

New Occurrences of Agaricomycetes (Fungi) to the Brazilian Pampa Biome

Novas Ocorrências de Agaricomycetes (Fungi) para o Bioma Pampa Brasileiro

Nuevas Ocurrencias de Agaricomycetes (Hongos) en el Bioma Pampa Brasileño

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Abstract

The study of Agaricomycetes in the Brazilian Pampa Biome is still incipient, but it is very necessary for the valorization and preservation activities in this area. In terms of fungal diversity, it is the least studied Brazilian biome, with only 84 species already described for the site, despite the great diversity of fauna and flora that this biome presents. The objective of this work is to know more about the mycodiversity of this biome, contributing to a better understanding of the distribution and ecology of the Agaricales. The collections were made following the usual methodology for Agaricales (Basidiomycota). Thus, specimens were collected for studies to identification to species level by macro- and microscopic characters. Descriptions of seven species collected in the rural region surrounding the city of São Gabriel, state of Rio Grande do Sul, Brazil are presented, which are cited for the first time for the Pampa Biome, namely: *Macrolepiota mastoidea*, *Protostropharia alcis* ssp. *austrorbrasiliensis*, *Trogia buccinalis*, *Crepidotus euterpicola*, *Entoloma depluens*, *Lepiota subincarnata* and *Neopaxillus echinospermus*. These new citations reinforce the importance of studies on the biodiversity of the Pampa biome for its preservation.

Keywords: Distribution; Species; Agaricales; Southern brazil; Pampa biome.

Resumo

O estudo de Agaricomycetes no Bioma Pampa brasileiro ainda é incipiente, mas muito necessário para a valorização e pelas atividades de preservação nesta área. Em se tratando de diversidade fúngica, é o bioma brasileiro menos estudado apresentando apenas 84 espécies já descritas para o local, mesmo sabendo-se da grande diversidade de fauna e flora que este bioma apresenta. O objetivo deste trabalho é conhecer mais acerca da micodiversidade deste bioma, contribuindo para um melhor entendimento da distribuição e da ecologia dos Agaricales. As coletas foram feitas seguindo-se a metodologia usual para Agaricales (Basidiomycota). Assim, foram reunidos espécimes para estudos macro e microscópicos e identificação ao nível de espécies por estes caracteres. São apresentadas descrições de sete espécies coletadas na região rural circunvizinha à sede do município de São Gabriel, estado do Rio Grande do Sul, Brasil, as quais são citadas pela primeira vez para o Bioma Pampa, a saber: *Macrolepiota mastoidea*, *Protostropharia alcis* ssp. *austrorbrasiliensis*, *Trogia buccinalis*, *Crepidotus euterpicola*, *Entoloma depluens*, *Lepiota subincarnata* e *Neopaxillus echinospermus*. Essas novas citações reforçam a importância dos estudos sobre a biodiversidade do Bioma Pampa para sua preservação.

Palavras-chave: Distribuição; Espécie; Agaricales; Sul do brasil; Bioma pampa.

Resumen

El estudio de Agaricomycetes en el Bioma Pampa Brasileño es aún incipiente, pero muy necesario para las actividades de valorización y preservación en esta area. En términos de diversidad de hongos, es el bioma brasileño

menos estudiado, con solo 84 especies ya descritas para el sitio, a pesar de la gran diversidad de fauna y flora que presenta. El objetivo de este trabajo es conocer más sobre la micodiversidad de este bioma, contribuyendo a una mejor comprensión de la distribución y ecología de los Agaricales. Las colectas se realizaron siguiendo la metodología habitual para Agaricales (Basidiomycota). Así, se colectaron especímenes para estudios macro y microscópicos e identificación a nivel de especie por estos caracteres. Se presentan descripciones de siete especies colectadas en la región rural circundante al municipio de São Gabriel, estado de Rio Grande do Sul, Brasil, que se citan por primera vez para el Bioma Pampeano, a saber: *Macrolepiota mastoidea*, *Protostropharia alcis* ssp. *austrobrasiliensis*, *Trogia buccinalis*, *Crepidotus euterpicola*, *Entoloma depluens*, *Lepiota subincarnata* y *Neopaxillus echinospermus*. Estas nuevas citas refuerzan la importancia de los estudios sobre la biodiversidad del Bioma Pampeano para su preservación.

Palabras clave: Distribución; Especies; Agaricales; Sur de Brasil; Bioma pampeano.

1. Introduction

The Pampa biome, also known as Campos do Sul or Campos Sulinos is shared by three countries, and covers the entire territory of Uruguay, parts of Argentina, and parts of Rio Grande do Sul state in the extreme south of Brazil, where it occupies an area of 176,496 km², and is characterized by being the only Brazilian biome limited to only one state (Boldrini, 2010; IBGE, 2020). In Brazil, the Pampa Biome comprises 63% of the territory of Rio Grande do Sul state, and only 2% of the national territory (IBFloresta, 2020). In 2004, it was officially recognized as a biome, through a partnership between the Instituto Brasileiro de Geografia e Estatística (IBGE) and the Ministério do Meio Ambiente (MMA, 2010).

Perhaps it is the smallest and youngest Brazilian biome. Along, with the Pantanal, it is one of the least studied when it comes to fungal diversity, with only 84 species mentioned, in contrast to the 3,017 species known for the Atlantic Forest, which is surpassed, including by the Caatinga, where about 1,000 species have been recorded (Maia *et al.*, 2015). However, it is known that the natural fields of the Pampa have a great diversity of fauna and flora (Bilenca & Biñarro, 2004). For the Agaricomycetes, which includes parasitic, decomposing and ectomycorrhizal fungi (Hibbett, 2006), there are few studies.

The Agaricales comprise the largest order of the Fungi kingdom with roughly 20,000 species described, according to the Catalogue of Life (Roskov *et al.*, 2019). In this sense, pioneering reviews for Brazil, such as those of Putzke and Putzke (2017; 2019; 2022), introduced to the great mycological diversity of the Pampa biome and demonstrated the presence of several families such as Agaricaceae, Amanitaceae, Bolbitiaceae, Coprinaceae and Tricholomataceae among others.

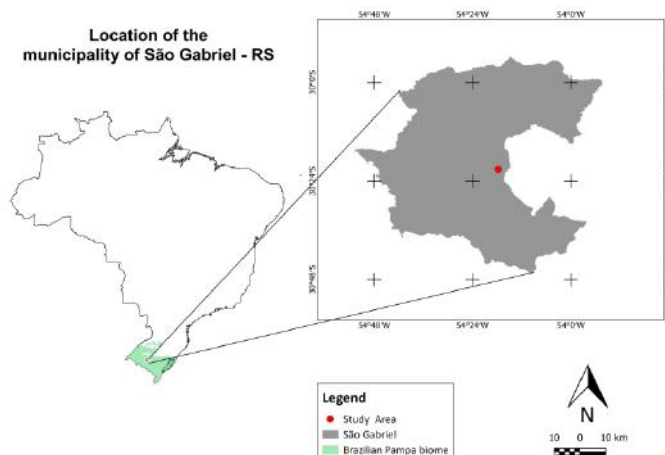
Knowing the diversity of this environment is essential to reinforce protection programs for this biome, especially in the sense of creating conservation areas, which are almost non-existent in this environment. Thus, we describe the occurrence of seven new species for the Pampa biome, contributing to a better understanding of the distribution and ecology of Agaricales.

2. Methodology

The species described here were collected in the municipality of São Gabriel, located in the southwest of the state of Rio Grande do Sul, 320 km from the capital Porto Alegre, with an area of 5,053.460 km² belonging to the Pampa biome, and at an altitude of 118 meters (IBGE, 2020). The region is formed by fields and low hills (São Gabriel, 2014). The climate in the region is humid and temperate subtropical, with well-defined seasons, classifying it as a steppe (IBGE, 2020).

The material was collected during field trips using the walking method, at the Morro do Sabiá location, an private property of about 70 ha, at an approximate distance of 5 km from the municipal center and an altitude of 124 meters (30° 21'20" S and 54°17'42" W) (Fig. 1). The region is composed of native fields, soybean plantations, and swamps that flows into the Vacacaí River. The soil in the area is of the Planossolo Hidromórfico Eutrófico Arênico type (Hasenack & Weber, 2010).

Figure 1: Map of the studied area (red dot), showing Brazil and the São Gabriel municipality.



Source: Authors.

On the map it is possible to observe the location of the collection area within the municipality of São Gabriel, as well as the location of the municipality in the state of Rio Grande do Sul at southern Brazil.

The specimens were collected and stored in plastic pots individually, and after taken for identification via macro and micromorphological analysis (Putzke & Putzke 2017; 2019). These procedures were performed at the Laboratory of Fungal Taxonomy (LATAF) of the Universidade Federal do Pampa (UNIPAMPA) São Gabriel campus, Rio Grande do Sul state. After the analyses, the material was dehydrated in an oven at an average temperature of 40°C to be later deposited in the Herbarium of the Universidade de Santa Cruz do Sul (HCB).

3. Results and Discussion

The study area presented a mycodiversity represented by seven species considered new citations to the Pampa Biome, members of five families, namely: *Macrolepiota mastoidea* (Fr. ex Fr.) Singer and *Lepiota subincarnata* J.E. Lange (both Agaricaceae); *Trogia buccinalis* (Mont.) Pat. (Tricholomataceae); *Crepidotus euterpicola* Senn-Irlet and De Meijer (Crepidotaceae); *Entoloma depluens* (Batsch) Hesler (Entolomataceae); *Neopaxillus echinospermus* (Speg.) Singer (Paxillaceae) and *Protostropharia alcis* (Kytöv.) Redhead, Thorn and Malloch., (Strophariaceae). The description of each species follows.

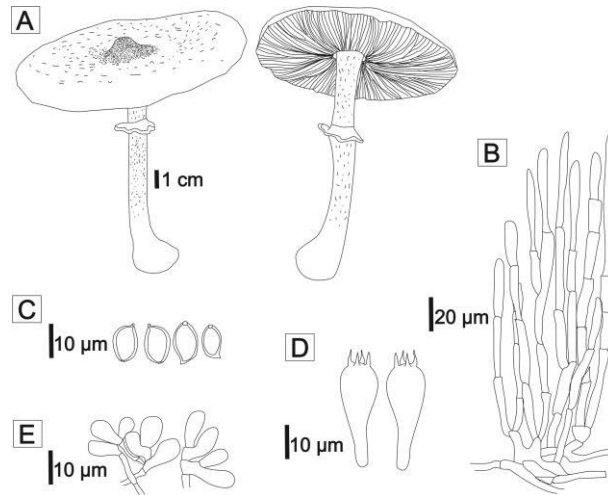
***Macrolepiota mastoidea* (Fr. ex Fr.) Singer, Lilloa 22: 417. 1951 (1949). (Figure 2)**

The species has a pileus surface with small squamules, pale ochraceous to brownish and a simple ring. Pileus 5 – 11 cm in diameter, fleshy, convex or flat-convex when mature. Lamellae free, broad, grayish brown. Stipe central up to 15 cm long, covered by scales and with an enlarged base. Spores 12 – 14 x 9 – 10 µm, ellipsoid to ovoid with thick walls, smooth, hyaline, with germinative pore. Basidia 32 – 44 x 12 – 14 µm, clavate, thin-walled, hyaline, tetrasporic. Pleurocystidia absent. Cheilocystidia 15 – 20 x 7 – 10 µm, clavate, hyaline, thin-walled. The cortical layer of the pileus is formed by a palisade of subcylindrical hyphae, without clamp connections.

They grow solitary in soil. The species is close to *M. excoriata* (Schaeff.) M.M. Moser, but differs in the coverage of the stipe without scales in the latter. GE *et al.* (2010) discuss this species from collections found in China. Cited to Brazil from Minas Gerais (Rosa & Capelari, 2009) and São Paulo states (Grandi *et al.*, 1984) for the Atlantic Forest domains. Alves *et al.*

(2016) cited the species following for the Pampa: *Macrolepiota gracilentata* (Krombh) Wasser, *Macrolepiota fuliginosquarrosa* Malençon, *Macrolepiota procera* (Scop.) Singer, *Chlorophyllum rachodes* (Vittad.) Vellinga and *Chlorophyllum molybdites* (G. Mey.) Masee. This therefore, is a new citation to the Pampa biome.

Figure 2: *Macrolepiota mastoidea* (Fr. ex Fr.) Singer. Basidiome with lamellated hymenophore (A). Elements of the cortical layer of the pileus (B). Basidiospores (C). Basidia (D). Cheilocystidia (E).



Source: Authors.

In the illustration it is possible to observe the macro and microscopic structures of *Macrolepiota mastoidea*, as well as its large basidiome as found in its natural habitat.

The species is also recorded in Australia (Vellinga, 2003), China (Ge *et al.*, 2010), Macedonia (Karadelev *et al.*, 2012), India (Pramanik & Chaudhuri, 2017), Korea (Cho *et al.*, 2019), Cuba (Alimammadova & Aghayeva, 2021) and Turkey (Keski *et al.*, 2022).

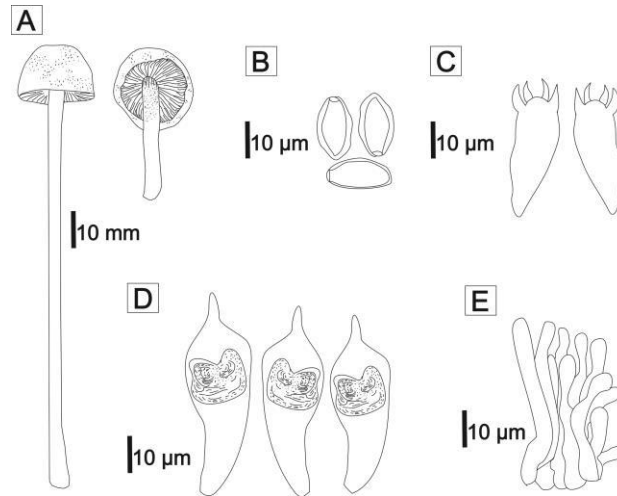
Material examined: Brazil, Rio Grande do Sul, São Gabriel, Morro do Sabiá, M. A. Heberle, 12/05/2021, HCB (18100).

***Protostropharia alcis* (Kytöv.) Redhead, Thorn and Malloch ssp. *austrobrasiliensis* (Cortez & R. M. Silveira) C. Hahn. (Figure 3)**

The species has pileus up to 30 mm in diameter, convex to slightly umbonate, dark yellow to olive yellowish or grayish. Lamellae adnate, pigmented with a brownish color, except for the serrated edge. Stipe 45 – 120 x 1 – 2.5 mm, glutinous, concolorous to pileus or weakly pigmented. Spores 11 – 15 x 6.5 – 9 µm, grayish brown, ellipsoid to oblong, thick-walled, smooth and with a central germinal pore, brownish under the microscope. Basidia 20 – 40 x 7 – 15 µm, tetrasporic. Pleurocystidia present as chrysocystidia of 20 – 60 x 8.5 – 15 µm, fusiform. Cheilocystidia present in the form of leptocystidia, measuring 17 – 40 x 3.5 – 10 µm, lageniform to cylindrical. Cortical layer of the pileus is formed by an ixotrichoderm, with thin hyphae, 1 – 6 µm in diameter in a gelatinous matrix. Caulocystidia are similar to cheilocystidia. Acanthocystidia absent. Clamp connections present.

They grow solitary or sparsely in dung in forest or pastures. The species is described for the states of Paraná, Santa Catarina and Rio Grande do Sul (Brazil) in the Atlantic Forest domain (Silva *et al.*, 2006, 2008; Calaça *et al.*, 2014; Seger *et al.*, 2017). This is the first citation of the species to the Pampa biome.

Figure 3: *Protostropharia alcis* ssp. *austrobrasiliensis* (Cortez & R.M. Silveira). Basidiome with lamellated hymenophore (A). Basidiospores (B). Basidia (C). Pleurocystidia of the chrysocystidia type (D). Cheilocystidia (E).



Source: Authors.

In the illustration above it is possible to observe the macro and microscopic structures of *Protostropharia alcis* ssp. *austrobrasiliensis*, as well as its basidiome as found in its natural habitat.

The species is cited as occurring in elk (*Alces alces* Linnaeus, 1758) dung in Estonia, Norway, Sweden, Finland (Kytövuori, 1999), and Poland (Kujawa *et al.*, 2012, Halama & Kudlawiec, 2014).

Kytövuori (1999) suggested that the distribution of *P. alcis* (Kytöv.) Redhead, Thorn and Malloch should continue eastward (Russia) and North America (Canada) due to the presence of elk being found in your dung, while being rare or absent in southern Europe. However, subsequent records of the species were done to Italy, on an unidentified dung, probably of deer (Noordeloos, 2011). This provides evidence of its broader substrate utilization capacity and more extensive global distribution. Cortez and Silveira (2008) describe *Stropharia alcis* var. *austrobrasiliensis* Cortez and R.M. Silveira, as morphologically identical to *P. alcis*, differing only in the substrate used, colonizing bovine manure. Wang & Tzean (2015) suggest that the species should be transferred to the genus *Protostropharia*.

The species has two occurrence records, one in Poland (Halama & Kudlawiec, 2014) and another in China (Wang & Tzean, 2015).

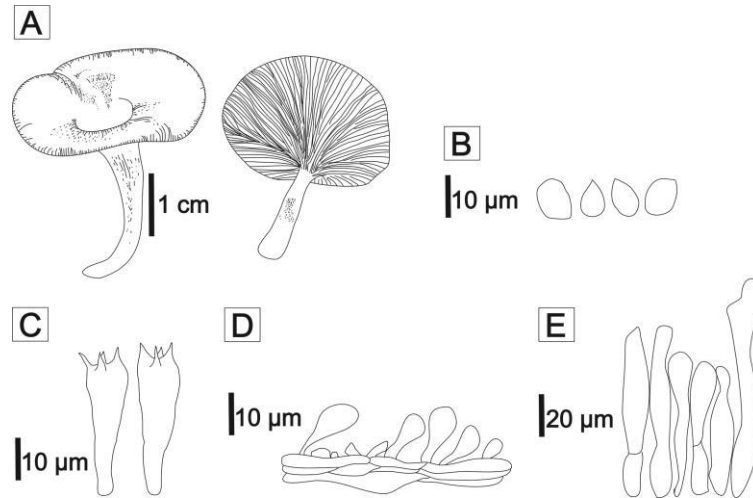
Material examined: Brazil, Rio Grande do Sul, São Gabriel, Morro do Sabiá, A. L. Costa, 13/05/2021, HCB (18101).

***Trogia buccinalis* (Mont.) Pat., Tab. Analyt. Fung. 7: 57. fig. 651. 1889. (Figure 4)**

The species has grayish-pale to pale-yellowish, umbilicated to infundibuliform pileus, ranging from 0.5 - 3 cm in diameter. Lamellae distant, deeply decurrent, concolor to the pileus or paler. Stipe 1.5 - 4 x 0.1 - 0.6 cm, short, central. Spores 5.5 - 7 x 4 - 5 µm, broadly ovoid to broad-ellipsoid, smooth and thin-walled. Basidia 25 - 35 x 4.5 - 5.5 µm, tetrasporic. Cheilocystidia 30 - 40 x 2 - 7 µm, cylindrical-sinuuous, often constricted, occasionally nodular at the apex, hyaline, thin-walled. Pleurocystidia absent. The cortical layer of the pileus is formed by more or less parallel and prostrate hyphae, 1 - 5 µm in diameter, with occasional diverticula.

Trogia citrina Corner is distinguished by the citrine color of the basidiome and the inflated dermatocystidia in the cortical layer of the pileus. It grows on trunks or branches, and can be solitary, but usually is aggregated. Pegler (1983) cite it for the Antilles and French Guiana. Pegler (1997) reports its occurrence to São Paulo, in the Atlantic Forest biome and for Rondônia in the Amazon biome. This is the first citation for the state of Rio Grande do Sul and to the Pampa biome.

Figure 4: *Trogia buccinalis*. Basidiome with lamellar hymenophore (A). Basidiospores (B). Basidia (C). Elements of the cortical layer of the pileus (D). Cheilocystidia (E).



Source: Authors.

In the illustration it is possible to observe the macro and microscopic structures of *Trogia buccinalis*, as well as its basidiome as found in its natural habitat.

The species is also cited for Mexico (Gomez-Hernandes & Williams-Linera, 2011).

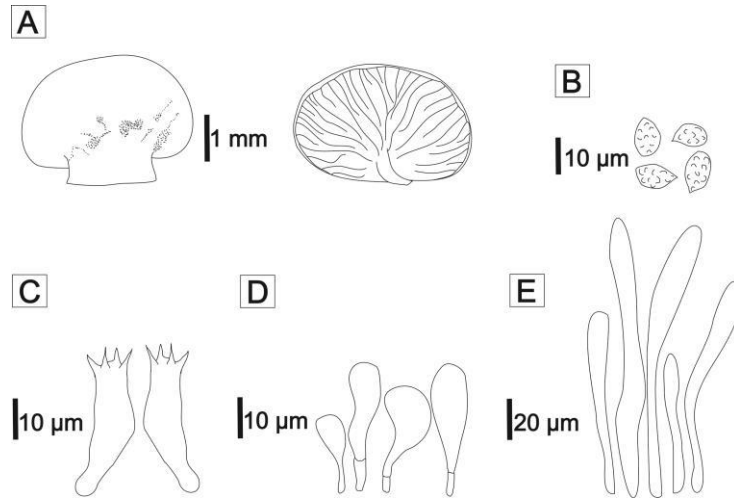
Material examined: Brazil, Rio Grande do Sul, São Gabriel, Morro do Sabiá, A. L. Costa, 13/05/2021, HCB (18102).

***Crepidotus euterpicola* Senn-Irlet & De Meijer, Mycotaxon 66: 191 (1998). (Figure 5)**

The species has a flabeliform, sessile pileus, beige to yellowish in color, with a smooth margin, ranging from 2 - 5 mm in diameter. Lamellae close, wide, radiating from the point of attachment. Stipe reduced to absent. Spores 8 - 12 µm ellipsoid, warty to rough. Basidia tetrasporic, thin-walled, hyaline. Pleurocystidia absent. Cheilosistidia 28 - 44 µm long, cylindrical, thin-walled hyaline. Cortical layer of the pileus formed by clusters of erect, thin-walled, spaced hyphae of 20 - 80 µm long.

It grows on branches and twigs, usually solitary to aggregated. Senn-Irlet & Meijer (1998) cited it to Paraná state (Brazil), growing on branches of *Euterpe edulis* Mart.. Therefore, this is the first citation of the species for the state of Rio Grande do Sul, in the Pampa biome. *C. euterpicola* is an endemic species to Brazil.

Figure 5: *Crepidotus euterpicola*. Basidiome with lamellated hymenophore (A). Basidiospores (B). Basidia (C). Cheilocystidia (D). Elements of the cortical layer of the pileus (E).



Source: Authors.

In the illustration it is possible to observe the macro and microscopic structures of *Crepidotus euterpicola*, as well as its basidiome as found in its natural habitat.

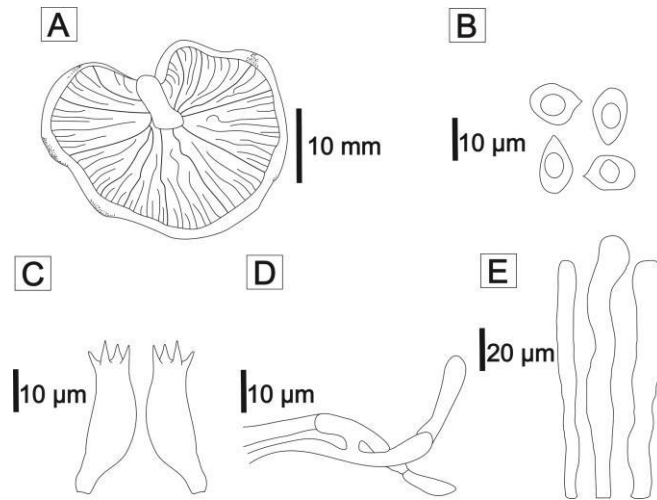
Material examined: Brazil, Rio Grande do Sul, São Gabriel, Morro do Sabiá, C. F. Lopes, 20/10/2021, HCB (18103).

***Entoloma depluens* (Batsch) Hesler, Beih. Nova Hedwigia 23: 16 (1967). (Figure 6)**

The species has tomentose, sessile to laterally or almost subcentrally attached pileus with a color ranging from pinkish to grayish, 1 - 5 mm in diameter. Lamellae adnate to decurrent, thick. Stipe laterally attached, present to absent. They present smooth, subellipsoid and pinkish spores of $8 - 7 \times 10 - 4 \mu\text{m}$. Basidia of $20 - 30 \times 7.5 - 14.5 \mu\text{m}$, tetrasperate. Hyphoid cheilocystidia 20 - 80 μm in diameter. The cortical layer of the pileus is formed by palisadoderm with elements 1 - 10 μm in diameter.

It grows solitary next to trunks or fallen wood branches on the ground. Pegler (1997) cites the species for the state of São Paulo (Brazil). Therefore, this is the first description of the species for the state of Rio Grande do Sul, domain of the Pampa biome. *E. depluens* also occurs in Scandinavia, Denmark (Noordeloos, 1982).

Figure 6: *Entoloma depluens*. Basidiome with lamellated hymenophore (A). Basidiospores (B). Basidia (C). Elements of the cortical layer of the pileus (D). Cheilocystidia (E).



Source: Authors.

In the illustration it is possible to observe the macro and microscopic structures of *Entoloma depluens*, as well as its basidiome as found in its natural habitat.

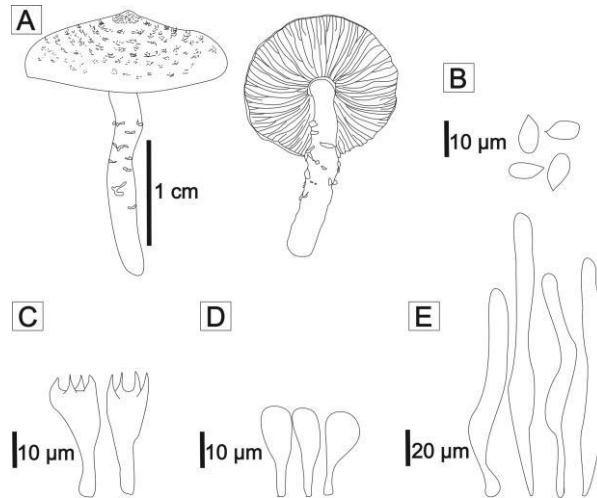
Material examined: Brazil, Rio Grande do Sul, São Gabriel, Morro do Sabiá, M. A. Heberle, 20/10/2021, HCB (18104).

***Lepiota subincarnata* J.E. Lange, Fl. Agar. Danica 5, Taxonomic Conspectus V (1940). (Figure 7)**

The species has basidiomes about 2 cm high, pileus 1 – 5 cm in diameter, campanulate, brown in the center becoming lighter at the edges, with brownish squamules on a light beige surface. Lamellae free, close, with smooth edges and white. Stipe 1.5 – 5.5 x 0.1 – 0.4 cm, central, cylindrical, light beige, covered with pinkish squamules and flocculent fragments remaining from the veil. Spores 5.3 – 7 x 3 – 4.5 µm, ellipsoid to subcylindrical, smooth, thin-walled, hyaline, pseudoamyloid. Basidia 14 – 25 x 4 – 5 µm, clavate, tetrasporic. Cheilocystidia 15 – 26 x 4 – 8 µm, cylindrical, slightly thick-walled. Pleurocystidia absent. Hymenophoral trama regular. Cortical layers formed by a trichoderm with relatively erect clustered elements, brown in color. Clamp connections present.

The species grows solitary or gregariously inside forests, along with wood humus. In Brazil the species is described by Albuquerque *et al.* (2007) from Rio de Janeiro and Wartchow (2005) from Pernambuco. In Europe it was described by Candusso and Lanzoni (1990), and from Germany by Enderle and Krieglsteiner (1989). This is the first description of the species for the state of Rio Grande do Sul in the Pampa biome.

Figure 7: *Lepiota subincarnata*. Basidiome with lamellated hymenophore (A). Basidiospores (B). Basidia (C). Cheilocystidia (D). Elements of the cortical layer of the pileus (E).



Source: Authors.

In the illustration it is possible to observe the macro and microscopic structures of *Lepiota subincarnata*, as well as its basidiome as found in its natural habitat.

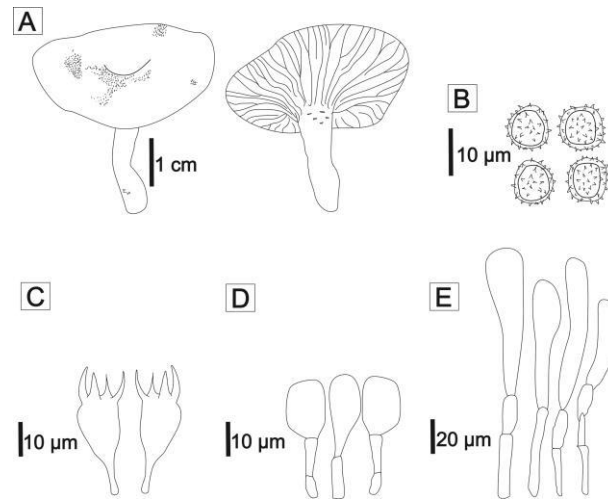
Material examined: Brazil, Rio Grande do Sul, São Gabriel, Morro do Sabiá, J. R. P. Velloso, 20/10/2021, HCB (18105).

***Neopaxillus echinospermus* (Speg.) Singer, Lilloa 22: 633 (1951) [1949]. (Figure 8)**

The species has a pileus up to 40 mm in diameter, with a depressed or flattened to infundibuliform shape, yellowish-brown to golden brown or reddish-orange color. Lamellae decurrent, subdistant, with a brownish central region and concolor edge. Stipe 25 – 40 x 2 – 3.5 mm, cylindrical or tapering, with bulbous base, yellowish-white to pale yellowish or orange coloration, smooth or slightly striated, without rhizomorphs. Spores 6.5 – 9 x 6.5 – 8.5 µm, brownish, with spines, subglobose to globose, thick-walled, inamyloid. Basidia 28 – 52 x 7.5 – 11 µm, tetrasporic. Pleurocystidia absent. Cheilocystidia 18.5 – 32 x 6.5 – 16 µm, versiform, from clavate to piriform, thin-walled and hyaline. Cortical layer hymeniform, with clavate or cylindrical and sometimes subcapitate cells, 33 – 49 x 7 – 10 µm in size, with sub-hymenium of thin hyaline hyphae up to 2.5 µm in diameter. Cortical layer of the stipe with prostrate hyphae with tufts of caulocystidia measuring 27 – 56 x 5 – 14 µm, cylindrical to clavate or spheropedunculated, thin-walled and hyaline. Oleiferous hyphae present. Clamp connections present.

They grow solitary or gregarious, almost cespitose on litter soil inside forest. Silva-Filho *et al.* (2016) redescribed this species based on material from Brazil. The species is described for the states of São Paulo, Paraná and Rio Grande do Sul in the Atlantic Forest domain (Singer & Digilio, 1951; Singer, 1964; Watling & Meijer, 1997). This is the first description of the species from the Pampa Biome. It also occurs in North America, being cited to Mexico (Guzmán, 1986).

Figure 8: *Neopaxillus echinospermus*. Basidiome with lamellated hymenophore (A). Basidiospores (B). Basidia (C). Cheilocystidia (D). Elements of the cortical layer of the pileus (E).



Source: Authors.

In the illustration it is possible to observe the macro and microscopic structures of *Neopaxillus echinospermus*, as well as its basidiome as found in its natural habitat.

Material examined: Brazil, Rio Grande do Sul, São Gabriel, Morro do Sabiá, C. F. Lopes, 20/10/2021, HCB (18106).

4. Conclusion

In this study seven new citations of species belonging to five families of Agaricales mushrooms were reported to the Pampa Biome, *Macrolepiota mastoidea* and *Lepiota subincarnata* (Agaricaceae); *Trogia buccinalis* (Tricholomataceae); *Crepidotus euterpicola* (Crepidotaceae); *Entoloma depluens* (Entolomataceae); *Neopaxillus echinospermus* (Paxillaceae) and *Protostropharia alcis* ssp. *austrobrasiliensis* (Strophariaceae). New citations such as those reported here reinforce the great diversity existing in the Brazilian Pampa biome and emphasize the importance of future studies, as there is much to be discovered, both in terms of fauna, flora and the fungi, increasing the appreciation and preservation of this biome.

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