First report of *Calvatia fragilis* Fr. (Basidiomycota) in Central-West Brazil

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Abstract

Calvatia fragilis, a species belonging to the Basidiomycota phylum, is noted for its presence in tropical and neotropical regions. However, this fungus has been scarcely documented in natural habitats in Brazil. The present study aims to enhance the understanding of the geographical distribution of *Calvatia fragilis*, reporting the first record of this species in a *Cerrado* ecosystem, specifically in the Brazilian Midwest, in the State of Goiás. The discovery site is an area dedicated to agricultural production, characterized by a red dystroferric clay soil. In December 2023, specimens of *Calvatia fragilis* were collected in this region. The gathered basidiomes were meticulously catalogued and identified using a specific identification key for the *Calvatia* genus. Prior to this study, records of the species were limited to the States of Rio de Janeiro, São Paulo, and Rio Grande do Sul. This research expands the knowledge on the distribution of *Calvatia fragilis*, now including the State of Goiás among the Brazilian areas where this fungus occurs.

Keywords: *Calvatia* genus, mushrooms, Agaricaceae, Agaricales, cosmopolitan fungi.

Primeiro relato de *Calvatia fragilis* Fr. (Basidiomycota) no Centro-Oeste do Brasil

Resumo

Calvatia fragilis, uma espécie pertencente ao filo Basidiomycota, é notória por sua presença em regiões tropicais e neotropicais. Este fungo, no entanto, tem sido pouco documentado em habitats naturais no Brasil. O presente estudo visa aprimorar o conhecimento sobre a distribuição geográfica de *Calvatia fragilis*, reportando o primeiro registro dessa espécie em um ecossistema de Cerrado, especificamente no Centro-Oeste brasileiro, no Estado de Goiás. O local do achado é uma área dedicada à produção agrícola, caracterizada por um solo vermelho distroférrico de textura argilosa. Em Dezembro de 2023, foram coletados exemplares de *Calvatia fragilis* nesta região. Os basidiomas recolhidos foram cuidadosamente catalogados e identificados empregando uma chave de identificação específica para o gênero *Calvatia*. Antes deste estudo, registros da espécie se limitavam aos Estados do Rio de Janeiro, São Paulo e Rio Grande do Sul. Com esta pesquisa, amplia-se o conhecimento sobre a distribuição de Calvatia fragilis, incluindo agora o Estado de Goiás entre as áreas brasileiras onde este fungo ocorre.

Palavras-chave: gênero *Calvatia*, cogumelos, Agaricaceae, Agaricales, fungos cosmopolitas.

1. Introduction

The genus *Calvatia* Fr. encompasses a diversity of mushrooms belonging to the Agaricaceae (Agaricales) family, with this genus forming mushrooms. The name *Calvatia* derives from the word *calvus* (Latin) meaning "bald" (Hedawoo, 2020). The genus is soft and is characterized by stalked-sessile, globose-subglobose, pyriform-subpyriform sporophore. Fruiting body dehisces by irregular fragmentation of the peridium. According to Verma et al. (2018) *Calvatia* species are popularly known as "puff balls". There are around 140 records of *Calvatia* species, classified by Swedish mycologist Elias Magnus Fries in 1849, with a single species *Calvatia craniiformis* originally described as *Bovista craniiformis* Schwein (Fries, 1849).

The species of Calvatia are cosmopolitan where make up a well-defined group within the Lycoperdaceae (the "Calvatia complex" by Kreisel, (1989)), differing from other genera by the way the peridium ruptures, which is irregular and never displayed by a single delimited pore, and by the exclusive presence of eucapillitium, generally exhibiting dilated pores also observed in the genera *Gastropila* and *Langermannia* (Miller; Miller 1988).

The genus *Calvatia* presents several controversies regarding its position (Zeller & Smith, 1964; Kreisel, 1989, 1992, 1994; Lange, 1993), all as described by Cortez et al. (2012), who present good arguments reflecting the great diversity of this genus of mushrooms. In some studies, researchers classify all species into a single *Calvatia sensu lato*, while other studies have divided the genus into different, but related, genera (*Gastropila*, *Handkea* Kreisel, *Langermannia*, and *Lanopila* Fr.). In this study, where we report the first study of C. fragilis, we followed the generic concepts of Calonge (1998), Calonge & Martín (1990), and Cortez et al. (2012), where they discuss its limits under morphological criteria, including peridium, gleba, and structure of basidiospores examined under scanning electron microscopy (SEM), and in our study, by optical microscopy (OM).

Phylogenetic studies conducted by Krüger & Gargas (2008) and Larsson & Jeppson (2008) have yielded valuable data on the systematics of *Calvatia* and related genera. However, these studies have been inconclusive; in general, the taxa are considered to be exclusively from the European continent and temperate climates. Following the same reasoning as Cortez et al. (2012), in our first report for the State of Goiás, Brazil, we classify Calvatia, specifically C. fragilis, based on morphology. This approach will be maintained until Neotropical species are more extensively sampled.

Most of the species described in *Calvatia* are edible when young, but some species such as *Calvatia fumosa* have a very pungent odor and should be avoided. Many *Calvatia* mushrooms are consumed by many cultures around the world, although they must be known and consumed safely. Species such as *Calvatia gigantean* (Batsch) Lloyd (giant puffball) and *Calvatia utriformis* (= *Bovistella utriformis* (Bull.) Demoulin & Rebriev) were reported as edible mushroom species by William & Arora (2008).

This article reports the first report of *Calvatia fragilis* Fr. in the Brazilian *Cerrado* area in the State of Goiás, Brazil, this being a new record of the fungal fauna for the Brazilian Central-West region.

2. Materials and Methods

2.1 Study area

The study area, georeferenced with the coordinates 17º43'13.2"S and 50º52'51.4"W, is a remnant of *Cerrado* in its restricted sense. Currently, it is under significant anthropic influence due to over 20 years of rural production. This area is located in the Rural Unit owned by Antônio Menezes & Filhos, in the municipality of Rio Verde, Goiás, Brazil. It was mapped using a Global Positioning System (GPS) and analyzed with Idrisi Software, revealing an area of 30 m² at an altitude of 780 meters.

2.2 Collection and identification

The collection occurred on December 18, 2023. The basidiomata were photographed during this time. Specimens of C. fragilis were transported to the Technological Chemistry Laboratory at the Agrochemistry Department of the Goiano Federal Institute, located in Rio Verde, Goiás, Brazil. Here, they were identified by the last author of this study. Two samples of basidiomes were preserved in an ultrafreezer for further analysis and subsequent studies and were cataloged with the voucher ANFI-01. For the identification process, microscopic data and the identification key for *Calvatia*, as described by Cortez et al. (2012), were utilized.

3. New occurrence

Individuals of the *Calvatia fragilis* mushroom (n = 10) were found germinating in red dystrophic soil in an agricultural crop area in January 2024 with a rainy seasonal period. Fruiting bodies were collected from leaf litter beneath a fruiting *Persea americana*. In the study by Gupta et al. (1974) researchers described *C. fragilis* inhabiting

areas in Rajgadh, HP, India, on humicous soil in coniferous forest. Hedawoo (2020) describes the presence of *C. fragilis* in areas of Amravati, India. In Brazil Cortez et al. (2012) describes basidiomes for *C. fragilis* scattered throughout the field, on grassy soil.

Key to *Calvatia*, *Gastropila* and *Langermannia* from Rio Grande do Sul and Goiás State, Brazil (Cortez et al., 2012) and this study (2024).

Calvatia fragilis (Vittad.) Morgan, J. Cincinnati Soc. Nat. History 12:168 (1890). Species description: Cortez et al. (2012). Material examined: Brazil. São Paulo: Angatuba, Teodoros neighborhood, rural area, 08-V-2018, Trierveiler-Pereira, L., & Baltazar, J. M. s.n. (SPSC); ibid., 15-V-2018, L. Trierveiler-Pereira & J.M. Baltazar s.n. (SPSC).

1. Basidiomata without or with a very rudimentary sterile base……..…….……..……2
  2. Basidiospores smooth, gleba disintegrating at maturity…….…….……5. *G. fragilis*
  2. Basidiospores echinate, gleba lanose and persistent……..……..3
    3. Eucapillitium single, without curled threads…….…….…..……6. *L. bicolor* var. bicolor
    3. Eucapillitium with curled (tendril-like) hyphae……..……..……..7. *L. bicolor* var. cirrifera
1. Basidiomata with a conspicuous, compact to cellular sterile base……4
  4. Mature gleba and subgleba ochraceous to olivaceous brown…….……..…….4. *C. rugosa*
  4. Mature gleba and subgleba greyish brown to biolaceous…….…….5
    5. Subgleba reduced (<20 mm), compact……..3. *C. fragilis*
    5. Subgleba large (>50 mm), cellular……..…….……..6
      6. Basidiospores 3.5 – 4.5 µm diam…….…….…....……..2. *C. cyathiformis* var. chilensis
      6. Basidiospores 6-7.5 µm diam…….…….....……*C. cyathiformis* var. cyathiformis

Description: Basidiomata 26 - 72 mm high, 44 - 104 mm diam., depressed subglobose, usually with a short rooting and rugose to plicate base, with scattered rhizomorphs. Exoperidium brownish grey to dark blond, membranous and dry, thin and fragile, cracked and falling away as irregular patches. Endoperidium smooth, very thin, greyish brown, dehiscence irregular. Gleba pulverulent, dark brown to greyish-brown when mature. Subgleba mostly compact and poorly developed, but in some basidiomata it can be spongy and short (about ¼ of the height). Basidiospores 4.5 – 6.0 µm diam., globose, echinate, with spines < 0.5 µm long, shortly pedicellate (< 1 µm), yellowish-brown under KOH; under SEM the ornamentation is seen to arise from an irregular basidiospore surface as short columns or spines.

Eucapillitium 2.5 – 5.0 µm diam., smooth and thick-walled, septate, with rare dichotomous branches, pores small and numerous or scattered and large, in both cases circular, pale yellowish brown under KOH. Exoperidium composed of purplish to yellowish-brown elements, 18.5 – 42 × 9.0 - 21.0 µm, of variable shape (subglobose, short cylindrical, pyriform), walls thick (< 3 µm) and smooth by according (Wartchow; Silva, 2007; Cortez et al., 2012).

Distribution: Known from all subtropical and tropical areas of the world (Kreisel, 1994; Bates et al., 2009). Brazil: known from the States of Rio de Janeiro (Silveira, 1943), Rio Grande do Sul (Guerrero; Homrich, 1999), São Paulo (Cortez et al., 2012; Fernandes et al., 2021) and now in Goiás State (2024). Habitat: Among grasses, on earth or sandy soil, in open areas (native meadows, coastal vegetation) (Cortez et al., 2012), and in high-productivity red dystrophic argisol soil described in this report. In research carried out by Zeller (1947) and Zeller & Smith (1964), researchers consider it as a form of *Calvatia cyathiformis*. However, this taxon *C. fragilis* differs considerably from the latter due to the absence or reduced size of the sterile base, smaller basidiospores and habitat in open areas as suggested by Cortez et al. (2012).

Figure 1. Individual of *Calvatia fragilis* in natural habitat in the *Cerrado* area in the State of Goiás, Brazil (**A**); In (**B**) top view of the vegetative part of *Calvatia fragilis*; In (**C**) individuals with different sizes and diameters collected, and in (**D**) horizontal section (longitudinal section) demonstrating the cuticle and pileus of the vegetative part in *Calvatia fragilis*. Photographs: **A**, **B**, **C** and **D** Menezes Filho, A. C. P. (2024).

Calvatia fragilis reported in this study has not yet been described for the Central-West region of Brazil and for the State of Goiás, Brazil, and represents an unprecedented record of the diversity of *Calvatia* in this tropical region of the Brazilian *Cerrado* area, thus, how can it contribute to studies on regional, national and international estimates for the genus *Calvatia* and its global dispersion.

4. Conclusions

This study constitutes the inaugural documentation of *Calvatia fragilis* Fr. in the *Cerrado* region of Brazil. It underscores the critical importance of preserving and encouraging further research on this species of basidiomycete mushroom within the Brazilian *Cerrado*. Such endeavors are essential for fostering a comprehensive understanding of this species and contributing to the development of a globally recognized mycological community. This research not only expands the ecological knowledge of *Calvatia fragilis* but also highlights the rich biodiversity of the *Cerrado* biome, emphasizing the need for its conservation and study.

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6. Authors' Contributions

Antonio Carlos Pereira de Menezes Filho: collection, initial analysis of the species, article writing, translation and publication. *Matheus Vinícius Abadia Ventura*: substantial contribution to the study planning and specimen

collection. *Aurélio Ferreira Melo*: verification of the correctness of the final study. *Marconi Batista Ferreira*: substantial contribution to the study planning. *Frederico Antônio Loureiro Soares*: substantial contribution to the study planning. *Carlos Frederico de Souza Castro*: substantial contribution to the planning of the study and coordinator of the Technological Chemistry Laboratory. *Porshia Sharma*: corrections to the English translation and structuring of the text. *Tullyo Henrique Lima Machado*: identification of the specimen, text corrections and translation into English.

7. Conflicts of Interest

No conflicts of interest.

8. Ethics Approval

Not applicable.

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