BRIEF REPORT

Unusual occurrence of rare Geastrum species with an abnormal stoma development found in Madurai, Tamil Nadu, India [version 1; peer review: 1 approved with reservations]

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Abstract

Unusual occurrence of Geastrum species fungi, belonging to the class Basidiomycetes, found in riverbank regions of Madurai, Tamil Nadu, India. Its detailed illustrations and morphological characterization analysis are reported in this study.

Keywords

Basidiomycetes, Geastrum and Characterization

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Author roles: Karthikeyan V: Conceptualization

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Introduction
The species of the genus *Geastrum*, classified under order Geastrales, is rare and the species are red listed (Piętka & Kujawa in 2012; Wojewoda et al., 2006). Although in the distant past they were recorded in several locations worldwide, sightings of this attractive earthstar in plains are now highly limited due to anthropogenic activity. Kirk et al. (2008) and Perez (2009) described 50 species of *Geastrum* in a dictionary of fungi. Zamora et al. (2014) described 100 to 120 species of *Geastrum* based on field and herbarium studies.

There have been many reports about the pluralistic distribution of *Geastrum* species in Western Ghats by Karun & Sridhar, 2014, in Central India by Verma et al. (2018) and in Tamil Nadu by Meenakshi & Selvam (2020). *Geastrum* species share morphology with *Myriostoma* species, as reported by Alexov et al. (2012); Esqueda (2009); Outcoumit et al. (2009) and Pawłowski & Adamska (2008).

Many mycologists classify *Myriostoma* in between Geastraceae and Astraeaceae. However, currently it is classified under Geastraceae (Dring, 1973; Sunhede, 1989). Using rDNA and other genetic markers, its molecular taxonomic position within Geastraceae was confirmed, and it is closely associated with the genus of *Geastrum* (Hosaka et al., 2006; Krüger et al., 2001).

These inedible fungi are widely distributed in Africa, Asia, North and South America, and Europe, where they grow in humus-rich forests. *Geastrum* are rather rare fungi; they have been characterized as critically endangered, red listed and threatened in many European countries like Poland, Montenegro, Bulgaria, Slovakia, Sweden, Switzerland and Czech Republic (Wojewoda et al., 2006 and Piętka & Kujawa, 2012). Consistent and periodical surveys of macrofungi result in identification of new fungi. The present objective of this work is to report the *Geastrum* species found in the riverbank canal of Madurai, Tamil Nadu, India with an abnormal stoma development.

Methods
Specimen collection
The specimens were collected from our college campus: Thiagarajar college, Teppakulam, Madurai, Tamil Nadu, India. The soil texture of the collection site is highly humus and moist, with shady regions provided by large trees in the nearby area. The collected specimens were photographed, subcultured in potato dextrose medium (pH 6.5) and preserved in formalin bottles (10%).

Specimen storage and analysis
Collected specimens were dried using a glass desiccator and analysed morphologically and anatomically. Morphological characters were observed using a stereomicroscope (Labline, model number 3923061230; 20X & 40X with flat field) and measurements were taken with a ruler. Microscopic features (e.g., basidiospores, capillitium, hyphae of peridium) were studied under light microscopy at high magnification (40X). Lacto-phenol (0.1%) mixed with trypan blue was used to study the gleba. Microscopic images were taken using Camera Lucida (HLP-3 Model). Standard specimen identification described by Smith (1963) was followed for the anatomical and morphological features based microscopic examination.

Results and discussion
The specimen (TMC 1001) has been deposited in the collection of Gasteroid fungi at the Department of Botany, Thiagarajar college, Teppakulam, Madurai, Tamil Nadu, India.

The fruiting body size measures 40 mm in width by 20 mm in height, is solitary, and consists of an upper stalked spore case and radiating rays below (Figure 1 and Figure 2). The exoperidium has 10–12 rays, which are revolute, thin, united in the base and free at the tips, and triple layered, with a blackish brown mycelial layer (Figure 3). The tips curve towards the basal position. The peridium is bilayered. The endoperidium body

Figure 1. *Geastrum* dorsal view.

Figure 2. *Geastrum* ventral view.
spore case is greyish brown, up to 19 mm in diameter, with many stalks attached. The spore case opens by many pores through which spores escape.

The fungus has brown basidiospores with a globose and verrucose structure. Brown eucapillitium threads are thin walled, straight, aseptate and up to 3μm in diameter. The exoperidium is composed of hyaline, aseptate to rarely septate, unbranched, thick walled, tightly packed hyphae, up to 4μm in diameter, wall thickness up to 1.7 μm. The endoperidium is unbranched, aseptate, brown in colour, with tightly packed hyphae measuring up to 3μm in diameter.

Although these fungi are cosmopolitan in distribution, due to the periodical environmental disruption nowadays, occurrence of these fungi is highly limited. According to Signalarter (2000) and Benkert (2003), the Geastrum species serve as indicators of specific habitats in need of conservation.

Data availability
All data underlying the results are available as part of the article and no additional source data are required.

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I grateful to our beloved management of Thiagarajar College for the permission to carry out this study. I deeply acknowledge to all my students of I-M.Sc., Botany for their valuable help during collection of this rare fungi.
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Open Peer Review

Current Peer Review Status: ?

Version 1

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The presence of more than one peristome and more than one stalk are reported by Sunhede, S. 1989. Geastraceae (Basidiomycotina). Morphology, ecology and systematics with special emphasis on the North European species. Synopsis Fungorum 1: 1-534.

The description need improvements:
1. Add details about Peristome.
2. Add spores measurements.
3. Add photos of microstructures.

Please add some discussions/conclusions.

Is the work clearly and accurately presented and does it cite the current literature?
Yes

Is the study design appropriate and is the work technically sound?
Partly

Are sufficient details of methods and analysis provided to allow replication by others?
Partly

If applicable, is the statistical analysis and its interpretation appropriate?
Not applicable

Are all the source data underlying the results available to ensure full reproducibility?
Yes
Partly

**Competing Interests:** No competing interests were disclosed.

**Reviewer Expertise:** Taxonomy, Sistematics, Mycology.

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard, however I have significant reservations, as outlined above.

Author Response 04 May 2021

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As per the reviewer, we accept the revision required. We here describe the:
1. Peristome size and structure peristome are up to 3μm in diameter.
2. The spore print was not taken at that time we are unable to determine the spore size.

**Competing Interests:** No competing interests were disclosed.