J. Bryol. (1992) **17**, 275–283

The taxonomic status of *Tortula muralis* var. *baetica* (Musci, Pottiaceae): a comparative study

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INTRODUCTION

Tortula muralis Hedw. is morphologically a very variable taxon, of which many varieties have been described (Wijk *et al.*, 1969). Some of them are very difficult to name in practice because the morphological variation of the relevant characters (hair-point length, colour of the plant, leaf size, stem height, size of the capsule) is more or less continuous. Even plants with different ploidy levels ($n = 26/27$ and $n = 50/52$) can be distinguished only in part by their size; hence, taxonomic recognition of two cytotypes is not justified (Newton, 1968).

In the Iberian Peninsula, six varieties of *T. muralis* have been recognized, including var. *muralis*, var. *aestiva* Brid. ex Hedw., var. *baetica* Casas & Oliva, var. *incana* B., S. & G., var. *obcordata* (Schimp.) Limpr. and var. *rupestris* K. F. Schultz (Casas, 1981). Of these we believe that only three can be distinguished easily and with certainty from each other and from the var. *muralis*: the var. *obcordata* by its strongly emarginate leaves, the var. *baetica* by its peculiar type of papillosity and the var. *aestiva*, widely recognized in Europe (Smith, 1978; Nyholm, 1956; Frahm & Frey, 1983). Smith (1978) noted that the 'var. *aestiva* is distinctive in its appearance and is probably more than just a habitat form'. Correct determination of the other varieties is uncertain because of the continuous wide range of variation they display.

The purpose of this paper is to present the results of a comparative study based on the varieties *obcordata* and *baetica*, both described from samples collected on the Iberian Peninsula (Schimper, 1876; Casas & Oliva, 1982), and the type variety, *muralis*, the aim being to determine the true taxonomic status of these taxa. This study was prompted by the knowledge that other infraspecific taxa of *T. muralis* have proved to warrant specific recognition. *Tortula muralis* fa. *gypsophila* (Amann, 1918), for example, is now recognized as *T. brevissima*, as described by Schiffner (1913) (Reimers, 1941; Boudier, 1988).

No type specimen of *T. muralis* Hedw. var. *muralis* Hedw., *Sp. Musc.* 123. 1801, was designated by Hedwig (1801), but we were able to study the lectotype from Geneva (G).

In the original description of *T. muralis* var. *obcordata* (Schimp.) Limpr. (Schimper, 1876), two localities were mentioned and samples from both are in BM. One of them (pr. Hyéres, terrasse du Grand Hôtel, Reuter leg.) does not fit the original description. The other (In ponte Guadalaviar prope Valenciam Hispaniae, 21 June 1847, Schimper leg., lectotype in BM), is therefore selected as the lectotype.

The type of *T. muralis* Hedw. var. *baetica* Casas & Oliva, España, Sevilla, Morón de la Frontera (holotype in Herb. Oliva, Córdoba), was also available for study.

METHODS

The biometric study of leaf cells involved three samples each of *T. muralis* var. *baetica*, var. *obcordata* and var. *muralis*. Two adult leaves were taken from 3 different shoots of each sample (see Appendix). The length and width of 5 cells in the upper half of each leaf were measured and the mean values for each leaf were calculated.

Leaves were washed with distilled water, fixed with glutaraldehyde, impregnated in Spurr resin and stained with toluidine blue and semifine transverse sections of leaves were made using a Reichert Jung Ultramicrotome.

For scanning electron microscopy, selected leaves were fixed in 3% glutaraldehyde with 0.1 M cacodylate buffer at 4°C, submitted to critical point drying in 100% acetone and liquid CO_2, sputtered with a 250–300 Å thick gold layer, and observed and photographed with a JEOL JSM T-300 operated at 10–20 kV.

For transmission electron microscopy, the spores were similarly fixed in 3% glutaraldehyde and post-fixed in 1% osmic acid in the same buffer mentioned above. After washing and dehydration in a graded ethanol series, the material was embedded in epoxy resin. Sections were made using a Reichert Jung Ultramicrotome and stained with uranyl acetate and lead citrate before studying with a Zeiss EM 10c electron microscope operated at 60 kV.

RESULTS AND DISCUSSION

Size of the leaf cells

In the genus *Tortula*, the size of the middle and upper leaf cells has been used as a character to differentiate between similar species — for example, between *T. intermedia* (Brid.) De Not. and *T. princeps* De Not., and between *T. muralis* var. *aestiva* Hedw. and *T. vahliana* (Schultz) Mont. (Smith, 1978) — as well as to separate *T. geheebiaeopsis* (C. Müll.) Broth. from *T. filaris* (C. Müll.) Broth. and *T. rubra* Mitt. (Lightowlers, 1985, 1986).

Data obtained during the present study (Fig. 1) show a certain degree of overlap in cell size of the three taxa. However, it can be seen that there is less overlap between the variety *baetica* and the other taxa, and the cells of the former are somewhat smaller than those of the latter. The mean length of the upper leaf cells is 7.75 μm and the mean width 8.25 μm. These values are noticeably lower than those of the varieties *obcordata* (mean length = 9.20 μm, mean width = 11.75 μm) and *muralis* (mean length = 12.2 μm, mean width = 13.1 μm). The last are practically identical to those given by Newton (1968) as ranges and means for *T. muralis*, which included the haploid cytotype with $n = 26/27$.

Anatomy of the leaf nerve

Detail of the leaf nerve, as seen in transverse section, has been regarded by Kramer (1980) as a character of the first order of importance in taxonomy of section *Rurales* De Not. It also serves to distinguish between species such as *T. mucronifolia* Schwaegr. and *T. subulata* Hedw. of the section *Tortula* Spruce. Similarly, the present study has shown that *T. muralis* var. *baetica* is quite distinct in

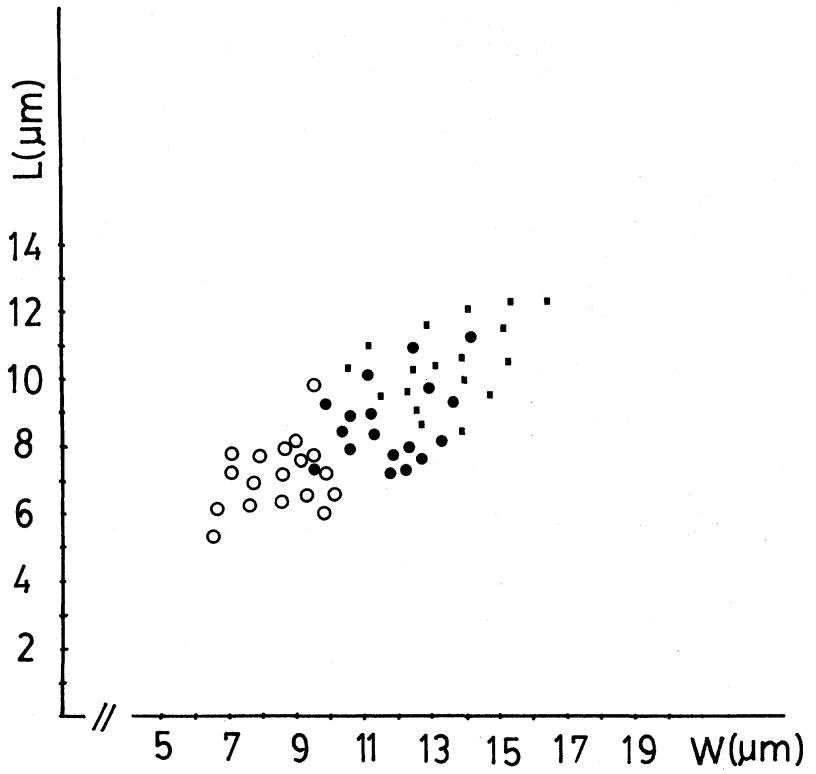

Fig. 1. Relative cell size at the uppermost part of the leaf. *Tortula muralis* var. *muralis* (squares), *T.m.* var. *obcordata* (solid circles), *T. m.* var. *baetica* (open circles). W = width, L = length.

this respect from the other two varieties. The differences are found in ventral cells, abaxial epidermal cells, stereid cells and nerve outline.

In var. *baetica* the ventral cells are markedly ampullose with 1(–2) protuberant mamillae in the upper and middle part of the leaf (Fig. 2*a*, *b*). In the other two taxa these cells are more or less cubical (Fig. 2*c*, *d*, see arrows). They are not ampullose but bear 3–4(5) papillae.

The abaxial epidermal cells, which in the var. *baetica* are generally ampullose in the upper third of the leaf, have one mamilla and thin walls (Fig. 2*a*). In the middle and the lower third of the leaf these cells lack mamillae and the walls are thickened considerably (Fig. 2*b*). In the other two taxa, the abaxial epidermal cells lack mamillae throughout the leaf and all their walls are thickened (Fig. 2*c*, *d*).

The stereid cells in the upper third of the leaf are considerably fewer in var. *baetica*, with 10–12 (Fig. 2*a*), than in the other taxa with 20–30(40) (Fig. 2*c*, *d*). In var. *baetica* the number increases considerably in the lower third, but in the other taxa they are numerous throughout.

The nerve outline is semicircular from the upper third to the base, in var. *baetica* (Fig. 2*a*, *b*). In the other two taxa it is usually elliptical from the middle to the base of the leaf (Fig. 2*c*, *d*), but is occasionally semicircular up to the apex.

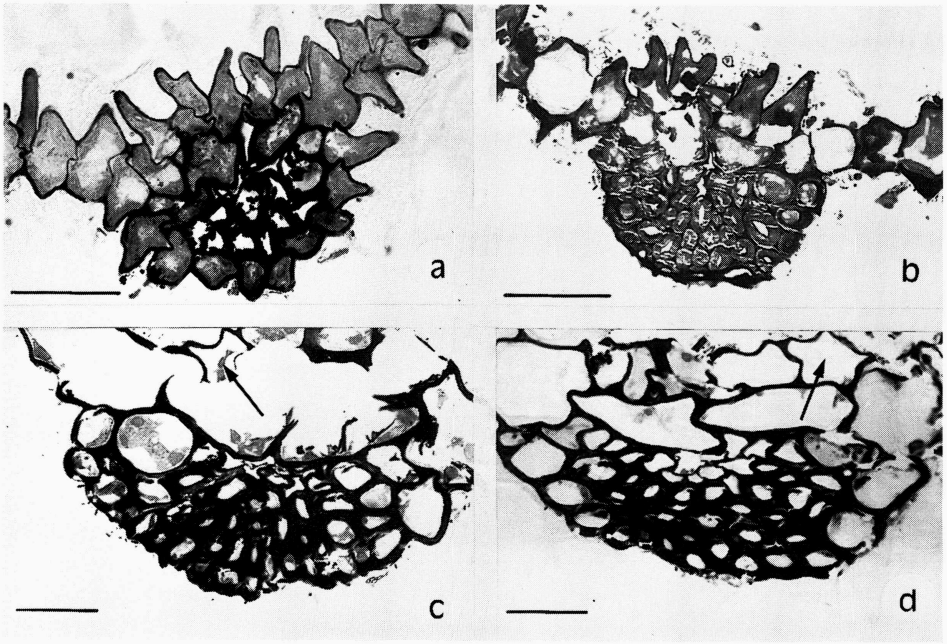

Fig. 2. Light microscope micrographs of semifine transverse sections of the leaves. *a*, *Tortula muralis* var. *baetica*, upper part. Bar = 25 μm; *b*, var. *baetica*, middle part. Bar = 25 μm; *c*, var. *muralis*, middle part. Bar = 20 μm; *d*, var. *obcordata*, middle part. Bar = 20 μm.

Laminal papillosity

The type of foliar papillosity is considered to be an important character and its usefulness in certain genera of the Pottiaceae (Delgadillo, 1975; Frey & Kürschner, 1984), including *Tortula* (Steere, 1940; Kramer, 1980), is widely recognized. Its phylogenetic implications have also been recognized (Mishler, 1986). In this respect, the differences between the var. *baetica* and the other two taxa treated here are highly significant and alone present strong evidence to support the elevation of the var. *baetica* to specific rank. The most frequent type of papillosity in the genus *Tortula* is that shown by the var. *muralis* and var. *obcordata*. On both the adaxial and abaxial surfaces, the upper leaf cells have 4–5(6) bi-or trifurcate papillae or mamillae (Fig. 3*e–h*). In the leaf of var. *baetica*, however, both surfaces of the upper and middle leaf cells possess single or occasionally bifurcate mamillae or very high papillae (Fig. 3*a–d*). The ornamentation is thus identical to that of the dorsal and ventral cells of the nerve (Fig. 3*d*).

The sporoderm

Even within a single genus, spores of the Pottiaceae sometimes exhibit taxonomically useful features (Lewinsky, 1974; Saito & Hirohama, 1974; Boros & Jarai-Komlodi, 1975; Scott & Stone, 1976; Stone, 1988, 1989; Carrión *et al.*, 1990; Casas *et al.*, 1990; Casas & Sérgio, 1990). In general, the exine contributes little to spore sculpturing, and the processes are made up almost exclusively of a very

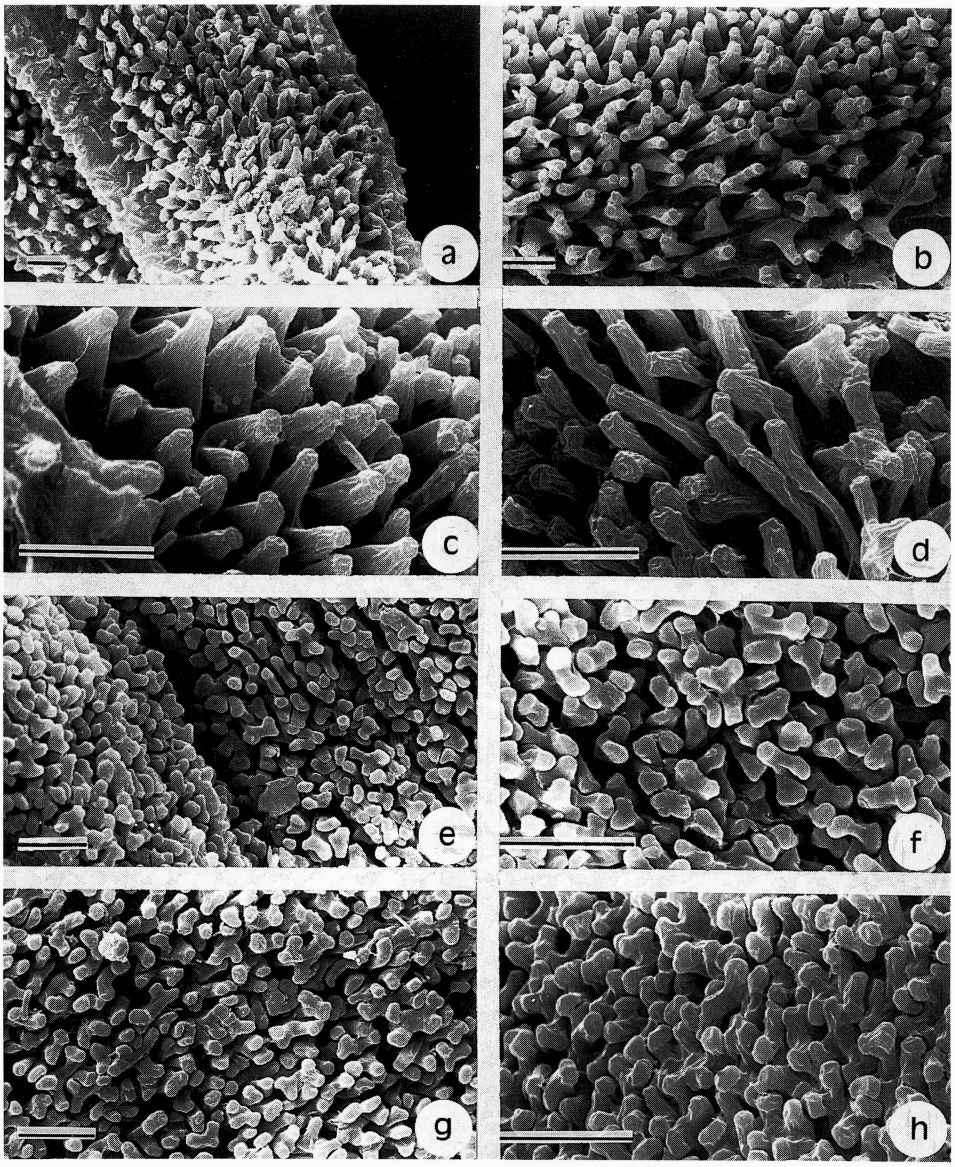

Fig. 3. Scanning electron microscope micrographs of leaf surfaces. *a*, *Tortula muralis* var. *baetica*, abaxial surface and nerve; *b*, var. *baetica*, adaxial surface near nerve; *c*, var. *baetica*, adaxial surface; *d*, var. *baetica*, adaxial surface of nerve; *e*, var. *muralis*, adaxial surface near nerve; *f*, var. *muralis*, adaxial surface; *g*, var. *obcordata*, adaxial surface near nerve; *h*, var. *obcordata*, adaxial surface. Bars = 10 μm.

electron-dense material of a perinic nature (McClymont & Larson, 1964; Carrión *et al.*, 1990).

Saito & Hirohama (1974) have previously discriminated between *T. muralis* and *T. princeps* on the basis of secondary patterns of ornamentation over the main processes of the former species. Surface-spore outlines were also studied in *Tortula*

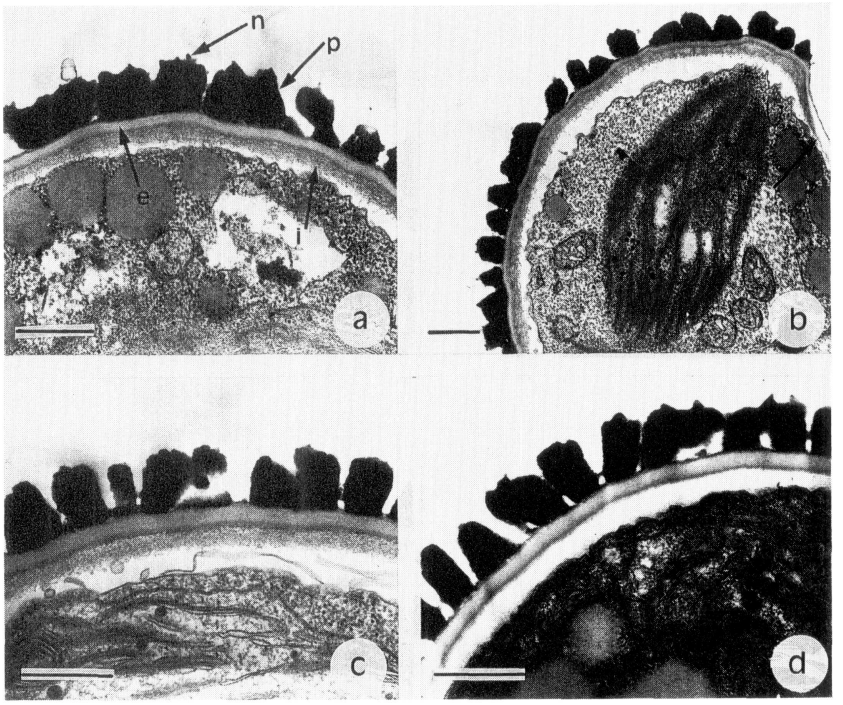

Fig. 4. Transmission electron microscope micrographs of spores showing inner sporoderm structure. *a*, *b*, *Tortula muralis* var. *baetica*; *c*, var. *muralis*; *d*, var. *obcordata*. Bars = 1.3 μm. Perinic elements (p) are proportionally broader in var. *baetica*. Relative thickness of exine (e) and intine (i) are similar in the three taxa. Nano-processes (*n*) covering surface of perine can be seen.

by Lewinsky (1974), although structural interpretation was inaccurate because sporomorphs were poured on a formvar/carbon coated grid and viewed under the microscope without previous preparation. Nevertheless, Lewinsky did demonstrate that the shape of secondary processes was of taxonomic value. In *T. muralis*, the papillae are conical or rounded and cover the outer surface completely (Fig. 4*a–d*). Perinic electron-opaque projections are very variable. Baculate, gemmate, clavate, pilum-like, large warts or irregular processes could be distinguished. Nano-spinules or nano-granulous elements are also visible. As in *Phascum* (Carrión *et al.*, 1990) and other mosses (Larson, 1964; Reighard, 1967; Olesen & Mogensen, 1978; Brown & Lemmon, 1980, 1981, 1985), internal polarity is displayed by *Tortula* spores (Fig. 4*b*, see arrow).

Although some intrapopulational variation was observed, proportionally broader processes and higher spinule-like nano-processes are noteworthy on the distal surfaces of mature spores of *T. muralis* var. *baetica* (Fig. 4*a*, *b*). These variations pertain to all populations studied of *T. muralis* var. *baetica* and are evident, although the taxonomic significance is debatable.

Ecological behaviour

It is interesting that ecological characteristics are identical in the taxa considered, all three of which colonize walls and rocks in nitrophilous sites. Frequently *T. muralis* var. *baetica* has been observed growing mixed with var. *muralis* and var. *obcordata*. On the other hand, populations morphologically intermediate between var. *muralis*, var. *obcordata* and var. *baetica* have not been found. These data support the hypothesis that genetic barriers isolate *T. muralis* var. *baetica* (see Grant, 1963, 1971).

Conclusions

The special type of leaf papillosity shown by *T. muralis* var. *baetica* is uncommon in the genus *Tortula*. This, together with a leaf nerve anatomy that is distinct from that of var. *muralis* and var. *obcordata*, the smaller size of the upper leaf cells and the configuration of the sporoderm, seems sufficient to warrant recognition of this taxon as a distinct species.

Tortula baetica (Casas & Oliva) Guerra & Ros, *stat. nov.*
Basionym: *Tortula muralis* Hedw. var. *baetica* Casas & Oliva, *Acta Bot. Malacitana* **7:** 104. 1982.

On the other hand, the characters studied are completely overlapping in *T. muralis* var. *muralis* and var. *obcordata*. Therefore it is considered that the latter taxon should retain varietal status or perhaps be regarded just as a form.

Summary

Tortula muralis var. *obcordata* and *T. muralis* var. *baetica*, two taxa described from samples collected in Spain, were compared with *T. muralis* var. *muralis* in order to establish their taxonomic status. The results obtained from a biometric study support maintenance of varietal status for var. *obcordata* and recognition of var. *baetica* as a species, *T. baetica* (Casas & Oliva) Guerra & Ros. *stat. nov.*

Acknowledgments

We are grateful to curators and directors of herbaria consulted for the loan of material, to Dr Martha E. Newton for revising the English manuscript and to the staff of the Electron Microscopical Service of Murcia University for technical assistance. This research was supported by the Council of Culture and Education of the Region of Murcia and DGICyT from Spain (Project PB90-0301-C02-01).

References

Amann, J. (1918). *Flore des Mousses de la Suisse*. Part 2. Lausanne.

Boros, A. & Jarai-Komlodi, M. (1975). *An atlas of recent European moss spores*. Akadémiai Kiadó, Budapest.

Boudier, P. (1988). *Tortula brevissima* Schiffner (Pottiaceae, Musci) nouveau pour la bryoflore de France et de Suisse. *Cryptogamie Bryol. Lichénol.* **9,** 219–230.

Brown, R.C. & Lemmon, B.E. (1980). Ultrastructure of sporogenesis in a moss, *Ditrichum pallidum*. III. Spore wall formation. *Am. J. Bot.* **67,** 918–934.

Brown, R.C. & Lemmon, B.E. (1981). Aperture development in spores of the moss *Trematodon longicollis* Mx. *Protoplasma* **106,** 273–287.

Brown, R.C. & Lemmon, B.E. (1985). Phylogenetic aspects of sporogenesis in *Archidium*. *Monogr. Syst. Botany Missouri Botanical Garden* **11,** 25–39.

Carrión, J.S., Guerra, J. & Ros, R.M. (1990). Spore morphology of the European species of *Phascum* Hedw. (Pottiaceae, Musci). *Nova Hedwigia* **51,** 411–433.

Casas, C. (1981). *The mosses of Spain: an annotated checklist.* Treballs de l'Institut Botànic de Barcelona 7.

Casas, C. & Oliva, R. (1982). Aportación al conocimiento de la brioflora de Andalucía noroccidental (Huelva, Sevilla y Córdoba). *Acta Bot. Malacitana* **7,** 97–118.

Casas, C. & Sérgio, C. (1990). *Acaulon fontiquerianum* sp. nov. de la Península Ibérica. *Cryptogamie Bryol. Lichénol.* **11,** 57–62.

Casas, C., Sérgio, C., Cros, R.M. & Brugués, M. (1990). Datos sobre el género *Acaulon en la Península Ibérica. Cryptogamie Bryol. Lichénol.* **11,** 63–70.

Delgadillo, M.C. (1975). Taxonomic revision of *Aloina, Aloinella* and *Crossidium* (Musci). *Bryologist* **78,** 245–303.

Frahm, J.P. & Frey, W. (1983). *Moosflora*. Eugen Ulmer, Stuttgart.

Frey, W. & Kürschner, H. (1984). Studies in Arabian bryophytes 3: *Crossidium asirense* (Pottiaceae) a new species from Asir Mountains (Saudi Arabia). *J. Bryol.* **13,** 25–31.

Grant, V. (1963). *The Origin of Adaptations*. Columbia University Press, New York.

Grant, V. (1971). *Plant Speciation*. Columbia University Press, New York.

Hedwig, J. (1801). Species Muscorum frondosorum descriptae. . , Leipzig.

Kramer, W. (1980). *Tortula* Hedw. sect. *Rurales* De Not. (Pottiaceae, Musci) in der östlichen Holarktis. *Bryophytorum Bibliotheca* 21. J. Cramer, Vaduz.

Larson, D.A. (1964). Further electron microscopic studies of exine structure and stratification. *Grana* **5,** 265–276.

Lewinsky, J. (1974). An electron microscopical study of the genus *Tortula* Hedw., with special reference to exine ornamentation. *J. Bryol.* **8,** 269–273.

Lightowlers, P.J. (1985). A synoptic flora of south Georgian mosses: *Tortula*. *Br. Antarct. Surv. Bull.* **67,** 41–47.

Lightowlers, P.J. (1986). Taxonomy and distribution of the subantarctic species of *Tortula*. *J. Bryol.* **14,** 281–295.

McClymont, J.W. & Larson, D.A. (1964). An electron-microscopic study of spore wall structure in the Musci. *Am. J. Bot.* **51,** 195–200.

Mishler, B.D. (1986). Ontogeny and phylogeny in *Tortula* (Musci: Pottiaceae). *Syst. Bot.* **11,** 189–208.

Newton, M.E. (1968). Cyto-taxonomy of *Tortula muralis* Hedw. in Britain. *Trans. Br. bryol. Soc.* **5,** 523–535.

Nyholm, E. (1956). *Illustrated Moss Flora of Fennoscandia. II. Musci*. Fasc. 2. Swedish Natural Science Research Council, Stockholm.

Olesen, P. & Mogensen, G.S. (1978). Ultrastructure, histochemistry and notes on germination stages of spores in selected mosses. *Bryologist* **81,** 494–516.

Reighard, J.A. (1967). *Light and electron microscopic studies on spore germination and bud apical meristems in* Polytrichum juniperinum *Hedw. and* P. ohioense *Ren. & Card.* Ph.D. Thesis, University of Illinois, USA.

Reimers, H. (1941). *Tortula brevissima* Schiffn., ein neues vorderasiatisches Wüstensteppenmoos im Zechstein-Kyffhäuser. *Notizbl. Bot. Gart. Berlin-Dahlem* **15,** 402–405.

Saito, K. & Hirohama, T. (1974). A comparative study of the spores of taxa in the Pottiaceae by use of the Scanning Electron Microscope. *J. Hattori bot. Lab.* **38,** 475–488.

Schiffner, V. (1913). Bryophyta aus Mesopotamien und Kurdistan, sowie Syrien, Rhodos, Mytilini und Prinkipo. *Ann. K.K. Naturhist. Hofnus.* **27,** 472–504.

Schimper, W.P. (1876). *Synopsis muscorum europaeorum praemissa introductione de elementis bryologicis tractante*. Ed. II, Stuttgart.

Scott, G.A.M. & Stone, I. (1976). *The mosses of Southern Australia*. Academic Press, London.

Smith, A.J.E. (1978). *The Moss Flora of Britain and Ireland*. Cambridge University Press, London.

Steere, W.C. (1940). *Tortula* in North American North of Mexico. *Bryologist* **43,** 12–23.
Stone, I.G. (1988). *Acaulon granulosum*, a new species in the *Acaulon muticum* complex: a comparison and key to Australian species. *J. Bryol.* **15,** 257–268.
Stone, I.G. (1989). Revision of *Phascum* and *Acaulon* in Australia. *J. Bryol.* **15,** 745–777.
Wijk, R. van der, Margadan, W.D. & Florschütz, P.A. (1969). *Index Muscorum*. Vol. 5. Utrecht.

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APPENDIX (SELECTED SPECIMENS STUDIED)

Tortula muralis Hedw. var. *muralis*. PORTUGAL: Ribatejo, Torres Novas, *Casas et al.* (LISU s.n.); Estremadura, Setúbal, Serra da Arrabida, *Sérgio & Sim-Sim* (LISU s.n.); Beira Alta, Sortelha, *Melo* (LISU 148903); Algarve, Luolé, *Sérgio et al.* (LISU s.n.); Minho, Freixieiro do Soutelo, *Melo* (LISU s.n.); Baixo Alentejo, Segura, *Melo* (LISU s.n.); Ribatejo, San Antonio, *Casas et al.* (LISU s.n.). SPAIN: Albacete, Sierra de Alcaraz, Río de las Espineras, *Guerra & Ros* (Bryoth. MUB 2235); Asturias, Pravia, Agones, *Muñoz* (Herb. Muñoz 0339); Badajoz, Sierra de Alange, *Romero et al.* (Bryoth. MUB 937); Cantabria, Potes, *Muñoz* (Herb. Muñoz 340); León, Priaranza del Bierzo, *Muñoz* (Herb. Muñoz); Navarra, Puerto de Erro, *Fuertes* (Herb. Fuertes s.n.); Salamanca, Aldeadávila de la Ribera, *Muñoz* (Herb. Muñoz 1552); Sevilla, carretera Morón-Pruna, *Oliva* (Bryoth. MUB 3777).

Tortula muralis Hedw. var. *obcordata*. PORTUGAL: Beira Litoral, Anciao, Río Nabao, *Sérgio et al.* (LISU s.n.). SPAIN: Almería, Sierra de María, *Ochotorena* (Bryoth. MUB 901); Granada, Sierra Elvira, *Varo* (Herb. Fuertes s.n.); Murcia, Cartagena, *Ros* (Bryoth. MUB 3775); Murcia, Jumilla, Cerro de la Sal, *Ros* (Bryoth. MUB 3156); Murcia, Cartagena, Fuente de la Muela, *Ros* (Bryoth. MUB 3543); Murcia, Bullas, Salto Lucero, *Ros* (Bryoth. MUB 704); Murcia, Cehegín, Balsa del Pino, *Ros* (Broth. MUB 501); Murcia, Isla Grossa, *Ros & Aboal* (Bryoth. MUB 660).

Tortula baetica (Casas & Oliva) Guerra & Ros. SPAIN: Almería, Mojácar, *López et al.* (Herb. Fuertes s.n.); Cádiz, Jerez de la Frontera, *Oliva* (Herb. Oliva 1275); Cádiz, El Bosque, Las Lomas, *Oliva* (Herb. Oliva s.n.); Córdoba, Castillo de Almodóvar, *Oliva* (Herb. Oliva 1279); Córdoba, Entre Montilla y Montemayor, *Oliva* (Herb. Oliva 1268); Murcia, Cartagena, calle Alfonso X, *Guerra* (Bryoth. MUB 3779); Murcia, Cartagena, Cementerio San Antón, *Ros* (Bryoth. MUB 3778).