

Ascobolus scatigenus (Berk. & Curtis) Brumm: New to Eastern Uttar Pradesh and Revision of Checklist of *Ascobolus*

Dheeraj Pandey¹, Ovaidd Akhtar^{2,*}, Shweta Chaturvedi³, Mohd. Shahbaan², Ifra Zoomi⁴

Abstract

In this study, Ascobolus scatigenus (Berk. & Curtis) Brumm. was reported for the first time from a newly described habitat in Eastern Uttar Pradesh, India. So far, this coprophilous cup-fungus was known to be reported only from Saharanpur Garden, Sheikhpura, Hyderabad, and Punjab. External morphology and microscopic characteristics were very similar to those of Ascobolus scatigenus (Berk. & Curtis) Brumm. Many apothecial cups of the fungus were growing over the dung-rich surface singly or in groups of 3–5, 8 mm broad, lacking a stalk. These apothecial cups were distinguished into ectal excipulum, medullary excipulum, subhymenium, and hymenium. The hymenial layer consists of fertile asci and sterile paraphyses. Asci were cylindrical with a granular inner wall, a rounded apex, and slightly tapered at the base, containing 8 ascospores. These ascospores were hyaline, fairly obtuse at the poles, with two wall layers. The outer wall layer was smooth without any ornamentation. The other detailed description of this coprophilous fungus, along with measurement data, is presented in this research, and the voucher specimen is wet-preserved in the form of a museum specimen. With the newly described habitat for A. scatigenus (Berk. & Curtis) Brumm. and the revision of the checklist for Uttar Pradesh, only two reports for the genus Ascobolus remain valid from the whole of Uttar Pradesh, India. Thus, the present study is the third report for all the Ascobolus species and the second report of A. scatigenus (Berk. & Curtis) Brumm. from Uttar Pradesh. From Eastern Uttar Pradesh, this is the first-ever report of A. scatigenus (Berk. & Curtis) Brumm. growing on the manured soil of the Naini Region of Prayagraj. This research also presents a revised checklist of Ascobolus species in Uttar Pradesh, India.

Keywords: *Ascobolus*, coprophilous fungus, apothecium, ascus, ascospores

*Author for Correspondence

Ovaidd Akhtar

E-mail: ovaidd.akhtar@hotmail.com

¹Assistant Professor, Department of Botany, Prof. Rajendra Singh (Rajju Bhaiya) University, Prayagraj, Uttar Pradesh, India

²Assistant Professor, Department of Botany, Kamla Nehru Institute of Physical and Social Science, Sultanpur, Uttar Pradesh, India

³Research Scholar, K. Banerjee Center for Atmospheric and Oceanic Studies, Faculty of Sciences, University of Allahabad, Prayagraj, Uttar Pradesh, India

⁴Assistant Professor, Department of Botany, Government Degree College, Manikpur, Chitrakoot, Uttar Pradesh, India

Received Date: September 03, 2026

Accepted Date: September 09, 2026

Published Date: September 11, 2026

Citation: Dheeraj Pandey, Ovaidd Akhtar, Shweta Chaturvedi, Mohd. Shahbaan, Ifra Zoomi. *Ascobolus scatigenus* (Berk. & Curtis) Brumm: New to Eastern Uttar Pradesh and Revision of Checklist of *Ascobolus*. International Journal of Fungi. 2026; 3(2): 1–9p.

INTRODUCTION

The genus *Ascobolus* was introduced in 1796 by the Dutch mycologist Christiaan Hendrik Persoon [1]. This was *Ascobolus pezizoides*, a dung-inhabiting cup-fungus. Since its discovery, many new species, subspecies, varieties, and forms have been described over the years from different regions of the world. This has also led to many erroneous interpretations and identifications, which were rectified by J. Van Brummelen [2] in the monograph “A world monograph of the genera *Ascobolus* and *Saccobolus*”. *Ascobolus* is a saprobic and coprophilous cup-fungus that grows on the dung of cows, buffaloes, horses and sheep [3]. Some of its species also grow on manured and burnt soils [4]. *Ascobolus* is an important fungus involved in nutrient cycling and decomposition [5] and coprophilous succession [6]. The spores of

Ascobolus are highly resistant to degradation due to their thick and melanized walls [7]; therefore, frequently found preserved in fossil records as non-pollen palynomorphs to reconstruct past ecosystems. The gut enzymes of herbivorous animals help to break spore dormancy, therefore, promoting germination upon excretion [5].

It is because of the above important role in driving the ecosystem that *Ascobolus* has attracted the attention of mycologists to study the habitats and systematics across the different parts of the world. Antonin and Moravec [8] described the variation in characteristics of *Ascobolus scatigenus*. Melo et al. [9] reported *Ascobolus elegans* and *A. americanus* from Brazil. Doveri [10] described 19 species of *Ascobolus* from Italy. Niranjana and Sarma [11] described a new species, “*Ascobolus gomayapriya* Niranjana and Sarma, *sp. nov.*,” growing on cow dung near Port Blair, South Andaman, India. Yatoo et al. [12] reported *A. brassicae*, *A. furfuraceus*, *A. foliicola* and *A. sacchariferus* as new to Kashmir, India. These four species were earlier reported by various mycologists from different parts of India (Table 1).

Table 1. A consolidate list of *Ascobolus* sp. reported from India with updated list of *A. scatigenus* (Berk. & Curtis) Brumm. from Uttar Pradesh.

S. N.	Name of the species	Place from where the species is reported	References
1	<i>A. indicus</i>	Delhi	Sanwal [13].
2	<i>A. furfuraceus</i>	India, place not mentioned Sopore, Kashmir	Van Brummelen [2]. Yatoo et al. [12].
3	<i>A. denudatus</i>	Narkanda, Mahasu, Uttarakhand	Thind and Waraich [14].
4	<i>A. brassicae</i>	Varansi, Uttar Pradesh Sopore, Kashmir	Khare [15]. Yatoo et al. [12].
5	<i>A. behnitziensis</i>	Kolhapur, Maharashtra	Ghandge and Patil [16].
6	<i>A. crenulatus</i>	Satara, Maharashtra	Ghandge and Patil [16].
7	<i>A. sacchariferus</i>	Kolhapur, Maharashtra Sopore, Kashmir	Ghandge and Patil [16]. Yatoo et al. [12].
8	<i>A. minutus</i>	Karad Maharashtra	Ghandge and Patil [16].
9	<i>A. hawaiiensis</i>	Kolhapur, Maharashtra	Ghandge and Patil [16].
10	<i>A. immersus</i>	Patna	Ghandge and Patil [16].
11	<i>A. geophilus</i>	Mussoorie hills, Uttarakhand	Ghandge and Patil [16].
12	<i>A. foliicola</i>	Radhanagari, Maharashtra Sopore, Kashmir	Ghandge and Patil [16]. Yatoo et al. [12].
13	<i>A. gomayapriya</i>	Andaman Islands	Niranjana & Sarma [11].
14	<i>A. scatigenus</i>	Saharanpur Garden, Uttar Pradesh	Reported by Gollan (1900), described as <i>A. gollanii</i> by Hennings [17], considered as <i>A. scatigenus</i> by Van Brummelen [2].
		Ladbar, Sheikhpura	Reported by Ahmad (1942), monographed by Van Brummelen [2].
		Hyderabad	Reported by Unknown (1958), monographed by Van Brummelen [2].
		Dalhousie, Punjab	Reported by Batra (1995), monographed by Van Brummelen [2].
		Prayagraj, Uttar Pradesh	Newly described habitat in this research (Voucher specimen: PHKNIPSS-317).

Recently, a species of *Ascobolus* was observed by the author in the agricultural field of the Naini region of Prayagraj in the last week of July 2025. Based on morphological, microscopic, and extensive literature surveys, it was found that, so far, no similar species is described for this region. Therefore, this taxonomic work is done to fully describe this *Ascobolus* species new to eastern Uttar Pradesh of India, and to update the checklist of valid *Ascobolus* so far known from Uttar Pradesh as well as from this country.

MATERIALS AND METHODS

Living Material and Field Observation

Freshly growing apothecia were observed over the manured soil in a suburban field in the Naini region of Prayagraj during the month of July 2025. The coordinates of the study site are 25.4024895N, 81.8827256E. In the field, numerous cups of the fungus were growing over the dung-rich surface. Characteristics related to external morphology were noted in the field diary on 23 July 2025.

Collection and Preservation

Enough samples were collected and fixed in fixative and preserved in the preservative solution. The recipe for the fixative solution for 100 ml is 80 ml of 50% ethyl alcohol (aqueous), 10 ml of formaldehyde (40%), and 10 ml of acetic acid.

After 24 hours of fixation, the cups were transferred to 70% alcohol as a preservative. A few of the samples were processed to study microscopic characteristics, while the remaining were wet-preserved in the form of museum specimens in the specimen jar. The specimen jar is available in the Plant Herbarium of this institute.

Microscopic Examination

Vertical sections (VS) of the apothecial cup were cut by the hand-sectioning method. The sections were mounted on a glass slide. Polyvinyl-Lacto-Glycerol (PVLG) was used as a mounting medium, which was prepared with 16.6 g of polyvinyl alcohol, 100 ml of lactic acid, 10 ml of glycerol, and 100 ml of distilled water. When needed, gentle pressure was applied to spread the section over the glass slide. To enhance the contrast, sections were stained in 0.05% trypan blue (prepared in lactic acid, glycerol, and water in a ratio of 2:2:1). The slides were kept in hot air oven at 50 °C for 24 hours to remove any air bubble trapped under the coverslip. After microscopic examination, the slides were kept in the Plant Herbarium along with the museum specimen. A compound microscope (Olympus Magnus CH20i) was used for all the microscopic observations. All the photomicrographs were captured by using MagVision camera. The scales in the photomicrographs were also inserted in the same software.

RESULTS

- *Ascobolus scatigenus* (Berk. & M.A. Curtis) Brumm., Persoonia, Suppl. 1: 159. 1967 (IF number 326523).
- *Sphaeria scatigena* (Berk. & Broome), Annals and Magazine of Natural History 7: 452. 1861.
- *Hypocopa scatigena* (Berk. & Broome) Sacc., Sylloge Fungorum 1: 243. 1882.
- *Coniochaeta scatigena* (Berk. & Broome) Cain, Studies of *Coprophilous Sphaeriales* in Ontario: 62. 1934.
- *Ascobolus notatus* (A.C. Batista & A.F. Vital), Anais Soc. Biol. Pernambuco 13(2): 61. 1955. ≡ *Ascobolus magnificus* (B.O. Dodge), Mycologia 4(4): 218. 1912.

Systematic Position

The systematic of taxon is in accordance with Species Fungorum, 2026 (www.speciesfungorum.org).

- Fungi.
- Dikarya.
- Ascomycota.
- Pezizomycotina.
- Pezizomycetes.
- Pezizomycetidae.
- Pezizales.
- Ascobolaceae.
- *Ascobolus*.

Habitat

Coprophilous, growing on manured soil in a suburban agricultural field.

Material Examined

Many apothecial cups.

Voucher Specimen

The voucher specimen was submitted to the Plant Herbarium of the Department of Botany, Kamla Nehru Institute of Physical and Social Sciences (PHKNIPSS), Sultanpur, Uttar Pradesh – 228118, India, with accession number PHKNIPSS-317. A complete macro-morphological as well as microscopic detail of the fungus studied is presented below.

Apothecium

Apothecia single or in groups of 3–5 (Figure 1a, b), round when single, irregular in groups, maximum 8 mm broad, sessile, lacking a stalk, cup-shaped, shallow upper surface (Figure 1a), smooth margin when young, slightly ruptured at maturity, slightly wrapped inward, hymenial surface olivaceous yellow colored when young (Figure 1a), tan–brown at maturity (Figure 1b), hymenial surface smooth when young (Figure 1a), showing yellow–green color, which disappeared upon fixation in fixative. Surface becomes slimy when mature (Figure 1b), dust particles collected, outer surface smooth when young (Figure 1a), become slight velvety at maturity, beige color (Figure 1a). In cross section (Figure 2a), cup distinguished into ectal excipulum (Figure 2b), medullary excipulum (Figure 2d), subhymenium (Figure 2e) and hymenium (Figure 3a).



Figure 1. Apothecia of *A. scatigenus* (Berk. & Curtis) Brumm. in the habitat. (a): Young developing and fully developed cup showing olivaceous yellow colored hymenium, (b): Mature apothecial cups in group showing tan–brown colored hymenium. bar = 1.0 cm.

Ectal Excipulum

Maximum 313 μm in depth, cells compactly arranged (Figure 2b), hyphoid, hyaline to light brown color, loosely tangled hyaline hyphoid hairs 5–7 μm wide, textura epidermoidea, emerge on the outer surface for the anchorage of cup to the substratum, entangled with sand particles and blue–green alga (Figure 2c).

Medullary Excipulum

Maximum 1260 μm in depth, cells in loose to compact arrangement, globose when loose, textura globulosa, hyaline to light brown color (Figure 2d).

Subhymenium

Maximum 160 μm in depth, hyaline to light brown color, cytoplasm granular, reactive to trypan blue stain (Figure 2e).

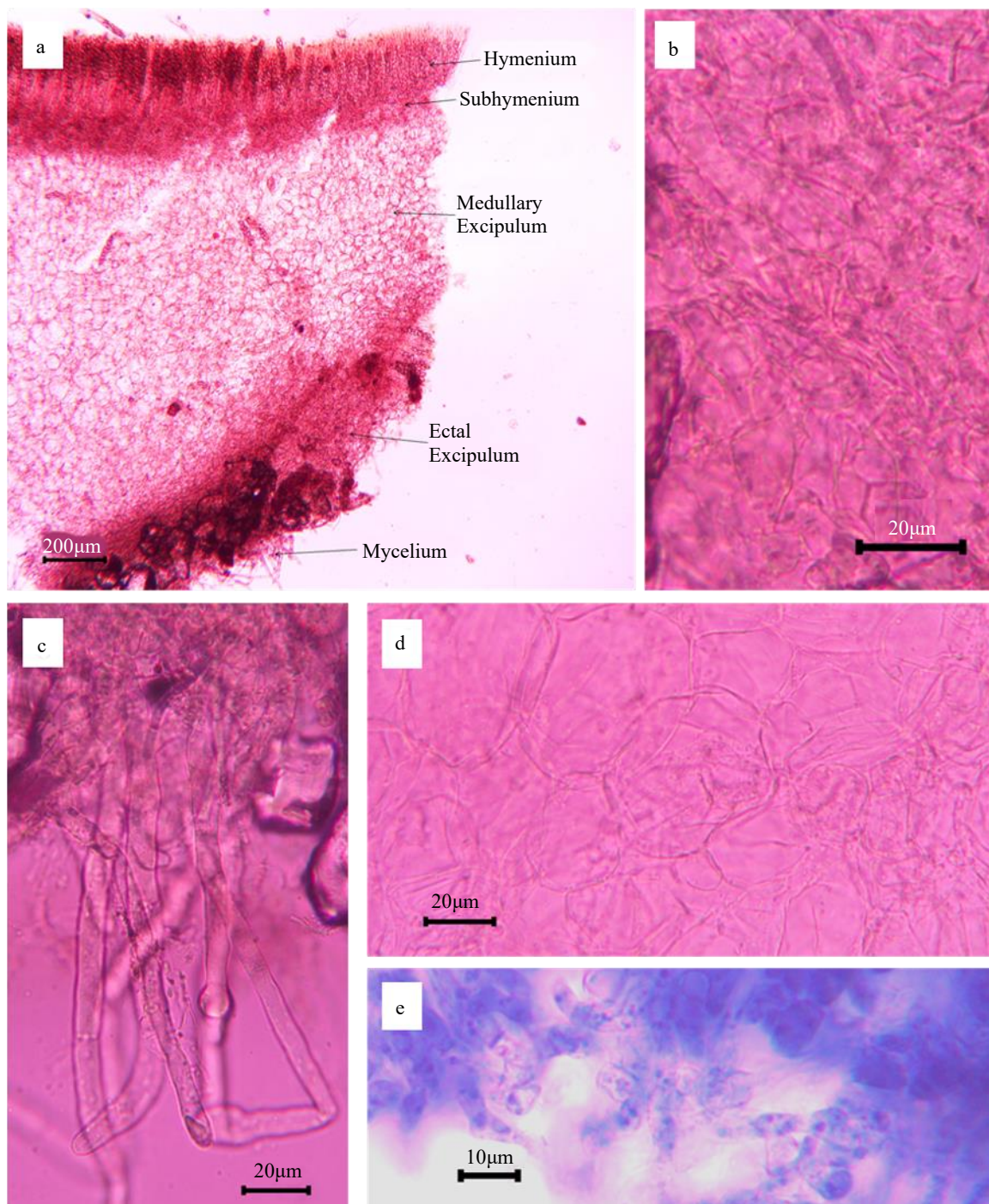


Figure 2. Photomicrographs of V.S. of the apothecia of *A. scatigenus* (Berk. & Curtis) Brumm. (a): V.S. of the half cup showing various tissue differentiation, (b): Cells of the ectal excipulum, (c): Hyphoid hair, (d): Medullary excipulum, (e): Subhymenium (stained in trypan blue).

Hymenium

Maximum 270 µm in depth, consists of fertile asci and sterile paraphyses (Figure 3a).

Ascus

Cylindrical 198–250 µm × 20–24 µm, inner side of wall granular, rounded apex, slightly tapered the base, 8-spored (Figure 3b, c), operculate (Figure 3d).

Paraphysis

205–232 μm long, cylindrical, filiform, 4 μm thick, slightly swollen at apex up to 6 μm , hyaline, reactive to trypan blue (Figure 3e), septate, sometimes irregular wall (Figure 3f).

Ascospore

8 in number, obliquely arranged in ascus (Figure 4a), shape oblong–ellipsoid, measuring 24–26 μm $\times$ 11–13 μm , hyaline, fairly obtuse at the poles (Figure 3b), wall 1.0 μm thick bilayered, outer wall layer smooth without ornamentation (Figure 3c).

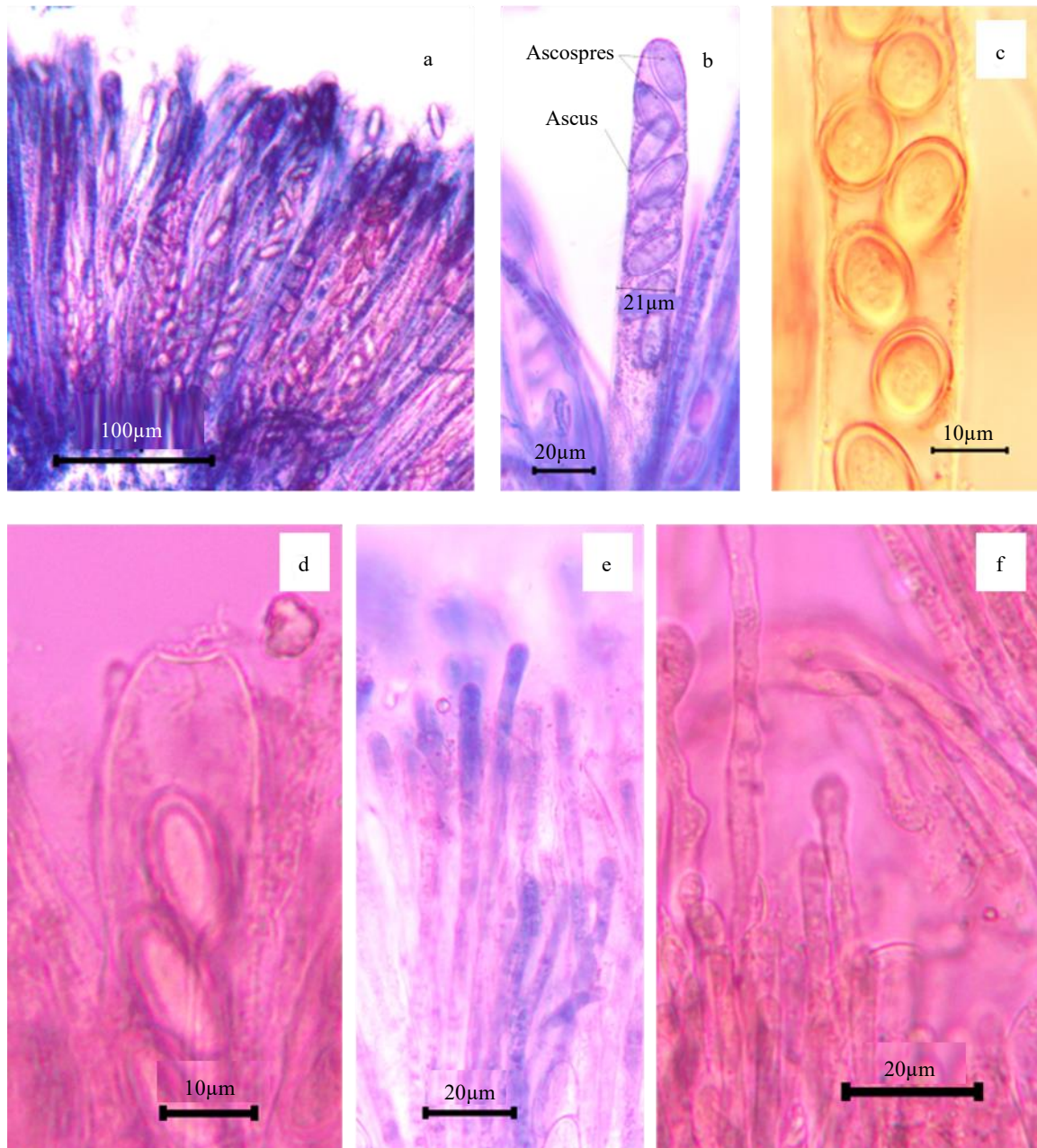


Figure 3. Photomicrographs of V.S. of hymenial layer of *A. scatigenus* (Berk. & Curtis) Brumm.

(a): V.S. of hymenium showing fertile asci intermingle with sterile paraphyses, (b): A single ascus showing obliquely arranged 8-ascospores, (c): Ascospores showing wall characteristics without stain, (d): Operculum at the tip of mature ascus, (e): Paraphyses showing swollen tips reactive to trypan blue stain, (f): Paraphyses showing septa and irregular wall.

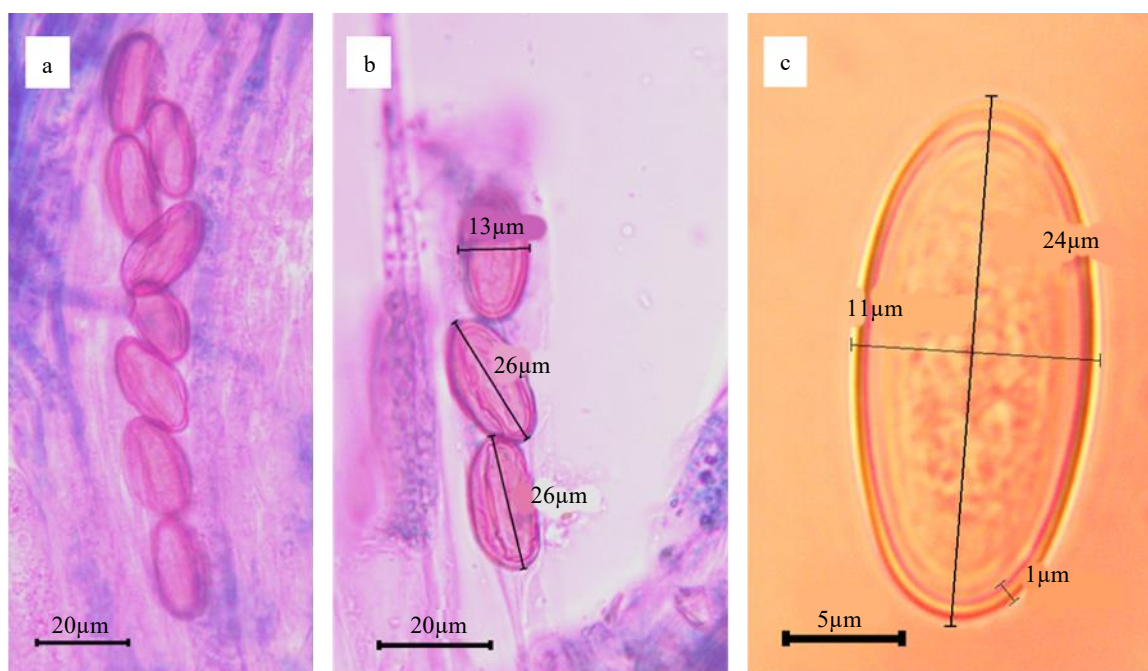


Figure 4. Photomicrograph of the ascospores of *A. scatigenus* (Berk. & Curtis) Brumm. (a): 8-ascospores inside a single ascus, (b): Inner wall layer of the ascospores is shrunken in the mounting medium, (c): Wall characteristics of a single ascospore.

DISCUSSION

Ascobolus scatigenus (Berk. & Curtis) Brumm., comb. nov., is well-described by J. Van Brummelen in his monograph [2] (IF number 326523). The shape, size and color of apothecium (Figure 1a, b), details of the cross sections of hymenium (Figures 2a and 3a), subhymenium, medullary excipulum, ectal excipulum (Figure 2a–e), asci and paraphyses (Figure 3a–f) are almost similar to the characteristics described for *A. scatigenus* by J. Van Brummelen [2]. Yellow–green color of upper surface of hymenium was observed at the time of collection, which was vanished with fixation in the fixative. Antonín and Moravec [8] observed the fast-changing color of this fungus at the time of spore discharge, and they have described this behavior as “chameleon-like changing of color”. Number, arrangement of ascospores in the asci (Figure 4a, b) are also in line with the Van Brummelen [2]. Shape and size of mature ascospores are, however, constant (Figure 4b, c). Further under light microscopic observation wall of the ascospores had two layers and the outer appeared smooth with one to two longitudinal or oblique fissure [2]. In the samples studied in the research, however, no fissure could be seen (Figure 4a–c). This may be due to the impact of the external environment or edaphic factors. Phenotypic plasticity is also a very common phenomenon exhibited by most of the fungi. Therefore, variation in the longitudinal fissure alone cannot be considered to create a new species. Also, morphological observation in the field (Figure 1) as well as microscopic examination of every part of the apothecium (Figures 2 and 3) and detailed characterization of the ascospores (Figure 4) are so clear that there is no further need for molecular characterization. However, to create a DNA barcode of the *A. scatigenus*, ITS-sequencing could be done soon.

“A world monograph of *Ascobolus* and *Saccobolus*” was published by Van Brummelen [2], which included four species of *A. scatigenus* from India. Recently, Yattoo et al. [12] published a checklist of the *Ascobolus* and *Saccobolus* reported from India. This included 15 species of *Ascobolus* reported from the Indian subcontinent. According to this checklist, *A. scatigenus* has been reported from Sheikhpura, Dalhousie, Hyderabad and Saharanpur Garden by Van Brummelen [2] and from Uttar Pradesh by Leelavathy and Flower [18]. In the original monograph by Van Brummelen [2], he has listed the specimens of *A. scatigenus* reported by (i) Ahmad from Ladbar, Sheikhpura in 1942, (ii) Batra from

Dalhousie, Punjab in 1955, (iii) Gollan from Saharanpur Garden in 1900, and (iv) an unknown collector from Hyderabad in 1958. Accordingly, *A. scatigenus* has been reported from two places in Uttar Pradesh, one from Saharanpur (listed by Van Brummelen) and another by Leelavathy and Flower [18]. As per the reference provided in Yatoo et al. [12], Leelavathy and Flower [18] published “Additions to South Indian Agaricales–I” in the journal “Kavaka” and reported three genera, viz., *Alboleptonia stylophora*, *Pouzaromyces lasius* and *P. myodermus* only. All these three were gilled mushrooms and the inclusion of this research in the list of *Ascobolous* is thought to be erroneous; therefore, omitted from the checklist. In view of the above facts, a revised checklist is presented in Table 1, mentioning only two reports of *A. scatigenus* from Uttar Pradesh with a newly described habitat from the Prayagraj of Eastern Uttar Pradesh.

Other species of *Ascobolus* recorded from various parts of India are listed in Table 1. From Uttar Pradesh, India, holotype S-A517 was collected by Gollan from Saharanpur Garden and described as *A. gollanii* by Hennings [17]. Another species, *A. brassicae* Crouan was reported growing on dung of various animals and on rotten stems of cabbage by Khare in 1976 [15]. Khare [15] reported this species growing on the soil behind Banaras Hindu University Press on 28 January 1968. He has deposited the specimen BHUPP 680 to the Banaras Hindu University Plant Pathology Herbarium. The holotype of *A. gollanii* (S-A517) originally described by Hennings [17] was considered to be *A. scatigenus* by Van Brummelen [2].

CONCLUSION

With the newly described habitat for *A. scatigenus* (Berk. & Curtis) Brumm. and the revision of the checklist for Uttar Pradesh, only two reports for the genus *Ascobolus* remain valid from the whole Uttar Pradesh of India. Thus, this is the third report for all the *Ascobolus* species and the second report of *A. scatigenus* from Uttar Pradesh. From Eastern Uttar Pradesh, this is the first-ever report of *A. scatigenus* growing on the manured soil of the Naini Region of Prayagraj. This new report and revision of the checklist of the genus *Ascobolus* of Uttar Pradesh opens the vistas to visit the holotype and revise the genus *Ascobolus* reported from the entire regions of the Indian subcontinent.

Acknowledgment

All the authors are thankful to Dr. Manoj Kumar, in charge of the Plant Herbarium at Kamla Nehru Institute of Physical and Social Sciences (PHKNIPSS), for submitting the wet-preserved apothecium cups along with permanent slides as voucher specimen. Ovaaid Akhtar is thankful to the Kamla Nehru Institute of Physical and Social Sciences for providing laboratory facilities. Dheeraj Pandey is thankful to Prof. Ashutosh Kumar Singh and Prof. N. B. Singh, Prof. Rajendra Singh (Rajju Bhaiya) University. All the authors acknowledge Dr. Achuyta Nand Shukla, Additional Director/Scientist-E, Ministry of Environment, Forest and Climate Change, for his valuable suggestions during the investigation.

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