

ANTHORRHIZA ECHINELLA C.R.Huxley & Jebb

An ant-plant of the Rubiaceae

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Summary: The rare ant-plant *Anthorrhiza echinella* C.R.Huxley & Jebb from Papua New Guinea is described and illustrated. The outlook of the species and the importance of *ex situ* conservation strategies for this and other ant-plants are discussed in the context of botanic garden collections. Based on the plant's likely current distribution we have assessed the species under the IUCN Red List as Endangered (EN) under criteria B1ab(iii)+2ab(iii).

Plants and ants have interacted for at least 140 million years (Moreau et al., 2006), leading to the evolution of many complex mutualistic relationships (Huxley and Cutler, 1991). Ant-plants (myrmecophytes) are a diverse group of plants, occurring in at least 50 plant families (Chomicki and Renner, 2015) which produce specialised structures called domatia ('ant-houses') which their mutualist ant partners utilise for nesting. Domatia can be derived from various plant structures including leaves, stems and roots (Chomicki and Renner 2015). In addition to ants benefiting from a nesting space, some ant-plants provide food rewards (Chomicki and Renner 2015). In return for these services ants provide defence against herbivores or nutrition – sometimes both. The sub-tribe Hydnophytinae (Rubiaceae) represents the largest group of ant-plants with over 100 species (Huxley, 1980; Chomicki and Renner 2016). Species within the Hydnophytinae are exclusively epiphytic and possess modified hypocotyl derived domatia which contain complex labyrinths of cavities, connected to the external environment by entrance holes, which can be occupied by mutualistic ants. Ants forming symbioses with Hydnophytinae epiphytes provide nutrition by defecating inside the domatia, as well as by bringing organic debris and sometimes defend the plant from herbivory (Huxley, 1978; Chomicki and Renner, 2017; 2019).

The genus *Anthorrhiza* was first described by Huxley and Jebb (1991) following fieldwork in Papua New Guinea on the rubiaceae tribe Psychotriaceae (subtribe Hydnophytinae), upon finding that several specimens they observed did not belong to the genera described at the time (*Hydnophytum* Jack, *Myrmecodia* Jack, *Myrmephytum* Becc., *Myrmedoma* Becc. and *Squamellaria* Becc.). These new specimens shared the common characteristic of a single inflorescence borne in a leaf axil at each node, rather than the paired inflorescences observed in the genera *Hydnophytum* and *Myrmecodia* (Huxley and Jebb 1991). The generic name alludes to the fact that in some species, including *A. echinella*, there are ridges of tissue running between the flower-bearing areas that are surmounted by root-derived spines. The initial description of the genus *Anthorrhiza* included eight species, with the ninth species, *A. camilla*, described two years later (Jebb 1993).

The first specimens of *Anthorrhiza* were collected long before the formal description of the genus in the 1990s. In fact they were first collected in 1935 by Cedric Carr, at Boridi in Central Province of

Papua New Guinea. He noted in his diary: "an interesting find was a simple plant, to which I gave no number, of a small very spiny *Myrmecophilum* [sic] with large snow white flowers." (MS in Natural History Museum, London). Unfortunately, his collection was not assigned a number, but is of the species *A. chrysacantha*. Just two months later Mary Clemens collected *Anthorrhiza echinella* near Finschhafen, in Morobe Province of Papua New Guinea. It was this species that Camilla R. Huxley recognised as belonging to a distinct genus when she collected specimens on Perakles Pass on the road between Lae and Bulolo in 1975. The specific epithet of the species describes the spine-covered inflorescences that have the appearance of small sea-urchins or pin-cushions.

ECOLOGY The Hydnophytinae is the largest group of ant-plants. Much like the rest of the subtribe Hydnophytinae, the nine known species in the genus *Anthorrhiza* show a high degree of variation in their mutualistic strategy and level of dependency on ants. The ant-plant mutualist strategies in the genus can be classified broadly into three groups. Firstly, generalist species (*A. echinella* [Figs. 1-3], *A. mitis*, *A. recurvispina* and *A. stevensii*) are often occupied by ants, however they do not display a high level of specificity regarding which species of ant they host. There is a high level of variation within the generalist strategy ranging from species that are often ant-occupied to those that are only occasionally ant-occupied (Huxley and Jebb, 1991). Second, specialist species (*A. chrysacantha*, *A. caerulea* and *A. camilla*) display a higher level of specialisation, and are usually inhabited by just one species of ant, typically the dolichoderine species *Anonychomyrma scrutator* (formerly *Iridomyrmex scrutator*). *Anthorrhiza chrysacantha* and *A. caerulea* provide post-anthetic sugar rewards in addition to the benefit of providing the ants a nesting space (Chomicki et al. 2016). The ants provide an additional service to nutrition whereby ants act as 'bodyguards', protecting against herbivorous insects. Importantly, specialist *Anthorrhiza* species appear to be farmed by ants in 'ant-gardens'. Ant-gardens arise from ants' seed planting behaviour, in which ants deposit the seeds of various plant species, including many myrmecophytes, within their nutrient-rich carton nests (nests actively built by most arboreal ants, made up of plant materials, debris and secretions) (Maeyama and Matsumoto 2000; Chomicki et al., 2017). These ant-gardens benefit the plant by creating a nutrient-rich environment, and the ants benefit by strengthening their carton nests, and increasing their nest space capacity (Kaufmann 2002). Seed planting behaviour has been confirmed experimentally for *A. caerulea* (Maeyama and Matsumoto 2000). With the exception of seed planting observations, the cultivation of specialized *Anthorrhiza* species by *Anonychomyrma* has not been studied. It remains unknown whether this system is closer to the Southeast Asian ant gardens (e.g. Kaufmann and Maschwitz, 2006) or to the true agriculture involving the closely related Fijian *Squamellaria* farmed by *Philidris nagasau* ants (Chomicki and Renner, 2016; Chomicki et al., 2019); the latter are also close relatives of *Anonychomyrma* ants that are established to inhabit *Anthorrhiza*. Lastly, two species of *Anthorrhiza* have lost their mutualism with ants (Chomicki and Renner, 2017). In *Anthorrhiza bracteosa*, the tuber is usually inhabited by cockroaches, beetles and other invertebrates; it is only rarely inhabited by ants (Huxley and Jebb 1991). In *A. areolata*, the tuber morphology has uniquely large, funnel-shaped entrance holes that lead to cavities filled with rainwater. As a result, the tuber of this species is never ant-occupied, but frequently contains cockroach egg cases, and occasionally geckoes (Huxley and Jebb 1991). Whether these non-ant inhabitants provide nutritional benefits, and if so to what extent, remains an open question.

CONSERVATION

No *Anthorrhiza* species have yet been assessed for the IUCN red list.. Their limited range (Huxley and Jebb 1991) and the extent of deforestation in Papua New Guinea (Bryan and Shearman 2015) suggest that this species is likely to be endangered. An *in situ* IUCN assessment of the species' distribution, population numbers and quantitative decline data is hampered by challenges associated with working in Papua New Guinea, such as the lack of road connectivity (Gibson and Rozelle 2003) and socio-political issues. Therefore here, we provide an assessment informed by geographic extent (the IUCN B category). The village of Patep, close to the type collection site, has expanded since the

plant's original collection in the 1980s. However the riverine forest where it grew is apparently intact, judging by Google Earth imagery; moreover *Anthorrhiza* is a genus more typical of montane, dwarf or semi-dwarf forest, or open *Casuarina* savannas, riparian, disturbed or coastal habitats than typical timber concession-forest.

Using the GeoCAT tool (Bachman et al. 2011), an IUCN Red List (IUCN 2001) compliant software, distribution data was used to calculate the extent of occurrence (EOO), and area of occupancy (AOO) for the species. A conservation rating was generated using the methodology of Willis et al. (2003). We determined the species as being Endangered under category B1 (EOO being less than 5000km²) a (severely fragmented and with no more than 5 locations) b(iii) (continuing decline in quality of habitat), as well as under category B2 (AOO being less than 500km²) a (severely fragmented and with no more than 5 locations) b(iii) (continuing decline in quality of habitat). In summary, we recommend that the species be treated as Endangered (EN) under criteria B1ab(iii)+2ab(iii).

The specimen of *A. echinella* at the University of Oxford Botanic Garden (Figs. 1-3) was collected by M. H. P. Jebb in May 1983. It was displayed without a label in a public glasshouse until in 2014, G. Chomicki, at the time PhD student at the University of Munich, identified the plant and highlighted its international conservation value. Other species of ant-plant existed in the Oxford Botanic Garden collection but were lost in decades subsequent to collection in the 1970s and '80s due to the challenge of maintaining adequate conditions for long periods under glass. The surviving specimen of *A. echinella* has since been a source of DNA for research and is now represented in the current phylogeny of the genus (Chomicki et al., 2017); its genome sequencing is ongoing. The specimen at Oxford does not set seed, meaning that its conservation value is limited beyond a source of cloned material and DNA or RNA data. Manual pollination of its heterostylous flowers (Thrum only) has not been successful so far, probably because of self-incompatibility. It has, however, been successfully propagated through layering; however plants propagated in this way lack the charismatic domatium. The other species of *Anthorrhiza* are apparently absent from botanic garden's conservation collections, but are known to exist in private collections. In summary, the genus' representation in conservation collections is highly sporadic and incomplete. Given the proven ability of botanic gardens to maintain specimens of ant-plant over long periods of time under glass, *ex situ* conservation should be a priority for these rare and vulnerable plants. Efforts should also be made to better reflect the genetic diversity of populations to improve the conservation value of these collections.

Description:

Anthorrhiza echinella Huxley & Jebb, *Blumea* 36 (1991) 25.

Tuber suborbicular, slightly flattened, to 18 cm tall, 25 cm diameter, smooth, grey. Spines numerous, evenly scattered on surface, forming a more or less closed canopy, irregularly stellate, 1.0-1.8 cm long, side branches c. 6, 0.3-0.6 cm, sharp, yellow to blackish. Holes scattered over tuber surface, 0.2-5 cm in diameter, with raised rims, often surrounded or occluded by spines. Tuber tissue drying pinkish. Stems few, rarely branched, to 100 x 2.5 cm, semi-pendulous, upcurving, with 2 rounded ridges descending from stipules, grey green; internodes 5-8 cm long. Spines largely restricted to inflorescences, but also a few, 0.4(-1.4) cm long, in lines along stem ridges, or rarely scattered. Leaves spreading. Lamina 15x6-29x10 cm, lanceolate to obovate, apex acute, base tapered, leathery to brittle, dark glossy green above, pale below. Midrib prominent below, caniculate above, yellow-white. Veins 12-17, almost perpendicular to midrib, with alternate, very faint veins. Petiole 0-2(-3.5) cm, rounded below, yellowish. Stipules triangular, to 1 cm long, fairly persistent, brown. Inflorescence oval to oblong, often raised in the centre, to 4 x 1.5 cm, densely covered and surrounded by branched spines. Up to 15-20 flowering-bearing areas, groups of 1-3 being surrounded by a narrow wall of tissue 0.2-0.5 cm high and 0.1-0.2 cm thick, surmounted by spines; spines to 2 cm long, and irregularly or stellately branched. Bracts inconspicuous, papery, to 2 mm. Flowers heterostylous. Calyx 2-3 mm, level with or slightly above disc, membranous at margin, with flaky scales. Corolla

15-18 mm, white; lobes greenish white, c. 4 mm. A ring of hairs below the middle of the tube. Longistyle flowers with anthers at level of hairs, stigma exerted. Brevistyle flowers with anthers at mouth of tube, stigma at level of hairs. Anthers cream. Pollen with 3 small vesicles; 60-72 µm in diameter, with large irregular pores, reticulation medium to coarse. Fruit unknown. Pyrenes 4 to 6, 3 x 1.3 mm.

Distribution

Endemic to Morobe Province of Papua New Guinea, with collections from near Finschhafen, at Patep Creek, Buso, Lasanga Island and Fly Island. The source of the living material and that for the plates all comes from the type locality on the Lae-Bulolo road just north of Perakles Pass.

Habitat

Collected from coastal, riparian and disturbed forests at sea level to 1000 m. The plant was reported to grow on *Casuarina* and other open-canopied trees as a low to middle level epiphyte in which trees contained a few scattered individuals. The plant was found always to be ant-inhabited, but not with any great specificity. It was reported to co-occur with *Myrmecodia tuberosa* Jack.

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Figure 1. A cross-section through the tuber of *Anthorrhiza caerulea* (a species closely related to *A. echinella*), showing the warted and smooth cavities which are differentially used by ants: the warted cavities for detritus and the smooth cavities for the brood. Specimen: UPNG 3498 coll. J. Dodd Dec. 1975 Mt Kaindi, Papua New Guinea.

Figure 2. Illustration of *Anthorrhiza echinella* C.R.Huxley & Jebb. Illustration by Rosemary Wise.

Figure 3. Illustration of *Anthorrhiza echinella* C.R.Huxley & Jebb. Clockwise from top right: habit; root-derived spines; cross-section of corolla; corolla (lateral view); immature corolla (lateral view). Illustration by Rosemary Wise.

Figure 4. The specimen *Anthorrhiza echinella* at Oxford Botanic Garden, habit of a mature specimen.

Figure 5. *Anthorrhiza echinella* in cultivation at the University of Oxford Botanic Garden. (A) Flowers. (B) Groups of root-derived spines on the stem where flowers are borne. (C) Domatium surface, covered in root-derived spines.