Geastrum hyalinum (Basidiomycota, Geastraceae), a new species from Brazilian Southern Amazon

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Abstract
The genus Geastrum Pers. has cosmopolitan distribution and can be found in humid, arid or semi-arid climates. Geastrum hyalinum is a new species found in the Brazilian Southern Amazon in the Rio Ronuro Ecological Station. This new species can be recognized by its hyaline microscopic structures, such as peridium hyphae, capillitium and spores; indeed, this character is unusual for the genus Geastrum. Description, discussion and photographs of this new taxon are given.

Key words – Biodiversity – Earthstar – Gasteromycetes – Neotropics – Taxonomy

Introduction
The genus Geastrum Pers. was proposed in 1794, mainly characterized by gasteroid basidiomata opened in a star shape. Some Geastrum species have interesting metabolites, such as G. saccatum Fr., which has polysaccharides with antioxidants and anti-inflammatory action (Guerra et al. 2007). This potential reinforces the importance of knowing and studying this fungus. The genus has a global range and is well-distributed in diverse climates, from humid to arid or semi-arid environments, such as the Caatinga and the Brazilian Cerrado (Sunhede 1989, Baseia & Galvão 2002).

The Brazilian Amazon is the largest Tropical Forest in the world, known for its high biodiversity. Even so, this Forest comprises many organisms still unknown to science (MMA 2007). In Brazil, the Southern region of the Amazon Forest includes an area where the agricultural frontier advances towards preserved areas and the rates of deforestation are higher than in the other Amazon areas. This corroborates the urgency of research in the Southern Amazon, both to describe the still unknown biodiversity and to delimit conservation strategies that will be effective for this region (INPE 2018, Oliveira-Filho & Metzger 2006).

Based on the most recent studies about the genus Geastrum in Brazil, about 60 species of Geastrum have been recorded (Cabral et al. 2017, Crous et al. 2018), a very significant number, since about 100 to 120 species are known to the world (Zamora et al. 2014a). For the state of Mato Grosso, located in the Central-West Region of Brazil, even composed as it is by extensive forest
areas (Cerrado and Amazon domain), only one record of *Geastrum* species was previously made (Trierveiler-Pereira et al. 2011). So, this work aims to expand the knowledge of the genus *Geastrum* through the description of a new species for the state of Mato Grosso, Brazil.

**Materials & Methods**

**Collection Location**

The specimens were collected during the rainy season in the Rio Ronuro Ecological Station, which is located in the municipality of Nova Ubiratã, central region of Mato Grosso state, between coordinates 12º 46’ 00”, 14º 07’ 00” south (latitude) and 55º 15’ 00”, 54º 19’ 00” west (longitude) (Fig. 1). The Ecological Station covers an area of 102, 000 ha and belongs to the Amazon domain. The climatic character is determinant for the creation of a transition strip that separates the northern part of the Amazonian region from the southern part, which includes Cerrado areas (Cepemar 1998, Silva et al. 2009).

![Fig. 1 – Details of collection site. Red circle represents the type location of *Geastrum hyalinum*.](image)

**Collection, herborization and analysis of specimens**

Two specimens were found, following the methodology proposed by Bascia et al. (2014) where, at the time of collection, they were photographed using a ruler as a measurement parameter. Observations about substrate, habit and geographic coordinates were also noted. After collection, the material was dehydrated following the guidelines from the mentioned literature. For the morphological study, the macroscopic and microscopic features were observed following the literature on the genus: Sunhede (1989) and Zamora et al. (2014b, 2015). The macroscopic observations were made under a Leica EZ4 magnifying glass, and staining was observed according to Kornerup & Wanscher (1978).

Microscopic analyses were performed using the Nikon Eclipse Ni (LM) with Nikon DS-Ri1 camera coupled using the NIS-Elements Ar v.4.51.00 software for the measurements. Slides with portions of the gleba and peridium were assembled in 5% KOH, and a Shimadzu SSX-550 was
used for scanning electron microscopy (SEM). For this, the preparation of the material examined in MEV was carried out based on Silva et al. (2011). All measurements include the ornamentation of basidiospores. The abbreviations of the basidiospores followed Sousa (2015) and Bates (2004): n = number of basidiospores measured at random; \( \bar{x} = \text{mean} \pm \text{standard deviation of basidiospore diameter and height (including ornamentation)}; \) Qm = average quotient of height / width. The collected specimens were deposited at the Herbarium of the Federal University of Rio Grande do Norte (UFRN), Natal city, and the isotype was deposited at the CNMT Herbarium of the Federal University of Mato Grosso, both in Brazil.

Results

Taxonomy

**Geastrum hyalinum** Freitas-Neto, N.M. Assis, J.O. Sousa, Baseia

MycoBank number: MB830897; Facesoffungi number: FoF06131

Etymology – given name referring to the microscopic structures, which are hyaline.

Holotype – Brazil, Mato Grosso State, Nova Ubiratã city, Rio Ronuro Ecological Station (S 13° 06' 22.9" W 54° 25' 42.3" 318 m), on leaf-litter, 23 Feb 2017, Assis NM, Drechsler S, Magnago A, NMA_M07 (UFRN Fungos – 3016, isotype; CNMTf76)

Diagnosis – Basidiomata saccate until 28–34 mm wide; endoperidium orange white to pale orange, sessile with protruding hyphae; hyphae of the exoperidium layers hyaline; gleba white; Basidiospores hyalines 3.6–5.0 μm diam.; eucapillitium hyaline 2.7–4.7 μm diam.

Description – Unexpanded basidiomata not observed. Expanded basidiomata saccate, 10–16 mm high (including peristome) × 28–34 mm wide. Exoperidium splitting into 5–7 triangular rays, revolute, non-hygroscopic. Mycelial layer yellowish white (2A2), cotonos e surface encrusted with debris. Fibrous layer grayish yellow (4B4), coriaceous. Pseudoparenchymatous layer dark brown (6F4), non-rimose, persistent, collar absent. Endoperidial orange with to pale orange (5A2; 5A3), globose to subglobose, 06–08 × 14–17 mm, sessile, with protruding hyphae, non-pruinose. Apophysis absent. Peristome fibrillose, flat, delimited, delimitation lighter than endoperidium <1 mm high. Gleba yellowish white (1A2). Basidiospores hyaline, globose to subglobose (sometimes flattened with a semi-elliptical shape), 3.6–5.0 × 3.6–4.9 μm \([\bar{x} = 4.1 \pm 0.3 \times 4.0 \pm 0.3, \text{Qm} = 1.02, n = 30]\), ornamentation conspicuous under LM. Warts cylindrical (0.3-0.7 μm high). Eucapillitium hyaline, thin-walled, 2.7–4.7 μm diam., surface encrusted, covered by small warts, lumen evident (Fig. 1 Mycelial layer composed of hyaline, some sinuous, thin-walled (0.3-0.7 μm) hyphae, 2.0–3.0 μm diam., surface non-encrusted, non-branched and lumen evident. Fibrous layer composed of hyaline, thin-walled (0.5-0.8 μm) hyphae, 3.7–5.7 μm diam., surface non-encrusted, non-branched and lumen evident. Pseudoparenchymatous layer composed of hyaline, thick-walled (0.7-1.2 μm) hyphal cells, subglobose, oval to elongated, 12.7–32.6 × 18.1–44.1 μm.

Habitat – Growing on leaf-litter.

Habit – Gregarious.

Material Examined – Brazil, Mato Grosso State, Nova Ubiratã city, Rio Ronuro Ecological Station, 23 Feb 2017, S 13° 06' 22.9" W 54° 25' 42.3" 318 m, Assis, NM; Drechsler, S; Magnago, A., NMA_M07 (UFRN Fungos – 3016, isotype; CNMTf76).

Remarks – *Geastrum hyalinum* is mainly characterized by the color of the structures, which are orange white to pale orange endoperidium, white gleba and hyaline microscopic structures: basidiospores, eucapillitium, mycelial layer, fibrous layer and pseudoparenchymatous layer. This species is also recognized by delimited fibrillose peristome, and hyaline basidiospores with 3.6–5.0 × 3.6–4.9 μm diam.

In the first view, *G. hyalinum* can be confused with *G. fimbriatum* Fr. by the saccate basidiomata, mycelial layer encrusted with debris, non-hygroscopic rays and fibrillose peristome. Furthermore, *G. fimbriatum* has three layers of exoperidium, all constituted by hyaline hyphae. However, *G. hyalinum* differs from the latter due to the presence of white gleba and peristome.
lighter than endoperidium, as well as smaller basidiospores (up to 5 μm) and hyaline eucapillitium (4.7 μm) (Trierveiler-Pereira et al. 2011).

Fig. 2 – *Geastrum hyalinum* (UFRN Fungos – 3016, from holotype). a Basidiomata on host. b Eucapillitium in MOP. c Eucapillitium in SEM. d Basidiospores in MOP. E Basidiospores in SEM. Scale bars: a = 1 cm, b,d = 5 μm, c,e = 10 μm
Geastrum hyalinum can be similar to G. rusticum Baseia, B.D.B. Silva & T.S. Cabral, by the saccate basidiomata, mycelial layer encrusted with debris, fibrous layer coriaceous and fibrillose peristome non-delimited. Layers of hyaline exoperidium differ from G. hyalinum by the semi-hygroscopic rays, papyraceous mycelial layer, mamiform peristome and dark brown gleba (Cabral et al. 2014).

Geastrum ishikawae Accioly, J.O. Sousa, Baseia & M.P. Martin is another similar species that also has saccate basidiomata, cotonose mycelial layer encrusted with debris and fibrillose peristome. However, the new species, G. hyalinum, does not have pruinose endoperidium and flattened peristome, unlike G. ishikawae; furthermore, the last one has bigger basidiospores with up to 7 μm (Crous et al. 2016).

Geastrum hyalinum can also be compared to G. hieronymi Henn. Both have cotonose mycelial layer encrusted with debris, coriaceous fibrous layer, light-colored endoperidium with protruding hyphae and layers of hyaline exoperidium. However, it differs from G. hyalinum because it presents stalked endoperidium, exoperidium with hygroscopic rays and large basidiospores (up to 7 μm), which are densely verrucose (Leite et al. 2007).

Other species that can be compared is G. saccatum Fries for present non-hygroscopic rays, sessile endoperid, basidiomata saccate and peristome fibrillose. However, it differs from G. hyalinum by the absence of incrustations with debris in the mycelial layer and protruding hyphae in the endoperidium, in addition to basidiospores and brownish eucapilitium (Baseia et al. 2003).

Another species that resembles morphologically with G. hyalinum is Geastrum litchiform Desjardin & Hemmes, because it presents light-colored endoperidium, sessile, with protruding hyphae and layers of hyaline exoperidium, non-hygroscopic rays and basidiomata saccate. It differs from the new species because it's not distinctly limited, smaller basidiospores (up to 3.8 μm), often form a collar surrounding the endoperidial body, besides its peculiar mycelial layer composed of tufts of subparallel hyphae irregular in outline to aculate inside of dehiscent cups with loosely interwoven hyphae, covered with an amorphous layer of crystalline matter (Hemmes & Desjardin 2011).

Some basidiospores of G. hyalinum are flattened, reaching semi-elliptical form. In this case it can be compared to Geastrum ovalisporum Calonge & Moreno-Arroyo that has oval-shaped spores and the three layers of exoperidium with hyaline hyphae, as well as cotonose mycelial layer encrusted with debris, fibrillose peristome and non-hygroscopic rays. However it is distinguished from the new species by presents basidiomata arched, stalk, apophysis, conical peristome and brown basidiopores (Calonge et al. 2000, Cortez et al. 2008).

Discussion
The Rio Ronuro Ecological Station is in a zone of ecological transition between the biomes of Cerrado and Amazon Forest. These transition zones tend to shelter biodiversity hotspots (Araújo & Williams 2001, Gaston et al. 2001) and, given the poor information about fungi in these regions, G. hyalinum is an important record to expand the knowledge of this little-known biodiversity. Allied to it, the ecological station is located in an area that suffers from the imminence of deforestation and agricultural expansion. These factors show the urgency of further research that seeks to fill gaps in a body of knowledge that can be extinguished before it is even discovered.

Lodge & Sourell (2015) show the occurrence of numerous species of fungi in southern amazon, corroborating the biodiversity richness of the region. However, such specimens do not have vouchers and morphological descriptions. According to the guide made by those authors, most of the identifications were made through photographs. Although the guide indicates which specimens may occur in the area, it cannot be used as a concrete source of data for occurrence of the species, since the data cannot be confirmed.

Until now, only one species of Geastrum is known for Mato Grosso state (Trierveiler-Pereira et al. 2011). In addition to being a new species for science, this is the second record for the genus throughout the state.
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