Llimonaea sorediata, a new lichen (Ascomycota), widely distributed in western Europe

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Abstract: The first sorediate species in the genus *Llimonaea*, *L. sorediata* is described from coastal areas of western Europe. This new species is easily mistaken for *Dirina massiliensis* f. sorediata with which it often co-occurs. It grows on vertical or overhanging acidic cliffs. Fertile specimens were recently collected in Portugal. A description of the species together with notes on distribution, ecology and chemistry are provided and a key to the species of *Llimonaea* is also provided.

Key words: Arthoniales, coastal habitats, corticolous, saxicolous, species nova, Trentepohlia

Introduction

A detailed survey of lichens and lichenicolous fungi in western Portugal on a small rocky hill (São Bartolomeu) led to the discovery of several new records for the country (van den Boom 2006). One fertile sorediate specimen was identifiable only to genus level. Its hyaline, halonate ascospores, which become brown with age, in combination with immersed to semi-immersed, \pm roundish apothecia and richly branched and anastomosed paraphysoids, indicated that it belonged to the recently described Llimonaea Torrente & Egea, a genus easily mistaken for Lecanographa (Torrente & Egea 1991). According to Egea & Torrente (1994). Llimonaea shows affinities with the family Roccellaceae, however the genus Llimonaea is currently placed in the Arthoniales incertae sedis (Eriksson 2006). According to the key to the species of Llimonaea (Sparrius 2004) there are four known species, all of which are non-sorediate. Following the discovery of the fertile material from Portugal, several additional specimens were located in the herbarium of the second

Material and Methods

This study is based on lichen specimens collected recently by A. M. Brand and P. P. G. van den Boom and herbarium material from E and MUB. Microscopic measurements of ascospores, hyphae, algae, etc. were made in water at × 400 or × 1000 magnification. Amyloid reactions were examined using Lugol's iodine solution (K/I). The secondary metabolites present in most specimens were identified by TLC (Culberson & Ammann 1979; Culberson & Johnson 1982; Elix & Ernst-Russell 1993) and/or by HPLC (Elix et al. 2003).

Additional comparative specimens examined. Llimonaea occulta Egea & Torrente. **Portugal:** Estremadura: Sintra, Cabo da Roca, 20–140 m, 1987, Egea 13619 (MUB, hb. v.d. Boom).

Llimonaea californica (Tuck.) Sparrius. **USA:** California: Monterey, Pacific Grove, Ocean View Blvd., coastal area from Berwick Park to Lovers Point, granite outcrops and Cupressus trees, on C. macrocarpa, 10 m, 2002, P. & B. van den Boom 29276 (hb. v.d. Boom); San Luis Obispo, S of Morro Bay, State Park road, N of Museum of Natural History, mature trees at beach side, on Cupressus macrocarpa, c. 2 m, 2002, P. & B. van den Boom 29477 (hb. v.d. Boom).

author. These specimens were collected in recent years from several coastal localities in western Europe, including Ireland, Brittany (France), north-western Spain and western Portugal. Additional material from southern England which had the unpublished name *Dirina 'zonata'*, was sent to us. One sterile corticolous species from California (USA) appears to be related to this new taxon.

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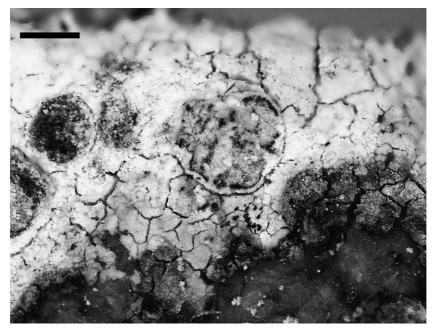


Fig. 1. Llimonaea sorediata, habitus with apothecia (holotype). Scale=0.5 mm.

Taxonomy

Llimonaea sorediata van den Boom, M. Brand & Elix sp. nov.

Thallus saxicola, crustaceus, corticatus, soralia formans, albus, prothallo fusco vel nigrocinctus, areolatus, areolis angulatis, isodiametris 0·1-0·5 mm latis vel elongatis magnitudine ad 0·15 × 0·4 mm; soralia punctiformia vel confluentia, farinosa; algae ad Trentepohlias pertintes. Ascomata immersa vel subimmersa, dendritica, 1 mm diam. vel ellipsoidae $0.8-1.2 \times 0.4-$ 0.7 mm, marginibus thallinis instructa, disco nigro, pruinoso. Paraphysoides ramosae et anastomosantes, $1 \cdot 5 - 2 \cdot 5 \; \mu m$ crassae. Asci cylindrici vel subclavati, $50\text{--}80\times10\text{--}20~\mu\text{m},~\text{octospori},~\text{ascosporis}~\text{elongate}$ fusiformibus $35-50 \times 4.5-5.5(-6) \mu m$ (sine perispora), 8-14-septatis, septis incrassatis, primum hyalinis, demum brunneis, perispora gelatinoso. Pycnidia non visa. Thallus atranorinum, erythrinum, isoerthyrinum et acida lecanorinum et placodiolicum, continens.

Typus: Portugal, Estremadura, E of Nazaré, São Bartolomeu, on top of small rocky hill, on E exposed overhanging outcrops, 9°03.7′W 39°35.5′N, 150 m, 15 July 2001, *P. & B. van den Boom* 27645 (LG—holotypus; hb. v.d. Boom—isotypus).

(Figs 1 & 2)

Thallus crustaceous, forming delimited circular patches up to 3.5 cm wide, areolate

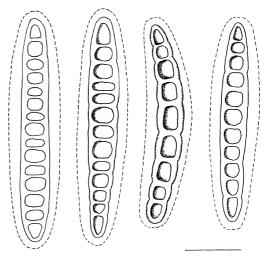


Fig. 2. Llimonaea sorediata, ascospores (holotype). Scale= $10 \mu m$.

to rimose areolate; areoles angular, 0.1–0.5 mm wide, or elongate and up to 0.15×0.4 mm, somewhat radiating, rather thin, up to 0.25(-0.3) mm thick, sorediate; *upper surface* smooth to slightly uneven, pale cream, to creamy white, sorediate parts pure

white, slightly brightish pink when fresh. Soralia initially punctiform and discrete, developing from the edge or on top of the areoles, soon becoming confluent, forming a farinose thallus in the centre; soredia 25–35 μm diam., containing 1 or a few algal cells of c. 12(-15) µm diam. and loose, thin, intricate hyphae. Cortex not well developed, with intricate hyaline hyphae; epinecral layer not developed; surface layer c. 15 μ m thick, without algae, hyaline, except at the top where the hyphae are pale brownish pigmented, homogeneously filled with many crystals (erythrin) which are also abundantly present in the soredia. Medulla mostly completely filled with algae and intricate, elongate, smooth, $2.5-3.5 \mu m$ thick hyphae; Photobiont Trentepohlia, cells roundish, up to 12(-15) µm diam. or ellipsoid and up to $20 \times 12 \,\mu\text{m}$. Prothallus nearly always present, conspicuous, visible as a thick, dark brown to blackish rim around the thallus.

Apothecia immersed to semi-immersed, roundish and up to 1 mm diam. or ellipsoid and $0.8-1.2 \times 0.4-0.7$ mm. Thalline margin level with disc, concolorous with the thallus, thin, 0.03-0.07 mm wide, often flexuose and deformed, soon excluded, inspersed with crystals. Disc plane to often uneven, dark brown to black, whitish pruinose. Proper exciple up to 80 µm wide laterally, continuous with the apothecial base, dark brown to blackish, hyphae with strongly gelatinized walls, inner part brownish-olive pigmented. Epihymenium mid-brown and slightly olive pigmented in the lower part, K+ greenish intensifying, abundantly inspersed with hyaline crystals. Hymenium hyaline, 110–140 um. Hypothecium dark brown, up to 25 µm high, continuous with the excipulum. Paraphysoids richly branched and anastomosing, $1.5-2.0 \,\mu m$ wide, apical cells not widened nor pigmented. Asci cylindrical-clavate, fissitunicate, 8-spored, $50-80 \times 10-20 \, \mu m$ of Grumulosa-type (Torrente & Egea 1989). Ascospores elongate-fusiform, rarely slightly curved, 35- $50 \times 4.5 - 5.5(-6) \mu m$ (excluding perispore), 8–14-septate, hyaline when young, mature spores often brownish pigmented (especially the outer wall), thick-walled, with thickened septa and usually with a perispore, up to c. 2 µm thick.

Pycnidia not seen.

Chemistry. Thallus C+ red, K-, KC-, P-. Erythrin (major), isoerythrin (minor), lecanoric acid (trace), placodiolic acid (trace), atranorin (trace), determined by TLC in solvent C & HPLC.

Distribution and ecology. Llimonaea sore-diata is known from five countries along the Atlantic coast in western Europe (Ireland, England, France, Spain and Portugal). In Portugal (type locality) it was not relocated two years later (2003), during a second visit to the same locality by the first author. During a visit to that locality by the second author in 2003, only sterile material of the new species was collected.

The new species is found mainly on vertical to overhanging cliffs in coastal areas, on acidic outcrops in west-exposed situations, and more rarely on rock walls and on one occasion, on a very old *Quercus* tree.

At the type locality, associated species growing close to L. sorediata, included Buellia leptoclinoides (Nyl.) J. Steiner, Dirina massiliensis Durieu & Mont. s.str., D. massiliensis f. sorediata (Müll. Arg.) Tehler, Peterjamesia circumscripta (Taylor) Hawksw. and Rinodina beccariana Bagl. s.lat. In some localities in France, additional associated species were Diploicia canescens (Dicks.) A. Massal., Ramalina lacera (With.) J. R. Laundon and *Roccella phycopsis* (Ach.) Ach. In one French locality the new species was found on a very old Quercus together with Arthonia pruinata (Pers.) Steud. ex A. L. Sm., Cresponea premnea (Ach.) Egea & Torrente, Opegrapha atra Pers., O. varia Pers. and O. viridis (Ach.) Nyl. In a collection of Llimonaea occulta Egea & Torrente from Portugal, Cabo da Roca (Egea 13633), L. sorediata was found together with Lecanographa werneri. This specimen of L. sorediata was also fertile, with three apothecia. Peterjamesia sorediata (Sparrius, P. James & M. A. Allen) D. Hawksw. has also been found as an accompanying species but is easily mistaken for L. sorediata, although the former differs chemically, as it contains only psoromic acid.

Notes. The relationship of the genus Llimonaea with genera such as Dirina, Lecanactis, Lecanographa and Peterjamesia (syn. Sclerophyton) is treated in Torrente & Egea (1991). Llimonaea sorediata can easily be mistaken for Dirina massiliensis f. sorediata. Both taxa have been found growing side by side and forming mosaics in most of their habitats. The thalli of both species are generally sterile and sorediate, initially with punctiform soralia but then becoming completely sorediate. When growing on acidic rocks the *Dirina* even has a somewhat paler brown prothallus (but not on calcareous rock). In contrast to L. sorediata, the thallus of D. massiliensis f. sorediata is not pure white, but pale brownish cream to pale cream, the soralia are not clearly white, and the thallus is generally much thicker (0.6-1.6 mm). The soralia in the *Dirina* are larger, 0.5-1.8 mm wide, level with the thallus, or slightly excavate and irregularly confluent; the soredia are grey to brown and 30–40 µm diam. The areoles of the Dirina are wider, 1.0-2.0 mm wide and more knobbly. Mature apothecia in D. massiliensis f. sorediata have only rarely been observed, but aborted apothecia are not rare. Pycnidia are sometimes present in the Dirina but were not found in L. sorediata (all specimens were carefully checked). The apothecia of L. sorediata resemble those of Lecanographa werneri (Faurel, Ozenda & Schotter) Egea & Torrente, and even the ascospores are somewhat similar. However the thallus of L. werneri is smooth to uneven rugose, not sorediate, thicker and darker cream with a more brownish tinge in comparison to L. sorediata.

So far, two chemical profiles have been reported for the genus *Llimonaea*, erythrin with lecanoric acid (in *L. occulta*) and 'lecanoric/gyrophoric acids' in *L. flexuosa* Egea, Torrente & Mies (Egea *et al.* 1995). Additional metabolites have now been detected including isoerythrin, placodiolic acid and atranorin. There is also a remarkable chemical similarity between *L. sorediata*

and *Dirina massiliensis* f. *sorediata*, as both species contain erythrin, isoerythrin, lecanoric acid and atranorin in very similar proportions.

In south-western USA, a sterile *Llimonaea* specimen has been found at Pacific Grove (Monterey) in a coastal area, on the bark of *Cupressus macrocarpa* where it forms a mosaic with *Lecanographa subdryophila* (Follmann & Vězda) Egea & Torrente. A second specimen from that area has been found associated with *Arthonia pruinata*. Anatomically the American material is identical with the European collections of *L. sorediata*, however their chemistry differs (with lecanoric acid as the major component). This most probably represents an undescribed taxon but further study and fertile material is required.

Specimens of Llimonaea sorediata examined: Ireland: Connemara: 13 km ENE of Clifden, near NW end of Kylemore Lough, on vertical to overhanging schistose rocks, shaded by trees, 9°52.2′W 53°33.6′N, 2000, M. Brand 40648 (hb. Brand).—British Isles: England: V. C. 9 Dorset, Isle of Portland, N of Rufus Castle, Penn's Weare, 30(SY)69.71, on underside of a large boulder, 2005, B. J. Coppins 22103, B. Edwards & V. J. Giavarini(E).-France: Manche: Cotentin, 13 WNW of Cherbourg, Gruchy, Rocher du Castel-Vendon, 1°46.9′W 49°40.9′N, NW-exposed overhanging granite rock, high above sea, 1997, M. Brand 36117 (hb. Brand); ibid., 10 km WNW of Cherbourg, Urville-Nacqueville, Manoir Dur Ecu, 1°45.2'W 49°40.6'N, on very old Ouercus in vard of mansion, 1997, M. Brand 36089 (hb. Brand); ibid., 2 km SE of Fermanville, Carneville, 1°26.6'W 49°40.2'N, on N-exposed granite of wall of house, 1997, M. Brand 36050 (hb. Brand); ibid., 10 km ENE of Cherbourg, Pointe du Brick, 1°29.5'W 49°40.2'N, on NEexposed vertical and overhanging granite rock above shore, 1997, M. Brand 36072 (hb. Brand). Seine-Maritime: 2 km SE of St. Valery-en-Caux, Manneville, 0°45′W 49°51′N, church in village, on brick, 1999, M. Brand 37795 (hb. Brand); 10 km S of Granvilles, Carolles, N of Cabane Vauban, 1°34'W 48°44'N, on steep W-exposed granite above sea, 1996, M. Brand 33977 (hb. Brand). Finistère: 6 km W of St Pol de Leon, Sibiril, Manoir de Kérouzère, 4°3.8′W 48°40.2′N, on Quercus in forest at N edge, 1997, M. Brand 36341(hb. Brand); Crozon Peninsula, Roscanvel, headland 0.3 km N of Anse de Fraternité, N-exposed slope, on overhanging quartzitic rock, 1997, M. Brand 36598b (hb. Brand); ibid., Pointe de Penhir, 4°36.9'W 48°15.5'N, on quartzite rock, overhang on E slope, 1978, M. Brand 30575 (hb. Brand); ibid., Pt. de Dinan, Château de Dinan, 4°34.2′W 48°14.1′N, 1996, M. Brand 34705 (hb. Brand); N of Pointe du Raz, Pointe du Van (N

side), 4°42.6'W 48°03.8'N, 1997, M. Brand 36867 (hb. Brand); 10 km SE of Concarneau, Pointe de Trévignon, 3°51.2'W 47°47.4'N, on coarse granite on W-exposed overhanging coastal rock, 1999, M. Brand, 38585 (hb. Brand). Côtes Armor: Perros-Guirec, N-coast, W of Sphinx, 3°26.5' W 48°49.1' N, 1996, M. Brand 34186 (hb. Brand); 16 km W of st. Malo, Pointe de St. Cast, 0.5 km E of P. de la Corbière, 2°14.9'W 48°38.6'N, migmatite of low N-exposed overhanging rock, 1999, M. Brand 38857 (hb. Brand); 2 km SSE of Concarneau, coastal area 1 km SE of Pointe de Cabellou, 3°54.6'W 47°51'N, coastal slope, underside of large block of granite, 1999, M. Brand 38597 (hb. Brand).—Spain: Cantabria: Pechon, mouth of Rio Deva, overhanging sandstone rocks at shore, 2003, M. Brand 48762b (hb. Brand). Lugo, Faro de Roncadoiro, E side, NE vertical steep rock face, 2003, M. Brand 49111b (hb. Brand). Galicia: c. 48 km W of La Coruña, 0.5 km NE of Punta Roncudo, granite on gentle NE slope above sea, vertical side of N-exposed standing rocks, 2003, *M. Brand* 49232b (hb. Brand).— **Portugal:** *Estremadura*: Sintra, Cabo da Rocha, on volcanic rock, 20–140 m, 1987, *J. Egea* 13633b (MUB, hb. v.d. Boom); *ibid.*, S side of cape, vertical sides of large blocks of rock, on ridge, 30 ix 2003, *M. Brand* s.n. (hb. Brand); E of Nazaré, São Bartolomeu, on top of small rocky hill, on E-exposed coarse overhanging granite, 9°03.7′W 39°35.5′N, 150 m, 2001, *P. & B. van den Boom* 26968, 26971 (hb. v.d. Boom); *ibid.*, 2003, *M. Brand* 49642 (hb. Brand).

Specimens of Llimonaea sp. examined. USA: California: Monterey, Pacific Grove, S of Ocean View Blvd., near lighthouse, coastal area with scattered Cupressus macrocarpa trees, on C. macrocarpa, 2002, P. & B. van den Boom 29333 (hb. v.d. Boom); ibid., Ocean View Blvd. coastal area from Berwick Park to Lovers Point, granite outcrops and scattered Cupressus macrocarpa trees, on C. macrocarpa, 2002, P. & B. van den Boom 29567 (hb. v.d. Boom).

Key to the species of Llimonaea

1	Thallus sorediate
2(1)	Thallus containing erythrin (major); apothecia rare, immersed; mostly saxicolous, occasionally corticolous (western Europe)
3(1)	Apothecia lirelliform to dendritic (Cape Verde) \dots L. flexuosa Apothecia roundish, short lirelliform, or forming lecanorine apothecia \dots 4
4(3)	Ascospores 6–10-septate, occasionally submuriform; apothecia lirelliform (southwestern USA to north-western Mexico)
5(4)	Thallus yellow; excipulum 15–25 μm wide laterally; saxicolous and corticolous (south-western America)

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