

SEED MORPHOLOGY OF TWO DISTINCT EUROPEAN SPECIES OF *ERICA* L. (ERICACEAE)

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Recibido el 21 de mayo de 2008, aceptado para su publicación el 30 de junio de 2008
Publicado "on line" en septiembre de 2008

ABSTRACT. *Seed morphology of two distinct european species of Erica L. (Ericaceae). Erica spiculifolia* is a distinct species within the genus *Erica*, considered by some authors as part of a monospecific genus: *Bruckenthalia*. The second species studied, *E. umbellata*, is the only European species of section *Pyronium*. Seed morphology of the two species was studied by means of SEM techniques. Seeds from different populations were used. Characters concerning size and shape of seeds, primary and secondary ornamentation were observed and measured. The seeds of *E. spiculifolia* are close to those of other species within the genus, supporting the inclusion in *Erica*. *E. umbellata* seeds have some exclusive characters within the European species of the genus: a verrucate secondary ornamentation and surface cells with "omega type" cell boundaries. The systematics of genus *Erica* is still unresolved, and the taxonomic position of these species has to be clarified. As found in previous studies, seed characters provide useful taxonomic characters that should be used in the interpretation of the taxonomic position of these species within *Erica*.

Key words. *Erica*, Ericaceae, seed morphology, SEM, testa sculpture, taxonomy.

RESUMEN. *Morfología de las semillas de dos especies europeas de Erica L. (Ericaceae). Erica spiculifolia* es una especie con características únicas dentro del género *Erica* en Europa, considerada por algunos autores como perteneciente al género monoespecífico *Bruckenthalia*. La segunda especie estudiada, *E. umbellata*, es la única especie de la sección *Pyronium* dentro del núcleo norte del género. Se estudia la morfología de las semillas mediante la fotografía de microscopía electrónica. Se miden semillas de diferentes poblaciones de cada especie. Se estudian y miden diversos caracteres del tamaño y la forma de las semillas, la ornamentación primaria y secundaria. Las semillas de *E. spiculifolia* coinciden en la mayor parte de los caracteres con otras especies del género, lo que apoya su pertenencia a *Erica*. Las semillas de *E. umbellata* presentan algunos caracteres exclusivos que permiten diferenciarla de otras especies europeas del género: una ornamentación secundaria verrucosa y las uniones entre las células de la testa de "tipo omega". La taxonomía del género *Erica* no está aún resuelta, y la posición sistemática de estas especies debe clarificarse. Como se desprende de éste y otros estudios publicados, la morfología de las semillas aportan caracteres de diagnóstico que deben utilizarse para una correcta interpretación de las relaciones taxonómicas dentro del género.

Palabras clave. *Erica*, Ericaceae, morfología de las semillas, M.E.B., ornamentación de la testa, taxonomía

INTRODUCTION

Erica L. (Ericoideae, Ericaceae) is a large genus of more than 800 species with a wide distribution from Northern Europe to South Africa (Oliver, 2000). Systematics of this genus is still unresolved, because even though a high morphological variation is found for a large set of characters, researchers have failed to establish a consistent classification that would include all this data. The position of the two species included here are not clear and both have been isolated in previous classification attempts.

Bruckenthalia was a monotypic genus described by Reichenbach (1831) for its only species *B. spiculifolia* (Salisb.) Reichenb. This genus was characterized by the absence of bracteoles, the partially fused sepals and a distinct caryology type (Webb, 1972; Stevens, 1978; Oliver, 1989, 1996, 2000). Recently, Oliver (1989, 2000) claims for the inclusion of this species in *Erica* as was originally described by Salisbury (1802).

This proposal is based mostly in the existence of african *Erica* species with fused sepals and bracteoles absent because these are substituting two of the sepals. *E. spiculifolia* occurs in mountain shrublands on acid soils, from 1400 to 2500 metres (Barbero *et al.*, 1975; Browicz, 1983). It is found in the Balkans, Transylvania mountains in Romania and some isolated populations in northern Turkey (Stevens, 1978; Browicz, 1983; fig. 1).

Erica sect. *Pyronium* Salisb. ex Benth. is formed by one European species, *E. umbellata* L. and about twenty species from South Africa (Schuman and Kirsten, 1992). Although this classification is doubtful, *E. umbellata* is a distinct species within the European group, characterized by its inflorescence with terminal whorls of flowers at the end of secondary branches, urceolate corolla and exerted anthers (Bentham, 1839; Hansen, 1950; Bayer, 1993). A few varieties have been described and one raised to subspecies: *E. umbellata* subsp. *major* (Coss. ex Willk.) Pinto da Silva

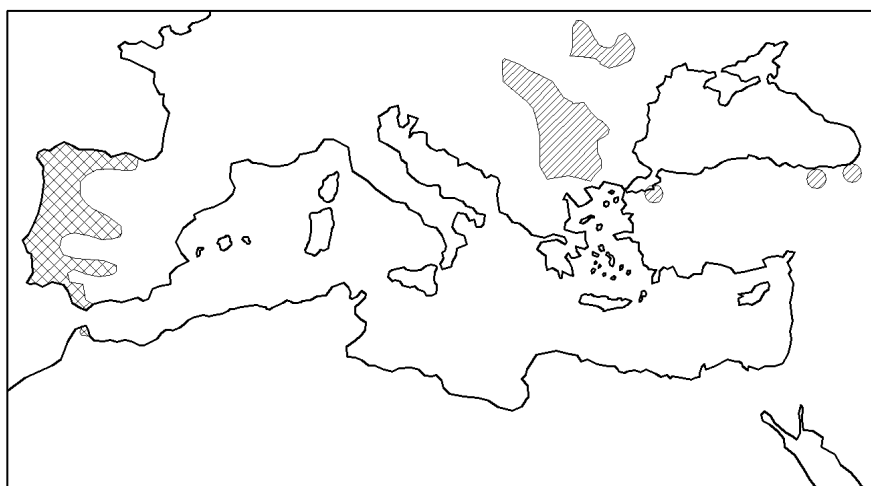


Figure 1. Approximate geographical distribution of *E. spiculifolia* (straight lines) and *E. umbellata* (crossed lines). *Distribución aproximada de E. spiculifolia* (rayado) y *E. umbellata* (cruzado).

& Teles, with larger corollas and less exerted anthers (Pinto da Silva & Teles, 1971). Some other varieties such as var. *subcampanulata* Benth.ex DC., var. *anandra* Lange and var. *filiformis* Merino have been described (Bentham, 1839; Willkomm, 1870; Colmeiro, 1887; Benito, 1948; Hansen, 1950; Bayer, 1993) but with low taxonomic value. This species occurs in degraded shrublands on acid soils, in the western half of the Iberian Peninsula and north Morocco (Benito, 1948; Hansen, 1950; Webb & Rix, 1972; Bayer, 1993; Díaz, 1998; fig. 1).

Seed morphology has recently been used as an aid to systematics in *Erica* (Fagúndez & Izco, 2003, 2008) or its related genera *Calluna* (Fagúndez & Izco, 2004a) and *Daboecia* (Fagúndez & Izco 2004b). In this study, seed morphology was analyzed with the purpose of the establishment of taxonomic relationships with the other European *Erica* species. The systematics of the genus should be revised as Bentham's (1839) classification is poorly supported. The position of these two distinct species is still uncertain and more taxonomic characters as those found in seed morphology will be useful to clarify it.

MATERIALS AND METHODS

Seeds were obtained from seedbanks, field surveys or commercial (tab. 1). Seeds collected in the field were dried in a drying chamber using silica-gel and put into glass tubes with a certain amount of silica-gel with a colour component to detect possible humification. These tubes are then stored in a 4°C chamber. Seeds might be used for different studies or long time preservation (Fagúndez & Izco 2003, 2004a, 2004b, 2008).

When available, 25 to 30 seeds were measured for each population. Seeds were measured with a binocular scope connected to a computer, images were captured and treated with UTHSCSA Image tool (San Antonio, Texas 1997). Characters measured were area (a), perimeter (p), major axis length (M), minor axis length (m), elongation (M/m) and sinuosity ($4\pi a / p^2$). Mean and standard deviation was calculated for each population. Weight of 100 seeds was measured for some populations only of *E. umbellata*. A Münsell colour chart for soils was used for colour determination.

For SEM pictures, 5-10 dry seeds from

Population	Locality	Date	From
spi355	unknown	2005	Comercial
spi356	unknown	2005	Comercial
spi376	RUMANIA: Mt. Bucegi	2005	C. D. Bitá-Nicolae
spi390	GREECE: Makedonia	1999	R.B.G. Kew Seedbank
umb33	SPAIN: A Coruña, Ames	1998	J. Fagúndez SANT 41449
umb68	SPAIN: Pontevedra, Faro	1999	J. Fagúndez SANT 41462
umb74	SPAIN: Lugo, Quiroga	1999	J. Fagúndez SANT 41463
umb81	SPAIN: Huelva, Andévalo	1999	J. Fagúndez & D. Reyes SANT 41475
umb141	SPAIN: Pontevedra, Oia	1999	J. Fagúndez & A. Aguilar
umb264	PORTUGAL: Algarve, Alportel	2000	J. B. Tapada da Ajuda
umb278	PORTUGAL: Beira Litoral, Buçaco	2002	J. B. Tapada da Ajuda

Table 1. Data of studied populations. *Poblaciones estudiadas*.

five populations were mounted on metal stubs using double-sided adhesive tape and gold-coated. Pictures were taken of whole seeds and of specific details for three seeds from each population. Three testa cells were randomly chosen and measures taken in the same way as for the whole seed.

Nomenclature used is that of Berggren (1981) for shape description and Barthlott (1981, 1984) for the ornamentation.

RESULTS

Values of size and shape of seeds are summarized in table 2. Both species have seeds around 0.5 mm, minor axis of 0.3 mm and elongation close to 1.75 (axis ratio 2:1 to 3:2). Shape of the seeds is variable, either simmetrical from hilum, ellipsoid or oblong, or with hilum laterally displaced, curved to kidney-shape, mostly in *E. spiculifolia* (fig. 2 E). In this species, a lateral wing is sometimes observed (fig. 2 A).

The primary ornamentation is reticulate in both species, with elongated surface cells. *E. spiculifolia* has prominent outer anticlinal

cell walls, mostly totally fused, while cell boundaries are channelled in *E. umbellata*. This species has strongly sinuate cell margins described as “omega type” (Barthlott, 1981, 1984). The secondary sculpture of *E. umbellata* has surface cells covered with micropapillae in both outer anticlinal and outer periclinal walls (fig. 3 D). In *E. spiculifolia*, surface is rugulose, with no clear pattern (fig. 2 B, D and F).

The interpopulations variability for the measured characters is high (tab. 2) for both species, but mostly for *E. umbellata* with populations that almost do not overlap, as populations “umb 33” and “umb 278”. Weight of 100 seeds was 1.9 mg for “umb 33” but 2.5 to 2.8 for the other populations. Colour of mature seeds was dark to very dark brown in *E. umbellata*, while *E. spiculifolia* has lighter seeds, reddish brown or dark reddish brown.

No seed characters would support the existence of *E. umbellata* subsp. *major*, that would eventually correspond to populations “umb81” and “umb264”. These populations are similar to all the rest according to the characters observed.

Population	N	Area	Perimeter	Main axis	Minor axis	Elongation	Sinuosity
spi355	29	0.1±0.01	1.32±0.06	0.49±0.03	0.26±0.02	1.87±0.18	0.72±0.04
spi356	25	0.12±0.02	1.44±0.08	0.53±0.04	0.3±0.03	1.78±0.18	0.74±0.03
spi376	13	0.11±0.02	1.37±0.09	0.51±0.03	0.29±0.03	1.75±0.17	0.73±0.05
spi390	9	0.12±0.01	1.45±0.05	0.53±0.02	0.3±0.02	1.8±0.13	0.72±0.05
Total <i>E. spiculifolia</i>	76	0.11±0.02	1.38±0.09	0.51±0.04	0.28±0.03	1.81±0.18	0.73±0.04
umb33	25	0.11±0.02	1.46±0.17	0.49±0.04	0.3±0.02	0.68±0.11	1.63±0.14
umb68	30	0.14±0.01	1.59±0.11	0.57±0.05	0.31±0.03	0.69±0.07	1.88±0.3
umb74	31	0.15±0.02	1.68±0.13	0.56±0.05	0.33±0.03	0.65±0.05	1.72±0.16
umb81	31	0.14±0.02	1.61±0.17	0.56±0.06	0.34±0.03	0.74±0.06	1.64±0.2
umb141	24	0.15±0.01	1.61±0.09	0.57±0.04	0.33±0.03	0.71±0.05	1.75±0.18
umb264	28	0.14±0.02	1.52±0.09	0.55±0.03	0.32±0.03	0.76±0.04	1.73±0.16
umb278	32	0.19±0.02	1.78±0.09	0.64±0.04	0.38±0.03	0.76±0.03	1.72±0.15
Total <i>E. umbellata</i>	201	0.15±0.03	1.61±0.16	0.56±0.06	0.33±0.04	1.72±0.21	0.71±0.07

Table 2. Mean ± standard deviation of seed characters. All values in mm except for area (mm²), elongation and sinuosity. *Media ± desviación típica de los caracteres seminales. Todos los valores en mm excepto area (mm²), elongación y sinuosidad.*

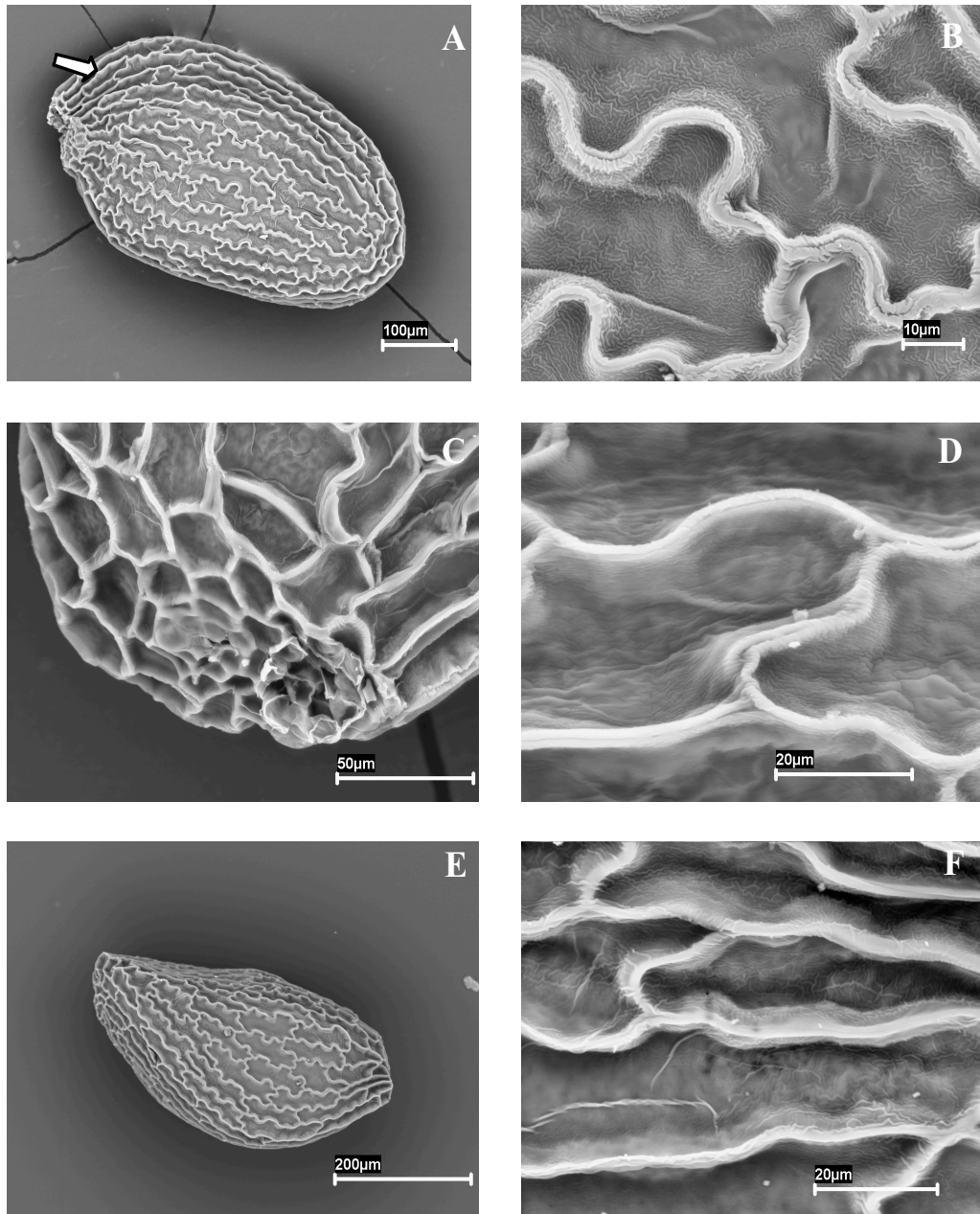


Figure 2. SEM pictures of the seeds of *E. spiculifolia*. **A** and **B**: spi390, **C** and **D**: spi355, **E**: spi356, **F**: spi376. **A**: whole seed, the winged side is pointed. **B**: detail of the secondary sculpture. **C**: detail of hilum region. **D** and **F**: detail of surface cells. **E**: whole seed. *Fotografías de M.E.B. de las semillas de E. spiculifolia. A y B: spi390, C and D: spi355, E: spi356, F: spi376. A: semilla completa. Se señala el ala lateral. B: detalle de la ornamentación secundaria. C: detalle de la región hilar. D y F: detalle de las células de la testa. E: semilla completa.*

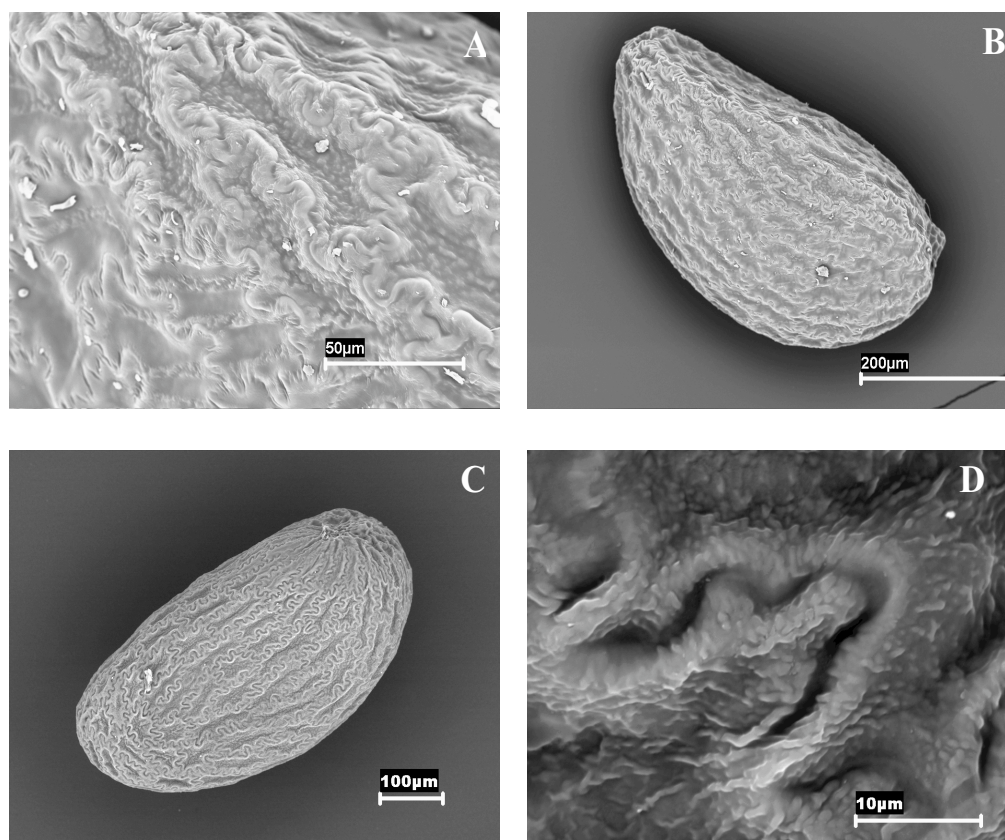


Figure 3. SEM pictures of *E. umbellata*. **A** and **B**: umb264, **C**: umb68, **D**: umb81. **A**: detail of elongated testa cells with «omega type» cell boundaries. **B** and **C**: whole seed. **D**: detail of channelled cell boundaries and surface of outer anticlinal and outer periclinal cell walls, with micropapillae. *Fotografías de M.E.B. de E. umbellata. A y B: umb264, C: umb68, D: umb81. A: detalle de las células alargadas de la testa con uniones intercelulares de «tipo omega». B y C: semilla completa. D: detalle de las uniones intercelulares acanaladas y de las paredes anticlinales y periclinales externas de las células de la testa, con micropapilas.*

Seed description

Erica spiculifolia Salisb.

Seeds reddish brown to dark reddish brown. Shape ellipsoid or kidney-shape. Outline elliptic or curved on the hilum region and apiculated on the distal end. Symmetrical from hilum or this displaced to one side. Cross section flat, sometimes with a lateral wing. 0.45-0.55 mm long. 0.25-0.35 mm

wide. Axis ratio 2:1 to 3:2. Appendages absent. Primary ornamentation reticulated, with large cells in the direction of the main axis. 6-10 cells in main axis, elongated but gradually isodiametric towards the polar areas. Cells 100-150 µm long 20 µm wide, with sinuate margins. Outer anticlinal walls prominent, continued by the outer periclinal walls, these smooth or continuing the ornamentation of the outer periclinal walls,

these rugulate. Cell boundaries anastomosing or rarely partially anastomosing.

Erica umbellata L.

Seeds dark to very dark reddish brown. Weight of 100 seeds 2-3 mg. Seed shape variable, ovate or curved, sometimes almost kidney-shape, outline with an oval or oblong base and elliptic apex, cross section elliptic with second axis significantly larger than third axis. 0.45-0.6 mm long. 0.3-0.35 mm wide. Appendages absent. Terminal hilum displaced to one side, sometimes protruding. Primary ornamentation irregularly reticulate, formed by cells of 100-150 µm long 50-100 µm wide, isodiametric towards the polar areas. 8-12 cells in main axis. Very sinuate cell margins, imbricate, "omega type". Outer anticlinal walls thick, continued by the outer periclinal walls. Secondary ornamentation strongly verrucate. Cell boundaries channelled, with occasional fusion points.

DISCUSSION

Bruckenthalia or *Erica*

Seeds of *E. spiculifolia* have been studied by Wilson *et al.* (1973) for the purpose of fossil identification, described as of 0.53-0.61 mm, reticulate with elongated surface cells and curve to sinuate, partially or totally divided cell boundaries. Its seeds are also considered similar to those of *E. lusitanica*. Our results mostly agree with this description, but no similarities were found with *E. lusitanica* seeds, which are smaller, rounder and with isodiametric surface cells (unpub. results).

The taxonomic value of genus *Bruckenthalia* has been strongly discussed and it is now considered as part of *Erica* (Oliver, 1989, 1996, 2000; Kron *et al.*, 2002; Nelson 2007). Absence of bracteoles is shared with some African *Erica* species, and

European species such as *E. scoparia* also have a fused calyx. However, the caryology type, the distinct syninflorescence type and other characters results in *E. spiculifolia* as a very distinct species within the northern nucleus of the genus. We have also observed some original characters in the study of herbarium material, such as occasionally fused stamen filaments at its half length, another exclusive character for the northern species.

Seed morphology has differences and similarities with the other European *Erica* species. The presence of winged seeds, result of the embryo lateral position, is found in *E. spiculifolia* and *E. arborea*. Shape of the seeds is the most variable of the observed characters, as sometimes seeds are curved and apiculated as found in *E. terminalis* or *E. maderensis* (Fagúndez & Izco, 2008), but sometimes regular, symmetrical, elliptic or oblong in its outline. Surface cell pattern is similar to the *E. scoparia* type (Fagúndez & Izco, 2003), with elongated cells, sinuate margins and cell boundaries anastomosing. None of these characters are conclusive about the systematic affinities of these species, although it seems that *E. arborea* and *E. scoparia* are the most likely to be the closest species according to seed morphology.

A recent study on the phylogenetic relationships of the African and European *Erica* species based on chloroplast and nuclear DNA (McGuire & Kron, 2005) resulted in *E. spiculifolia* as a sister species of the rest. This would suggest that seed characters such as winged seeds or a reticulate ornamentation could be primitive states within *Erica*.

Erica umbellata and sect. *Pyronium*

The *E. umbellata* seeds have been analyzed also for fossil studies by Huckerby *et al.* (1972), and for descriptive purposes

by Fraga (1984). Size of the seeds is 0.5-0.6 mm, and the verrucate sculpture pattern is highlighted by both authors. Our results mainly agree with their description.

Section *Pyronium*, represented by *E. umbellata* in the European nucleus, has clear distinct characters. The muticous, exerted anthers, glabrous ovary and pink, urceolate corolla are shared with *E. multiflora* and the other species of the sect. *Gypsocallis*, whereas the syninflorescence type with whorls of flowers at the end of secondary branches are closer to *E. australis* (sect. *Tylospora*) and other species. Seed morphology has two exclusive characters: the “omega type” surface cells boundaries and a strongly verrucate (micropapillate) secondary sculpture. Both characters should be considered as with a high taxonomic importance as stated by Barthlott (1984). This supports the existence of an exclusive section for this species, although infrageneric classification in this large genus needs a thorough review (Oliver, 2000). Comparisons with the African species of *Pyronium* will help to clarify it. Because of its original seed morphological type, no relations with other European sections or species can be established from seed morphology of *E. umbellata*.

ACKNOWLEDGEMENTS. We would like to thank every person and institution that has provided seeds for this study (see table 1). Partially supported by the project: CGL2006-06890, Ministerio de Educación y Ciencia.

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