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Source: Willdenowia, 44(3) : 393-398

Published By: Botanic Garden and Botanical Museum Berlin (BGBM)

URL: <https://doi.org/10.3372/wi.44.44309>

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Abstract

Windisch P. G., Lorscheitter M. L. & Nervo M. H.: *Isoetes naipiana* (Isoetaceae), a new species from southern Brazil. – Willdenowia 44: 393–398. 2014. – Version of record first published online on 10 November 2014 ahead of inclusion in December 2014 issue; ISSN 1868-6397; © 2014 BGBM Berlin-Dahlem.

DOI: <http://dx.doi.org/10.3372/wi.44.44309>

Isoetes naipiana (Isoetaceae), a new species from southern Brazil, is described, illustrated and compared to similar species. So far it is known only from the original locality in the open grasslands of the upper highlands in the NE region of the State of Rio Grande do Sul (Brazil). It is characterized by megaspores with an irregular reticulum on the surface, presenting deposition of thin parallel silica strands; laevigate microspores; and arcuate microphylls with shortly alate bases, forming a compact rosette around the erect bilobate corm.

Additional key words: aquatic lycophytes, quillworts, floristics, conservation

Introduction

Worldwide the genus of *Isoetes* L. (Isoetaceae) is represented by c. 350 aquatic to terrestrial species, from temperate to tropical climates (Hickey & al. 2003). For South America, Fuchs-Eckert (1986) listed 75 species, while Hickey (1986) suggested 47. The diversity in Brazil is still far from being known in appropriate detail, due to poor collecting, difficulties in the preservation of samples, lack of fixed material for morphological and anatomical analysis, samples restricted to type material for many species, apparently narrow distributions, lack of easily observable characters, hybridization and polyploidy. Progress is being made due to a rising interest in the group. Prado & Sylvestre (2010) recorded 14 species (12 endemic) for Brazil, but that number was soon increased to 21 (Prado & al. 2012); most of these species occur in coastal moun-

tain ranges in the S and SE regions. The relationships of the species occurring in Brazil still need further research.

A study of the genus for southern Brazil in the “Flora Ilustrada Catarinense” by Fuchs-Eckert (1986) included six species for the states of Paraná, Santa Catarina and Rio Grande do Sul. Lorscheitter & al. (1998, 2009) presented a detailed palynological study including distributions, habitats and short descriptions of eight species from the state of Rio Grande do Sul, two listed as “species ineditae”. One of these was described as *Isoetes maxima* Hickey, Macluf & Link-Pérez (Hickey & al. 2009), and the second as *I. mourabaptistae* J. B. S. Pereira, P. G. Windisch, Lorscheitt. & Labiak (Pereira & al. 2012). Pereira (2012) presented a dissertation on *Isoetes* for southern and southeastern Brazil, with 15 species, including palynology (SEM and LM) and cytology (ploidy levels correlated with spore sizes).

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Extensive field work in the grassland formations in the upper highlands of the NE region of the state of Rio Grande do Sul revealed an interesting and distinctive species here described along with comments on the remaining species of the local flora.

Material and methods

Morpho-anatomical analyses — Material was collected and pressed in the traditional method for herbarium vouchers, and some samples were fixed in 70% ethanol for morphological and anatomical analyses. Structural terms follow Lellinger (2002).

Spore analyses — The Scanning Electron Microscopy (SEM) images were generated by transferring the spores to aluminium stubs coated with an asphalt adhesive. The stubs were then coated with gold palladium and photographed with a Zeiss EVO MA-10 electron microscope. A light microscope (DIAPLAN Leitz) was used for the measurement of acetolysed microspores. Spore size descriptions include the variation and the arithmetic average of 25 measurements, the first value corresponding to the polar axis and the second to the equatorial axis. The terminology used to classify the spore surface follows that of Hickey (1986) and Tryon & Lugardon (1990).

Results and Discussion

Isoetes naipiana P. G. Windisch, Lorscheitt. & Nervo, **sp. nov.** — Fig. 1–3.

Holotype: Brazil, state of Rio Grande do Sul, municipality of Jaquirana, Tainhas river drainage, 900–950 m, submersed aquatic in rock crevices, high-altitude grasslands, 14 Jan 2014, Windisch & Nervo 11063 (ICN; isotypes: B, ICN, RB, US).

Diagnosis — The general morphology of the plants differs markedly from the other species occurring in the region by the small size of the microphylls (up to 2.2 cm long) in mature plants, while at least twice as long in the remaining species. Furthermore, the microphylls are rigid in the living material, arched and disposed forming a flat rosette (2–3 cm in diam.), half-terete with a complanate adaxial surface (basal $\frac{2}{3}$), distally terete and tapering to a pointed apex (Fig. 1A). The microphylls present short (3–4 mm long) abruptly ending lateral alae at the base, not extending along the margins as in *Isoetes mourabaptistae* (up to the basal $\frac{1}{4}$). In *I. weberi* Herter, these alae may extend beyond the middle of the microphyll. In *I. fuscomarginata* H. P. Fuchs, *I. ramboi* Herter, *I. sehnemii* H. P. Fuchs, *I. smithii* H. P. Fuchs, *I. spannagelii* H. P. Fuchs (from a locality in the neighbouring state of Santa Catarina, Lages municipality) and *I. weberi* the alae usually taper off gradually.

The reticulate megaspores of *Isoetes naipiana* have an equatorial diameter greater than 600 μm , which is similar to *I. fuscomarginata*, *I. mourabaptistae*, *I. ramboi* and *I. smithii* in size. It differs from those species by having thin, dense, transversely oriented and parallel silica strands forming the muri of the reticulum, this parallel disposition being more pronounced in the deeper lumina. The muri of the megaspores are a good distinctive character as they frequently cross over the trilete arms. The reticulate species occurring in Rio Grande do Sul have smaller megaspores (*I. brasiliensis* H. P. Fuchs, *I. sehnemii*), or are verrucate (*I. weberi* and *I. maxima*).

Four species of *Isoetes* from Rio Grande do Sul have megaspores with equatorial diameters greater than 600 μm , but only *I. naipiana* has smooth to slightly undulate microspores (SEM, especially the proximal face). *Isoetes mourabaptistae* has microechinate microspores. The microspores (SEM) are laevigate-granulate in *I. fuscomarginata* and *I. smithii*, and micropapillate-granulate in *I. ramboi*. The structure of the parallel silica strands forming the muri of the megaspores is quite distinctive in *I. naipiana*.

Description — *Plants* aquatic, submerged, growing in rock crevices in current. *Corm* erect, bilobed, up to 10 mm long, 3–4 mm in diam. *Roots* conspicuous, dichotomous. *Microphylls* 20–35, 1–2.2 cm long, 0.4–0.6 mm wide at middle, 0.7–1 mm wide at base above sporangium (fixed material); 0.8–1.8 cm long, 0.2–0.4 mm wide at middle, 0.4–0.8 mm wide at base above sporangium, 1.2–1.4 mm wide at sporangium (dried material), rigid in living material, arched and disposed forming a flat rosette (2–3 cm in diam.), half-terete with a complanate adaxial surface (basal $\frac{2}{3}$), distally terete and tapering to a pointed apex. *Alae* only at base, wings membranous, hyaline, 3–4 mm long, clasping at widened base (0.6–0.9 mm wide on each side), reduced to 0.4–0.6 mm (middle), ending abruptly shortly beyond sporangium. *Ligule* deltate, c. 0.7 mm long, up to 0.6 mm wide, frequently eroded, base weakly auriculate on either side. *Velum* cover c. $\frac{1}{2}$ or more with an oval opening at base. *Labium* not clearly visualized, a short projection observed in some cases. *Sporangium* roundish-elliptic, c. 1.8 mm long, c. 1.2 mm broad, covered by velum. *Microspores* (18–)20(–26) $\times$ (24–)27(–34) μm (including perispore/paraexospore), (15–)16(–18) $\times$ (21–)23(–27) μm (excluding perispore/paraexospore), reniform, monolete, laesura extending more than $\frac{3}{4}$ of microspore length; paraexospore detached from exospore, laevigate perispore (in SEM sometimes with irregular, slightly undulating surface, especially on proximal face). *Megasporangia* with 4 or 8 megaspores. *Megaspores* (500–)550(–590) $\times$ (620–)655(–690) μm , spheroidal, equatorial limb subtriangular-convex, trilete, arms equalling radius, equatorial view with largely convex proximal and distal faces and a distinct equatorial flange, curved between arms; reticulate with large irregular reticulum distributed on entire surface.

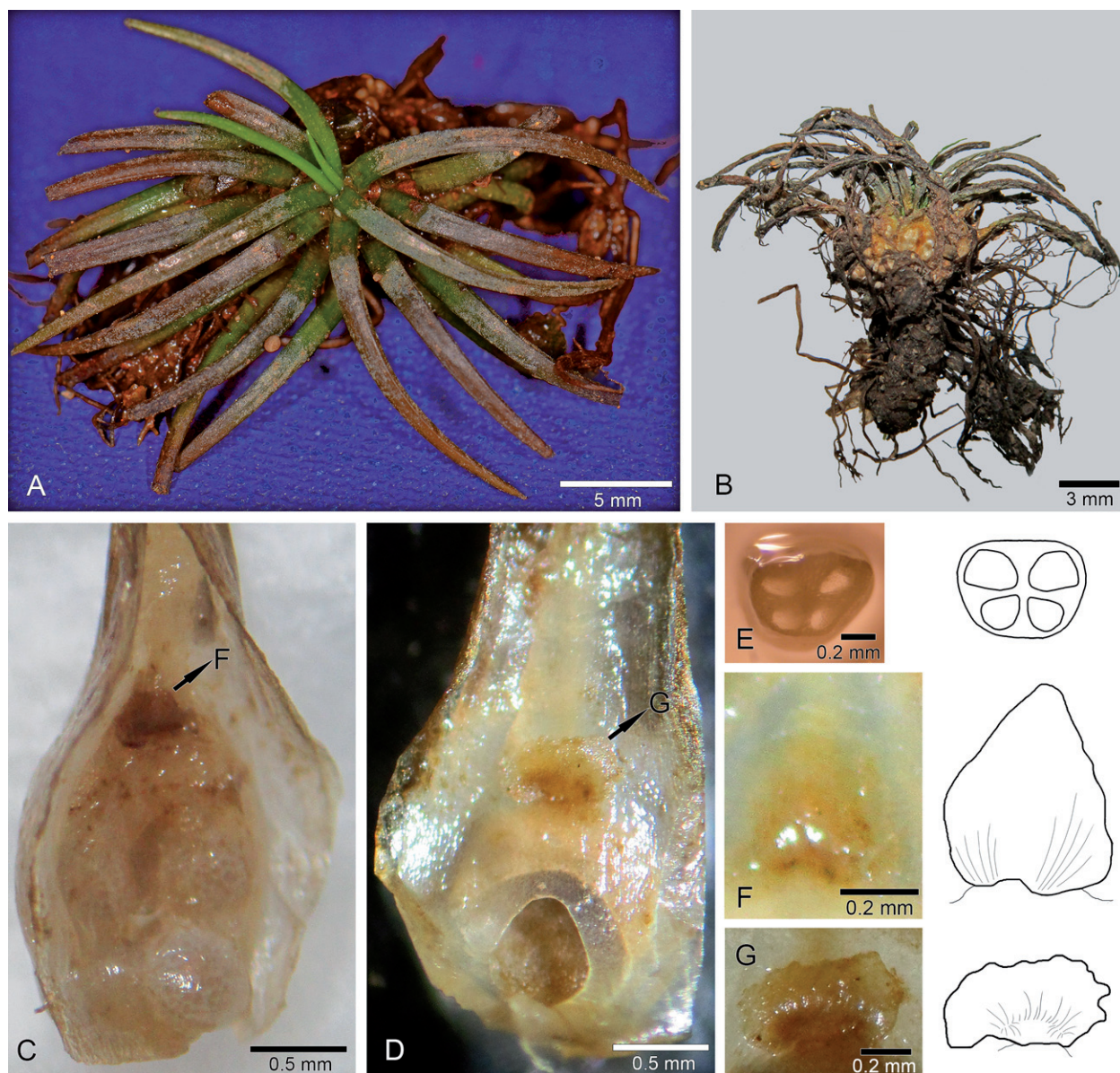


Fig. 1. *Isoetes naipiana* – A & B: habit; C: adaxial view of base of megasporophyll with megaspores; D: adaxial view of base of microphylls with velum covering more than $\frac{1}{2}$ of sporangial surface; E: microsporophyll in transverse section, with accompanying illustration; F: deltate ligule, with accompanying illustration; G: deltate eroded ligule, base weakly auriculate, with accompanying illustration. – All from Windisch & Nervo 11063.

Distribution and ecology — *Isoetes naipiana* is currently known only from the original locality, in high-altitude grassland formations, at c. 950 m, where it grows in the rapid moving stream current, at 20 to 30 cm depth (dry season) in basaltic rock crevices.

Conservation status — The inclusion of *Isoetes naipiana* in the threatened species listings as Vulnerable VU D1+2 (IUCN 2012) is proposed here based on small population size with restricted occupancy, with two known locations in an area prone to the effects of human occupancy. The environmental changes due to the current use of the high-altitude grasslands for extensive forestry and agricultural activities require urgent action.

Etymology — The original inhabitants of the NE highlands of Rio Grande do Sul were the Kaingang, a widespread ethnic group of southern to southeastern Brazil. In their mythology, Naipi, the extremely beautiful daughter of a chief, was to be consecrated to the divinity M'boi (son of the highest divinity Tupã) who reigned in the depths of rivers in the form of a giant serpent. Just before her consecration, she escaped in a canoe with her beloved one. M'boi became furious, the canoe was destroyed in a waterfall and Naipi transformed into a rock, to be eternally battered by the running water, like this species of *Isoetes*.

Additional remarks — The size of the megaspores of *Isoetes naipiana* is quite remarkable for such diminutive

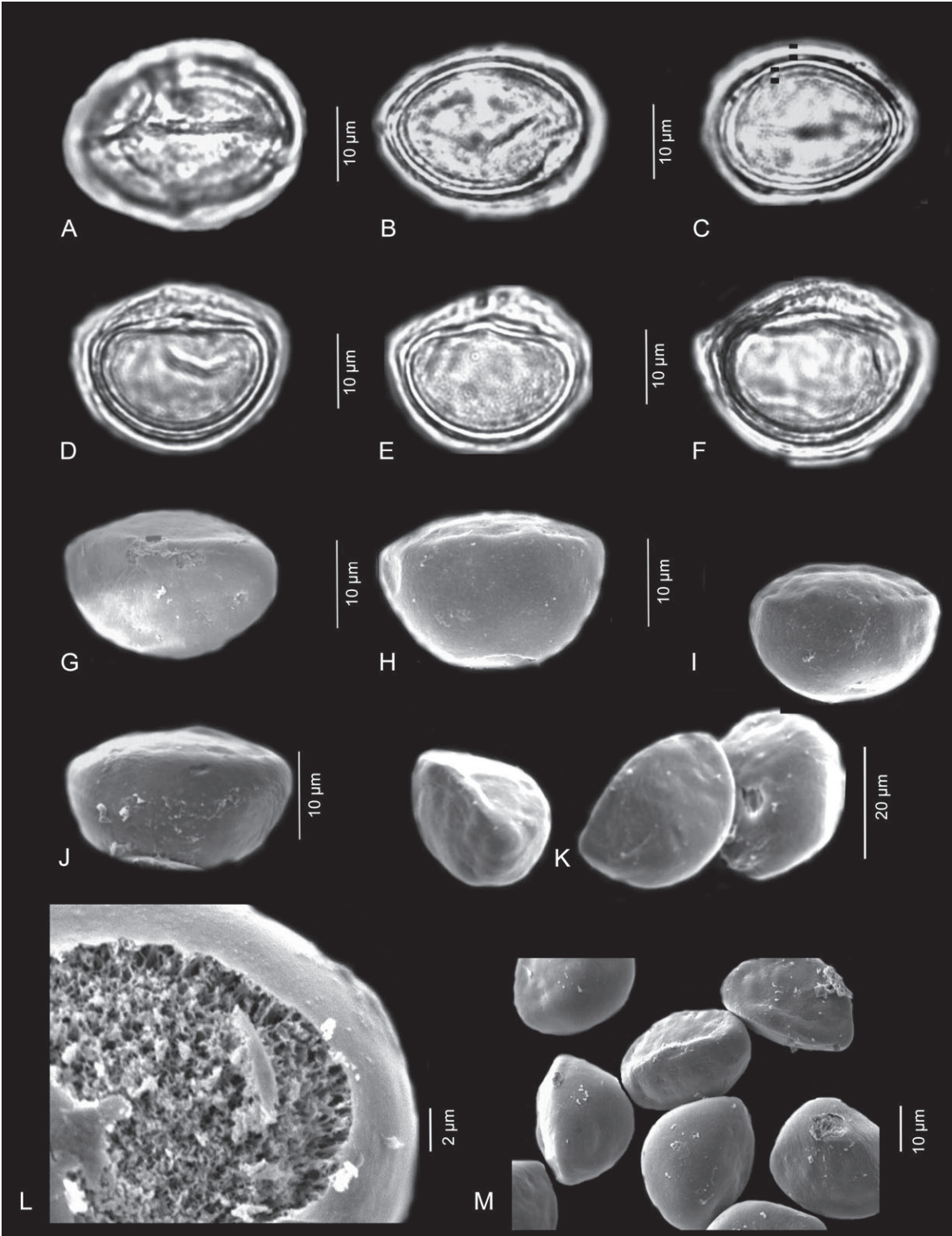


Fig. 2. *Isoetes naipiana*, microspores – A–F: LM images of paraexospore detached from exospore; A: proximal face; B & C: distal face; D–F: equatorial view; G–M: SEM images of laevigate perispore surface; G: proximal face; H–J: equatorial view; K: grains in distinct positions; L: detail of eroded surface, irregular fused strands of inner perispore/paraexospore; M: general view of microspores. – All from Windisch & Nervo 11063.

plants; the total length of the microphylls corresponds to 35–40 times the diameter of the megaspores. The megaspores are quite characteristic, showing an irregular

reticulum, with fine, parallel fused strands, in a transverse orientation, forming the high muri with alveolar content and denser strands in the bottoms of the areolae. Gener-

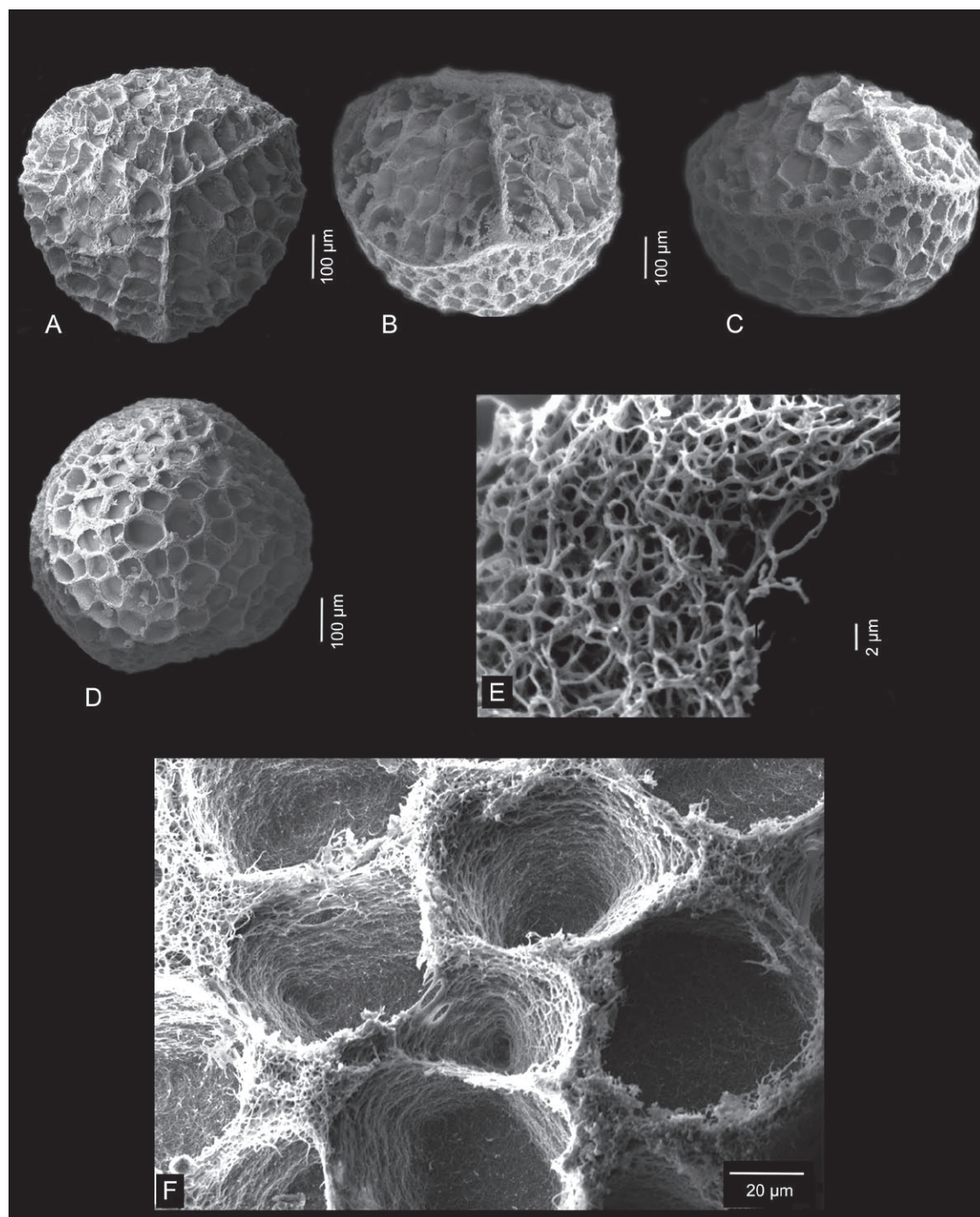


Fig 3. *Isoetes naipiana*, SEM images of megaspores – A: proximal face, longer areolae close to arms, muri crossing over trilete arms; B & C: oblique and equatorial views showing distinct equatorial flange; D: distal face; E: magnified detail of reticulum surface with alveolar strands; F: magnified view of reticulum showing fine fused strands deposited in transverse and parallel fashion forming high muri and denser disposition of strands in bottoms of areolae. – All from Windisch & Nervo 11063.

ally longer areolae can be observed close to the arms, the muri frequently crossing over the trilete arms.

The structure of the ligule and labium (vestigial or as a short projection) was variable, as observed by Budke & al. (2005) in a detailed study of *Isoetes tennesseensis* Luebke & Budke.

Acknowledgements

The authors acknowledge support of the Universidade Federal do Rio Grande do Sul, Programa de Pós-Graduação em Botânica, as well as the granting agencies CNPq (Brazilian National Research Council), CAPES

(Coordenação de Aperfeiçoamento do Pessoal do Ensino Superior – Ministry of Education) and FAPERGS (State of Rio Grande do Sul Research Foundation). Prof. Werner Greuter and Prof. Brigitte Zimmer provided important suggestions, as did two anonymous reviewers on an earlier version of this paper.

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