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Research Article

Pollen Studies on Some Species of the Genus *Convolvulus* L. (Convolvulaceae) from Morocco

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Abstract: The pollen grains of nine species of *Convolvulus* L. from Morocco were studied using scanning electron microscopy and light microscopy. The species belong to three sections and four subsections: sections *Convolvulus*, *Acanthocladi* Boiss. and *Inermes* Boiss. and subsections *Convolvulus*, *Spinescentes* Boiss., *Oleifolii* Peter, and *Diffusi* Boiss. The pollen grains in the genus examined are all 3-zonocolpate. The pollen is suboblate, oblate-spheroidal, prolate-spheroidal and prolate. The nexine is thinner than the sexine and the tectum is distinctly perforated in all species. The pollen grains show variation within the genus in polar and equatorial axes, colpus length and shape. These variations sometimes correspond with the morphological features of the species and, therefore, can be used to distinguish between some closely related species.

Key Words: *Convolvulaceae*, *Convolvulus*, pollen.

Bazı Fas *Convolvulus* L. (Convolvulaceae) Cinsi Türlerinde Polen Çalışması

Özet: Fas'ta yayılan, *Convolvulus* cinsine ait dokuz türün polenleri Taramalı Elektron Mikroskobu ve ışık mikroskobu kullanılarak çalışılmıştır. Polenleri incelenen türler üç seksiyon ve dört alt seksiyona aittir; seksiyonlar *Convolvulus* L., *Acanthocladi* Boiss. ve *Inermes* Boiss. ve altseksiyonlar *Convolvulus* L., *Spinescentes* Boiss., *Oleifolii* Peter ve *Diffusi* Boiss.'tir. İncelenen bütün polenlerin 3-zonocolpate olduğu görülmüştür. Pollen şekilleri suboblate, oblate-sferoyid, prolat-sferoyid ve prolat'dır. Bütün türlerde nexin sexin'den daha ince ve tektum açık bir şekilde perforat'dır. Polar ve ekvatoral eksen, kolpus uzunluğu ve polen şekli cins içindeki türler arasında varyasyonlar göstermektedir. Bu varyasyonlar türlerin morfolojik karakterleriyle bazen uyum göstermektedir ve bu nedenle birbirlerine yakın akraba olan bazı türlerin ayrımında kullanılabileceği görülmüştür.

Anahtar Sözcükler: *Convolvulaceae*, *Convolvulus*, pollen.

Introduction

There have been some attempts to use pollen features in the delimitation of the genera in the family *Convolvulaceae*. Hallier (1893) was the first to divide the family on the basis of pollen features into two groups, Echinoconiae and Psiloconiae. The genus *Convolvulus* L. was put in *Psiloconiae*, in which the pollen has either a psilate or granulated surface. In the division of Gamble (1923), the family was divided into two groups on the basis of echinate and non-echinate pollen grains with the genus *Convolvulus* included in the latter one. Erdtman (1952) separated the *Convolvulaceae* pollen grains into two groups, namely *Ipomoea* type and other types, in which he included the genus *Convolvulus*. The *Ipomoea* type possesses pollen grains which are polyporate, with a thick nexine and are echinate, whereas the *Convolvulus* type is distinctly perforate. O'Donell (1955) separated the genera *Convolvulus* and *Calystegia* R.Br. on the basis of Hallier`s aperture descriptions, but he reported that in *Convolvulus* the pollen is 3-colporate. Lewis & Oliver (1965) stated that their findings agree with those of Hallier, but not those of O'Donell. Sengupta (1972) carried out a comprehensive study in the family and divided the *Convolvulaceae* into four main pollen types based on the number and distribution of apertures. His division was not followed by Cronk & Clarke (1981).

The genus *Convolvulus* is represented by 20 species in Morocco (Menemen & Jury, unpub.). These species fall into three sections and six subsections. The aims of this study are to document the pollen features of some Moroccan *Convolvulus* species, and to determine whether the pollen characters support the delimitation of the taxa within the genus *Convolvulus* from Morocco. In the present study, nine species belonging to three sections and four subsections were studied.

A synopsis of the *Convolvulus* species in Morocco

A: Section *Acanthocladi* Boiss., *Fl. Or.* 4: 84 (1875).

Spiny shrub or subshrub, with branching shoots.

Subsection 1. *Spinescentes* Boiss., *Fl. Or.* 4: 84 (1875).

Flowers sessile or subsessile, solitary or in a capitulum.

C. trabutianus Schweinf. & Muschler

B: Section *Inermes* Boiss., *Fl. Or.* 4: 84 (1875).

Shrubs or subshrubs, without spines, erect or prostrate, but not twining.

Subsection 2: *Floridi* Sa'ad, *The Convolvulus Species of the Canary Isles, the Mediterranean region and the Near and Middle East*, 288 (1967).

Shrubs with numerous flowers at the top of the branches in the form of a panicle.

C. floridus L.

Subsection 3: *Lanuginosi* Peter in Engler and Prantl, *Naturl. Pflanzenfam.* 4 (3a): 33 (1897).

Flowers numerous, terminal in heads; base of the shoots ligneous.

C. lanuginosus Desr.

Subsection 4: *Oleifolii* Peter in Engler & Prantl, *Naturl. Pflanzenfam.* 4 (3a): 33 (1897)

Shoots with axillary and terminal flowers at the top.

C. cantabrica L., *C. lineatus* L., *C. mazicum* Emb. & Maire

Subsection 5: *Diffusi* Boiss., *Fl. Or.* 4: 84 (1872).

Annual or perennial herbs, rarely subshrubs; shoots prostrate or ascending, never twining.

C. gharbensis Batt & Pitard, *C. humilis* Jacq., C. *pentapetaloides* L., *C. sabatius* Viv., *C. siculus* L., *C. supinus* Cossen & Kralik, *C. tricolor* L., *C. valentinus* Cav.

C: Section *Convolvulus*

Herbs or subshrubs; branches twining.

Subsection 6: *Convolvulus*

Annual or perennial herbs; shoots twining, never spiny.

C. arvensis L., *C. althaeoides* L., *C. elegantissimum* Mill., *C. dryadum* Maire, *C. glauorum* Br-Bl. & Maire, *C. pitardii* Batt.

Materials and Methods

The pollen material was obtained from herbarium specimens (Table 1). Several anthers from mature flowers were placed in a watch glass and squashed with the addition of a few drops of wetting agent. Then the floral fragments were drawn to the side of the watchglass with fine forceps and a mounted needle under a dissecting microscope, leaving just the pollen grains to dry. An acetolysis mixture was made by mixing nine parts of acetic anhydride with one of conc. sulphuric acid (acetolysis time: 9 to 15 seconds). This was added with a bulb pipette to the dry pollen in the watchglass on the heating block. When the pollen grains darkened, they were allowed to cool for a few minutes and methylated spirit was added drop by drop to the centre of the remaining acetolysis mixture. The acetolysis mixture formed a ring around the rim of the watchglass and was wiped away with a tissue. The pollen grains were transferred to the stubs, which were already prepared with double-sided adhesive tape for scanning electron microscopy (SEM) study. For preparing light microscope slides, the remaining pollen grains in the watch glasses were transferred onto the slides on a small block of glycerine jelly with safranine stain added. When the glycerine jelly melted on the heating block, cover slips were added. For the SEM study, stubs were coated with gold for 5-6 minutes. The measurements were carried out using light microscopy and based on 20 readings for each specimen. Pictures of the pollen grains were taken by a JEOL T20 SEM and using a Zeiss light microscope. The terminology used in the present study is according to Punt et al. (1994).

Table 1. Measurements (µm) of the pollen grains examined

Taxa	Polar axis (P)	Equatorial axis (E)	P / E	Shape	Nexine (N)	Sexine (S)	N / S	Colpus (C) length	Herb.	Coll. and number
C. trabutianus	36.1-(46.3) 55. 2	42.7-(52.2)-62.4	0.84-0.89	Suboblate or oblate-spheroidal	0.9- (1.0)- 1.1	1.7-(1.9)-2.1	0.53-0.52	38.3-(43.4)-47.8	RNG	Miller et al. 422
C. cantabrica	56.7-(65.3)-71.6	54.4-(62.3)-68.5	1.04-1.05	Prolate-spheroidal	0.9- (1.1)- 1.2	1.8-(2.1)-2.2	0.50-0.55	56.7-(63.2)-67.2	RNG	Davis 52091
C. humilis	45.8-(53.3)-60.2	45.3-(52.8)-59.1	1.01-1.02	Prolate-spheroidal	0.9- (1.0)- 1.1	1.8-(1.9)-2.0	0.50-0.55	43.3-(49.1)-54.1	E	Jahandiez 244
C. sabatius	27.1-(32.3)-40.1	26.2-((28.3)-36.4	1.03-1.10	Prolate-spheroidal	0.7-(0.8)-0.9	1.2-(1.4)-1.5	0.58-0.60	26.5-(29.1)-32.3	E	Spence S.84
C. supinus	51.3-(57.7)-65.2	62.3-(68.2)-75.3	0.82-0.87	Suboblate	0.9-(1.0)-1.1	2.0-(2.3)-2.5	0.44-0.45	49.3-(53.3)-57.6	E	P.&J. Davis D.49063
C. valentinus	60.5-(70.3)-74.2	60.1-(69.5)-72.4	1.01-1.02	Prolate-spheroidal	0.8-(1.0)-1.1	1.7-(1.8)-2.0	0.47-0.55	51.1-(56.5)-60.8	E	Davis 51405
C. arvensis	62.3-(72.4)-75.2	60.7-(70.2)-72.2	1.03-1.04	Prolate-spheroidal	0.9- (1.0)- 1.1	1.8-(2.1)-2.3	0.50-0.48	57.7-(63.9)-68.5	E	Spence S. 74
C. dryadum	48.7-(57.8)-65.1	32.7-(40.6)-46.8	1.39-1.49	Prolate	0.9- (1.1)- 1.3	1.7-(1.9)-2.1	0.53-0.62	44.0-(48.9)-51.9	RNG	Jury 11467
C. pitardii	53.7-(59.3)-68.9	52.4-(58.1)-66.0	1.02-1.04	Prolate-spheroidal	0.9- (1.1)- 1.2	1.8-(2.0)-2.1	0.50-0.57	46.1-(49.8)-54.6	E	Davis 557

Results

The pollen grains in the *Convolvulus* species examined are all 3-zonocolpate (Figs. 1-3). The shape of the pollen is suboblate, oblate-spheroidal, prolate-spheroidal, subprolate and prolate (Table 1). *C. arvensis*, *C. valentinus, C. humilis, C. pitardii, C. sabatius* and *C. cantabrica* are prolate-spheroidal; *C. trabutianus*, suboblate or oblate-spheroidal; *C. supinus*, suboblate; and *C. dryadum*, prolate.

The polar axis (P) ranges from 27.1 µm to 75.2 µm, the equatorial axis from 26.2 µm to 74.6 µm and the length of the colpus from 26.2 µm to 67.3 µm (Table 1). The smallest pollen grains belong *C. sabatius*. The colpus is long and broad, and varies in length. The nexine is thinner than the sexine; the ratio of the nexine to sexine ranges from 0.44 to 0.62 in the species. The SEM study showed that the tectum is distinctly perforate (Figs. 3 and 4). Perforations are approximately circular and dense. There are also micro-echinate sculpturing with blunt apices in all species. Perforations and spines are irregularly distributed in all species (see Fig. 4).

Descriptions

1. C. trabutianus

Shape class: Suboblate or oblate-spheroidal.

Dimensions: P = 36.1 - (46.3) 55.2, E = 42.7 - (52.2) - 62.4, P / E = 0.84 - 0.89.

Pollen class: Tricolpate.

Appertures: Simple, colpi tapering at both ends, C = 38.3 - (43.4) - 47.8.

Exine: Tectate, tectum perforate, perforations approximately circular, dense, distinct, irregularly distributed, sexine thicker than nexine, N = 0.9 - (1.0) - 1.1, S = 1.7 - (1.9) - 2.1, N / S = 0.53 - 0.52.

Sculpturing: Micro-echinate, spines with blunt apices, irregularly distributed.

2. C. cantabrica

Shape class: Prolate-spheroidal

Dimensions: P = 56.7 - (65.3) - 71.6, E = 54.4 - (62.3) - 68.5, P / E = 1.04 - 1.05.

Pollen class: Tricolpate

Appertures: Simple, colpi tapering at both ends, C = 56.7 - (63.2) - 67.2.

Exine: Tectate, tectum perforate; perforations approximately circular, dense, distinct, irregularly distributed, sexine thicker than nexine, N = 0.9 - (1.1) - 1.2, S = 1.8 - (2.1) - 2.2, N / S = 0.50 - 0.55.

Sculpturing: Micro-echinate, spines with blunt apices, irregularly distributed.

3. C. humilis

Shape class: Prolate-spheroidal.

Dimensions: P = 45.8 - (53.3) - 60.2, E = 45.3 - (52.8) - 59.1, P / E = 1.01-1.02.

Pollen class: Tricolpate.

Appertures: Simple, colpi tapering at both ends, C = 43.3 - (49.1) - 54.1.

Exine: Tectate, tectum perforate; perforations approximately circular, dense, distinct, irregularly distributed, sexine thicker than nexine, N = 0.9 - (1.0) - 1.1, S = 1.8 - (1.9) - 2.0, N / S = 0.50 - 0.55.

Sculpturing: Micro-echinate, spines with blunt apices, irregularly distributed.

4. C. sabatius

Shape class: Prolate-spheroidal

Dimensions: P = 27.1 - (32.3) - 40.1, E = 26.2 - (28.3) - 36.4, P / E = 1.03 - 1.10.

Pollen class: Tricolpate

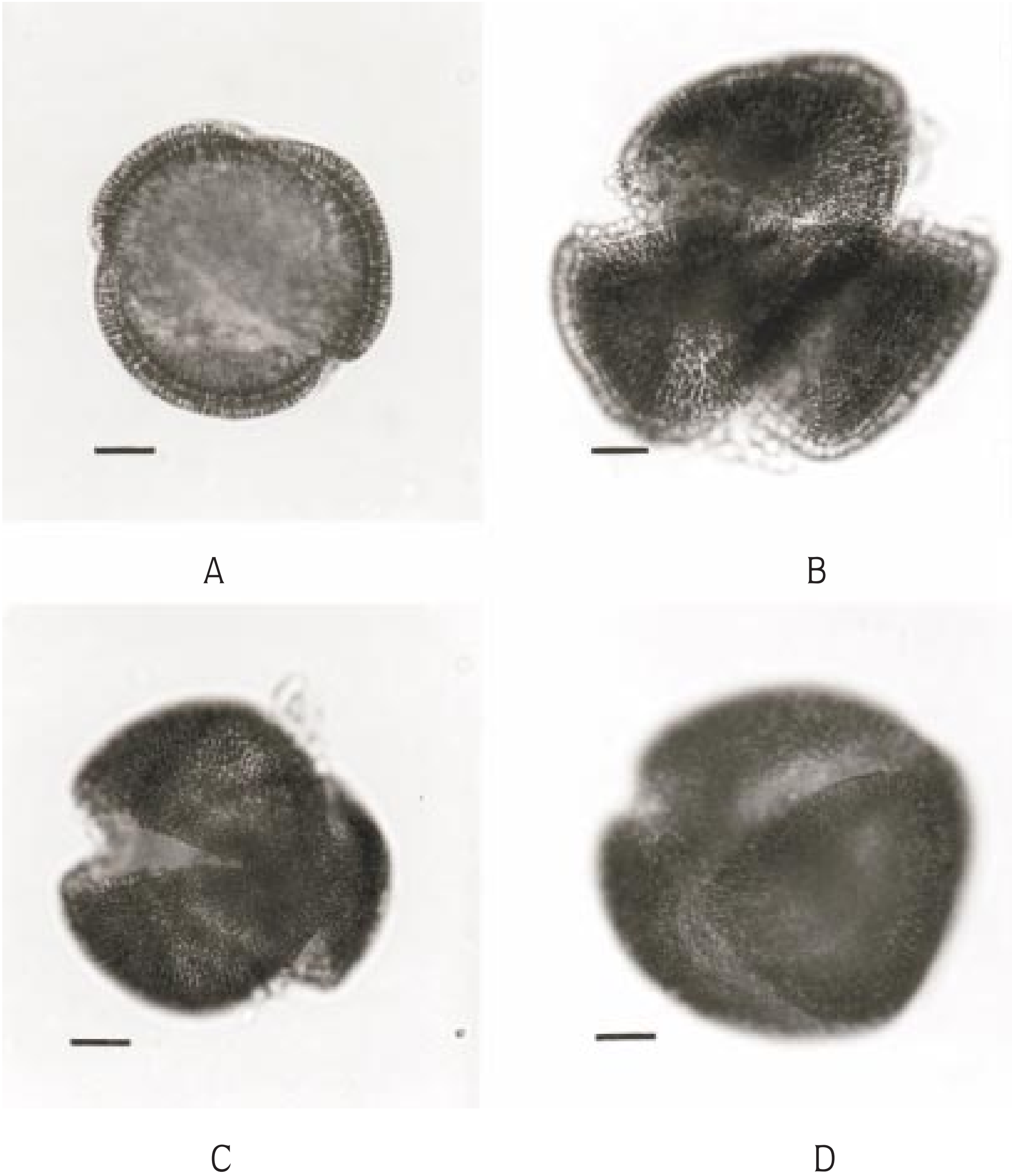


Figure 1. Light micrographs of the pollen grains of *C. humilis* (A), *C. supinus* (B), *C. pitardii* (C) and *C. cantabrica* (D). Scale bar 10 µ.

Appertures: Simple, colpi tapering at both ends, C = 26.5 - (29.1) - 32.3.

Exine: Tectate, tectum perforate; perforations approximately circular, dense, distinct, irregularly distributed, sexine thicker than nexine, N = 0.7 - (0.8) - 0.9, S = 1.2-(1.4)-1.5, N / S = 0.58 - 0.60.

Sculpturing: Micro-echinate, spines with blunt apices, irregularly distributed.

5. C. supinus

Shape class: Suboblate

Dimensions: P = 51.3 - (57.7) - 65.2, E = 62.3 - (68.2) - 75.3, P / E = 0.82 - 0.87

Pollen class: Tricolpate

Appertures: Simple; colpi tapering at both ends, C = 49.3 - (53.3) - 57.6.

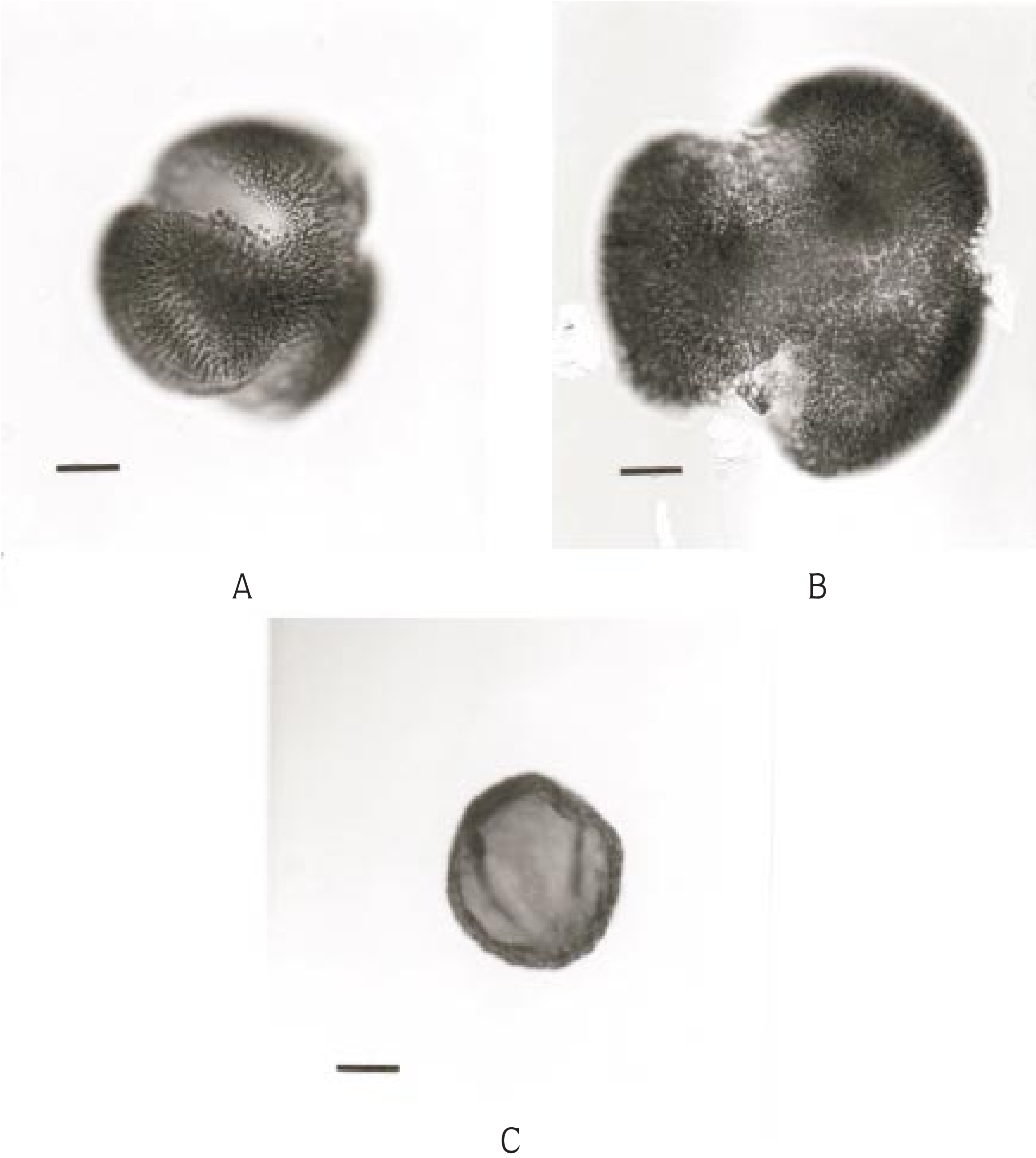


Figure 2. Light micrographs of the pollen grains of *C. trabutianus* (A), *C. valentinus* (B), *C. sabatius* subsp. *sabatius* (C). Scale bar 10 µ.

Exine: Tectate, tectum perforate; perforations approximately circular, dense, distinct, irregularly distributed, sexine thicker than nexine, N = 0.9 - (1.0) - 1.1, S = 2.0 - (2.3) - 2.5, N / S = 0.44 - 0.45.

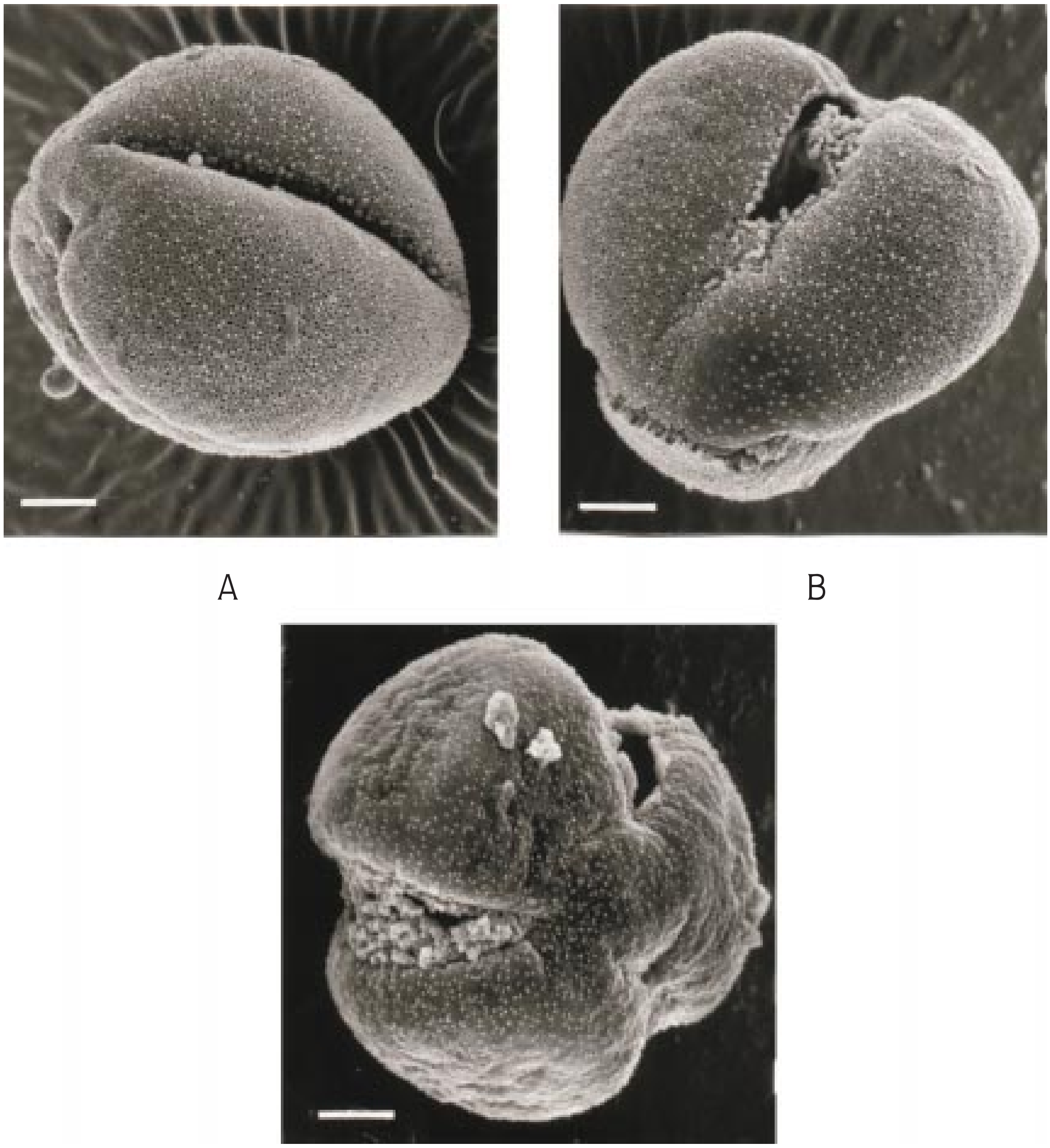
Sculpturing: Micro-echinate, spines with blunt apices, irregularly distributed.

6. C. valentinus

Shape class: Prolate-spheroidal

Dimensions: P = 60.5 - (70.3) - 74.2, E = 60.1 - (69.5) - 72.4, P / E = 101-102.

Pollen class: Tricolpate.



C

Figure 3. SEM micrographs of the pollen grains of *C. dryadum* (A), *C. trabutianus* (B) and *C. arvensis*. Scale bars 10 μ.

Appertures: Simple, colpi tapering at both ends, C = 51.1 - (56.5) - 60.8.

Exine: Tectate, tectum perforate; perforations approximately circular, dense, distinct, irregularly distributed, sexine thicker than nexine, N = 0.8 - (1.0) - 1.1, S = 1.7 - (1.8) - 2.0, N / S = 0.47 - 0.55.

Sculpturing: Micro-echinate; spines with blunt apices, irregularly distributed.

7. C. arvensis L.

Shape class: Prolate-spheroidal.

Dimensions: P = 62.3 - (72.4) - 75.2, E = 60.7 - (70.2) - 72.2, P / E = 1.03-1.04.

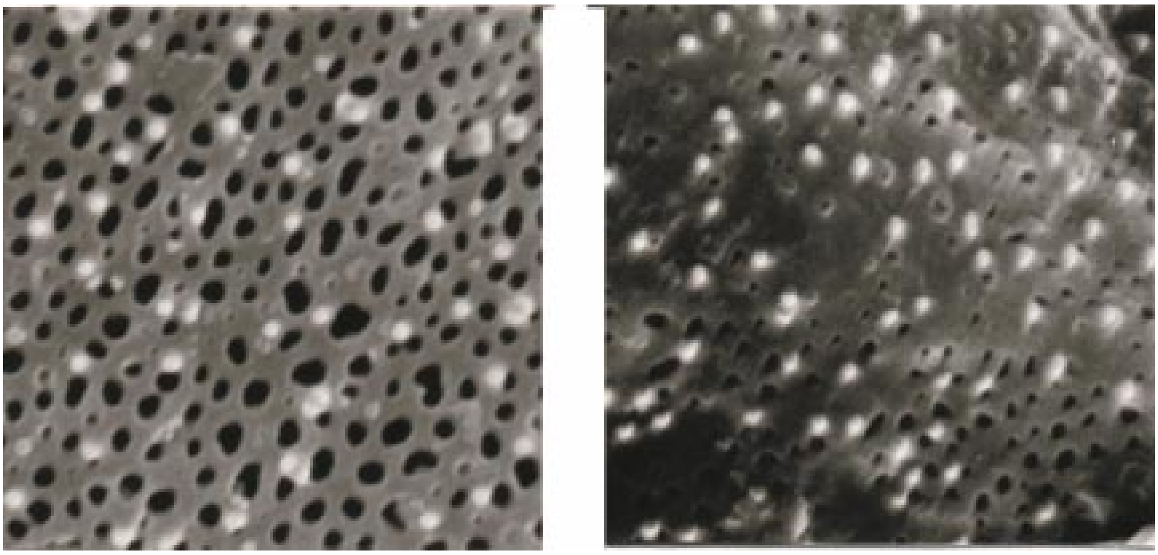


Figure 4. SEM micrographs of the pollen grains surfaces of *C. dryadum* (A) and *C. arvensis* (B) with 10,000 magnification.

Pollen class: Tricolpate

Appertures: Simple, colpi tapering at both ends, C = 57.7 - (63.9) - 68.5.

Exine: Tectate, tectum perforate; perforations approximately circular, dense, distinct, irregularly distributed, sexine thicker than nexine, N = 0.9- (1.0)- 1.1, S = 1.8 - (2.1) - 2.3, N / S = 0.50 - 0. 48.

Sculpturing: Micro-echinate, spines with blunt apices, irregularly distributed.

8. C. dryadum

Shape class: Prolate

Dimensions: P = 48.7 - (57.8) - 65.1, E = 32.7 - (40.6) - 46.8, P / E = 1.39-1.49

Pollen class: Tricolpate

Appertures: Simple, colpi tapering at both ends, C = 44.0 - (48.9) - 51.9.

Exine: Tectate, tectum perforate; perforations approximately circular, dense, distinct, irregularly distributed, sexine thicker than nexine, N = 0.9- (1.1)- 1.3, S = 1.7 -(1.9) - 2.1, N / S = 0.53 - 0.62.

Sculpturing: Micro-echinate, spines with blunt apices, irregularly distributed.

9. C. pitardii

Shape class: Prolate-spheroidal

Dimensions: P = 53.7 - (59.3) - 68.9, E = 52.4 - (58.1) - 66.0, P / E =1.02 -1.04.

Pollen class: Tricolpate

Appertures: Simple; colpi tapering at both ends, C = 46.1 - (49.8) - 54.6

Exine: Tectate, tectum perforate; perforations approximately circular, dense, distinct, irregularly distributed, sexine thicker than nexine, N = 0.9- (1.1)- 1.2, S = 1.8 - (2.0) - 2.1, N / S = 0.50 - 0.57.

Sculpturing: Micro-echinate; spines with blunt apices, irregularly distributed.

Discussion

Lewis & Oliver (1965) studied the pollen grains of the genus *Convolvulus*, of which some species grow in Morocco, but only two taxa endemic to Morocco were included (*C. arvensis* L. and *C. floridus* L. from the Canary Islands, *C. cantabrica* L. from Romania, *C. fruticosus* Pall. and *C. lineatus* L. from the U.S.S.R., *C. gharbensis* Batt. & Pitard and *C. sufruticosus* Desf. var. *oranensis* Pom. from Morocco and cultivated *C. siculus* L and *C. tricolor* L. species). They described the *Convolvulus* pollen grains as 3- or rarely 4-zonocolpate and prolate to subspheroidal. El Ghazali (1993) stated that *C. arvensis* pollen grains are tricolpate with a perforate tectum, ectexine thicker than the endexine and micro-echinate sculpturing. The present study of *Convolvulus* agrees with Lewis & Oliver (1965) and El Ghazali (1993) and does not agree with O`Donell (1955), who stated that the *Convolvulus* pollen grains are tricolporate.

The pollen grains of the genus *Convolvulus* from Morocco show variation between the species that sometimes corresponds with the morphologic features. Therefore, the variations can be used to distinguish between some closely related species. For example, *C. supinus* is morphologically very similar to *C. valentinus* and they were separated on the base of the existence of hairs on both sides of the leaf and slight variations in their leaf shapes (Sa`ad, 1967). The shape of the pollen grain shows another difference in these two species and supports the delimitation of them. Similarly, *C. pitardii* is morphologically very close to *C. dryadum*, from which it is also separated in terms of pollen-grain shape, *C. pitardii,* prolate-spheroidal and the other prolate. In addition, *C. sabatius* is separated from the rest of the taxa examined in having the smallest pollen grains.

However, the pollen characteristics cannot be used to distinguish the sections or subsections.

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