A New Subspecies of *Isoetes coromandelina* from Northern Australia

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Abstract

Marsden, C. R. A new subspecies of *Isoetes coromandelina* from northern Australia. *Contrib. Herb. Aust.* 24: 1–10, 1976. *Isoetes coromandelina* L. f. is recorded for the first time for Australia, where it is known only from the northern part. The Australian populations of this species differ from the Indian ones in their megaspore morphology and they are here segregated as a new subspecies, ssp. macrotuberculata. Various vegetative features of *I. coromandelina* are discussed.

INTRODUCTION

Isoetes coromandelina L. f. was the second species referred to Isoetes (Linnaeus 1781). This species has been well documented, with detailed investigations of its anatomy (Bhambie 1957, 1962, 1963a, 1963b, 1971) and cytology (Verma 1960, 1961; Pant and Srivastava 1965) having been published. However, no comprehensive account of the morphology of *I. coromandelina* has been published since Pfeiffer (1922), except for megaspore morphology, which has been one of the main characters used in delimiting this and other Indian species of *Isoetes* (Pant and Srivastava 1962; Goswami and Arya 1970).

I. coromandelina has previously been considered endemic to the Indian subcontinent where it is widespread (Pant and Srivastava 1962). Recently, however, several collections apparently closely related to *I. coromandelina* were noted during examination of northern Australian specimens of *Isoetes*. Subsequent comparison between Australian and Indian plants revealed differences in the ornamentation of the megaspores, and the former are here described as a new subspecies.

MATERIALS AND METHODS

Dried, spirit-preserved and fresh plant material was examined for morphological features and hand sections of fresh leaves were used for anatomical investigation.

Spores were examined by light and scanning electron microscopy. To reduce coating and charging difficulties, megaspores for electron microscopy were exposed to the vapours of a 2% solution of osmic acid for several hours prior to mounting, as suggested by Pfefferkorn (1970). Specimens were then coated with gold, or gold-palladium alloy, and examined and photographed using an ETEC Autoscan scanning electron microscope.

DIAGNOSIS

The original description of I. coromandelina L. f. was very brief, and additional details were published by Pfeiffer (1922) in her monograph of Isoetaceae. However, as further information is now available, a more detailed description is included here.

Isoetes coromandelina L. f., Suppl. Plant.: 447 (1781).

Corm 3(-4-5) lobed. Leaves 15-60, up to 60(-80) cm long, bright green, erect, flexible, with four strongly developed peripheral fibrous strands and several small strands between (Fig. 18). Stomata present on apical portion of leaves (Fig. 22). Internal hairs present, projecting into the four lacunae. Labium conspicuous, hemiorbicular, covering all but the apex of the lanceolate ligule (Figs 23, 24). Ligule often lost as leaf develops. Outer sporangia orbicular (Fig. 23), up to 7 mm in diameter; inner sporangia obovate (Fig. 24), up to 12 mm long. Mature sporangial wall pale buff in colour, cell walls not thickened (Fig. 21). Velum absent. Megaspores white when dry, grey when wet, tuberculate, dimorphic in each sporangium with two main size classes plus a few joined or double megaspores, or both, also present. Smaller megaspores flattened, larger megaspores almost spherical. Microspores rare, reddish or buff in colour, smooth, rugose to papillate or echinate.

Ssp. coromandelina

Megaspores 350-460 and 470-660 μ m, with short blunt tubercles. Larger megaspores with few to numerous tubercles on each proximal face (Figs 2, 4) and numerous tubercles on distal faces (Fig. 6).

Holotype: Linn 1256.2, König (annotated by L. f.). Photograph seen.

Additional specimens examined

Dabra, near Gwalior, H. K. Goswami Dab/Gos 1973, Dab/Gos 1974; Kalvari, Mizapur district, M. B. Ralzada (DD); Khandala, C. McCann (BLAT); Khandala, H. Santapau (DD, BLAT); Meerut, G. D. Tyogi (DD); Puri Coast, Y. A. Rao 5923 (CAL); Shivpuri (DD).

Distribution

This subspecies is confined to the Indian subcontinent, where it is widespread (Pant and Srivastava 1962).

Ssp. macrotuberculata Marsden, ssp. nov.

Megasporae 330-410 et 420-530 μ m, tuberculis globulis. Megasporae majores uno vel aliquot tuberculis magis et aliquot tuberculis parvioribus per superficiem proximalem, tuberculis numerosis in superficie distali.

Type: Northern Territory, Mt Bundey Station 13°03'S., 121°17'E., 26.iv.1974, *C. Dunlop 3193.* (Holotype: AD 97522176; isotypes: AD, BM, BRI, CANB, DNA, NT.)

Megaspores 330-410 and 420-530 μ m, with globular tubercles. Larger megaspores with one to a few large and several smaller tubercles on each proximal face, numerous tubercles on distal faces (Figs 3, 5, 7).

The subspecific epithet *macrotuberculata* refers to the large tubercles on the megaspores which distinguish this subspecies from ssp. coromandelina.

Additional specimens examined

NORTHERN TERRITORY: Arnhem Highway, C. Dunlop 3474 (DNA); Berrimah, Darwin, C. Dunlop 3593 (DNA, NT); Jabiru, C. Dunlop 3688 (DNA); c. 3.5 km N. of Katherine, L. G. Adams 1705 (MEL); Survey Creek, N. Byrnes 658 (BRI, NT); Survey Creek, N. Byrnes 1812 (AD, MEL); Survey Creek, N. Byrnes 2072 (AD, NT); South Brolga, A. O. Nicholls (NT). QUEENSLAND: Cooktown, S. T. Blake 21834 (BRI); Iron Range, Cape York Peninsula, L. J. Brass 19218 (BRI, TNS). WESTERN AUSTRALIA: Kimberleys, Galvin's Gorge, A. C. Beauglehole 47901A.

Distribution

This subspecies is known only from northern Australia (Fig. 1), but it will probably be found to occur more widely than now recorded. At present, *Isoetes* is poorly known throughout Australia, possibly in part because of difficulty in field recognition of the genus.



Fig. 1. Distribution map of Isoetes coromandelina ssp. macrotuberculata.

DISCUSSION

Anatomy and morphology

Megaspores. The ornamentation of the megaspores is the main feature used to distinguish the two subspecies of *I. coromandelina* and has been investigated in detail using scanning electron microscopy (Figs 2-16). The larger megaspores of ssp. macrotuberculata (Figs 2-7) differ from those of ssp. coromandelina in having dimorphic tubercles on the proximal faces, markedly larger tubercles on the distal faces and commissural ridges which are thicker and more irregular. The smaller megaspores of ssp. macrotuberculata also have larger tubercles and thicker, more irregular, ridges than those of ssp. coromandelina (Table 1). However, the ultrastructure of the megaspores was found to be similar in both subspecies (Figs 8-11). The apex of each tubercle is covered by a close reticulum (Figs 10, 11), which is densely packed so as to appear closed on some of the tubercles of megaspores of ssp. coromandelina (Fig. 10). Between the tubercles, the surface pattern is an open, cross-linked network of threads overlaying a pattern similar to that found on the tubercle apices (Figs 8, 9). The megaspores of ssp. macrotuberculata are slightly smaller (330-410 and 420-530 μ m) than those of ssp. coromandelina (350-460 and 470-660 μ m), although there is considerable overlap in the size ranges.



	Ssp. macrotuberculata	Ssp. coromandelina
	Larger megaspores	
Spore diameter (µm)	420-530	470-660
Ornamentation		
Proximal face	Tubercles dimorphic: large globular tubercles $1-3$ (rarely 4), each 50-80 μ m diameter, or if only one present, 100-150 μ m diameter; smaller, low tubercles up to 18, each 20- 45 μ m diameter	Tubercles all similar: low tubercles 10–15, each 30–70 μm diameter
Distal face	Numerous globular tubercles, most 70–140 µm diameter	Numerous low tubercles, most $40-90 \ \mu m$ diameter
Ridges	Triradiate and commissural ridges irregularly corrugate	Triradiate and commissural ridges almost smooth
	Smaller megaspores	
Spore diameter (µm)	330-410	350-460
Ornamentation		
Proximal face	1 (rarely 2 or 3) globular tubercle, often almost spherical, 55-80 μm diameter (Fig. 13)	Up to 5 (commonly 3 or 4) low tubercles 40-65 μ m diameter (Fig. 12)
Distal face	Numerous globular tubercles $50-100 \ \mu m$ diameter crowded in the centre of the face; tubercles mostly higher than broad (Fig. 15)	Numerous shallow tubercles $35-75$ μ m diameter crowded in the centre of the face; tubercles broader than high (Fig. 14)
Ridges	Commissural and triradiate ridges irregularly corrugate (Figs 13, 15)	Commissural and triradiate ridges almost smooth (Figs 12, 14)

Table 1. Comparison of the megaspores of the two subspecies of Isoetes coromandelina

Microspores. No microspores of ssp. *macrotuberculata* have been observed, but they have occasionally been found in ssp. *coromandelina*. Microspores of this latter subspecies have previously been recorded as smooth (Pfeiffer 1922) or rugose to papillate (Knox 1950). Those examined in the scanning electron microscope during this study (Khandala, C. McCann, BLAT) were found to be covered with somewhat rough, conical spines (Fig. 17) and were pale in colour although Pfeiffer (1922) recorded the microspores of *I. coromandelina* as being sometimes reddish brown.

Figs 2-7. Scanning electron micrographs of large megaspores of I. coromandelina. Scale = $200 \ \mu m$.

Fig. 2. Ssp. coromandelina, side view. (Dabra, Goswami Dab/Gos 1973.) Fig. 3. Ssp. macrotuberculata, side view. (Dunlop 3474.) Fig. 4. Ssp. coromandelina, proximal faces. (Dabra, Goswami Dab/Gos 1973.) Fig. 5. Ssp. macrotuberculata, side view. (Holotype.) Fig. 6. Ssp. coromandelina, distal face. (Kalvari, DD 99422.) Fig. 7. Ssp. macrotuberculata, distal face. (Isotype, AD.)

Ligule and labium. Pfeiffer (1922) recorded the ligule in *I. coromandelina* as 'conspicuous, very wide and short, often appearing truncate in older leaves but pointed in young'. In this description, it is likely that Pfeiffer referred to the labium, which covers most of the ligule (Figs 23, 24), rather than to the ligule itself. The ligule in both subspecies is generally lanceolate and much narrower than the labium. The ligule is often lost or damaged on mature leaves, although this tendency is less apparent in ssp. coromandelina.

Internal hairs of the leaves. Internal hairs as described by Hall (1971) were observed on the walls of each of the four lacunae in both subspecies of *I. coroman*delina (Fig. 18). These hyaline cells (Fig. 20) arise directly from the chlorenchymatous



Figs 8-11. Scanning electron micrographs of ultrastructure of distal face of large megaspores of *I. coromandelina.*

Fig. 8. Ssp. coromandelina, surface structure between tubercles. Scale = $20 \ \mu m$. (Dabra, Goswami Dab/Gos 1973.) Fig. 9. Ssp. macrotuberculata, surface structure between tubercles. Scale = $20 \ \mu m$. (Isotype, AD.) Fig. 10. Ssp. coromandelina, apex of tubercle. Scale = $10 \ \mu m$. (Dabra, Goswami Dab/Gos 1973.) Fig. 11. Ssp. macrotuberculata, apex of tubercle. Scale = $10 \ \mu m$. (Isotype, AD.)



Figs 12-15. Scanning electron micrographs of smaller megaspores of *I. coromandelina*. Scale = $150 \ \mu m$.

Fig. 12. Ssp. coromandelina, proximal face. (Dabra, Goswami Dab/Gos 1973.) Fig. 13. Ssp. macrotuberculata, proximal face. (Holotype.) Fig. 14. Ssp. coromandelina, distal face. (Kalvari, DD99422.) Fig. 15. Ssp. macrotuberculata, distal face. (Isotype, AD.)

Fig. 16. Scanning electron micrograph of 'double' megaspore of *I. coromandelina* ssp. macrotuberculata, distal face. Scale = $200 \ \mu m$. (Holotype.)

Fig. 17. Scanning electron micrograph of microspore of *I. coromandelina* ssp. coromandelina. Scale = $10 \mu m$. (Khandala, C. McCann, BLAT.)

tissue of the leaves but themselves lack chloroplasts. The side and outer walls of the hair cells are heavily thickened (Fig. 20) and bear conspicuousl, sometimes curved, spines on the projecting parts. The function of these cells is unknown. They



Figs. 18-24. I. coromandelina ssp. macrotuberculata.

Fig. 18. T.S. leaf (from middle of leaf) with peripheral fibre strands (pfs), stele (ls), internal hairs (ih) and the four lacunae (la). Fig. 19. Two cells of a translacunar diaphragm with acicular spines (sp) projecting into the air spaces (as). Fig. 20. Internal hair with thick walls and spines on projecting part of cell. Fig. 21. Sporangial wall cells. Fig. 22. Epidermal cells and stomata (s) from apical part of leaf. Figs 23, 24. Base of outer and inner sporophylls respectively, showing rounded sporangium (s), ligule (li), labium (lab), and broad membranous wings (w).

apparently do not serve a support function (Hall 1971) and the thickening of the cell walls suggests that they are not involved in absorption or gas exchange.

Translacunar diaphragms. Hall (1971) also noted, in some species of *Isoetes*, minute acicular spines projecting from walls of the cells which make up the diapgragms traversing the lacunae at intervals along the leaves. Similar spines were observed in both subspecies of *I. coromandelina* (Fig. 19).

Leaf bases. Leaves of I. coromandelina are expanded at the base into membranous wings, which narrow at about the level of the ligule and gradually taper over several centimetres along the leaf. These wings are much broader on the first-formed leaves of each season (Fig. 23) than on later leaves (Fig. 24); a corresponding change occurs in shape of sporangia from orbicular to obovate.

Corms. Although the corms of ssp. *coromandelina* are usually trilobed, plants with four or five lobes have been recorded. Only plants with trilobed corms have been thus far recorded for ssp. *macrotuberculata*.

Subspecific relationships

The ornamentation of megaspores has been widely used in taxonomy of *Isoetes*. Pfeiffer (1922) subdivided the genus into four sections, Tuberculatae, Echinatae, Cristatae and Reticulatae, to which a fifth section, Psilatae, was added by De Vol (1972).

I. coromandelina, which Pfeiffer (1922) placed in section Tuberculatae, varies slightly in megaspore morphology within each of the two subspecies, mainly in number and, to a lesser extent, size of the tubercles. Nevertheless, the ornamentation of the megaspores for each subspecies has been found to be consistently distinct (Figs 2-7, 12-15; Table 1).

In contrast to differences in overall ornamentation, the similarity in surface ultrastructure of the megaspores (Figs 8-11) probably indicates a close relationship between the two subspecies.

It is on the basis of differences in megaspore ornamentation contrasting with similarities in megaspore surface ultrastructure and general morphology that subspecific rank has been given to the Australian material of *I. coromandelina*. The geographic isolation of the Australian and Indian localities supports this conclusion.

ACKNOWLEDGMENTS

I wish to thank Dr E. M. Wollaston for her valued assistance and encouragement, and helpful comments on the manuscript. I am grateful to Dr H. K. Goswami, Mr C. D. Dunlop, and Mr A. C. Beauglehole for sending specimens. I am also indebted to Mr R. J. Chinnock and Dr J. Jessop for helpful suggestions, and to the Directors of the South Australian State Herbarium, the Herbarium of the Northern Territory, the Queensland Herbarium, the Blatter Herbarium, the Bombay Herbarium, Central National Herbarium of the Botanical Survey of India, the Dehra Dun Herbarium and National Science Museum, Tokyo, for specimens made available for this study.

I also wish to thank Mr J. Carrick for checking and correcting the Latin description.

This study was assisted by a grant under the Australian Biological Resources Study.

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