

Standard Paper

A new epiphytic species of *Gyalolechia* with a maritime, Mediterranean-Macaronesian distribution

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Abstract

Gyalolechia fruticum, a new epiphytic species with a Mediterranean-Macaronesian distribution, is described. It differs in molecular data and in ecology from two closely related calcicolous species, *G. luteococcinea* and *G. marmorata*, both formerly called *Caloplaca subochracea* auct. The new species grows on coastal shrubs (rarely on wood), accompanied by species-rich communities of maritime epiphytic lichens. The typical habitat is undisturbed sand dunes, under dry conditions with frequent spells of humid air from the sea. The species is so far known from Greece, Italy, Tunisia, the Canary Islands and probably mainland Spain. We consider this conspicuous lichen an umbrella species of endangered and declining epiphytic communities of ancient shrub vegetation on well-developed sand dunes along Mediterranean coasts affected by touristic overexploitation.

Keywords: epiphytes; lichens; taxonomy; *Teloschistaceae*

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Introduction

The taxonomy of the calcicolous, Mediterranean lichen formerly known as *Caloplaca subochracea* auct. (*Teloschistaceae* Zahlbr.) was clarified by Arup *et al.* (2023) on the basis of morphological, chemical and molecular data. Their results showed that this lichen belongs to *Gyalolechia* A. Massal. as redefined by Arup *et al.* (2013), and that two closely related species can be recognized, formerly treated as varieties. In their phylogenetic tree, the two formed a fully supported clade but were separated on two distinct branches, indicating the existence of two saxicolous species: *G. marmorata* (Bagl.) Nimis & Arup (with a white thallus) and *G. luteococcinea* (Clauzade & Cl. Roux) Cl. Roux *et al.* (with a yellow thallus). Arup *et al.* (2023) had also included in the molecular analysis a sample from the Canary Islands (Tenerife) which was morphologically very similar to *G. luteococcinea*, but was epiphytic on *Euphorbia balsamifera* Aiton. That sample differed from *G. luteococcinea* in 17 bases in the ITS gene, suggesting that a third species could be present in the complex. This result reminded one of the authors of the present paper (PLN) of an epiphytic lichen that he had collected on coastal shrubs in several localities in Central and Southern Italy, which was very similar to *G. luteococcinea* but remained unidentified due to the very different substratum (Nimis 1993: 184). However, despite several attempts, these samples proved

to be too old for successful DNA extraction. Fresh epiphytic samples collected by JV in Rhodes (Greece) were molecularly analyzed, and proved to cluster together with the sample from Tenerife, prompting the search for fresh material of epiphytic samples from Italy; this was finally obtained from four localities in Latium and Sardinia in 2024. The molecular analysis of these samples confirmed the existence of a third, epiphytic species, which is formally described in this paper.

Materials and Methods

Anatomical measurements

Anatomical measurements were carried out in water, except those of paraphyses and excipular hyphae, which were observed and measured after treatment with 10% potassium hydroxide (K). Crystals were observed in polarised light (POL). Spot tests were carried out using K, commercial bleach (C) and paraphenylenediamine (P). Characters were measured according to the guidelines in Vondrák *et al.* (2013).

Secondary chemistry

Secondary metabolites were identified using HPLC according to Søchting (1997). Thallus and apothecia were analyzed separately, but fragments of apothecium initials could not be avoided completely in the thallus sample. The relative composition of the secondary compounds was calculated based on absorbance at 270 nm, according to Søchting (1997).

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Sequence alignment

ITS sequences from 19 *Gyalolechia* specimens of *G. luteococcinea*, *G. marmorata* and related taxa were downloaded from GenBank (Table 1). The ITS sequences were aligned together with six newly produced sequences of our new species in Geneious v. 11.1.15 using the MAFFT option (auto). Unalignable ends, indels and ambiguously aligned parts were excluded from the alignment. *Gyalolechia flavorubescens* (Hudson) Søchting *et al.* was used as outgroup.

Phylogenetic analysis

Phylogenetic relationships were inferred using maximum likelihood (ML) as implemented in IQ-TREE 2 (Minh *et al.* 2020), and Bayesian tree inference was carried out using Markov chain Monte Carlo (MCMC) as implemented in MrBayes v. 3.2 (Ronquist *et al.* 2012). The most suitable likelihood model, SYM + G, was selected

Table 1. Voucher information and sequences of *Gyalolechia* species used in the analysis (Fig. 1). Newly produced sequences are in bold and the remainder were downloaded from GenBank.

Species	Country, collector, collector no., herbarium	nrITS
<i>Gyalolechia epiphyta</i> 1	China, Vondrák JV14462, PRA	KU926995
<i>G. epiphyta</i> 2	China, Hur CH 130112, KoLRI	KY614429
<i>G. epiphyta</i> 3	China, Vondrák JV12411, PRA	ON028635
<i>G. flavorubescens</i>	Iran, Vondrák JV5575, PRA	KT804969
<i>G. fruticum</i> 1	Italy, Giordani 44810, TSB	PQ778908
<i>G. fruticum</i> 2	Italy, Nascimbene & Nimis 44807, TSB	PQ778905
<i>G. fruticum</i> 3	Italy, Nascimbene & Nimis 44809, TSB	PQ778907
<i>G. fruticum</i> 4	Italy, Nascimbene & Nimis 44808, TSB	PQ778906
<i>G. fruticum</i> 5	Greece, Vondrák 27405, PRA	OR490797
<i>G. fruticum</i> 6	Greece, Vondrák 27463, PRA	OR490798
<i>G. fruticum</i> 7	Greece, Sipman & Raus 63382, B	MN172443
<i>G. fruticum</i> 8	Tunisia, Guttová s. n., SAV	PQ778909
<i>G. fruticum</i> 9	Tunisia, Guttová s. n., SAV	PQ778910
<i>G. fruticum</i> 10	Tenerife, Klepsland JK14–1220, priv. hb.	OQ235067
<i>G. fruticum</i> 11	Greece, Sipman & Raus 63172, B	MN172442
<i>G. luteococcinea</i> 1	France, Bertrand & Roux 10, LD	OQ235060
<i>G. luteococcinea</i> 2	France, Bertrand & Roux 10, LD	OQ235061
<i>G. luteococcinea</i> 3	France, Bertrand & Roux 10, LD	OQ235059
<i>G. marmorata</i> 1	France, Arup L19416, LD	OQ235066
<i>G. marmorata</i> 2	France, Bertrand & Roux 9, LD	OQ235062
<i>G. marmorata</i> 3	France, Bertrand & Roux 9, LD	OQ235063
<i>G. marmorata</i> 4	Malta, Rabat 1 Jan 2020, TSB	OQ235065
<i>G. marmorata</i> 5	France, Bertrand & Roux 9, LD	OQ235064
<i>Gyalolechia</i> sp.	Portugal, Sipman 63007, B	MN586937
<i>Gyalolechia</i> sp.	Portugal, Sipman 63040, B	MN586938

using the Bayesian information criterion (BIC) as implemented in the software jModelTest v. 2.1.4 (Guindon & Gascuel 2003; Darriba *et al.* 2012), evaluating only the 24 models available in MrBayes (Ronquist *et al.* 2012). Three parallel runs with 20 000 generations, starting with a random tree and employing six simultaneous chains, were executed, five of which were incrementally heated with a temperature of 0.10. Analyses were diagnosed every 1000 generations in the last 50% of the tree sample and automatically halted when convergence was reached. Convergence was defined as an average standard deviation of splits (of frequency 0.1) between runs below 0.01. Every 2000th tree was sampled. A majority-rule consensus tree was constructed from the post-burn-in tree samples. The consensus trees were visualized using FigTree v. 1.4.4 (Rambaut 2018) and redrawn in Adobe Illustrator. The maximum likelihood analyses used the same evolutionary models as those used in the Bayesian analyses. Branch support values were computed via 1000 non-parametric bootstrap replicates.

Results

Molecular analysis

We generated six new nrITS sequences for this study and the alignment consisted of 25 sequences including the outgroup of 528 aligned nucleotide sites, of which 78 were parsimony-informative. The Bayesian analysis halted after 2 320 000 generations. The trees resulting from the Bayesian and maximum likelihood (ML) analyses were almost identical and did not differ significantly. In the ML tree (Fig. 1) the outgroup, *Gyalolechia epiphyta* (Lynge) Vondrák, and a branch holding *G. luteococcinea*, *G. marmorata*, an unknown epiphytic taxon from Madeira called *Caloplaca* cf. *flavovirescens* in the original paper by Sipman & Aptroot (2020), and our new species *G. fruticum* are placed as sister clades. On the latter branch, *G. fruticum* is sister to the taxon from Madeira, both are sister to *G. luteococcinea* and finally *G. marmorata* is sister to all of them. All taxa are well supported. These results suggest the existence of at least one epiphytic species in the complex, represented by the samples from Greece, Italy, Tunisia and Tenerife. A second epiphytic species could be represented by the two samples from Madeira, although we have seen no material. This taxon could have been included in the new species since they both sit on a well-supported branch and the epithet cf. *flavovirescens* suggests similarities with the other epiphytic samples. We choose to provisionally keep them as two distinct taxa because there is a difference of 10–13 bases between the sequenced specimens of *G. fruticum* and those from Madeira, and the clades are distinctly separated from one another in the phylogenetic tree. The new species is formally described and discussed below.

The mtSSU sequence from the type specimen is 99.8% similar to *G. luteococcinea*, the only available sequence in NCBI (OQ248479), and 98.7% and 98.8% similar to *G. marmorata* (OQ248480, OQ248481).

Secondary chemistry

The apothecia analyzed contain emodin and parietin as major compounds, and citreorosein, emodin, emodic acid, fallacinal and fragilin as minor compounds. The thallus contains mainly fragilin but also low amounts of emodin and parietin that probably originate from apothecium initials embedded in the thallus.

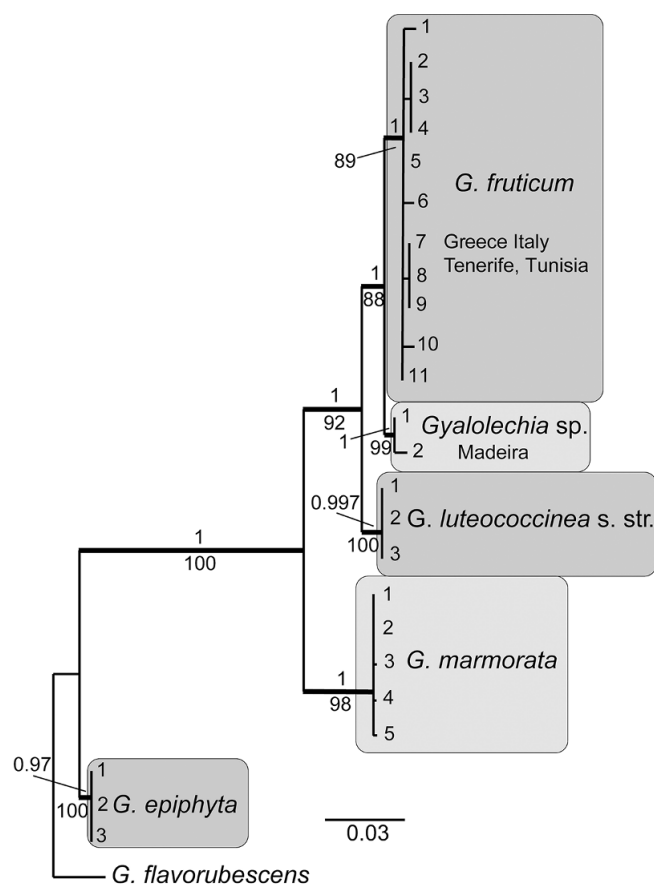


Figure 1. Majority-rule consensus tree based on a maximum likelihood analysis of nrITS data showing the position of the new species *Gyalolechia fruticum* in relation to its close relatives. Branches with posterior probabilities (PP) ≥ 0.95 are shown in bold. Bootstrap values and PP are presented below and above the branches, respectively. Voucher information and GenBank Accession numbers are available in Table 1.

Taxonomy

Gyalolechia fruticum Arup, Nimis & Vondrák sp. nov.

MycoBank No.: MB 857037

Differing from the calcicolous *Gyalolechia luteococcinea* in the epiphytic growth on littoral shrubs, and in molecular data.

Type: Italy, Sardegna, Prov. Sud Sardegna, St Anna Arresi, Loc. Porto Pino, 38°57'36"N, 08°36'39"E, alt. 10 m, sand dunes, on roots of *Juniperus macrocarpa*, 4 May 2024, coll. Giordani (TSB 44810, holotype). DNA sequences from the holotype: ITS (PQ778908), mtSSU (PQ778904); *rbcl* from the photobiont (PQ851754).

(Figs 2 & 3)

Thallus crustose, up to 3(–4) cm diam., at first very thinly epibiotic and c. 0.1 mm thick, then 0.2(–0.4) mm thick, bright orange in sun-exposed specimens to yellow (Fig. 2D) or almost white (Fig. 2A–C) in shade forms (but the peripheral areoles usually have a yellowish hue), continuous and smooth to rimose-areolate, often with a rough surface, usually surrounded by a pale prothallus (a dark, incomplete prothalline line was observed in a single specimen from Tenerife). **Photobiont** trebouxoid, the mature cells up to 15 µm wide. Based on the *rbcl* DNA barcode, the photobiont in the

type specimen has the closest (99.5% identical) NCBI Blast match with *Trebouxia decolorans* (AJ969660, photobiont of *Xanthoria parietina*). The algal layer is c. 50 µm thick, discontinuous (Fig. 3A) in young, thin thalli, continuous in well-developed thalli, overlain by an up to 80 µm thick epinecral layer formed of cell remnants (Fig. 3B), which is, in yellow-orange thalli, interspersed with anthraquinone crystals, POL+ bright orange (POL+ white, glowing crystals, probably of calcium oxalates, present in a single specimen with a whitish thallus); true cortex and medulla absent.

Apothecia biatorine, at first semi-immersed, then sessile (Fig. 3A & C), circular to finally irregular in outline, (0.2–)0.3–0.8(–1.1) mm wide, with an initially deep orange, finally often dark rusty red to dark orange-red (rarely blackening with age), slightly concave to slightly convex, epruinose disc; proper margin slightly paler orange but never yellow (Fig. 2A–D), smooth, at first c. 0.1 mm thick and slightly raised, finally level with disc or sometimes excluded. **Proper exciple** prosoplectenchymatous, with orange anthraquinones at surface, colourless inside, but in the specimen from Tenerife, interspersed with anthraquinone crystals also inside. Excipular hyphae arranged in a fan-shaped way, the cells c. 2 µm wide and thin-walled in water, but lumina very thin and cell walls thickened and adglutinated in K; outermost cells of exciple almost spherical, up to 4 µm wide. **Epithecium** brownish orange, K+ purple, C–. **Hymenium** colourless, (40–)50–70(–80) µm high, not interspersed with oil droplets. **Paraphyses** simple or forked in upper part, not anastomosing, c. 1.5–2 µm thick at base, the apical cells (2–)2.5–3.5(–4) µm wide. **Hypothecium** colourless, 90–110 µm high, formed of a thin prosoplectenchymatous tissue below a strongly developed subhymenium, usually densely interspersed with very small oil droplets (Fig. 3C) not dissolving in K, without POL+ bright white crystals (Fig. 3E), or rarely with POL+ crystals. **Asci** 8-spored (Fig. 3G), *Teloschistes*-type (Fig. 3H), 40–60 × 12–15 µm. **Ascospores** polarilocular (Fig. 3F), hyaline, ellipsoid, (9–)10–14 × (4–)5–8(–9) µm, the septum 4–6 µm wide.

Pycnidia usually numerous, c. 0.1–0.2(–0.25) mm diam., half- to fully immersed in the thallus (Fig. 3D), visible as orange-red to dark red (rarely almost black) dots (Fig. 2A & B) due to concentrated anthraquinones around the ostiolum (K+ purple). Conidiophores of *Umbilicaria*-type (*sensu* Vobis 1980; Fig. 3I). **Conidia** short-bacilliform, 3–4 × 0.8–1(–1.3) µm.

Chemistry. Thallus and apothecia with unchlorinated anthraquinones. Parietin and emodin predominate in apothecia, whereas fragilin predominates in the thallus. Spot tests: thallus and apothecia K+ purple-red, C–, KC–, P–.

Etymology. The epithet refers to the occurrence of the new species on shrubs.

Similar species. *Gyalolechia fruticum* can be confused with other species which may occur in the same habitat: *Gyalolechia flavorubescens* also has a yellow thallus, but differs in having larger (up to 3 mm across), lecanorine apothecia with a prominent, yellow proper margin, an orange (never rusty red) disc reacting C+ red due to the presence of chlorinated anthraquinones in the epithecium, an oil-interspersed hymenium, and slightly larger spores. Occasional specimens of *G. fruticum* without a yellow thallus could be confused with *Blastenia xerothermica* Vondrák *et al.*, or with *Caloplaca aegatica* Giralt *et al.*, both of which differ from *G. fruticum* in having dark grey to black pycnidia reacting K–, without anthraquinones; the latter species, which probably

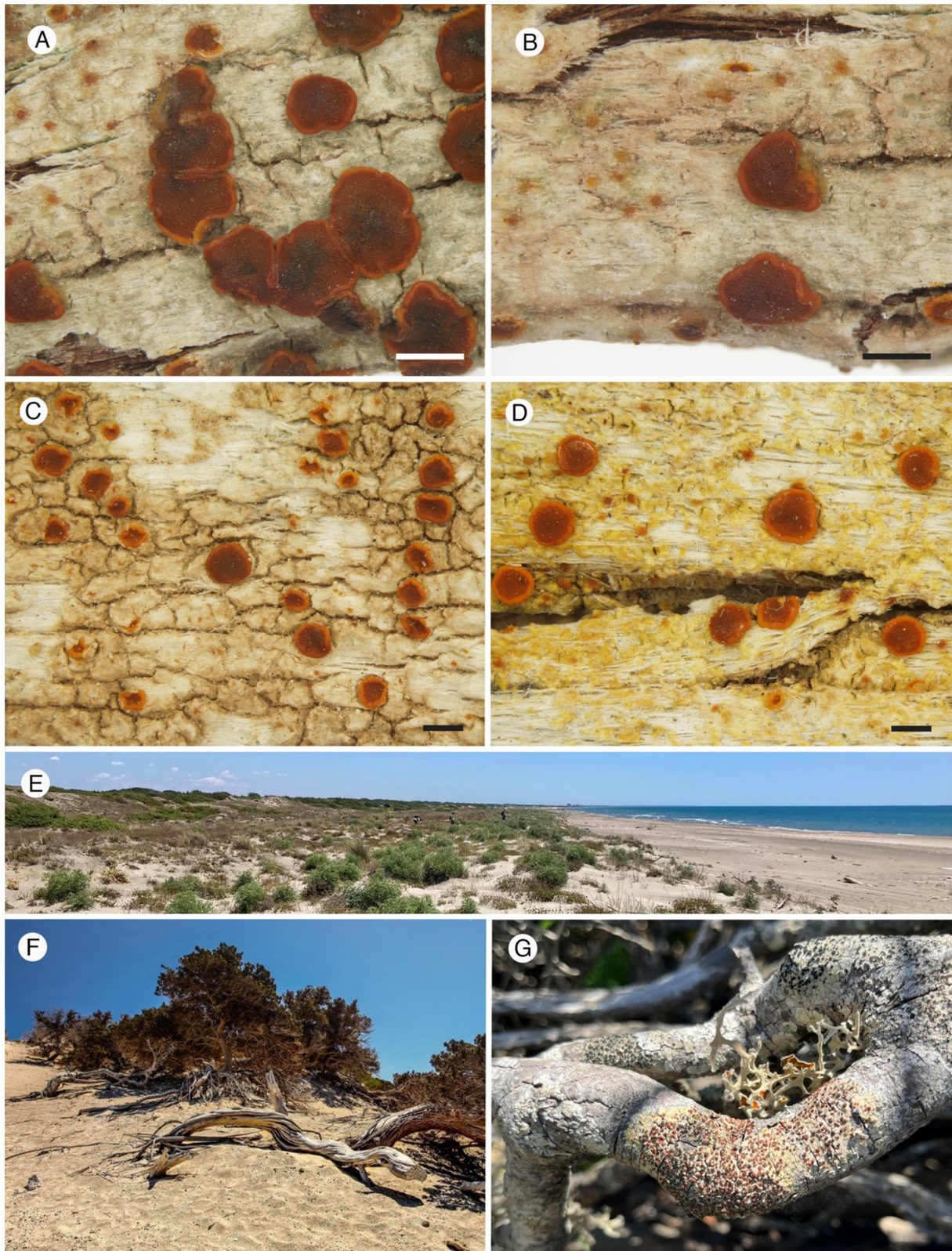


Figure 2. Morphology and habitat of *Gyalolechia fruticum*. A & B, the type specimen. A, with predominant apothecia. B, with predominant pycnidia (red dots). C, whitish thallus without anthraquinones (Vondrák 27405). D, yellowish thallus with anthraquinones (Vondrák 27463). E, the dunes of Castelporziano (Italy), with old shrubs in the background. F, old *Juniperus macrocarpa* with suitable microhabitats for *G. fruticum* (Greece, Chrissi) (source: freepic.com). G, *G. fruticum* with *Seiophora villosa* on a twig of *Pistacia* (Italy, Castelporziano). Scales = 0.5 mm. In colour online.

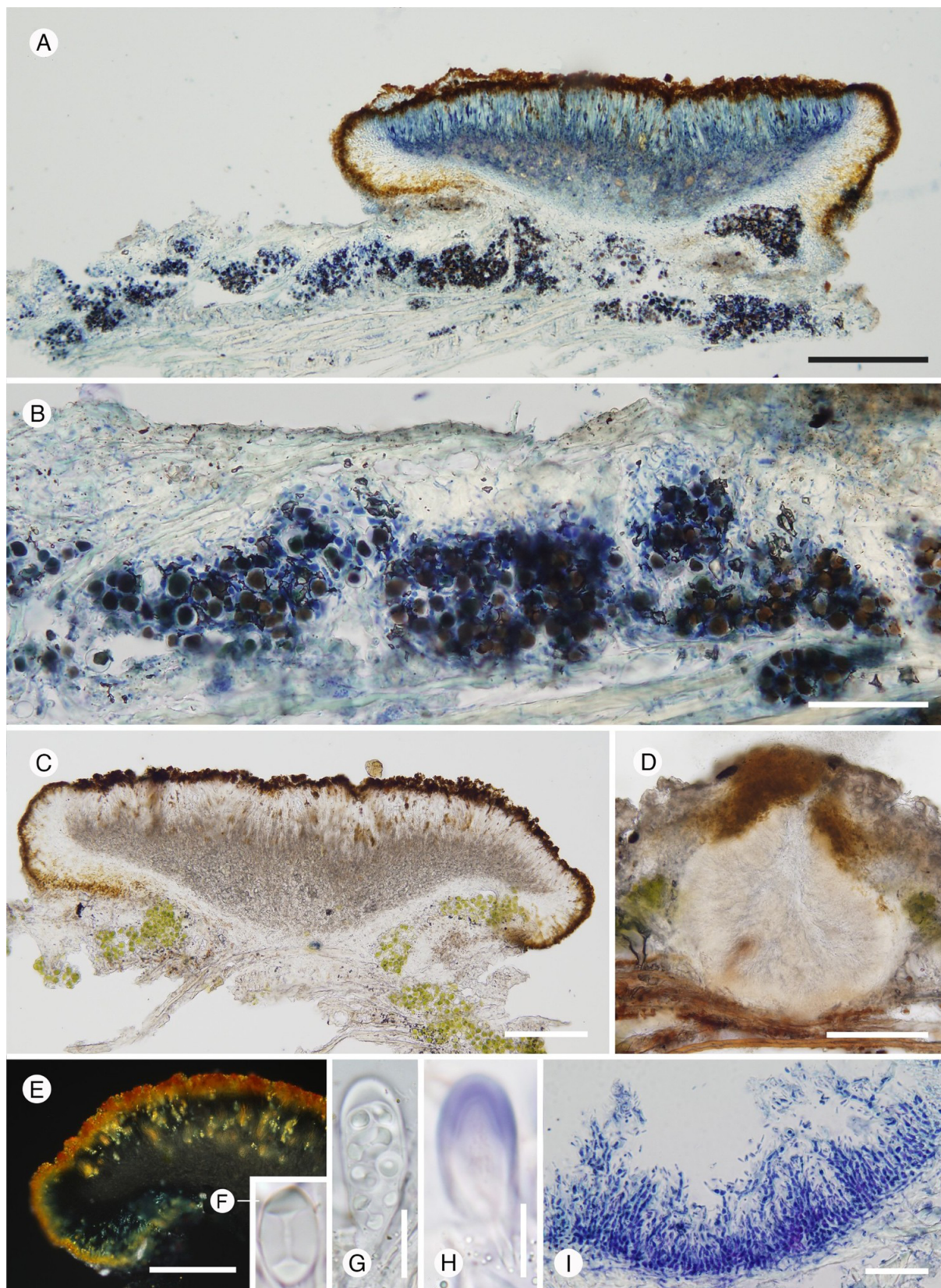


Figure 3. Anatomy of *Gyalolechia fruticum* in vertical sections. A, thallus and apothecium, algal cells forming colonies in a discontinuous layer between phorophyte bark tissue (below) and an epinecral layer (above). B, detail of thallus; vivid fungal cells (stained) are mostly restricted to algal colonies, whereas the epinecral layer is without living mycobiont cells. C, biatorine apothecium, with inspersed hypothecium and occasional algal colonies in lower exciple. D, pycnidium with distinct anthraquinone pigmentation around the ostiolum. E, apothecium in polarized light with glowing anthraquinones, but without POL+ crystals in the hypothecium. F, polarilocular ascospore (13 μm in length). G, ascus with ascospores. H, *Teloschistes*-type ascus. I, conidiophores and conidia. A, B & I, in lactoglycerol cotton blue; C–G, in water; H, in Lugol's solution after K treatment. Scales: A, C–E = 100 μm ; B = 50 μm ; G & H = 10 μm ; I = 20 μm . In colour online.

belongs in *Ikaeria* (Sipman & Aptroot 2020), also differs in having broadly ellipsoid to subglobose ascospores.

Ecology and distribution. All samples were growing in coastal localities subject to humid maritime winds, mostly on the twigs of shrubs (*Euphorbia balsamifera*, *Juniperus macrocarpa* Sibth. & Sm., *Phyllirea* spp., *Pistacia lentiscus* L. and *P. terebinthus* L.) on sand dunes, but in one case also on wooden planks. The new species rarely occurs in very sun-exposed situations, preferring slightly shaded conditions with plenty of diffuse light; this was particularly evident in the case of specimens colonizing erect wooden planks on the dunes of the Castelporziano Estate (Italy), which were present only on the north-facing sides of the planks, while the south-exposed flanks were colonized by other lichens.

Frequent accompanying species, both in Greece and in Italy (Castelporziano), were *Amandinea maritima* Giralt et al., *Bacidia parathalassica* Llop & Gómez-Bolea, *Bactrospora patellarioides* (Nyl.) Almq., *Caloplaca aegatica* Giralt et al., *Diploicia canescens* (Dicks.) A. Massal., *Lecidella elaeochroma* var. *flavicans* (Ach.) Hazsl., *L. elaeochroma* var. *juniperina* Poelt & Nimis, *Physcia adscendens* H. Olivier, *Pyrrhospora quernei* (Dicks.) Körb., *Ramalina canariensis* J. Steiner and *Xanthoria parietina* (L.) Th. Fr. At Castelporziano (Italy), *Ramalina lacera* (With.) J. R. Laundon, *Seiophora villosa* (Ach.) Frödén and *Tornabea scutellifera* (With.) J. R. Laundon were also present and often abundant. The most typical lichen vegetation in which the new species occurs along the Tyrrhenian coasts of Italy was described by Nimis & Schiavon (1986) as Teloschisto-Tornabeniopsidetum atlanticae, a rare community very much resembling the one described by Hernández Padron & Pérez de Paz (1980) for the Island of Hierro (Canary Islands), on twigs of *Juniperus phoenicea* L. subjected to humid winds from the ocean. In Italy, such vegetation is restricted to a small number of coastal, protected areas in the Mediterranean biome, having been largely destroyed by the touristic exploitation of beaches, with the consequent disappearance of well-developed, undisturbed sand dunes. The climate of the Italian stations can be characterized as Mediterranean-suboceanic (Nimis & Schiavon 1986), and has clear affinities with that of the Atlantic coasts of south-western Europe (Giacobbe 1949). The microclimatic conditions are generally dry, but with frequent spells of humid air from the sea. This is well in accordance with the peculiar ecology of the two most conspicuous accompanying species in the most undisturbed stands in Italy that was underlined by Nimis & Poelt (1987). These authors subdivided the lichen biota of Sardinia into several biogeographical groups, with the smallest group including only two species: *Tornabea scutellifera* and *Seiophora villosa*, designated as 'species of fog-deserts', well adapted to long, dry and warm periods but needing a periodic supply of high atmospheric moisture.

The lichen communities with *G. fruticum* observed in Rhodes, on coastal dunes with ancient *Juniperus macrocarpa* shrubs, were rather different from those in Italy. The two flagship lichens present at the Italian sites (*Seiophora* and *Tornabea*) were not observed, perhaps due to the slightly drier climate. Macrolichens were rarely observed, only *Physcia adscendens*, *Xanthoria parietina* and a small number of *Ramalina canariensis* thalli. However, microlichens co-occurring with *G. fruticum* formed a very specific maritime community. Frequent species included *Alyxoria ochrocincta* (Werner) Ertz, *Arthonia albopulverea* Nyl., *A. tenellula* Nyl., *Catillaria servitii* Szatala, *Coppinsiella ulcerosa* (Coppins & P. James) S. Y. Kondr. & Lőkös, *Dirina ceratoniae* (Ach.) Fr., *Haloplaca* sp., *Lecanora lividocinerea* Bagl., *Melaspilea oleae* J. Steiner, *Naevia*

pinastri (Anzi) Thiyagaraja et al., *Ocellomma picconianum* (Bagl.) Ertz & Tehler, *Polycauliona phlogina* (Ach.) Arup et al., *Rinodina furfuracea* H. Magn., *Scutula effusa* (Rabenh.) Kistenich et al., *Thelenella melanospora* Etayo & H. Mayrhofer, *Thelopsis isiaca* Stizenb. and *Toninia* aff. *populorum*.

The distribution of the new species (Fig. 4) is clearly Mediterranean-Macaronesian, extending from the easternmost islands of Greece (Rhodes) to the Canary Islands, through Italy and Tunisia, and possibly mainland Spain (Almeria, El Ejido, 5 m, on twigs of *Juniperus turbinata* Guss.; see good images posted at <http://liquesdesdealmeria.blogspot.com/search/label/Caloplaca%20subochracea%20var.%20luteococcinea>). In Italy, it has a mainly western, Tyrrhenian distributional pattern.

Additional specimens examined. **Greece:** Rhodes: Apolakkia, 36° 04' 11" N, 27° 45' 41" E, alt. c. 5 m, on wood of *Juniperus macrocarpa*, 18 iv 2023, Vondrák (PRA 27405); *ibid.*, on bark of *Pistacia lentiscus*, Vondrák (PRA 27422); *ibid.*, 36° 04' 43" N, 27° 45' 42" E, on bark and wood of *Juniperus macrocarpa*, 15 iv 2023, Vondrák (PRA 27463, 27479).—**Italy:** Toscana: Prov. Livorno, Parco Nazionale Arcipelago Toscano, Pianosa, 42° 35' 2.3" N, 10° 04' 28" E, alt. 15 m, on bark, 2005, Tretiach & Muggia (TSB 38194). Lazio: Prov. Roma, Torvaianica, 41° 36' 46" N, 12° 28' 44" E, alt. 4 m, on *Juniperus*, 1987, Nimis & Tretiach (TSB 9972); *ibid.*, Presidential Estate of Castelporziano, 41° 41' 32" N, 12° 22' 45" E, alt. 20 m, on bark of *Juniperus macrocarpa*, 1985, Nimis (TSB 6369); *ibid.*, 41° 41' 25.1" N, 12° 21' 59.2" E, alt. 11 m, coastal sand dunes, on wooden planks, N-exposed, 27 v 2024, Nascimbene & Nimis (TSB 44808); *ibid.*, 41° 41' 32.5" N, 12° 21' 47.6" E, alt. 10 m, coastal sand dunes, on *Juniperus macrocarpa*, 27 v 2024, Nascimbene & Nimis (TSB 44807); *ibid.*, 41° 41' 14.8" N, 12° 22' 17.7" E, alt. 10 m, coastal sand dunes, on *Phillyrea*, 27 v 2024, Nascimbene & Nimis (TSB 44809); Prov. Latina, Circeo National Park near Sabaudia, 41° 14' 39" N, 13° 02' 8.6" E, alt. 2–3 m, on *Juniperus macrocarpa* on coastal dunes, 1984, Nimis (TSB 4162). Puglia: Prov. Foggia, Tremiti Islands, San Domino, 42° 06' 44" N, 15° 29' 33" E, alt. c. 30 m, on *Pistacia* along the coast, 1986, Nimis (TSB 7589). Sicilia: Prov. Agrigento, Pelagie Islands, Linosa, Mt Nero, 35° 51' 31" N, 12° 52' 12" E, alt. 80 m, on bark of *Pistacia*, 1992, Nimis (TSB 17325); Prov. Trapani, Egadi Islands, Marettimo, 37° 58' 52" N, 12° 03' 29" E, alt. 30 m, on bark, 1988, Ottonello (TSB 12174).—**Spain:** Canary Islands: Tenerife, Anaga, between Chamorga and Roque Bermejo, 28° 34' 51" N, 16° 08' 27" W, alt. 255 m, on old



Figure 4. Distribution map of *Gyalolechia fruticum*. The triangle refers to a specimen identified from a photograph (see main text).

Euphorbia balsamifera in *Euphorbia* habitat, xii 2014, J. Klepsland (JK14-1220).—**Tunisia:** Les Mogods, Corniche - La Grotte, alt. 27 m, 37°19.956'N, 9°50.593'E, on *Juniperus* twigs, 29 iii 2014, A. Guttová s. n. (SAV, as *Gyalolechia flavorubescens* in Guttová *et al.* (2015)). Nabeul: Cap Bon, Djebel Haouaria Mtn, 37°04.282'N, 11°02.456'E, alt. 367 m, on wood of *Erica* and *Phillyrea* branches growing on the cliff top, 26 iii 2014, A. Guttová s. n. (SAV, as *Blastenia* cf. *hungarica* in Guttová *et al.* (2015)).

Discussion and Conclusion

The new species is morphologically very similar to the calcicolous *Gyalolechia luteococcinea*, the only substantial differences being the higher frequency in the latter of a black prothalline line delimiting the thallus (which is, however, present in the epiphytic sample from Tenerife), and the presence of POL+ crystals (probably of calcium oxalates) in the hypothecium, most likely due to the growth on calcareous rocks (such crystals are, however, present in an epiphytic sample with a pale thallus from Greece). Other important differences are in the substratum, the new species being consistently epiphytic, and in the different molecular data.

The ITS sequences of *G. fruticum* differ from those of known related species in at least 15 positions, allowing convenient identification of the species by ITS-barcoding. There are numerous closely related species in *Teloschistaceae* that differ in fewer than five bases, as for example *Flavoplaca communis* (Vondrák *et al.*) Arup *et al.* and *F. rudorum* (Malbr.) Arup & Søchting (see Arup *et al.* 2024). Some morphologically defined species are even undifferentiated by ITS; for example, *Variospora aurantia* (Pers.) Arup *et al.* and *V. flavescens* (Huds.) Arup *et al.*, are phenotypically well separated (Šoun & Vondrák 2008), but are indistinguishable from each other in the ITS-barcode (T. Hauser & J. Vondrák, unpublished data). In this context, the differentiation of *G. fruticum* at the species level is sufficiently reliable.

Gyalolechia luteococcinea and *G. fruticum* are a further example of a species pair in which substratum requirements may have caused speciation, the substratum allowing their distinction in the simplest way, as discussed by, for example, Vondrák *et al.* (2020) for *Blastenia*, Kantelinen *et al.* (2022) for *Micarea*, and Kosecka & Oset (2024) for the *Varicellaria lactea*–*V. hemisphaerica* species pair.

The typical habitats for *G. fruticum* are old-growth *Juniperus macrocarpa* stands on coastal sand dunes which have been heavily disturbed and partly damaged in many places along the Mediterranean coasts (Muñoz-Reinoso 2003, 2004; Picchi 2008). Due to the disappearance of its natural habitat (various undisturbed coastal shrubs), the new species is currently quite rare and probably further declining, apparently being highly sensitive to human disturbance: for example, in the Presidential Estate of Castelporziano near Rome, it is still rather frequent but only in the strictly protected area, while it seems to be completely absent in the adjoining area open to the public. Due to its conspicuousness, *G. fruticum* may represent an umbrella species for species-rich and threatened epiphytic communities growing on ancient shrubs along the Mediterranean–Macaronesian seashores.

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Data Accessibility. Newly generated sequences have been deposited in GenBank. Nomenclatural novelties have been deposited in MycoBank.

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