NEW RECORDS OF CREPIDOTACEAE (AGARICALES) FROM SEASONAL SEMIDECIDUAL FOREST OF PARANÁ STATE, BRAZIL

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ABSTRACT

Crepidotus crocophyllus and *Simocybe tucumana* were identified in a survey of agaricoid fungi from seasonal semideciduous forests, western Paraná State, South Brazil. *Crepidotus crocophyllus* is new record from Paraná and *Simocybe tucumana* is firstly reported from this ecosystem. Detailed macro- and microscopic descriptions and illustrations are presented for these species, including scanning electron microscopy of the basidiospores of *C. crocophyllus* and a brief discussion on their taxonomy and distribution.

Key world: Agaricomycetes, Atlantic forest, taxonomy

RESUMO

Crepidotus crocophyllus e *Simocybe tucumana* foram identificados em um levantamento de fungos agaricoides em fragmentos de Floresta Estacional Semidecidual no Oeste do Paraná, Sul do Brasil. *Crepidotus crocophyllus* é um novo registro para Paraná, e *Simocybe tucumana* é reportada pela primeira vez para este ecossistema. São fornecidas descrições, ilustrações macroe microscópicas, microscopia eletrônica de varredura dos basidiósporos de *C. crocophyllus* e uma breve discussão taxonômica e de distribuição.

Palavras chave: Agaricomycetes, Floresta Atlântica, taxonomia

INTRODUCTION

Crepidotaceae (S. Imai) Singer includes a heterogeneous group of mostly pleurotoid, but also collybioid and omphalinoid fungi, which produce pale yellow to dark-brown sporeprint from pip-shaped, ellipsoid, or globose basidiospores without germ pore (Singer, 1986). Recent phylogenetic studies placed the genera *Crepidotus* (Fr.) Staude, *Episphaeria* Donk, *Inocybe* (Fr.) Fr., *Neopaxillus* Singer, *Pleuroflammula* Singer, and *Simocybe* P. Karst. in this family, although some of these were previously classified in other families (Aime *et al.*, 2005, Petersen *et al.*, 2010). Currently, species of *Crepidotus*,

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Inocybe, *Neopaxillus*, *Pleuroflammula* and *Simocybe* are known in the Brazilian mycobiota (Maia *et al.*, 2015).

Crepidotus has been fairly studied in Brazil by Singer (1973b, 1989), and recent investigations resulted in new species and records from the states of Rio Grande do Sul (Pereira, 1990), Paraná (Senn-Irlet & de Meijer 1998) and São Paulo (Capelari 2007). *Inocybe* has been reported from all Brazilian regions, and includes both native and exotic species (Sulzbacher *et al.*, 2013, Wartchow, 2020). *Simocybe* is a poorly known genus, with three species reported from Brazil: Sobestiansky (2005) listed *S. cf. tucumana* from Rio Grande do Sul State; de Meijer (2006) listed from Paraná *S. aff. alachuana* (Murrill) Singer, *S. cf. semiglobata* (Murrill) Singer and *S. tucumana* Singer; *Neopaxillus echinospermus* (Speg.) Singer is the only known species of this genus in Brazil and is distributed along the Neotropical zone, including the forests of Paraná State (Silva-Filho *et al.*, 2016). *Pleuroflammula* is represented in the country only by *P. fluminensis* Singer, (Singer 1973a), after Horak (1978) considered a doubtful report due to lack of preserved material. Members of *Episphaeria* have not yet been reported from Brazil.

In a survey of the mycobiota from the seasonal semidecidual forest, western Paraná State (Silva-Filho *et al.*, 2016, Silva-Filho & Cortez, 2017, Silva-Filho *et al.*, 2018a, 2018b, Silva-Filho *et al.*, 2019) members of *Crepidotaceae* were collected, which are reported and described as follows.

MATERIAL AND METHODS

Specimens were collected in two localities: Parque Estadual São Camilo, municipality of Palotina (24°18'34.23"S; 53°54'22.24"W); and Universidade Federal do Paraná, Campus Palotina (24°17'35.96"S; 53°50'30.19"W). These comprise fragments of seasonal semidecidual forest (Atlantic Forest Biome) in the western region of Paraná state, South, Brazil. Morphological macro- and microscopical analysis followed standard procedures for agaricoid fungi (Largent, 1986, Largent et al., 1977). Colour names and codes used in the macroscopic descriptions are from Kornerup & Wanscher (1978). Microscopic features are noted from 3% KOH mounts. Micrographs were taken from an Olympus CX31 optical microscope with a Toup Cam FMA050 digital camera, and the measurements were taken through software Toup tek Toup View. In the basidiospore description, L(W) = length (width) average from a single basidioma, Q is the quotient between length and width, Qm is the median value of Q, n is the number of measured basidiospores/number of analyzed basidiomata/number of collections. Scanning electron micrographs (SEM) were performed at the Center of Electron Microscopy of the Universidade Federal do Paraná (CME/UFPR), Curitiba, under a Jeol JSM-6360LV scanning electron microscope. Specimens were dried in an open air drier (±40°C) and preserved at the Herbarium of the Universidade Federal do Paraná, Setor Palotina (HCP).

RESULTS AND DISCUSSION

Crepidotus crocophyllus (Berk.) Sacc., Sylloge Fungorum 5: 886 (1887)

Figs. 1, 2

■ Agaricus crocophyllus Berk., London Journal of Botany 6: 313 (1847)

Pileus 17–39 mm diam., dimidiate to flabelliform, convex to broadly convex, surface fibrillose to adppressed squamulose in younger specimens, sometimes smooth in older specimens, margin slightly involute, non-striate to slightly translucent striated, pastel yellow (3A4) to dull yellow (3B4) with yellowish brown (5D5) to brown (5EF) scales (Fig. 1a). Context thin, up to 2 mm thick at the insertion point, white (1A1), consistency fleshy.

Lamellae subdistant, with three-sized lamellulae, margin smooth to slightly serrate, concolor with the sides, yellowish white (1A2) to orange grey (5B2) (Fig. 1b). Stipe absent. Mycelial pad sometimes present, white (1A1). Spore print not obtained.

Basidiospores 5–6 (-7) × (4.5–) 5–6 (-6.5) μ m, L= 5.4, W= 5.2, Q= 1–1.05(-1.08), Qm= 1.02, n= 60/3/3, globose to subglobose, spinulose-vertuculose, slightly thick-walled, reddish brown to orange (Fig. 1c), under SEM the ornamentation is composed of cylindrical spines with flattened tips and rare acute tips (Fig. 2). Basidia 19-32 × 6.5-8 µm, clavate, tetrasporic, hyaline, some with granular content. Pleurocystidia absent. Cheilocystidia 36–62 (-78) \times 4.5–7 (-8) μ m, cylindrical to cylindrical clavate, flexuous, less commonly catenulate, clamped at base, thin-walled, hyaline, sometimes vellowish (Fig. 1d), numerous, crowded at gill edge. Lamellae trama composed of parallel hyphae, 3-5.5 µm diam., thin-walled, smooth and hyaline. Lamellae edge fertile, with scattered basidia. Pileipellis a cutis, composed of interwoven hyphae, 6-16 µm diam., thin-walled, sometimes slightly thickened walls (up to 0.3 µm thick), brownish pigment encrusted wall, sometimes smooth, content hyaline to slightly brownish; terminal hyphae cylindrical 52-56.5 \times 6–9 µm, thin-walled (up to 0.3 µm thick), smooth to slightly encrusted, brownish (Fig. 1e-g). Pileus trama composed of spaced and interwoven hyphae, 5-18 µm diam., smooth and hyaline. Clamp connection present in all examined tissues. Oleiferous (thrombopleurous) hyphae absent.

Examined specimens: BRAZIL, PARANÁ: Palotina, P.E. São Camilo, 30.IX.2010, *A.J. Ferreira & R.L.Dias 8-6* (HCP 365); 24.VI.2015, *A.G.S. Silva-Filho 792* (HCP 1155); Campus UFPR, 10.XI.2015, *C. Bottke 04* (HCP 1156).

Habitat and distribution: Solitary on dead wood, in the forest. Known from Asia, Europe and America (Singer, 1973b, Ripková *et al.*, 2005, Bandala *et al.*, 2008, Kasuya *et al.* 2014). In Brazil, reported from Rio Grande do Sul (Rick, 1961, Pereira, 1990) and Paraná States.

Notes: Crepidotus crocophyllus belongs to Sect. Echinosporae Pilát, Subsect. Porpophorini Singer due its non-homogeneous basidiospore wall, presence of clamp connections and brown encrusted pileipellis hyphae (Singer 1973b). It is recognized in the field by yellowish pileus (and lamellae) with brown-pigmented fibrils or scales (Bandala et al., 2008). Microscopically, the following features are diagnostic: globose to subglobose basidiospores with spinulose-verruculose ornamentation, clavate or subclavate cheilocystidia and a pileipellis forming a cutis, composed of hyphae with yellow to brown pigments, sometimes thick-walled and encrusted (Bandala et al., 2008). Our sample agrees with the diagnostic features cited by Bandala et al. (2008) and produce predominantly cylindrical cheilocystidia, feature reported by Singer (1973b) in North American and Argentinean samples. In Brazil, the collections reported by Rick (1961) and Pereira (1990) from Rio Grande do Sul have pileus measuring 10-30 mm, with brownish scales, globose basidiospores measuring 4.5-7 µm, with spinulose ornamentation, pileipellis composed of brown and hyaline hyphae, sometimes encrusted. The cheilocystidia, wich measure $32-53 \times 5-10$ (12) µm, differ in the clavate, cylindricventricose shape sometimes slightly capitate (Pereira, 1990).

Crepidotus crocophyllus and its synonyms, *C. nephrodes* (Berk. & M.A. Curtis) Sacc. (Ripková et al., 2005), *C. appalachianensis* Hesler & A.H. Sm., *C. aureifolius* Hesler & A.H. Sm., *C. distortus* Hesler & A.H. Sm., *C. subaureifolius* Hesler & A.H. Sm., *and C. subnidulans* Hesler & A.H. Sm. (Bandala *et al.*, 2008) probably form a complex of species, composed by similar basidiomata with small morphological discrepancies, possibly related to different environmental conditions, such as vegetation (substrate type) and light incidence. Compared to the samples reviewed by Bandala *et al.* (2008) our sample is

morphologically closer to holotype (H. Forstinger 51492) due its cheilocystidia size (34–55 × 5–8 μ m), absence of sterile bodies on lamellar side, and the presence of a finely encrusting pigment on pileipellis hyphae. It is expected molecular studies with an integrative approach, comprising materials from all continents where the fungus has been reported, clarifying the degree of morphological variation and provides a more reliable specific delimitation of this species.

Crepidotus crocophyllus has a wide distribution in Europe (Senn-Irlet, 1995), North America (Hesler & Smith 1965), Neotropical America (Horak, 1964, Singer, 1973b, Bandala *et al.*, 2008) and Japan (Kasuya *et al.*, 2014). In Brazil, *C. crocophyllus* was reported from Rio Grande do Sul State in seasonal semidecidual forest and dense ombrophilous forests (Rick, 1961, Pereira, 1990). This is the first record from the Paraná State.

Simocybe tucumana Singer, Sydowia 15(1-6): 72 (1962)

Fig. 3

Simocybe junquillea var. *tucumana* (Singer) Raithelh., Nueva flora micológica Argentina: 289 (2004)

Pileus 3–22 mm, convex at first and broadly convex to plane when mature, velutinous at first becoming smooth when mature, margin conspicuously striate, then sulcate when mature, straight to slightly decurved, yellowish brown (5D5), light brown (5D8) and pale yellow (4A3) at the centre (Fig. 3a). Context thin (1.5–2 mm thick), pale grey (1B1), consistency fleshy. Lamellae adnexed to sinuate adnexed, close to subdistant, with 3 lamellulae, margin even to slightly crenate, concolor with the sides, yellowish brown (5D5–5E7) (Fig. 3b). Stipe $4–7 \times 1.5–3$ mm, eccentric, cylindrical, equal, pale orange (5A35), light orange (5A4), surface velutinous, fistulose, inserted base with white mycelial pad (Figs. 3a,b). Spore print brown (5E8).

Basidiospores 5.5-7 × 4-5.5 µm, L= 6.1, W= 4.4, Q= 1.22-1.51, Qm= 1.39, n= 40/4/2, ovoid, oblong to short ellipsoid, smooth, moderately thick-walled, light orange to reddish brown (Fig. 3c). Basidia 17–23.5(–27) × 5–6.5 µm, tetrasporic, hyaline. Pleurocystidia absent. Cheilocystidia 19-42 × 6-14.5 µm, mostly clavate, but also cylindro-clavate, lageniform or capitate, sometimes catenulate, clamped at base, thinwalled, hyaline, numerous in the gill edge (Fig. 3d). Lamellae trama composed of parallel and inflated hyphae, 7-15 µm diam., thin-walled, smooth and hyaline. Lamellae edge sterile. Pileipellis a cutis composed of interwoven hyphae, 2.5-11 µm diam., thin-walled, sometimes slightly thickened walls (up to 0.3 µm thick), smooth, sometimes brownish pigment encrusted wall (Fig. 3f). Pileocystidia 21.5-36 × 8.5-14 µm, short clavate to almost sphaeropedunculate, scattered or in small groups, forming fascicles, thin-walled, hyaline to pale brown (Fig. 3f). Pileus trama composed of interwoven hyphae, 5-11 µm diam., anticlinal, thin-walled smooth and hyaline. Stipitipellis a cutis composed of parallel hyphae, 2–5 µm diam., thin-walled, smooth, hyaline. Stipititrama composed of parallel hyphae, 4.8-7.5 µm diam., thin-walled, smooth and hyaline. Caulocystidia (11-)19-49.5(-57) × 8–12.5(–15.5) µm, cylindrical to clavate, sometimes catenulate, numerous, clamped at the base, thin-walled, hyaline to pale brown (Fig. 3e). Clamp connections present in all examined tissues. Oleiferous (thrombopleurous) hyphae absent.

Examined specimens: Brazil, Paraná: Palotina, UFPR, 11.XI.2015, *C. Bottke* 04 (HCP); 21.VI.2016, *C. Bottke* 23 (HCP 1159).

Habitat and distribution: In the board of forest trail, in seasonal semidecidal forest, on rotting wood of dicotyledonous and on *Merremia* sp. Known only from Argentina (Singer , 1973b) and Brazil (Sobestiansky, 2005, de Meijer, 2006).

Notes: *Simocybe tucumana* is morphologically characterized by its brownish yellow pileus, measuring up to 30 mm diam., eccentric stipe, ovoid to short ellipsoid smooth basidiospores, clavate cheilocystidia, pileocystidia and caulocystidia, and a pileipellis of cutis type with slightly encrusted hyphae (Singer, 1973b). All these morphological features support the identification of our samples that were found growing on the rotting wood of dicotyledonous tree and also on the rotting stems of the vine *Merremia* sp. (*Convolvulaceae*).

Simocybe tucumana was originally described by Singer (1962) in region of Yungas, in Tucuman, Argentina. However, the holotype of *S. tucumana* is currently lost (Perera *et al.*, 2017). Some several decades later, it was reported from South of Brazil by Sobestiansky (2005) in mixed ombrophilous forest of Rio Grande do Sul State and by de Meijer (2006), in the same ecosystem, but further north, in the Paraná State. This is the first report describing Brazilian collections and from seasonal semideciduous forest and the first paper to provide detailed photos of basidiomata and microstructures of the species.

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Figure 1 *Crepidotus crocophyllus* (*A.G.S. Silva-Filho* 792): **a-b** Basidiomata **c**. basidiospores **d**. cheilocystidia **e**. Pileipellis **f**. terminal hyphae of pileipellis. **g**. encrusted hyphae of Pileipe. Scale bar = **a-b** 10 mm, **c-g** 10 μm.



Figure 2 SEM of basidiospores: a,b. Crepidotus crocophyllus (A.G.S. Silva-Filho 742). Scale bar = 2 µm.



Figure 3 Simocybe tucumana (C. Bottke 04): **a-b** Basidiomata **c**. basidiospores **d**. cheilocystidia **e**. caulocystidia **f**. pileipellis, the arrow show a pileocystidium. Scale bar = **a-b** 10 mm, **c-f** 10 μ m.