
***Tylopilus temucensis* sect. *Oxydabiles* (Fungi, Basidiomycota, Boletaceae), new species and first record of the genus from Southamerican *Nothofagus* forest**

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Tylopilus temucensis, sect. *Oxydabiles* (Fungi, Basidiomycota, Boletaceae) is described as a new species and represents the first record for the genus from South American *Nothofagus* forests. Its systematic context and possible reasons for the low number of known boletaceous species in the Chilean *Nothofagus* area are discussed. A key for Chilean *Boletaceae*, including *T. temucensis*, is provided.

Key words: *Boletales*, Chile, Southern Beech, temperate forest, *Tylopilus*.

Introduction

Temperate forests of southern Chile between 35° and 55° s.l. are biogeographically isolated and rich in endemic biota (Armesto *et al.*, 1996). The dominant tree genus, *Nothofagus*, is represented with 10 species (Veblen *et al.*, 1996). Being strictly ectomycorrhizal, the andino-patagonian *Nothofagus* spp. are associated with a diverse community of symbiotic macromycetes (Garrido, 1988). Dominating fungal taxa in this southernmost ectotroph area are *Cortinarius* and related genera (Moser and Horak, 1975; Garnica *et al.*, 2003), contrasted by conspicuously few *Russulales* and *Boletales* (Garrido, 1988; Horak, 1979; Horak, 1977). However, considering that larger mycological surveys have been few and geographically limited compared with well-studied areas on the northern hemisphere (Garrido, 1988), the list of recorded species may still be far from complete. In particular, little is known about fungal species richness of the lowland Mediterranean *Nothofagus* forests in the central depression between 36° 30' and 41° s.l. which are now reduced to a few small and scattered relicts after extensive deforestation and subsequent

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plantation of introduced *Pinus* and *Eucalyptus* spp. or transformation in agricultural land during the last 150 years (Donoso, 1994). Previously, seven autochthonous *Boletaceae* had been reported from lowland *Nothofagus* forests between Chile's VIII. and X. Region (Garrido, 1988; Horak, 1977), six of them belonging to the genus *Boletus* (*B. araucarianus* Garrido, *B. bresinskyanus* Garrido, *B. chilensis* Singer, *B. loyita* Horak, *B. loyo* Phil. ex Speg. and *B. putidus* Horak) and one to *Gastroboletus* (*G. valdivianus* Horak). *Tylopilus temucensis* was discovered in native forest relicts in the central depression of southern central Chile and represents the first record of the genus for the South American *Nothofagus* area.

Materials and methods

Macroscopical and microscopical characteristics of basidiomata were documented by digital colour photographs taken of fresh specimens *in situ* and in the laboratory with a Nikon Coolpix 950 camera. Microscopical features were studied on fresh material mounted in water and documented by colour microphotographs and line drawings at 1000X magnification (bright field, oil immersion objective) on a Leitz Dialux microscope equipped with a camera adapter and a drawing device (camera lucida). Daylight colours of fresh basidiomata and sporeprints were matched as close as possible to colour reference codes from Kornerup and Wanscher (1962). As references for the generic classification, the type descriptions of *Tylopilus* by Horak (1968) and Singer (1986) were studied. For preservation, the specimens were desiccated for 48 hours at 50°C in a ventilated oven (Memmert). Herbarium reference material has been deposited at the fungal collection of the Natural History Museum Santiago, Chile (SGO).

Results

Tylopilus temucensis Palfner, **sp. nov.** (Figs. 1-3)

Etymology: the epithet refers to the collection site near the city of Temuco.

Basidiomata gregaria. *Pileus* 35-130 × 15-35 mm, primo hemisphaericus dein convexus, centro depresso, fuscus vel fuscocastaneus, glabrus, leviter glutinosus, contexto albido, fractu rubescentis, odore inconspicuo, sapore miti. *Hymenophor* tubuliformis, tubis pallide luteis, 6-8 mm altis, poris roseoaurantiis vel roseobrunneis, 1-2(-3) pro mm. *Stipes* 40-85 × 25-45 mm, subventricosus vel latiter fusiformis, siccus, basi ferrugineus, flocculosus, apice luteus, laevis, contexto albido, basi luteolo, fractu rubescentis, mycelio basali albo. *Basidiosporae* (9-)10-13(-15,5) × 3,5-5(-6) µm, ellipsoidae vel fusiformes, fuscae, in cumulo roseo-fuscae. *Basidia* tetrasporigera, (30-)33-43(-45) × 4-8(-10) µm, cylindrica ad clavata. *Cystidia* 23-63(-67) × 4-10(-12) µm, clavata vel lageniformes. *Caulocystidia* clavata, 16-33(-36) × 4-8(-10) µm. *Epicutis* ex hyphis cylindricis vel cystidiiformibus, subcapitatis, fortiter

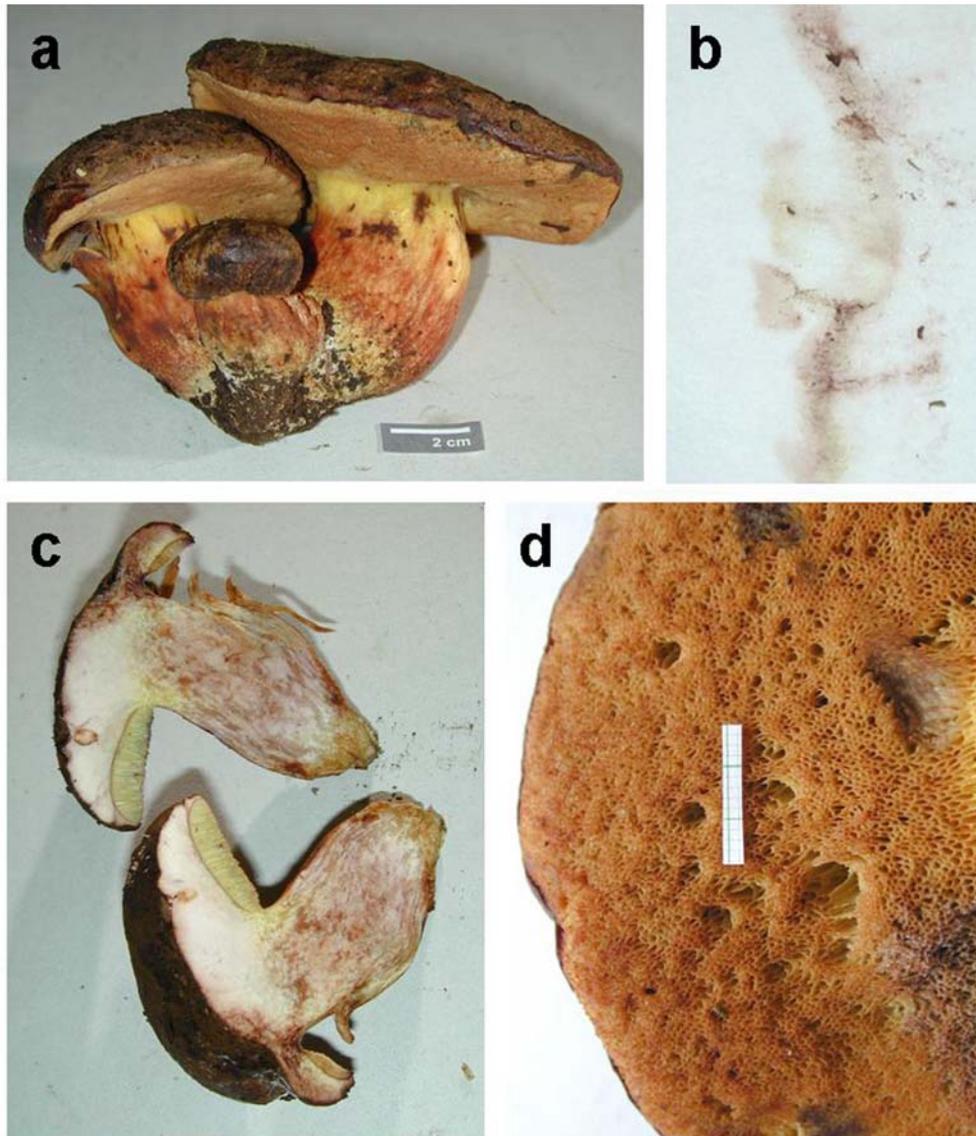


Fig. 1. *Tylopilus temucensis* (from holotype). **a.** Fresh basidiomata. **b.** Sporeprint. **c.** Autoxidation of context of freshly cut basidioma. **d.** Pores in surface view. Scale in mm.

incrustatis, $33-66(-80) \times 5-8(-10) \mu\text{m}$. *Septa* hypharum defibulata. *Habitat* in sylva ad terram et humum sub *Nothofagus obliqua* (Mirb.) Oerst. Chile, Temuco, Chivilcan, Rucamanque, $38^{\circ} 39'$ lat. austr., $72^{\circ} 35'$ long. occid., 17-IV-2004, leg. M.A. Casanova, det. G. Palfner, holotypus sub RU0052 in SGO conservatur.

Basidiomata (Fig. 1a) gregarious. *Pileus* (Figs. 1a, c) 35-130 mm in diam., 15-35 mm thick, convex, hemispherical when young, at maturity expanded, with a central depression, disk dark brown, also with reddish brown

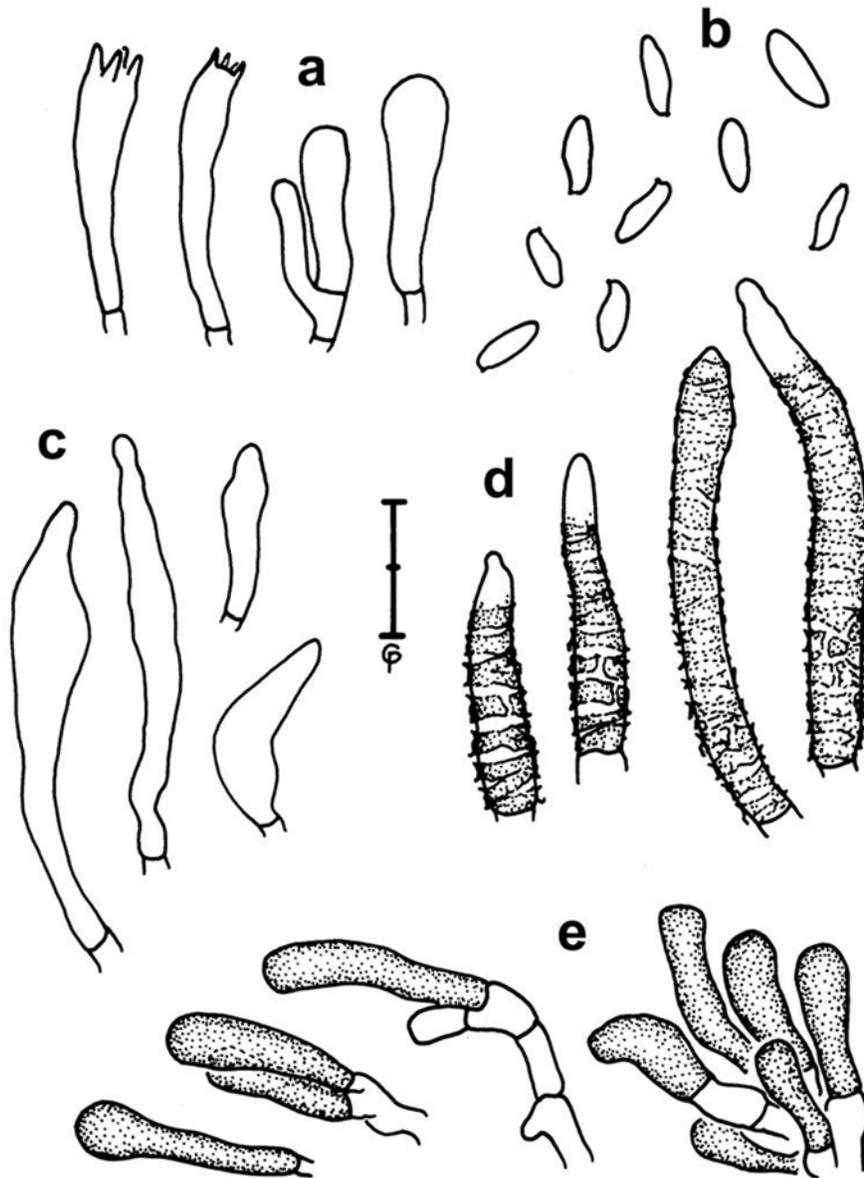


Fig. 2. *Tylopilus temucensis* (from holotype). **a.** Basidia and basidioles. **b.** Basidiospores. **c.** Hymenial cystidia. **d.** incrusted hyphal ends of the epicutis. **e.** Caulocystidia, dotted areas symbolizing the membranaceous pigment. Bar = 20 μ m.

or greyish brown tones (Kornerup and Wanscher: 8E8, 8F8 to 8F4), smooth, slightly glutinous, epicutis turning dark orange to red brown in KOH (5%), context whitish, turning reddish brown (K & W: 8D5 to 8D6) when cut, without idiosyncratic smell, taste mild. *Hymenophore* (Figs. 1c, d) tubulose,



Fig. 3. *Tylophilus temucensis*, microscopical features of type specimen. **a.** Basidiospores. **b.** Heilocystidia. **c.** Pigmented caulocystidia. **d.** Encrusted hyphal ends of epicutis. Bars = 10 μ m.

slightly depressed around stipe; tubes pale yellow (K & W: 1A3, 30A3), not changing when cut or bruised, 6-8 mm long, tube mouths brownish orange with a pinkish tint (K & W: 7B6 to 7B4), roundish, 1-2 (3) per mm. *Stipe* (Figs. 1a, c) 40-85 mm long, 25-45 mm in diam., broadly fusiform to ventricose, base slightly rooting, dry, middle and lower part reddish brown to brick red (K & W: 7B7 to 7D7), flocculose with minute red brown scales (best seen under a dissecting microscope), apex pale yellow (K & W: 3A3, 3A4), with scattered dark brown spots (K & W: 7E7, 7F7), reticulum lacking or very

Key for Chilean *Boletaceae* associated with *Nothofagus* spp.

This key is based on species descriptions by Garrido (1988), Horak (1977) and specimens collected by the author.

1. Basidiomata gastroboletoid, tube layer not or only partially exposed
..... *Gastroboletus valdivianus*
1. Basidiomata boletoid, with fully exposed tube layer 2
2. Stipe distinctly reticulate *Boletus chilensis*
2. Stipe not or very faintly reticulate 3
3. Tube mouths orange brown to pinkish brown, tubes yellow
..... *Tylophilus temucensis*
3. Tube mouths with yellow tones, more or less concolourous with tubes 4
4. Pileus large, up to 400 mm in diam., dark red, stipe bulbous, yellow *Boletus loyo*
4. Pileus smaller, stipe cylindrical, conical or clavate 5
5. Pileus and stipe with brown colours, spores 8-10.5 µm long *Boletus putidus*
5. Pileus and/ or stipe with dark red tones, spores longer 6
6. Mature pileus and stipe deep wine red, context yellow *Boletus loyita*
6. Mature pileus brown, stipe base red 7
7. Pileus large (75-200 mm in diam.), ochraceous brown, stipe apex yellow with red spots ...
..... *Boletus araucarianus*
7. Pileus smaller (47-115 mm in diam.), chestnut brown, stipe apex white.....
..... *Boletus bresinskyanus*

faint in a narrow zone next to the hymenium, interior solid, flesh whitish, yellowish in base, turning reddish when cut (K & W: 8D5, 8D6), single or fused, basal mycelium whitish. *Basidiospores* (Figs. 1b, 2b, 3a) in fresh deposit dark pink to purplish grey (K & W: 10B3 to 10D3, 13B2 to 13C2), under the microscope fuscous, elongated ellipsoid to fusiform, smooth, (9-)10-13(-15.5) × 3,5-5(-6) µm. *Basidia* (Fig. 2a) 4-spored, cylindrical to subclavate, colourless, (30-)33-43(-45) × 4-8(-10) µm. *Cheilo- and Pleurocystidia* (Figs. 2c, 3b) scattered, subclavate to lageniform, 23-63(-67) × 4-10(-12) µm. *Caulocystidia* (Figs. 2e, 3c) abundant, clavate, 16-33(-36) × 4-8(-10) µm, smooth, hyaline towards stipe apex, towards stipe base often with a fuscous to vinaceous brown membranaceous pigment which dissolves quickly in KOH (5%), often in clusters macroscopically seen as fine scales (see stipe). *Epicutis* (Figs. 2d, 3d) a trichoderm, terminal cells often cystidia-like, cylindrical or subcapitate, 33-66(-80) × 5-8(-10) µm, with thick, brown epimembraneous

incrustations typically peeling off in flaky patches or girdle-like bands and quickly dissolving in KOH (5%), hyphal tips often hyaline without incrustations. *Clamp connections* lacking.

Habitat: in *Nothofagus* forest on litter, in autumn.

Known distribution: Central Southern Chile.

Material examined: *Tylopilus temucensis*: holotype collection (three basidiomata) CHILE, IX Región, city of Temuco, sector Chivilcan, Rucamanque forest reserve, 38° 39' s.l., 72° 35' w.l., side of forest road under secondary growth of *Nothofagus obliqua*, 17 April 2004, leg. M.A. Casanova, det. G. Palfner, holotype RU0052 in SGO; other material: three basidiomata, Chile, X. Región, 18 km east of city of Valdivia, side of country road along northern slope of valley of Rio Calle-Calle, secondary forest of *Nothofagus obliqua*, 13 April 1996, leg. G. Palfner, GP3705, herb. G. Palfner; *Boletus loyo*: Chile, X. Región, 20 km east of city of Valdivia, country road along northern slope of valley of Rio Calle-Calle, Fundo “El Junco”, secondary forest of *Nothofagus obliqua*, 5 April 1998, leg. G. Palfner, GP5001, herb. G. Palfner; *Boletus putidus*: Chile, X. Región, 18 km east of city of Valdivia, side of country road along northern slope of valley of Rio Calle-Calle, secondary forest of *Nothofagus obliqua*, 23 March 1996, leg. G. Palfner, GP3701, herb. G. Palfner.

Discussion

The genus *Tylopilus* Karst. is widely distributed in temperate deciduous and coniferous forests of both hemispheres with its main centre of diversity in eastern North America (Bessette *et al.*, 2000). Its ectomycorrhizal status has been proven for the type species *T. felleus* (Bull.) Karst. by Uhl (1988, 1989). According to Singer (1986), distinctive generic features, matched by *T. temucensis*, are the pinkish brown spore print, the small pores, the depressed hymenium around the stipe, the maximum spore length of 15.5 µm, the abundant caulocystidia and the white context; however, *T. temucensis* does not share the reticulate stipe, unchanging context and bitter taste with *T. felleus* (Horak, 1968; Singer, 1986); instead it shows characteristics like the flocculose stipe which lacks a reticulum, the reddish autoxidation, mild taste and ectomycorrhizal association with *Nothofagus* and is therefore placed in sect. *Oxydabiles* (Singer, 1986).

In the field, *T. temucensis* may be confused with the similarly coloured *Boletus putidus* Horak which grows in the same habitat, however both species can be distinguished by the pinkish brown or orange brown pore mouths of the former which in *B. putidus* are concolourous with the pale yellow tubes (Horak, 1977) and by the larger and darker spores of *T. temucensis*. The rather vivid colour of the pore mouths of *T. temucensis*, coming close to some boletes from the *luridi* group, is not typical for most *Tylopili*, nevertheless the reddish autoxidation and the dark pink spore print clearly characterize it as a member of this genus.

The majority of *Tylopilus* species are known from the northern hemisphere, especially North America (Bessette *et al.*, 2000), however in recent years, several new species have been described from andean *Quercus* forests in Costa Rica and Colombia (Amtoft *et al.*, 2002, Halling and Mueller, 2002). According to Halling and Mueller (2002), these Central- and South American taxa may have evolved during migration processes from the North American subcontinent southwards across the Central American isthmus, reaching its natural southern limit with the andine Oak belt.

It is rather surprising that there have been no previous records of *Tylopilus* in the South American *Nothofagus* area, not only because this genus shows considerable diversity in other parts of the same continent, but also considering the gondwanean origin of many Chilean biota and the fact that *Tylopilus* and other boletaceous genera have been reported from the related ectotroph areas of Australia and New Zealand (McKenzie *et al.*, 2000; Grgurinovic and Simpson, 2001; Watling, 2001). Horak (1977) already commented on the discrepancy between the high ecological diversity of temperate forests in Chile and the low number of known native *Boletaceae*; by then, merely five species (*Boletus chilensis*, *B. loyita*, *B. loyo*, *B. putidus*, *Gastroboletus valdivianus*) had been described from the Chilean *Nothofagus* area (Horak, 1977; Mujica *et al.*, 1980), all from the narrow zone defined as lowland temperate rainforest between 37° 45' and 43° 20' s.l. in and around the Valdivian Province (Veblen *et al.*, 1996). Although later records from central Chile and the Andean slopes extended the known range of Chilean Boletoidae (Garrido, 1988), only three new autochthonous species (*Boletus araucarianus*, *B. bresinskyanus*, *T. temucensis*) have been reported since then, raising the total number of taxa to eight. Apart from *Gastroboletus valdivianus*, no other native secotioid or hypogeous *Boletales* have been accounted for.

Possibly, Quaternary glaciation events, approximately between 700 and 13 ka b. p. (Markgraf *et al.*, 1996) which substantially reduced the South American *Nothofagus* forests, may also have caused the extinction of some thermophilous fungal taxa; on the other hand, climatological and geographical barriers, especially the long arid belt on the Pacific side of the Andes ranging from Peru to northern Chile may have impeded species migration from the northern Oak forests to the southern *Nothofagus* area.

It should also be considered that in Chile nowadays only a few scattered and small relicts remain of the Mediterranean lowland forests between regions VIII and X, the distribution centre of endemic *Boletaceae*. The poor presence of boletoid taxa in this zone may thus not only be a result of long-term biogeographical isolation or climatic effects, but also a consequence of the recent and drastic anthropogenic destruction of their habitat.

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