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Two new alectoronic acid-containing *Parmotrema* species from the coast of São Paulo State, southeastern Brazil

MARCELO P. MARCELLI¹, MICHEL N. BENATTI² & JOHN A. ELIX³

Instituto de Botânica, Núcleo de Pesquisa em Micologia
Caixa Postal 3005, São Paulo / SP 01031-970, Brazil

³Research School of Chemistry, Building 33, Australian National University
Canberra, A.C.T. 0200, Australia

CORRESPONDENCE TO: ¹mpmarcelli@msn.com,

²michelbenatti@yahoo.com.br & ³John.Elix@anu.edu.au

ABSTRACT— Descriptions are presented for two new *Parmotrema* species containing alectoronic acid, *P. conidioarquatatum* and *P. pycnidioarpum*, resulting from a survey of *Parmeliaceae* species in natural ecosystems and urbanized coastal areas of southeastern Brazil.

KEY WORDS— *Parmotrema maraense*, *Parmotrema subrugatum*

Introduction

The genus *Parmotrema* A. Massal. is characterized by lobes with broad, rotund apices and naked lower margins, the absence of pseudocyphellae, the frequent occurrence of marginal cilia, simple rhizines, and thick-walled, ellipsoid ascospores (Brodo et al. 2001, Nash & Elix 2002). More than 300 species are known worldwide (Nash & Elix 2002), and about one third of them occur in Brazil.

Two new species containing alectoronic acid were discovered by the authors during research on the broad-lobed species of *Parmeliaceae* at the coast in São Paulo State, Brazil, at the same localities cited in Benatti et al. (2010).

Both new species lack vegetative propagules, and are corticolous in coastal mangrove or restinga forests. More morphological and chemical comparisons with other somewhat similar species can be found in Benatti (2005).

Material & methods

Specimens were distinguished by morphological characters using standard stereoscopic and light microscopes. Anatomical sections, including those of apothecia

and pycnidia, were made with a razor blade by hand. The chemical constituents were checked by spot tests with potassium hydroxide (K), sodium hypochlorite (C) and *para*-phenylenediamine (P), and also examined under UV light (360 nm). Chemical constituents were identified by thin-layer chromatography (TLC) using solvent C (Bungartz 2001), high performance liquid chromatography (HPLC) (Elix et al. 2003) and comparison with authentic samples.

We have encountered problems dealing with the many morphological terms present in the literature. For the purpose of this paper, we consider that lacinules represent adventitious, ribbon-like secondary outgrowths from the primary lobe margins (Marcelli et al. 2007). Lobules are similar, but short and rounded.

The diagnosis for each taxon refers exclusively to observations of the holotype specimen and the English descriptions and comments to all the material studied.

The species

Parmotrema conidioarcuratum Marcelli, Benatti & Elix, sp. nov.

FIG. 1

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Species cum thallo similis Parmotrematis subrugati sed lobis latioribus, lacinulis multum longioribus et ramosioribus, lobulis aggregatis saepe submarginalibus formans, ciliis saepe majoribus, margine inferne eburnea solum subter apothecia vel lacinulis, apotheciis cum margine semper eciliata, sporis minoribus et conidia variabiliter arcuatis differt. atranorinam, chloroatranorinam, acidum alectoronicum, acidum β -alectoronicum, acidum dehydrocollatolicum et acidum dehydroalectoronicum continens.

HOLOTYPE: Brazil, São Paulo State, Municipality of Ilha Comprida, near the raft to Cananéia, low “restinga” vegetation, corticolous, under partial sunlight, leg. A.A. Spielmann, M.P. Marcelli, L.S. Canêz, & M.N. Benatti 964, 03-IV-2004 (SP).

ETYMOLOGY: This species is named after the arcuate conidia.

THALLUS up to 23 cm in diameter, subcoriaceous, ramulicolous, pale greenish gray but becoming dark gray in the herbarium, lobed to sublobed. **LOBES** irregularly branched, (3.5–)8.0–18.0 mm wide, crowded, weakly to strongly ascending, generally unattached; apices \pm plane to subconvex, subrotund (primary lobes) to irregular and partially lacinulate; margin subcrenate to irregularly dissected, \pm plane to ascending as it begins to form lacinules and/or apothecia, irregularly weakly incised, ciliate. **UPPER SURFACE** continuous to irregularly cracked, smooth to subrugose; **MACULAE** distinct and punctiform to linear when present on the stipes and amphithecia of the apothecia, laminal maculae rare and weak. **LACINULES** usually long, 0.3–17.5 \times 0.2–2.3 mm, regularly spreading from lobe margins or apices, abundant at thallus center, initially simple but becoming irregularly branched, subcanaliculate, truncate or acute, ciliate, underside generally white, often mixed with tiny ciliate lobules which may develop into peculiar, scattered, submarginal, small, bouquet-shaped agglomerations, 3.0–8.0 mm wide. **SOREDIA**, **PUSTULES** and **ISIDIA** absent. **CILIA** black, simple or rarely furcate, 0.4–4.5 \times ca. 0.05 mm, abundant at the lateral margins of the lobes but sparse at the apices. **MEDULLA** white, with scattered

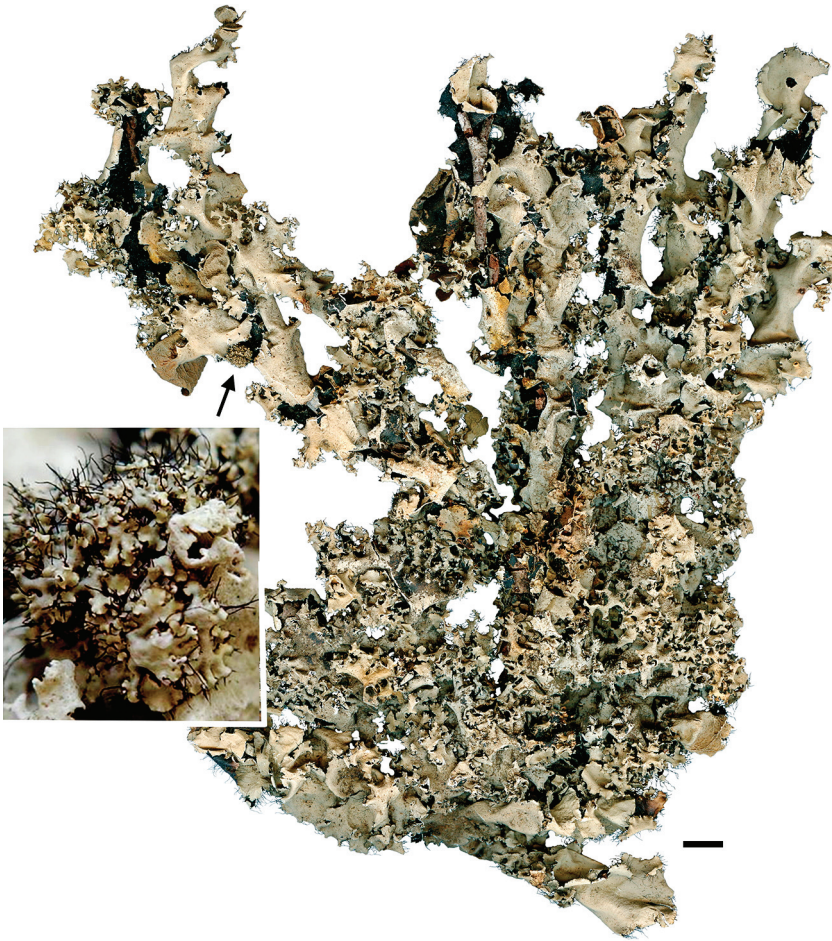


FIGURE 1. The holotype of *P. conidioarcuratum* and the bouquet of lobules typical of the species (arrow and detail). Bar = 1 cm.

orange, pigmented spots. LOWER SURFACE black, shiny, smooth to rugose or veined, free of rhizines when not contacting the substrate; MARGINAL ZONE shiny, usually brown, but turning white, cream or sometimes variegate under lacinules or apothecia, smooth to subrugose, 1.0–8.0 mm wide, naked; RHIZINES black, simple, furcate to irregularly branched, $0.10\text{--}2.30$ (3.80) \times 0.05 ($\text{--}0.10$) mm, frequent to abundant, in scattered groups. APOTHECIA submarginal or subapical, sometimes originating from subcanaliculate lobe apices, common, concave when young but becoming fissured and distorted with age, up to 25.2 mm wide, margins smooth to subcrenate, rarely lacinulate, always eciliate,

with very inflated stipes, amphithecia and stipe smooth at first then strongly veined and rugose or folded with age; disc brown, epruinose, imperforate; ASCOSPORES ellipsoid, $19.0\text{--}25.0 \times 9.5\text{--}12.5 \mu\text{m}$, episore $1.5\text{--}2.0 \mu\text{m}$ thick; PYCNIDIA submarginal, common, particularly abundant on the lacinules, with black ostioles; CONIDIA variable inside a single pycnidium, bacilliform or short filiform, often arcuate, sigmoid or contorted into several different shapes, $5.0\text{--}7.5 \times \text{ca. } 1.0 \mu\text{m}$.

COLOR REACTIONS: upper cortex K+ yellow, UV–; medulla K+ weakly yellowish, C–, KC+ rose, P–, UV+ bluish green, and scattered spots of an orange, K+ dark reddish pigment.

TLC/HPLC: cortical atranorin (minor) and chloroatranorin (minor); medullary alectoronic acid (major), β -alectoronic acid (minor), dehydrocollatolic acid (minor), and dehydroalectoronic acid (trace).

COMMENTS: This species produces no vegetative propagules, has densely ciliate margins with abundant, long and branched lacinules, a brown lower marginal zone that turns white or cream colored only under lobes with apothecia and/or lacinules. The apothecia have smooth to subcrenate, eciliate margins, with ascospores up to $25.0 \mu\text{m}$ long, and a white medulla containing frequent spots of an orange, K+ dark reddish pigment.

However, the most striking characteristics of *P. conidioarcuratum* are the formation of tiny ciliate lobules, especially at submarginal parts of the thallus, lobules which eventually aggregate into small bouquets, and, most notably, the bacilliform or short filiform conidia that are often variably arcuate (arched, sigmoid, sinuous, subcrenate, somewhat unciform and several other shapes) intermixed with some linear conidia. Together, these characters readily distinguish *P. conidioarcuratum* from other *Parmotrema* species containing alectoronic acid. It is also one of the species with the broadest lobes in the genus.

We found no reference to *Parmotrema* species bearing similar small bouquets of lobules, nor are they present in any other Brazilian species known to us. Initially, we considered the possibility that this was due to some kind of deformation of the lacinules, but these lobules are clearly very common and consistent, differing in shape from the lacinules and with such regularity on the upper cortex, that they constitute a very unique morphological characteristic of the species. In addition, we had access to other specimens collected from inland São Paulo State, which have the same lobular bouquets (in early development), arcuate conidia and the other specific characteristics.

At first glance, *P. conidioarcuratum* might be mistaken for *P. subrugatum* (Kremp.) Hale, which invariably has unciform conidia ($4.0\text{--}6.0 \mu\text{m}$) and larger ascospores ($26.0\text{--}39.0 \mu\text{m}$). Sterile specimens of *P. subrugatum* (M!) are readily distinguished by the much narrower, more sparingly ciliate lobes,

the continuously white borders under the margins of all lobes, the absence of the lobular bouquets, and the simple to dichotomously branched lacinules (Benatti et al. 2010). In contrast, the lacinules of *P. conidioarcuratum* are more canaliculate, irregularly branched, densely ciliate, and grow crowded along the lobe margins; in addition they are sometimes more rugose due the accumulation of pycnidia.

Because of the dense marginal lacinulae and cilia, *P. conidioarcuratum* is probably more closely related to the *P. wainioi* subgroup, as they have similarly sized ascospores and conidia as well as a brown marginal zone on the underside. The unusually shaped conidia present in *P. conidioarcuratum* are believed to be derived from bacilliform or short filiform conidia.

Parmotrema maraense Hale (Hale 1990) has narrower lobes, with margins bearing sparingly branched lacinules, as well as more lacinulate and ciliate apothecia (Benatti 2005, Benatti et al. 2010), and short bacilliform conidia (Klaus Kalb, pers. comm.).

***Parmotrema pycnidioarpum* Benatti, Marcelli & Elix, sp. nov.**

FIG. 2

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Species cum thallo simili Parmotrematis subrugati sed pycnidii absentibus in lobis, apotheciis bullatis et multum pycnidiatis, cum pigmento aurantiaco disposito sub hymeniis et ad marginem differt. Atranorinam, chloroatranorinam, acidum alectoronicum, acidum α -collatolicum, acidum β -alectoronicum, acidum β -collatolicum, acidum dehydrocollatolicum, methyl pseudoalectoronatum, et methyl pseudo- α -collatolatum continens.

HOLOTYPE: Brazil, São Paulo State, Municipality of Iguape, Barra do Ribeira, between Suamirim “River” and the ocean, low “restinga” vegetation growing on inundated soil, on small tree branch in the wood, leg. M.P. Marcelli & O. Yano 6663, 17-VII-1989 (SP).

ETYMOLOGY: This species is named after the peculiar distribution of the pycnidia.

THALUS up to 14.5 cm wide, submembranaceous, ramulicolous, pale grayish green, becoming darker in the herbarium, lobate to sublobate. **LOBES** 1.5–3.5(–5.0) mm wide, irregularly branched, imbricate to crowded, adnate, ascending when fertile, adnate to loosely adnate or unattached; apices \pm plane to subconvex when fertile, subrotund to irregular; margin smooth near the apices, \pm flat to ascending and subundulate, entire to irregularly incised, irregularly sublacinulate, ciliate. **UPPER SURFACE** continuous but becoming irregularly cracked with age, smooth to subrugose. **MACULAE** present only on some amphithecia and stipes, weak, punctiform, sometimes aggregated and linear. **ADVENTITIOUS LACINULES** sparse, simple or irregularly ramified, flat, 0.3–1.3 \times 0.2–1.1 mm, irregularly distributed along the lobe margins, truncate or acute, underside concolorous with the lower margin or cream on fertile lobes, occasionally intermixed with some small irregular lobules. **CILIA** black, simple to rarely furcate or irregular, 0.1–1.5 \times ca. 0.05 mm, frequent along the

margins but sparse at the lobe apices. MEDULLA white, invariably with orange pigmentation below the hymenium as apothecia mature, pigmented also at some apices and margins of older lobes. SOREDIA, PUSTULES and ISIDIA absent. LOWER SURFACE black, shiny, smooth to subrugose or weakly veined; MARGINAL ZONE shiny, brown, smooth to weakly veined, 0.5–3.5 mm wide, naked, turning cream under lobes with apothecia; RHIZINES black, simple, furcate or irregularly branched, $0.10\text{--}0.80\text{--}(1.30) \times \text{ca. } 0.05\text{--}(0.10)$ mm, frequent to abundant, occasionally becoming agglutinated, usually scattered but partly in groups. Apothecia submarginal or subterminal, in part originating from subcanaliculate lobe apices, concave to urceolate, becoming rugose and distorted with age, up to 5.2 mm wide, eciliate, stipes highly inflated and bullate, margins smooth when young, then crenate, amphithecia and stipes smooth when young but becoming rugose or vertically folded; disc brown, epruinose, imperforate; ascospores absent, no asci observed in hymenia; pycnidia frequent to very abundant on the stipes and amphithecia, rarely submarginal on apotheciate lobes, with black ostioles; conidia short, unciform, $3.0\text{--}5.0 \times \text{ca. } 1.0$ μm .

COLOR REACTIONS: upper cortex K+ yellow, UV–; medulla K–, C–, KC+ rose, P–, UV+ bluish green, with a K+ dark reddish, orange pigment (unknown anthraquinone) below the hymenia of all mature apothecia.

TLC/HPLC: cortical atranorin (minor) and chloroatranorin (minor); medullary alectoronic acid (major), α -collatolic acid (minor), β -alectoronic acid (trace), β -collatolic acid (trace), dehydrocollatolic acid (trace), methylpseudoalectoronate (trace) and methyl pseudo- α -collatolate (trace).

COMMENTS: This alectoronic acid-containing species is characterized by narrow, ciliate lobes, the absence of vegetative propagules, eciliate apothecia with highly inflated, bullate stipes, pycnidia mainly on the amphithecia and stipes of the apothecia, very small unciform conidia, and an orange-pigmented medulla around the hymenium.

The orange medullary pigment (K+ dark reddish, possibly an anthraquinone) which is frequently deposited under and around the hymenia of apothecia, appears also in the medulla of some of the older lobes (often those beginning to form apothecia).

Particularly notable is the uncommon distribution pattern of the pycnidia. They grow almost exclusively on the amphithecia and stipes of the apothecia. Only rarely some submarginal pycnidia are found on the lobes, precisely where apothecial initials are being formed, around the edge of the juvenile disc.

The shape of the highly inflated stipes somewhat resemble large “skirts” as they begin to form, with the resulting lobe apices becoming more convex, especially the subapical ones. In this aspect, they differ from all other Brazilian species with inflated apothecia (Hale 1965, Fleig 1997, Louwhoff & Elix 1999,



FIGURE 2. Both sides of the holotype of *P. pycnidiocarpum*.
Bar = 1 cm.

Eliasaro 2001, Kurokawa & Lai 2001, Canêz 2005, Spielmann 2005), where the apothecial pedicel is always \pm cylindrical.

The conidia shape and size place this species in the *P. subrugatum* subgroup (Benatti et al. 2010). However, the lower marginal zone of *P. pycnidiocarpum* varies from brown on the distal lobes to cream on those bearing apothecia, in contrast to the almost entirely white margins of *P. subrugatum* (Benatti et al. 2010).

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