
Isoetes orientalis (Isoetaceae), a New Hexaploid Quillwort from China

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ABSTRACT. *Isoetes orientalis* is a previously undescribed, rare, and endangered quillwort of southeast China with a hexaploid chromosome count of $2n = 66$. Megaspores of *I. orientalis* have a reticulate ornamentation consisting of irregularly anastomosing muri with ragged crests. Microspores have an echinate-tuberculate ornamentation. Herbarium specimens of *I. orientalis* previously have been misidentified as *I. sinensis*. *Isoetes orientalis* differs from *I. sinensis* in spore morphology and chromosome number.

Key words: China, East Asia, Isoetaceae, *Isoetes*, quillwort.

Three basic diploid species and one tetraploid species of *Isoetes* have been reported from China (Liu et al., 2002; Wang et al., 2002). These are *I. hypsophila* Handel-Mazzetti ($2n = 22$), *I. taiwanensis* DeVol ($2n = 22$), *I. yunguiensis* Wang Q. F. & W. C. Taylor ($2n = 22$), and *I. sinensis* Palmer ($2n = 44$). Over the last five years, field, herbarium, and laboratory studies have been conducted in the assessment of *Isoetes* populations over a large part of mainland China. As part of these studies, examination of herbarium specimens of *Isoetes* collected from Songyang County in southeast China revealed specimens that had been previously identified as *I. sinensis*. Closer inspection of these specimens showed that they differed from *I. sinensis* in spore ornamentation. Further analysis of these herbarium specimens and living specimens from Songyang County indicate that these collections are distinct from *I. sinensis* in spore morphology and chromosome number. They represent a unique, previously undescribed species of *Isoetes*, which is named and described as follows:

Isoetes orientalis H. Liu & Q. F. Wang, sp. nov.
TYPE: China, Zhejiang: Songyang County, ca. 0.15 km S of Anming Town, 12 Dec. 2002, Liu H. & Wang J. Y. WH20021214 (holotype, WH). Figures 1, 2.

Planta aquatica ex caudice trilobo emergens. Folia 20 ad 40, viridia, usque ad 10–20 cm longa, ca. 2.0 mm lata ad medium longitudinis, basi dilatata et complanata; fibris fibrolignosis praesentibus; ligula subtriangulata, 1.5–2.0 mm longa, 2.0–3.0 mm lata. Velum rudimentarium. Sporangium basale, 5–6 mm longum; megasporae superficiei reticulata ex porcis irregulariter anastomosantibus cristis laceratis coronatis constante, 350–450 μm diametro; microsporis pallide canis in massam dispositis, ellipticis, 19–29 μm longis, echinatis. Chromosomatum numerus $2n = 66$ (Fig. 1B).

Plant aquatic, emergent. Root stock 3-lobed. Leaves white basally, green above, spirally arranged, wide spreading, 10–20 cm long, ca. 2.0 mm wide at mid-length, in tufts of 20 to 40, flattened on adaxial side, rounded on abaxial side, base alate (Fig. 1A); peripheral fiber strands present; central intrastelar canal one; ligule ovate-subtriangular, 1.5–2.0 \times 2.0–3.0 mm. Sporangium basal, obovate, ca. 5–6 \times 3.8–4.5 mm; velum rudimentary, covering only distal edge of sporangium. Megaspores white when dry, gray when wet, ca. 350–450 μm diam. (mean = 420 μm), with reticulate texture composed of irregularly anastomosing muri with ragged crests, girdle obscure (Fig. 2A–D). Microspores gray in mass, elliptic, ca. 19–29 μm long (mean = 22 μm), tuberculate-echinate (Fig. 2F–I). Chromosomes: $2n = 66$.

Distribution. In China, Songyang County of Zhejiang Province.

Ecology. The marshes containing *Isoetes orientalis* are in a unique, swampy, loamy meadow between two mountains. The water in these marshes has a pH of 5.8. The reddish soil in which the plants are rooted has a pH of 5.4. Common associates include *Eriocaulon alpestre* Hooker f. & Thomson, *Juncus effusus* L., *Cyperus haspan* L., *Murdannia trigueta* (Wallich) Brückner, and *Oenanthе javanica* (Blume) DC.

Isoetes orientalis and *I. sinensis* both occur in Zhejiang Province, but they are separated by ele-

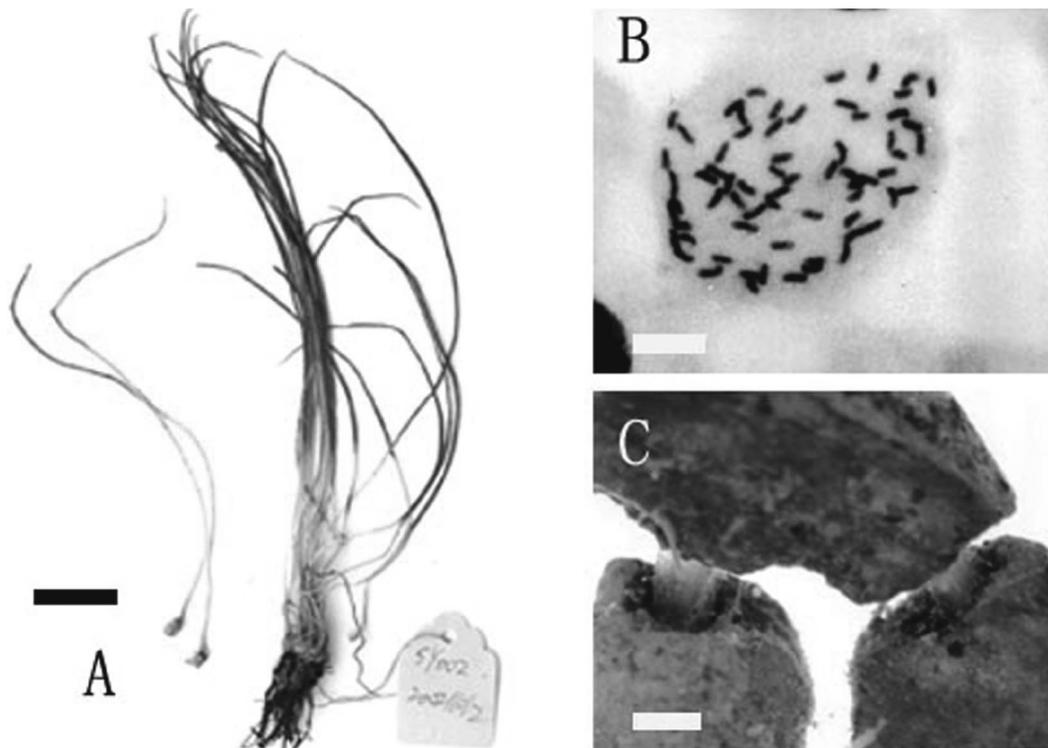


Figure 1. *Isoetes orientalis* H. Liu & Q. F. Wang. —A. Holotype: Liu H. & Wang J. Y. WH20021214 (WH). —B. Somatic chromosomes in mitotic root tip squash. —C. Roots with reddish rhizosphere. Scale bar: A = 2 cm; B = 10 μ m; C = 200 μ m. Figures B and C from isotype: Liu H. & Wang J. Y. WH20020215 (WH).

vation. The *I. orientalis* populations occur at 1200 m, whereas *I. sinensis* grows between 130 and 300 m.

Leaf features. In cross section, the leaves of *Isoetes orientalis* are oval and almost completely occupied by four, thin-walled, oblong air chambers with the two adaxial air chambers being slightly larger than the abaxial ones. In contrast, the air chambers in the leaves of *I. sinensis* appear to be smaller, more circular, and have thicker walls. Furthermore, *Isoetes orientalis* has one intrastelar canal, whereas *I. sinensis* has three or more intrastelar canals (Takamiya et al., 1997).

Root features. A noteworthy feature of *I. orientalis* is the reddish, sheathing mantle formed around its dichotomously branched roots (Fig. 1C). The reddish color of this mantle or rhizosphere is reportedly due to oxidation of iron in the soil effected by oxygen released from the roots (Tessenow & Baynes, 1975, 1978). Certain kinds of bacteria and fungi grow in the rhizosphere of *Isoetes* (Sharma, 1998). This feature has not been observed on the roots of *I. sinensis*, and it has not been previously reported for any species of *Isoetes* in Asia.

Spore size and texture. Spore measurements

and photomicrographs were made using a Hitachi S-800 scanning electron microscope at 15 Kv. Megaspores of *I. orientalis* are ca. 350–450 μ m diam. (mean = 420 μ m diam., N = 25), whereas, according to Huang et al. (1992), megaspores of *I. sinensis* are ca. 360–390 μ m diam. Microspores of *I. orientalis* are 19–29 μ m long (mean = 22 μ m long, N = 40), whereas, according to Huang et al. (1992), the microspores of *I. sinensis* are 19–20 μ m long and average 20 μ m long.

Megaspores of *I. orientalis* have an irregular cristate-reticulate ornamentation (Fig. 2A–D), whereas those of *I. sinensis* are cristate with fewer anastomosing ridges (Watanabe et al., 1996: 290, figs. 39–43). Megaspore ornamentation of *I. orientalis* resembles that of the octaploid *I. pseudojaponica* M. Takamiya, Mitsu. Watanabe & K. Ono (Watanabe et al., 1996: 288, figs. 15–18), but they differ with respect to microornamentation. At higher magnification, the fibrils on the megaspores of *I. orientalis* (Fig. 2E) are longer and larger than the fibrils on the megaspores of *I. pseudojaponica* (Watanabe et al., 1996: 288, fig. 19). Megaspore texture of *I. orientalis* is also similar to that of *I. japonica* A. Braun, but *Isoetes orientalis* differs from *I. japonica*

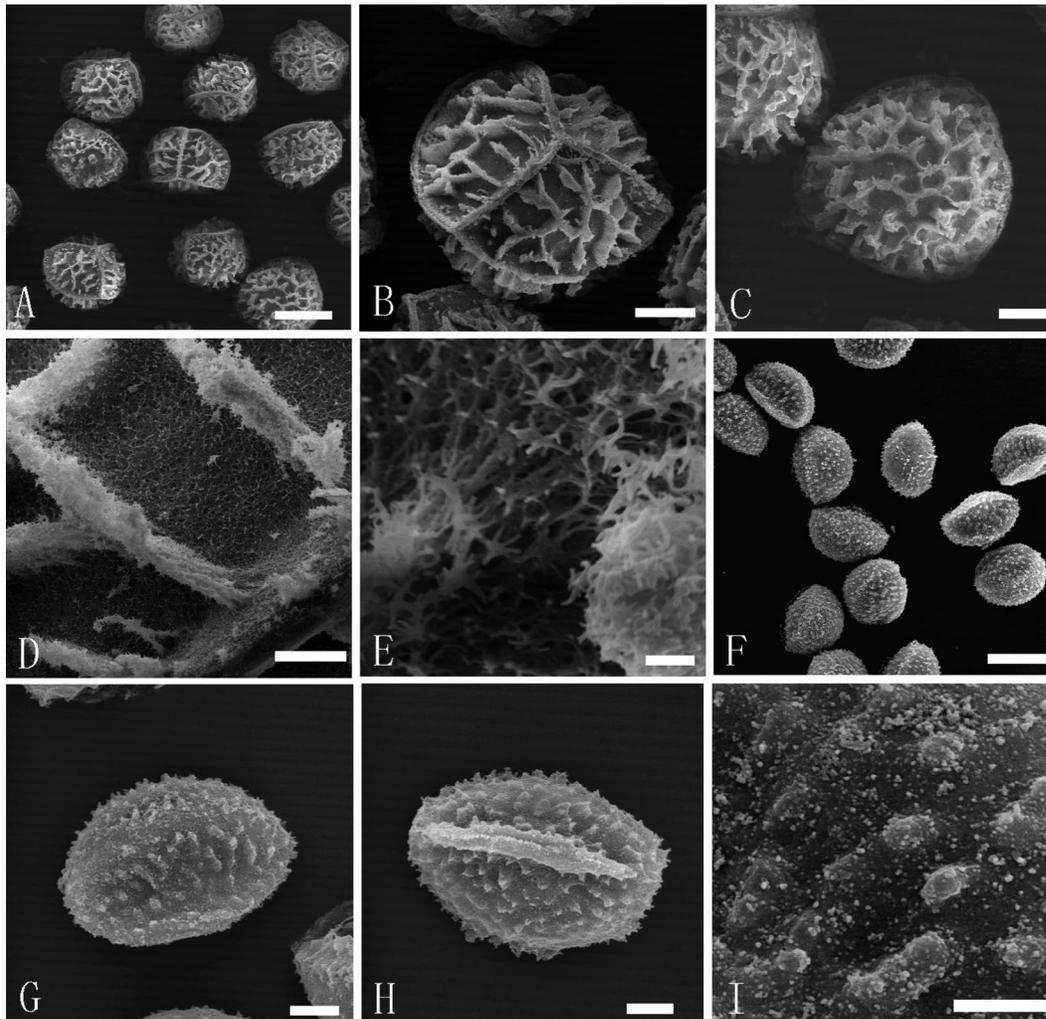


Figure 2. *Isoetes orientalis*. —A. Megaspores. —B. Proximal view of megaspore. —C. Distal view of megaspore. —D, E. Megaspore surface microornamentation. —F. Microspores. —G. Distal view of microspore. —H. Proximal view of microspore. —I. Microspore surface microornamentation. Scale bar: A = 200 μm ; B & C = 75 μm ; D & F = 15 μm ; E & I = 2 μm ; G & H = 4 μm . All from isotype: Liu H. & Wang J. Y. WH20020215 (WH).

in microspore texture. Microspores of *I. orientalis* have an echinate-tuberculate ornamentation (Fig. 2F–H), whereas *I. japonica* microspores are nearly laevigate (Watanabe et al., 1996: 284, figs. 8–13). At higher magnification, the short spines of *I. orientalis* look more tuberculate and a granulate microornamentation is apparent (Fig. 2I). In contrast, the microspores of *I. sinensis* are clearly echinate (Huang et al., 1992: 147, fig. 3A–C; Watanabe et al., 1996: 290, figs. 46–50).

Although *Isoetes orientalis* and *I. sinensis* differ in spore ornamentation, the plants are similar in general appearance and growth habit. Both species are much alike in leaf, sporangium, ligule, and ve-

lum characters, as well. Early descriptions of *Isoetes* species were based on general appearance and habit, and this might be the reason *I. orientalis* was originally identified as *I. sinensis*.

Cytology. Chromosome counts were made from root tips of plants collected at the type locality and prepared following the methods of Liu et al. (2002). Counts from a total of 80 cells found in root tip squashes of 12 individuals showed that *I. orientalis* is a hexaploid with a chromosome number of $2n = 66$ (Fig. 1B). The base number for *Isoetes* is $x = 11$ (Löve et al., 1977).

Takamiya et al. (1994) recognized $2n = 44, 65, 66,$ and 68 cytotypes for *I. sinensis*. Takamiya et al.

(1997) revised this earlier taxonomy recognizing the tetraploid cytotype as *I. sinensis* var. *sinensis* and the $2n = 65, 66,$ and 68 cytotypes as *I. sinensis* var. *coreana* (Y. H. Chung & H. K. Choi) M. Takamiya, Mitsu. Wantanabe & K. Ono. According to Takamiya et al. (1997), both of these varieties have cristate megaspores and echinate microspores. Their megaspore texture is distinct from *I. orientalis*, which has reticulate megaspores and echinate-tuberculate microspores.

Phenology. Megaspores of *Isoetes orientalis* mature from mid May to early September. Microspores develop from late June to mid October. By December, most sporophylls have decayed and only a few depauperate, green leaves remain, but like other emergent or amphibious quillworts in China, *I. orientalis* stays green, at least to some extent, through the winter.

Like all other quillworts in China, *I. orientalis* is a rare and endangered species. To date, it has been found at only two sites, one of which has been decimated. The last known extant *I. orientalis* population covers no more than 200 square meters and contains fewer than 400 individuals. Pollution and increasing urbanization continue to raise the risk of extinction for this species. Protection of the one remaining population of *I. orientalis* is crucial, and searches for additional populations are urgently needed.

Paratype. CHINA. **Zhejiang:** Songyang County, Anmin Town, Andaihou, *Lou L. H.* 227 (SY, ZJFC).

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Literature Cited

- Huang, T. C., H. J. Chen & L. C. Li. 1992. A palynological study of *Isoetes taiwanensis* DeVol. *Amer. Fern J.* 82: 142–150.
- Liu Xing, Y. Wang, Q. F. Wang & Y. H. Guo. 2002. Chromosome numbers of the Chinese *Isoetes* and their taxonomical significance. *Acta Phytotax. Sin.* 40(4): 351–356. [In Chinese with English abstract.]
- Löve, A., D. Löve & R. E. G. Pichi Sermolli. 1977. *Cytotaxonomical Atlas of the Pteridophyta*. J. Cramer, Vaduz.
- Sharma, B. D. 1998. Fungal associations in the root of three species of *Isoetes* L. *Aquatic Bot.* 61: 33–37.
- Takamiya, M., M. Watanabe & K. Ono. 1994. Biosystematic studies on the genus *Isoetes* (Isoetaceae) in Japan. I. Variations of the somatic chromosome number. *J. Pl. Res.* 107: 289–297.
- , ——— & ———. 1997. Biosystematic studies on the genus *Isoetes* (Isoetaceae) in Japan. IV. Morphology and anatomy of sporophytes, phytogeography and taxonomy. *Acta Phytotax. Geobot.* 48: 89–122.
- Tessenow, U. & Y. Baynes. 1975. Redox-dependent accumulation of Fe and Mn in a littoral sediment supporting *Isoetes lacustris* L. *Naturwissenschaften* 62: 342–343.
- & ———. 1978. Experimental effects of *Isoetes lacustris* L. on the distribution of Eh, pH, Fe and Mn in lake sediment. *Verh. Int. Vereinigung Theor. Limnol.* 20: 2358–2362.
- Wang, Q. F., Liu Xing, W. C. Taylor & He Zhao-Rong. 2002. *Isoetes yunguiensis* (Isoetaceae), a new basic diploid quillwort from China. *Novon* 12: 587–591.
- Watanabe, M., M. Takamiya, T. Matsusaka & K. Ono. 1996. Biosystematic studies on the genus *Isoetes* (Isoetaceae) in Japan. III. Variability within qualitative and quantitative morphology of spores. *J. Pl. Res.* 109: 281–296.