CANTHARELLUS ILICIS SP. NOV., A NEW SPECIES FROM THE MEDITERRANEAN BASIN COLLECTED IN EVERGREEN QUERCUS FORESTS

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ABSTRACT. Cantharellus ilicis sp. nov., a new species from the Mediterranean Basin collected in evergreen Quercus forests. In this article, a new species occurring in Mediterranean Quercus forests, on calcareous soil, is described. C. ilicis is characterized by its bright orange yellow pileus in wet conditions, whitish stipe and hymenophore, strong staining, as well as by the long basidiospores in comparison with other species. Due to the thick-walled pileipellis hyphae, it is accommodated in the Cantharellus sect. Cantharellus. Macroscopic and microscopic detailed descriptions are provided and the new species is compared to closely related species, such as C. ferruginascens and C. lilacinopruinatus. The variability of the pileus colour, the presence of coating and the staining, currently used as diagnostic characters, are discussed. A key is proposed for the species so far known in the Mediterranean Basin.

Key words: Basidiomycota, Cantharellales, taxonomy, ecology, systematics.

RESUMEN. Cantharellus ilicis sp. nov., una nueva especie mediterránea recolectada en bosques perennes de Quercus. En este artículo se describe una nueva especie hallada en bosques mediterráneos de Quercus, en suelo calcáreo. C. ilicis es una especie caracterizada por su píleo amarillo naranja vivo en condiciones húmedas, estípite e himenóforo blanquecinos, fuerte enrojecimiento y esporas largas en comparación con otras especies. Debido a la pileipelis formada por hifas de pared gruesa, C. ilicis se incluye en Cantharellus sección Cantharellus. Se aportan descripciones macroscópicas y microscópicas detalladas, y la nueva especie es comparada con especies próximas, como C. ferruginascens y C. lilacinopruinatus. Se discute la variabilidad de algunos caracteres actualmente usados como diagnósticos en el grupo, tales como el color del píleo, la presencia de pruina y el enrojecimiento. Se propone una clave para la identificación de las especies conocidas en el área mediterránea.

Palabras clave: Basidiomycota, Cantharellales, taxonomía, ecología, sistemática.

INTRODUCTION

Chanterelles, the basidiomata of *Cantharellus cibarius* Fr.: Fr. and related species, are economically important edible fungi commercially harvested from wild populations (DANELL, 1999). Indeed, the basidiomata of *Cantharellus* are not only consumed worldwide, but they are one of the most important marketed wild mushrooms (PILZ *et al.*, 2003). For instance, Sweden sold 50 tons in 1993, the state of Washington sold 126 tons in 1990 (DANELL, 1994) and France more than 1000 tons in 1997 (COURVOISIER, 1997). The economical importance of *Cantharellus* species lies in some of their biological features. The basidiomata of this ectomycorrhizal genus are highly resistant to insect predation, an unusual feature amongst fleshy basidiomata (PILZ *et al.*, 2003). Thus, *Cantharellus* basidiomata are able to maintain a slow growth rate and persist for an average of 44 days, and even, occasionally more than 90 days in the field (LARGENT & SIME, 1995; NORVELL, 1995). Being so long-lived, *Cantharellus* basidiomata can resist long distance

transportation and storage. The presence of bicyclic carotenes, rare amongst the agaricoid fungi (GILL & STEGLICH, 1987), is responsible for the yellow colour of many *Cantharellus* species (ARPIN & FIASSON, 1971).

Despite its economical importance and having been the subject of research, the C. cibarius group is far from being taxonomically well-known. The absence of remarkable morphological differences between the species has probably discouraged some mycologists from describing new species. Thus, species with similar morphology were historically lumped under the binomen C. cibarius (EYSSARTIER & BUYCK, 2000; GUEVARA et al., 2004; DUNHAM et al., 2003). However, in recent decades, a different species-concept has begun to be adopted and new species have been described based on the colour, ecology and staining of the basidiomata (PILAT, 1959; ORTON, 1969; CORNER, 1966). More recent research, supported by molecular data, demonstrated that slight morphological differences can mask different species of Cantharellus (FEIBELMAN et al., 1996; DUNHAM et al., 2003). In contrast, REDHEAD et al. (1997) proved for Cantharellus formosus Corner that some of the characters currently used in taxonomy vary on account of the microhabitat and meteorological conditions, such as the scale formation, the staining or the colour. The latter feature is thought to be influenced by the light, since basidiomata not exposed to light are usually pale (PILZ et al., 2003; personal observations). In addition, other environmental factors such as carbon and nitrogen sources, pH and temperature might alter the expression of colour (DUNHAM et al., 2003). The phenotypical variability of a few species is well known, and interpreting many names remains difficult.

After the attempt of ROMAGNESI (1995) to key out the genus *Cantharellus* in Europe, EYSSARTIER & BUYCK's (2000) commented compilation of the European taxa is noteworthy. This paper originated a new interest in the taxonomy of the genus in Europe, and characters such as the pileus overlying coating (called pruine), staining of the basidiomata and the wall thickness of the pileipellis hyphae were proposed to possess high taxonomical significance. A few species of *Cantharellus*, however, have still been described from the Mediterranean area (MALENÇON & BERTAULT, 1975; HERMITTE *et al.*, 2005), where, being less prospected than other biogeographical regions, many new species are being currently described (MORENO *et al.*, 2000; MOREAU *et al.*, 2005).

During the revision of the genus *Cantharellus* in the Iberian Peninsula, a species not matching any other described species was encountered in several localities. Due to its unique combination of morphological characteristics, *C. ilicis* is proposed as a new species in this article.

MATERIAL AND METHODS

Herbaria are abbreviated according to HOLMGREN and HOLMGREN (1998). The collections examined are deposited in ARAN and BIO herbaria. The macroscopic descriptions are based on fresh material. Pileus diameter refers to the "projected diameter" described by JOSSERAND (1952). Fresh colours were standardized following THE ROYAL HORTICULTURAL SOCIETY (1995) and exsiccatum colours according to MUNSELL COLOR CORPORATION (1990). Dried specimens were used for the microscopic study. Fine sections were made using a razor blade under a stereomicroscope and mounted in Congo Red in KOH 5%. Microscopic studies were carried out under a light microscope Nikon Labophot. Measurements were made at 1250 × magnification. Measurements of the basidiospores exclude the apiculus. Letter abbreviations describing basidiospore size are: $L_m =$ mean length, $W_m =$ mean width and $Q_m = L_m / W_m$; 25 basidiospores were measured per collection. Basidia measurements exclude the sterigmata. Wall thickness and diameter of the pileipellis hyphae were measured in the terminal elements. The author names are abbreviated according to KIRK and ANSELL (1992, 2003).

DESCRIPTION

Cantharellus ilicis Olariaga & Salcedo sp. nov.

Figs. 1a to 1d, Photo. 1, 2.

Pileus 28-90 mm, convexus et umbilicatus, vivido luteo-aurantio colore, ad marginem saepe pallidiore, in siccitate luteo-ochraceo. Stipes $20-50 \times 5-15$ mm, primo albido aurantius pallidus, deinde ochraceo-aurantiacus, vulneratus luteus vel ferruginascens. Lamellae furcatae vel anastomosantes, ad initium albidae, post pallidae ochraceo-aurantiacae. Caro albida, sub cuticula aurantiaca, insipida, demum leviter acris, odor debilis. Sporae in cumulo pallida luteo-ochracea. Basidiosporae ellipsoideae, $(8,4)-9-10,5-(11,6)\times(4,2)-4,8-5,8-(6,4)$ µm. Basidia 4-6 sporigera. Hyphae cuticulae fibulatae, 4,5-7 µm latae, tunica incrassata. A Cantharellus ferruginascens differt pileo saturate aurantiaco et habitatione sub Quercus ilicis. A Cantharellus lilacinopruinatus differt pileo a colore lilacino et pruina albida destituto.

Holotypus: BIO-Fungi 11689, Hispania, Zaragoza, Arroyo de la Tejera, sub Quercus rotundifolia, Quercus humili et Pinus halepensis, ad solum calcareum, 11 November 2006. Isotypi: BIO-Fungi 12600, BIO-Fungi 12601.

Etymology: *ilicis*, Latin, referring to the habitat under evergreen oaks (*Quercus ilex* and *Q. rotundifolia*).

Basidiomata medium to large sized, growing isolate or sometimes fasciculate (Photo, 1, 2). Pileus 28-90 mm diam., initially convex, expanded and depressed in the centre, finally shallowly infundibuliform; surface initially finely velvety, remaining so in the involute margin, brighter afterwards, slightly but clearly hygrophanous, completely devoid of overlying coating. Under wet conditions, bright orange yellow (17A, 21A to 21C, 23A), seldom lighter orange yellow (14A, 15A), sometimes fading towards the margin (23B, 21B, 21C, 17C), whitish orange (19C, 20D) in unexposed zones and often at the margin of small basidiomata; in dry conditions egg-yellow (19A, 20B) to yellowish ochre (19B, 19C, 20C); few light purple patches (181C, 182C), rarely present on the surface; with scales seldom present in the centre, adpressed, concentric, non-contrasting to slightly brownish (165C, 165D); staining inconspicuous to slight when bruised. Margin initially slightly involute and regular, soon straight and finally sinuous, lobed and revolute. In exsiccatum, light ochre (2.5Y 8/2, 8/3, 8/4), sometimes with ochre brown (10YR 6/6, 6/8) patches. Stipe 20-50 × 5-15 mm, cylindrical, often turbinate, solid, smooth to slightly velutinous, initially white (155B), gradually becoming pale orange ochre (19B to 19D), sometimes with brownish (167B, 168C) maculae or purple patches (181C, 182C); staining conspicuous when bruised, weaker in old or extremely wet basidiomata, quickly bright yellow (8C), turning orange or reddish brown (169B, 169C); in exsiccatum, ferruginous brown (2.5YR 4/6, 4/8; 5YR 5/8), rarely light ochre (2.5Y 8/2). Hymenophore composed of well developed veins, initially forked, anastomosing afterwards, at first ochre white (11D, 158B), gradually becoming pale orange ochre (19B, 19C, 20C), seldom with light purple (181C, 182C) patches. In exsiccatum light ochre (2.5YR 8/3, 8/4) to ochre (10YR 8/6, 7/8). Sporeprint: light yellowish ochre (11B, 12C). Context white (155A), orange (19A, 21D) underneath the surface of the pileus, staining when bruised. Smell C. cibarius-like, weak; taste mild, finally slightly peppery. Macrochemical reactions: context FeSO₄ + reddish grey; context and stipe surface KOH -; pileus surface TL4 + green; context gayac + = slowly blue.

Basidiospores ellipsoidal in side view, rarely narrowly ovoid or broadly cylindrical, seldom constricted; apiculus conspicuous, thin-walled, (8,4)-9-10,5- $(11,6) \times (4,2)$ -4,8-5,8- $(6,4) \mu m$ ($L_m = 9,7$ -10,0; $W_m = 4,9$ -5,6; $Q_m = 1,84$ -1,99). Basidia claviform, mainly 4-6-spored, occasionally 2-3-spored, clamped, 62-114 \times 8,5-10 μm . Pileipellis composed of hyphae forming a plagiotrichoderm, more erect in young basidiomata or near the young margin, cylindrical, thick-walled (1-1,6 μm), hyaline to yellowish, clamped, 4,5-7 μm wide, with cylindrical and blunt ends. Stipitipellis composed of parallel arranged hyphae, cylindrical, thin-walled, hyaline to yellow, clamped, 3-12 μm wide. Hyphae of the context cylindrical, thin-walled, hyaline, clamped, 3-12 μm

wide. Basal mycelium white to yellowish, composed of sometimes slightly fascicled hyphae, cylindrical, thin-walled, 2,4-5 μm wide.

MATERIAL EXAMINED. SPAIN. BALEARIC ISLANDS: Mallorca, Campanet, Finca Son Pons, 31SD9803, 100 m, under *Quercus ilex* on calcareous soil, *leg.* J.C. Salom & J.L. Siquier, 28-9-2005, BIO-Fungi 12025. CÓRDOBA: Santa Maria de Trassierra, Los Boscos, 30SUH3200, 420 m, under *Quercus rotundifolia* on calcareous soil, *leg.* T. Illescas & I. Olariaga, 2-12-2003, BIO-Fungi 10075; ídem, BIO-Fungi 10001. NAVARRA: Arakil, Izurdiaga, 30TWN962506, 500 m, under *Quercus rotundifolia* on calcareous soil, *leg.* P.M. Pasaban, 14-11-2005, ARAN-Fungi 5073001. ZARAGOZA: Sigüés, arroyo de la Tejera, 30TXN521203, 550 m, under *Quercus rotundifolia*, *Q. humilis* and *Pinus halepensis* on calcareous soil, *leg.* I. Olariaga, 11-10-2006, BIO-Fungi 11689 (Holotype); ídem, BIO-Fungi 12600 (Isotype); íbídem, 20-10-2003, BIO-Fungi 9983. Sigüés, Venta Garrica, 30TXN6220, under *Quercus rotundifolia* on calcareous soil, *leg.* I. Olariaga, 20-10-2003, BIO-Fungi 9968.

DISCUSSION

Cantharellus ilicis differs from other taxa by a unique combination of features. Because of its fleshy basidiomata, whose pileipellis hyphae are thick-walled, together with the presence of clamps, C. ilicis belongs to the section Cantharellus, where most European species are included (EYSSARTIER, 2000). At the species level, C. ilicis is macroscopically characterized by its bright orange yellow pileus, devoid of overlying coating and an ochre white hymenophore and stipe in small basidiomata, along with the strong and rapid staining on the stipe. However, the pileus colour of C. ilicis is somewhat variable, due to its weak hygrophanous character. Two specimens collected in the same patch of the same locality exhibited fairly different colours; either yellowish ochre (holotype, dry conditions, Photo. 1) or bright yellow orange colour (BIO-Fungi 9983, wet conditions, Photo. 2). Furthermore, a hydrated and more brightly coloured basidiome can be observed in Photo. 1. Some small or young basidiomata have a narrow whitish pileus margin, as well as some discoloured parts if they have not been exposed to the light. Many basidiomata display a typically obconical stipe. The purple patches over the surface of some basidiomata are considered not to be distinctive, since similar patches have rarely been found in C. ferruginascens P.D. Orton and C. amethysteus Quél., according to our observations. Those patches often appear with age, by contrast with the overlying coating of some species, which is more striking in young basidiomata and progressively disappears. Concerning its ecology, all the localities where C. ilicis was found have in common the presence of evergreen Ouercus trees, with which it seems to be associated. However, the marcescent oak Quercus humilis and Pinus halepensis occur in the type locality and a possible association with them cannot be ruled out. C. ilicis seems to prefer calcareous soil. C. ilicis was previously reported as C. cibarius var. rufescens by PALAZÓN (2001), a not validly published name in accordance with the art. 36.1 and 37.1 (MCNEILL et al., 2006).

In the key given by EYSSARTIER & BUYCK (2000) a total of 17 staining taxa were mentioned in Europe, some of them are worth discussing. Amongst them, *C. ferruginascens* is probably the most allied species, which shares with *C. ilicis* the white stipe, pileus margin and hymenophore, at least in small or young basidiomata. However, it differs from *C. ilicis* in being less fleshy and possessing paler and citrine yellow colour (ORTON, 1969; COURTECUISSE, 1993; EYSSARTIER & BUYCK, 2000). Moreover, some basidiomata of *C. ferruginascens* can display either olive-green (ORTON, 1969) or brownish tones (EYSSARTIER & BUYCK, 2000) on the pileus. According to our experience in the Iberian Peninsula, their ecology and distribution are also different; while *C. ferruginascens* is widespread in the Atlantic area under deciduous trees, *C. ilicis* has only been found in the Mediterranean area under evergreen *Quercus* trees. *C. lilacinopruinatus* Hermitte, Eyssart. & Poumarat is a Mediterranean species described as allied to *C. ferruginascens* (HERMITTE *et al.*, 2005), which is associated with *Quercus*, on calcareous soil. Indeed, it has been found coexisting with *C. ilicis* in a locality of Mallorca (J.C. Salom and J.L. Siquier, pers. com.). The examination of 11 collections and several photographs of *C. lilacinopruinatus* revealed a high macroscopic variability on *C. lilacinopruinatus*. Young basidiomata of *C. lilacinopruinatus*

can possess entirely pink pileus, tones that often remain in the pileus margin or the centre of the pileus. The presence of a white overlying coating is the most constant of the features, in nearly all the collections, albeit sometimes not covering the whole pileus. Occasionally, aged basidiomata of *C. lilacinopruinatus* that have completely lost the whitish coating can be seen, making it difficult to discern it from *C. ilicis*. However, young basidiomata of *C. lilacinopruinatus* are clearly distinguishable from *C. ilicis* on account of their white pileus coating, along with sometimes possessing pink tones. In spite of having collected numerous basidiomata of *C. ilicis* in the type locality and surroundings, we have not observed any basidiomata with the typical pink tones of *C. lilacinopruinatus* there, nor with its highly constant white coating.

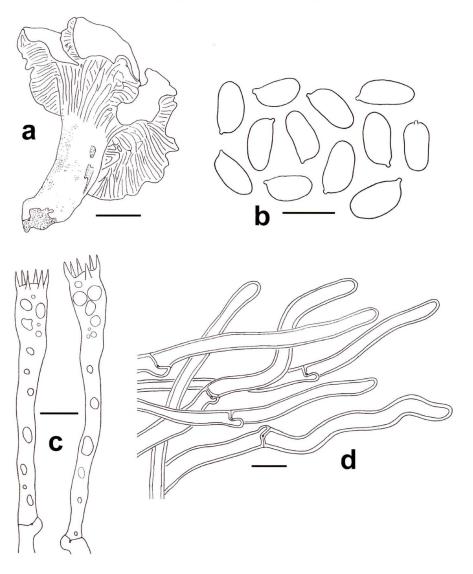


Fig. 1. *C. ilicis* (holotype). **a)** Silhouette of a basidiome (Bar = 10 mm). **b)** Basidiospores. **c)** Basidia. **d)** Pileipellis hyphae. (1b, 1c and 1d, $Bar = 10 \text{ }\mu\text{m}$).

C. rufipes Gillet has been interpreted as a Cantharellus cibarius-resembling taxon, that turns reddish when handled (COOKE, 1890; EYSSARTIER & BUYCK, 2000). It was originally described as a pale yellow fungus that is scaly and flesh-yellow in the centre of the pileus (jaune d'or clair, écailleux et jaune carné au centre) (GILLET, 1878). Slight lilac tones can be noticed in planche 142 of the above-mentioned study (lectotype, EYSSARTIER & BUYCK, 2000), a feature that does not match C. ilicis. C. cibarius subsp. flavipes R. Heim ex Eyssart. & Buyck is another staining taxon. Both the original description and the type designated by EYSSARTIER & BUYCK (2000) describe a fungus with pale pileus and hymenophore with more intense yellow-orange colour, which does not correspond with C. ilicis. Furthermore, the ecology of C. cibarius subsp. flavipes is different from that of C. ilicis, since it has been reported under Fagus and Corylus.

Often, strikingly staining specimens of *Cantharellus* are identified as *C. alborufescens* (Malençon) Papetti & S. Alberti in the Mediterranean area (PAPETTI & ALBERTI, 1998; PÉREZ-DE-GREGORIO, 2003). *C. alborufescens*, originally described from Morocco (MALENÇON & BERTAULT, 1975), is characterized by possessing a white to very pale yellow pileus, strong staining and habitat under evergreen *Quercus*. We were able to examine a colour copy of an original watercolour based on the type collection of *C. alborufescens*. In that drawing, 3 basidiomata are depicted. The older one has a yellow pileus, while the younger ones have their pileus covered by a white coating. In our opinion, due to the presence of this coating, *C. alborufescens* seems more closely related to *C. lilacinopruinatus* than to *C. ilicis*. In any case, *C. alborufescens* is described in the protologue (loc. cit.), as well as by EYSSARTIER & BUYCK (2000), as having longer basidiospores than those of *C. ilicis*.

C. pallens was described on the basis of its pileus with white coating, thick pileus margin and more robust basidiomata than C. cibarius (PILAT, 1959). No information concerning the staining on the stipe was given in the protologue. More recently, C. subpruinosus Eyssart. & Buyck was originally described from France, being characterized by its pileus with white coating and strong staining (EYSSARTIER & BUYCK, 2000). In the latter study, C. subpruinosus was not compared with C. pallens, since it was interpreted as a non-staining taxa. The protologues of the two names do not show any relevant difference, except for the strong staining of C. subpruinosus. According to our observations (36 freshly collected specimens examined), the staining was seen to vary depending on the age and hydration of the basidiomata. In consequence, we consider it that there is no clear limit between C. pallens and C. subpruinosus. In order to make the discussion clearer, we will refer to this species complex as C. pallens below. C. pallens is a widespread species in the Mediterranean area of the Iberian Peninsula. In that area, C. pallens is more common on acid soils, but it can also be found in *Ouercus* evergreen forests on calcareous soil. Young basidiomata of C. pallens are distinguished from C. ilicis in a more sturdy habit, thick and often involute margin and pileus covered with a white coating. Older basidiomata of C. pallens can become bright yellow orange once the coating is lost, but they differ from C. ilicis by often having an orange hymenophore near the margin (usually paler in the rest), i.e. in the youngest parts. Microscopically, both species can be identified by their non-overlapping mean basidiospore size; since the basidiospores are smaller in C. pallens $(7,1-8,9 \times 4,5-5,1 \mu m, 36 \text{ collections})$.

C. cibarius var. bicolor Maire and C. cibarius var. albidus Maire are two other taxa described from the Mediterranean area of the Iberian Peninsula (MAIRE, 1937). The description of the former does not match C. ilicis because of its yellow-orange hymenophore and its white pileus. The later shares with C. ilicis the pale hymenophore. According to EYSSARTIER & BUYCK (2000), no original material of C. cibarius var. bicolor nor var. albidus is known to exist. We visited Viladrau (type locality of C. cibarius var. albidus) and Sils (type locality of C. cibarius var. bicolor) and found that C. pallens is common in both localities. As C. pallens seems to be a very variable species concerning its pileus and hymenophore colour, the descriptions of both var. bicolor and var. albidus macroscopically fit within the variability of C. pallens. The microscopic examination of the basidiomata that we collected has small basidiospores that conform to those of C. pallens.

Outside Europe, some taxa with yellow orange pileus and whitish hymenophore have been described (EYSSARTIER, 2001). In North America, C. cibarius var. pallidifolius A.H. Sm. is the most remarkable species. Originally described as a sturdy variety, it is characterized by its "not hygrophanous, ...buff vellow to egg-vellow" pileus, "pallid" hymenophore when young and strong and readily staining (SMITH, 1968). C. cibarius var. pallidifolius therefore differs from C. ilicis in its paler colour that do not change depending on the hydration. Moreover, the spore-print of C. cibarius var. pallidifolius was described as "ochraceous salmon (flushed pink)", while that of C. ilicis has yellow tones. THIERS (1985) reported a collection from California that matches the original description of Cantharellus cibarius var. pallidifolius, also describing it as having pale vellow to dark vellow pileus and ochraceous to pinkish spore-print. PILZ et al. (2003) discussed an inadequately characterized form from British Columbia that could be related to C. cibarius var. pallidifolius. In agreement with EYSSARTIER (2001), this taxon might be related to C. ferruginascens. However, PETERSEN's (1979) interpretation of C. cibarius var. pallidifolius (as pallidofolius) differs in some aspects from its original description. He depicted a Cantharellus with a bright orange pileus, a very pale hymenophore and a conspicuous staining on the stipe, which macroscopically considerably resembles C. ilicis. Precise data concerning its geographical origin, ecology, basidiospore size and spore-print colour are missing. As explained above, the original description of C. cibarius var. pallidifolius refers, in our opinion, to another species, and therefore that name cannot be used to refer to C. ilicis.

C. subalbidus A.H. Sm. & Morse is another North American species with pale hymenophore and stipe, that stains when bruised. However, it possesses a whitish pileus and smaller basidiospores than C. ilicis, besides being associated with conifers (PILZ et al. 2003). More taxa that resemble C. cibarius have been described (CORNER, 1966; EYSSARTIER, 2001), but none of them exhibits the same combination of characteristics as the species here proposed as new. C. ilicis might be a widespread species throughout the Mediterranean Basin. It is even consumed in some areas (Balearic Islands, Córdoba) where it is regarded as a good edible species.

PROPOSED KEY TO THE SPECIES OF THE CANTHARELLUS SECT. CANTHARELLUS OCCURRING IN THE MEDITERRANEAN AREA

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Cantharellus ilicis. Holotype, BIO-Fungi 11689. Fresh basidiomata.



Cantharellus ilicis. BIO-Fungi 9983. Fresh basidiomata from the same patch as the holotype.

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