh, V. 2004: *Plant Systematic An integrated approach*. - Science publication, USA.