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# **Original Research Article**

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# Diversity and Distribution of *Calvatia* Species in India: A New Record from Central India

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# ABSTRACT

### Keywords

Calvatia, Calvatia pyriformis, Agaricaceae (Agaricales)

Article Info

Accepted: 18 August 2018 Available Online: 10 September 2018 An account of mushrooms belonging to *Calvatia* reported from different part of India is given. Total 16 species of were compiled from literature with their records of habitat, distribution and references. Jammu and Kashmir represents the maximum diversity of *Calvatia* and 5 species were reported followed by Himachal Pradesh (3 species), Assam, Madhya Pradesh and Uttarakhand (2 species each), West Bengal and Meghalaya (1 species each). One species *Calvatia pyriformis* was recorded for the first time from sal forest of central India.

# Introduction

*Calvatia* belong to family Agaricaceae (Agaricales) is a mushroom forming fungi. The members of this family are commonly known as 'puff balls'.

There are 140 records of Calvatia species available on website, out of them 58 are valid names under the genus (http://www. indexfungorum.org). Genus *Calvatia* was erected by Swedish mycologist, Elias Magnus Fries as early as in 1849, which includes a single species, Calvatia craniiformis and it was originally described as *Bovista* craniiformis Schwein (Fries, 1849). Most species in the genus are edible when young,

but some species for example, *Calvatia fumosa*, has a very pungent odour and should be avoided. Many wild species are collected and consumed around the world although species identified in the field and safely eaten vary widely from country to country. *Calvatia gigantean* (Batsch) Lloyd (giant puffball) and *Calvatia utriformis* (=*Bovistella utriformis* (Bull.) Demoulin & Rebriev) were reported as edible (William and Arora, 2008).

Several studies have been conducted on taxonomic works on the genus (Lange, 1993; Zeller and Smith 1964; Morales and Kimbrough, 1978; Martín, 1997). Some workers classify *Calvatia* as a species complex (Kreisel, 1989, 1992). Studies on

molecular phylogeny (Larsson and Jeppson, 2008; Bates *et al.*, 2009) have not been decisive in the elucidation of the relationship of the monophyly of *Calvatia*. Recently many macro-fungi from central India have been reported including some edible, ecto mycorrhizal and saprophytic mushrooms (Verma *et al.*, 2016a-c; Verma and Verma, 2017; Verma *et al.*, 2017a-e; Verma *et al.*, 2018a-b; Verma and Pandro2018a-b).

The present article reports distribution of 16 mushrooms belonging to genus *Calvatia* and reported from 8 states of India. Two species namely, *Calvatia cyathiformis* was recorded for the first time from central India (Madhya Pradesh) while and *Calvatia pyriformis* is new fungal record from India.

# Materials and Methods

Specimens of *Calvatia* mushrooms were collected from Jabalpur and Chada, Dindori, Madhya Pradesh during June-July. Some parts of collected samples were preserved in 70% alcohol just after collection for microscopic study. The fruit bodies of fungi were dried under the sun or in the wooden box lighted with 100W electric bulb. Microscopic slides were prepared by using stain, mountant, clearing and softening chemicals.

Slides were observed under advanced research microscope (Leica, Germany) using 5x, 10x, 20x, 40x objectives and 10x and 15x eyepieces. Observations under phase contrast and dark field were also made whenever required. Photomicrography was done with the help of a digital camera (make, Leica) attached to the advanced microscope.

Identification of fungi have been done with the help of published literature (Alfredo *et al.*, 2014; Bisht *et al.*, 2006; Chakraborty *et al.*, 2012; Dorjey *et al.*, 2016; Gogoi and Prakash, 2015; Gupta *et al.*, 1974; Khare, 1976; Kreisel, 1992; Syed Abrar *et al.*, 2008; Thind and Thind, 1982; Zeller *et al.*, 1964).

### **Results and Discussion**

### Taxonomic description

# Calvatia cyathiformis (Bosc) Morgan (Figs. 1-8)

*≡Lycoperdon cyathiforme* Bosc Mag. *Gesell.naturf. Freunde, Berlin* 5: 87 (1811)

=*Calvatia cyathiformis* (Bosc) Morgan, *J. Cincinnati Soc. Nat. Hist.* 12(4): 168 (1890) subsp. *cyathiformis* 

*=Lycoperdon fragile* Vittad., *Mém. R. Accad. Sci. Torino*, Ser. 2 5: 180 (1843)

*=Utraria fragilis* Quél., *Compt. Rend. Assoc. Franç. Avancem. Sci.* 14(2): 451 (1886)

=Calvatia fragilis (Quél.) Morgan, J. Cincinnati Soc. Nat. Hist. 12(4): 168 (1890)

=*Calvatia cyathiformis* subsp. *fragilis* (Quél.) Dring, *Mycol. Pap.* 98: 38 (1964)

*=Calvatia cyathiformis* f. *fragilis* (Quél.) A.H. Sm., *Lloydia* 27: 150 (1964)

Cyathiformis means flask shaped or like a flask. It is growing alone, scattered, gregariously in amongst grasses and pastures. Fruiting Body: 7-7.5 cm high and 6.5 cm wide when mature; ball-shaped when young, but soon developing a thick basal portion that is slightly more narrow than the upper portion, globose to turbinate or subpyriform, tapering abruptly into a large, well-developed, thick, stout rooting base, deeply wrinkled. Outer Surface: Tan to pale brown; the pigment breaking up into small, mosaic-like scales; eventually becoming very pale brown to gravish or nearly white, with a vague mosaic of sections punctuated by brownish dots; dry; the skin 1-2 mm thick. Interior: White and

firm when young; soon becoming twochambered texturally, with the basal portion distinct from the upper portion; upper portion becoming yellowish and finally deep brownish purple as it matures and turns into spore dust; basal portion turning yellowish, then olive with age. Spore dust, purple. Basidiospores 3-6  $\mu$ m (including ornamentation); globose; covered with spines 0.5-1  $\mu$ m long; hyaline in KOH; brownish in Melzer's reagent. Capillitial threads 2-5  $\mu$ m wide; globose to subglobose, echinulate, walls about 0.5  $\mu$ m thick, flexuous or straight, sparsely branched, slightly; hyaline in KOH; smooth or very minutely pitted; a little narrowed at septa.

Diagnostic characters include its habits of occurrence in grassy areas, a ball shaped when young, which on maturity develop a basal portion like inverted pear or a loaf of bread. The surface of mushroom can be fairly smooth and pale or, more commonly, brown and broken up into mosaic-like scales. It is similar species includes: *Calvatia fragilis, C. gigantean* and *C. craniiformis.* 

### **Collection examined**

On ground amongst herbaceous weeds and litter, TFRI campus, Jabalpur, Madhya Pradesh, 17/06/2013, Tropical Forest Research Institute TF-3434.

# Calvatia pyriformis (Lév.) Kreisel (Figs. 9-18)

### *≡Hippoperdon pyriforme* Lév. 1846

Fruit bodies roughly pear shaped, 7-9cm tall and 6 cm broad, very short false stalk, wrinkled at base and lower side of the cap, outer wall is first white then pale yellowish to grayish brown, scurfy surface breaking up in to polygonal areas, inner wall papery grayish brown, the upper part breaking away to expose the spore mass. Hyphae 2.5–6.2 $\mu$ m, swollen up to 5-15  $\mu$ m. Basidiospores globose to lemon shaped, centrally guttulate, olive – light brown, smooth, 3-5 x 2.5-3.5  $\mu$ m. Capillitial threads 2.5-10 $\mu$ m wide.

# Fig.1-2 Calvatia cyathiformis (1) habit (2) a single fruit body in side view



**Fig.3-4** *Calvatia cyathiformis* (3) preserved specimen in 70% alcohol and (4) cross section of preserved specimen



**Fig.5-6** *Calvatia cyathiformis* (5) hyphae and basidiospores (6) enlargement of above showing a basidium and basidiospores



**Fig.7-8** *Calvatia cyathiformis* (5) hyphae with pattern of capillitium characters and basidiospores (from preserved specimen)



7 8 **Fig.9-10** *Calvatia pyriformis*. 9 habit, 10 top view showing polygonal areas



Fig.11-12 Calvatia pyriformis. 11 fruit body with stalk, 12 over mature fruit body showing disintegrating stalk





Fig.13-14 Calvatia pyriformis. 13 preserved fruit body with stalk, 14 different types of hyphae

Fig.15-18 Calvatia pyriformis. 15-17 capillitium character, 18. Basidiospores







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Fig.19 Distribution of Calvatia species in different states shown on map of India with blackdots

Table.1 Distribution of Calvatia mushroom in India

S. No.	Name of fungus	Habit	Distribution	Reference
1.	Calvatia bovista (L.) Pers.	Onhumicolous soil, scattered forests of <i>Salix</i> <i>alba</i> and <i>Populusnigra</i>	Sumoor area, Nubra, Leh, Ladakh, Jammu and Kashmir	Dorjey <i>et al.,</i> (2016)
2.	<i>Calvatia caelata</i> var. <i>caelata</i> (Bull.) Morgan	Onground	Babeh pass and Bashahar state, HP	Ahmad (1941a)
3.	<i>Calvatia candida</i> (Rostk.) Hollós	From soil	Varanasi, UP	Khare (1976a)
4.	Calvatia craniiformis (Schwein.) Fr. ex De Toni ≡Bovista craniiformis Schwein.	In open grounds and meadows	Shanthi Nagar, Simoga, Karnataka	Syed Abrar <i>et</i> <i>al.</i> , (2008)

5.	<i>Calvatia cyathiformis</i> (Bosc) Morgan	From grassy soil; growing in grassy, moist and shady place; it is edible when young	Ahmedabad, Gujarat; Jorhat, Assam; Sankrail block, Howrah, West Bengal	Rao (1964); Khare (1976a);Gogoi and Prakash (2015); Chakraborty <i>et</i> <i>al.</i> , (2012); Present article
6.	Calvatia excipuliformis (Scop.) Perdeck=Calvatia saccata (Vahl) Morgan ≡Lycoperdon saccatumVahl	Onthe ground	Mussoorie, Uttarakhand; Baramulla, Jammu & Kashmir	Butler and Bisby (1931) Ahmad (1942)
7.	Calvatia fenzlii(Reichardt) Kawam. ≡LasiosphaerafenzliiReichardt	On manure	Unknown, India	Butler and Bisby (1931)
8.	Calvatia fragilis(Quél.) Morgan	On humicolous soil in coniferous forest	Rajgadh, HP	Gupta <i>et al.</i> , (1974)
9.	<i>Calvatia gardneri</i> (Berk.) Lloyd ≡ <i>Lycoperdongardneri</i> Berk.	On ground	Khasi Hills, Meghalaya	Lloyad (1904- 1919)
10.	Calvatia gigantea(Batsch) Lloyd ≡Lycoperdon giganteumBatsch	On ground	Sonamarg, Kashmir Kohima, Nagaland	Ahmad (1941a) Bhaben <i>et al.</i> , (2011)
11.	Calvatia lilacina(Mont. & Berk.) Henn. ≡Bovistalilacina Mont. &Berk.,	Onsandy soil	Gurdaspur, Punjab	Ahmad (1941b)
12.	Calvatia longicauda (Henn.) Lloyd	Onsoil	FRI campus, Dehradun, Uttarakhand	Bisht <i>et al.</i> , (2006)
13.	<i>Calvatia lycoperdoides</i> A.H. Sm.	Growingsolitary- gregarious in coniferous forests,	Bhadarwash, Athkhar, Jammu & Kashmir	Kumar and Sharma (2011a)
14.	Calvatia pachyderma(Peck) Morgan ≡Lycoperdon pachydermumPeck	Onhumicolous soil in coniferous forest	Khadrala, HP	Gupta <i>et al.</i> , (1974)
15.	Calvatia pyriformis (Lév.) Kreisel	On ground in sal forest	Chada, Dindori, Madhya Pradesh	Present article
16.	<i>Calvatia rubroflava</i> (Cragin) Lloyd	growing on humicolous scattered mixed forests of conifers and broad leaved trees	Kishtwar, Padder, Jamuu & Kashmir; from Jorhat, Assam	Kumar and Sharma (2011a); Gogoi and Vipin (2015)

# **Collection examined**

On ground in sal forest, Chada, Dindori, Madhya Pradesh, 24/07/2017, Tropical Forest Research Institute TF 3989.

The 16 mushrooms belonging to genus Calvatia were recorded from 8 different states of India. Out of them, five species were recorded from Jammu and Kashmir these include: C. bovista, C. gigantea, C. lycoperdoides, C. rubroflava and C. saccata (Ahmad, 1942; Dorjey et al., 2016; Kumar and Sharma, 2011a). Three species (C. caelata var. caelata, C. pachyderma and C. fragilis), were recorded from Himachal Pradesh (Ahmad, 1941a; Gupta et al., 1974). Two species each were reported from Assam and Uttarakhand these are: C. cyathiformis, C. excipuliformis rubroflava, С. and С. longicauda (Gogoi and Vipin, 2015; Kumar and Sharma, 2011a; Butler and Bisby, 1931; Bisht et al., 2006). One species each of Calvatiawas recorded from Puniab. Meghalaya and UP these are C. lilacina, C. candida, C. gardneri, respectively (Ahmad, Llovad, 1904-1919; 1941b; Khare, 1976a) while one species, C. fenzlii was reported from unknown place of India (Butler and Bisby, 1931) (Table 1).

Calvatia cyathiformis is distributed mostly in prairie or grasslands in North America, Illinois, Québec and Australia. It is edible and steroids were also isolated from this mushroom. While the second species described above, C. pyriformis is distributed in tropical forests of Africa, Mauritius, Sri Lanka, Java, Japan, Indonesia, Mexico, Panama, Costa Rica, Ecuador, Peru and Bolivia.

Species of *Calvatia* were recorded from different types of soils and substrates including, humicolous soil, grassy soil, sandy

soil, on manure, on soils of moist and shady places. The main habitat of this mushroom reported in literature includes forests of *Salix alba*, *Populus nigra*, broad leaved forests, coniferous forest and sal forest.

Some of Calvatia species were reported to be edible as long as the inside is pure white and it should be consumed within 24 hours of harvest. The fruit bodies of *Calvatia* cyathiformis are edible when voung (Chakraborty et al., 2012). An account on ethno-myco-medicines from central India was reported (Rai et al., 1993). Two steroids including. Calvasterols А  $(14\alpha$ hydroxyergosta-4, 7, 9, 22-tetraen-3, 6-dione) and B (9a, 14a-dihydroxyergosta-4, 7, 22trien-3, 6-dione) and a novel dimeric steroid and calvasterone were detected from the fungus, Calvatia cyathiformis (Kawahara et al., 1993; 1994, 1995). Calvacin a new antitumor agent was also reported (Roland et al., 1960). Ethnomycology and biotechnological potential species of genus *Calvatia*, namely С. cyathiformis, С. craniiformis, С. excipuliformis, С. gigantean С. and utriformisis reviewed and reported that these are sources of food and traditional medicine (Coetzee and van Wyk, 2009). Indigenous knowledge of ethnic tribes for utilization of wild mushrooms including C. gigantea as food and medicine in Similipal biosphere reserve, Odisha was studied by Sachan et al., (2013). This species was also tested for medicinal compounds and was determined that extracts of this species contains calvacin which is a strong tumor-retarding substance present in immature basidiocarp (Roland et al., 1960).

Mushrooms of genus *Calvatia* were distributed in 8 states of India (Assam, Himachal Pradesh, Jammu & Kashmir, Madhya Pradesh, Meghalaya, Uttarakhand and West Bengal). Jammu & Kashmir represents the maximum species diversity. Two species namely, *Calvatia cyathiformis* and *Calvatia pyriformis* were recorded for the first time from central India (Madhya Pradesh).

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