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BOLETUS SHARMAE, A NEW SPECIES FROM SIKKIM (INDIA)

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ABSTRACT

Boletus sharmae, characterized by red-brown coloured subvelvety to velvety (when dry) or sticky (when wet) pileus, yellowish tube with orange-red pore surface (tube-mouth), red stipe with red reticulum on upper half, a distinct yellowish stipe-base and basidiospores of $9.0\text{--}13.2 \times 4.0\text{--}5.8 \mu\text{m}$ is described as new to science with supporting illustrations. It is compared with its allied Indian and extralimital taxa.

Keywords: *Boletus sharmae*, *Boletaceae*, *Macrofungi*, *Taxonomy*, *Sikkim*, *India*

INTRODUCTION

The North-district covering 4226 sq km is the largest amongst the four districts of Sikkim, one of the small Himalayan states in India. It is surrounded by vast stretches of Tibetan plateau on the North, Nepal in the West, Bhutan in the east and three other adjoining districts of Sikkim (West-, South- and East-district) lie to its South. Since 2009 thorough macrofungal survey of this district had been undertaken by the senior author (KD) mainly focusing the temperate to subalpine forested areas and a couple of novel or interesting taxa has already been reported (Das *et al.*, 2012, 2013; Das, 2013a & b; Das and Chakraborty, 2013; Das and Zhao, 2013; Van *et al.*, 2012). Recently, while undertaking a routine survey to this district the senior author came across a species from two different localities: 1) Dombang valley, dominated by coniferous ectomycorrhizal trees like species of *Picea*, *Abies*, *Tsuga*, *Larix*, *Juniperus*; 2) between Yumthang and Shibmandir, dominated by *Abies* sp. After thorough examination of macro- and micromorphological features followed by literature study this species appeared as an undescribed taxon and proposed here as *Boletus sharmae*.

MATERIALS AND METHODS

Macromorphological characters were recorded from the fresh basidiomata in the field and basecamp. After recording the macromorphological characters, basidiomata were dried with a field drier. Photographs of these fresh and dry basidiomata were taken with the aid of Nikon D300s, Olympus C-5060 and DFC550 (attached with Leica S8APO) microscope. Colour codes and terms (mostly) are after Methuen Handbook of Colour (Kornerup and Wanscher, 1978).

Micromorphological characters were noted with the help of a light microscope: Olympus CX 41 from dry samples mounted in a mixture of 5 % KOH and phloxin, 30 % glycerol, and Melzer's reagent. Spore-measurements were recorded in side view from twenty basidiospores. Spore-size measurements and length/width ratios (Q) are given as: minimum–mean–maximum. Herbarium name is after Holmgren *et al.*, (1990). Scanning Electron Microscope (SEM) images of basidiospores were obtained from dry spores that were directly mounted on a double-sided adhesive tape pasted on a metallic specimen-stub and then scanned with gold coating of 5 nm at different magnifications in high vacuum mode (20 KV) to observe patterns of spore-ornamentation. SEM work was carried out with a ZEISS EVO 40EP model imported from Germany and installed at Wadia Institute of Himalayan Geology, Dehradun, India.

Taxonomy

Boletus sharmae K. Das & D. Chakraborty *sp. nov.*, Figures 1-3

Mycobank: MB 807633

Etymology: In recognition of Jai Ram Sharma for his contribution to Indian mycoflora.

Pileus 80–105 mm diam., convex when young, becoming plano-convex to slightly uplifted with maturity, surface sub-velvety to velvety when dry, viscid or sticky when wet, violet brown (11E7–10F8), gradually

Research Article

paler towards margin up to brownish red (10D7 to 10D8) or reddish brown (9D8) and finally more pale to orange red (8A6) at margin or reddish brown entirely, turning blue black immediately after bruising, brownish orange (5C5) with the application of KOH and unchanging with FeSO₄; margin entire, decurved with a narrow flap of sterile tissue.

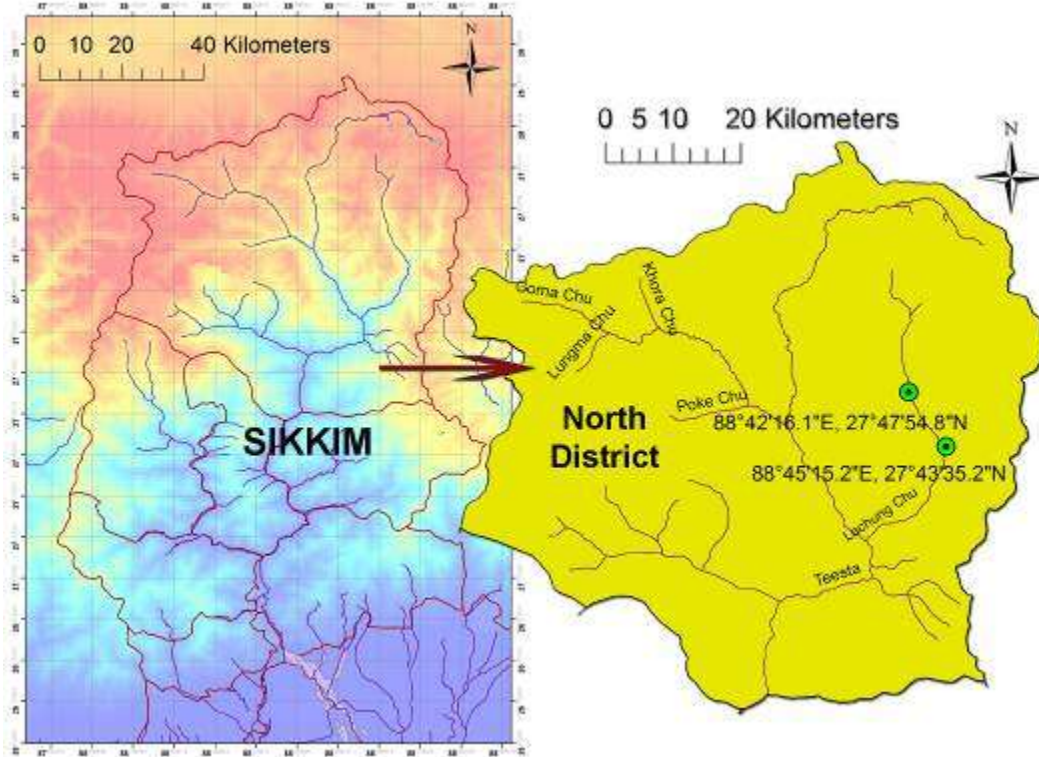


Figure 1: A map showing the geographical distribution of *Boletus sharmae* in the North district of the state of Sikkim (India).

Pore surface (tube-mouth) depressed near the juncture of stipe, light orange to orange, becoming brownish red (10D7) to greyish red or brownish violet (11D8) with age, turning blue black immediately after bruising; pore rounded, 2–3/mm. Tube sinuate, up to 13 mm long, yellowish when exposed, becoming greenish to bluish gradually. Stipe 80–100 × 18–23 mm, cylindrical with slightly bulbous base or subclavate, surface striated (up to half of the length), striations often anastomosing forming reticulation or net pattern, lower half almost smooth, light yellow (3A5) to yellow (3A6) at the juncture (of hymenophore and stipe), then red (10B8) to deep red (11C8) reticulum on concolorous background, yellowish white to pale yellow (3A3) or dull yellow (3B3) to ochraceous at or near base; entire surface turning blue black (12F3) when bruised or handled. Context up to 17 mm at pileus, solid, initially yellowish white to pale yellow (1A3) or light yellow (1A4), immediately becoming greyish turquoise (24E4) to greyish blue (23E6) when exposed, but 4A6 (yellow) at base, blued context turning light yellow (3A4) with FeSO₄, yellow (4A6) to yellowish orange with KOH, unchanging with Guaicol. Taste and smell indistinct. Spore print olive brown (4D7–4E7).

Basidiospores 9.0–11.0–13.2 × 4.0–4.6–5.8 μm (n = 20, Q = 2.04–2.38–3.00), subfusiform to elliptic, inequilateral, smooth (under light microscope and SEM), slightly thick-walled, greenish. Basidia 20–29 × 7–9.5 μm, 2–4 spored, subclavate to clavate; sterigmata 3–5 × 1.5–2 μm. Pleurocystidia 23–40 × 5–8 μm, emergent 9–13 μm, common, subfusiform to ventricose-rostrate or appendiculate, content dense, brown. Cheilocystidia 36–45 × 6–8 μm, common, ventricose-rostrate to subfusoid, content dense brown. Tube trama divergent, tramal hyphae 6–7 μm wide, septate. Pileipellis up to 175 μm thick, trichoderm to

Research Article

ixotrichoderm, composed of densely packed erect septate hyphae; terminal cells $44-80 \times 8.5-17 \mu\text{m}$, mostly cystidoid, content irregularly dense, brown, fusoid, subfusoid, ventricose to appendiculate. Stipitipellis up to $70 \mu\text{m}$ thick, fertile, composed of hyphae, basidia and cystidia embedded in a glutinous layer; caulocystidia $27-45 \times 6-13 \mu\text{m}$, subfusoid to ventricose or appendiculate with rounded to capitate apex, content irregularly dense, brown; hyphae $3-4 \mu\text{m}$ wide, erect with fusoid to subfusoid apex; caulobasidia similar to tube basidia. Clamp connections absent.

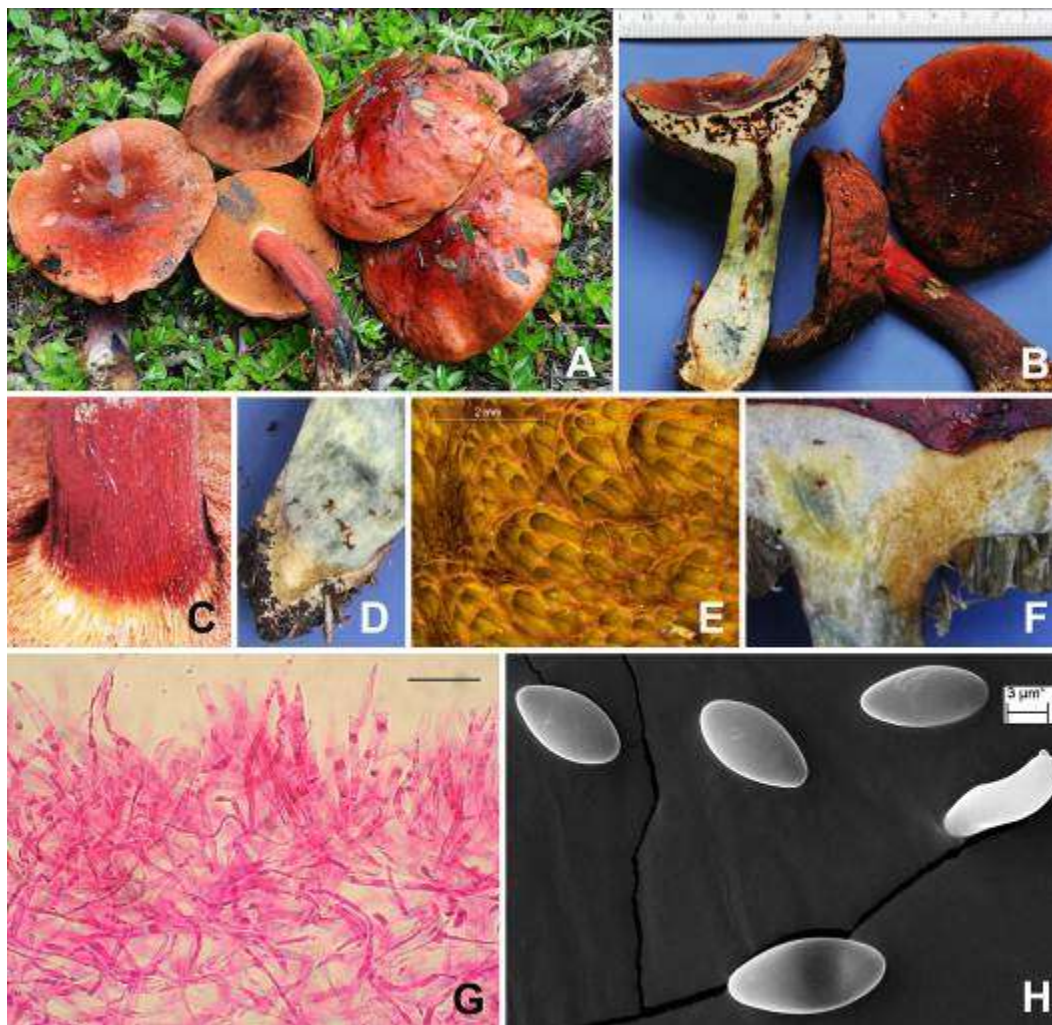


Figure 2: Macro- and micromorphological features of *Boletus sharmae* K. Das & D. Chakraborty (KD 13-013). A & B: Young and mature fresh basidiomata showing pileus, stipe and pore surface. C: Red reticulum on red stipe surface. D: Stipe base showing distinct yellow coloration. E: Dry sample showing yellowish tube with orange tube-mouth. F: Blued context turning light yellow with FeSO_4 and yellowish orange with KOH . G: Pileipellis showing hyphal arrangement in trichoderm pattern. H: SEM micrograph of basidiospores. Scale bars: E = 2 mm; G = 50 μm ; H = 3 μm .

Type: India, Sikkim, North district, Dombang, alt. 2920 m, N27° 43' 35.2" E88°45'15.2", under *Abies densa* Griff., subalpine coniferous forest, 24th July 2010, K. Das, KD 13-013, CAL 1150 (holotype); ibid., North district, near Yumthang, alt. 3644 m, N27°47'54.8" E88°42'16.1", under *Abies densa* Griff., subalpine coniferous forest, 24th July 2010, K. Das, KD 13-013, CAL 1151.

Research Article

Notes: *Boletus sharmae* can easily be characterized by orange-red to brownish viscid pileus that becomes sub-velvety to velvety when dry, presence of sterile flap of tissue at pileus margin, orange-red pore surface (tube-mouth), red colored stipe surface with red reticulum spreading on upper half of the stipe, stipe-base with yellow coloration, basidiospores of $9.0-13.2 \times 4.0-5.8 \mu\text{m}$ and trichoderm to ixotrichoderm nature of pileipellis. Unornamented spores, unmodified spore-apex, orange to brownish tube-mouth and reticulated stipe surface undoubtedly place this species under stirps *Luridus*, subsect. *Luridi*, sect. *Boletus* under the genus *Boletus* (Smith and Thiers, 1971).

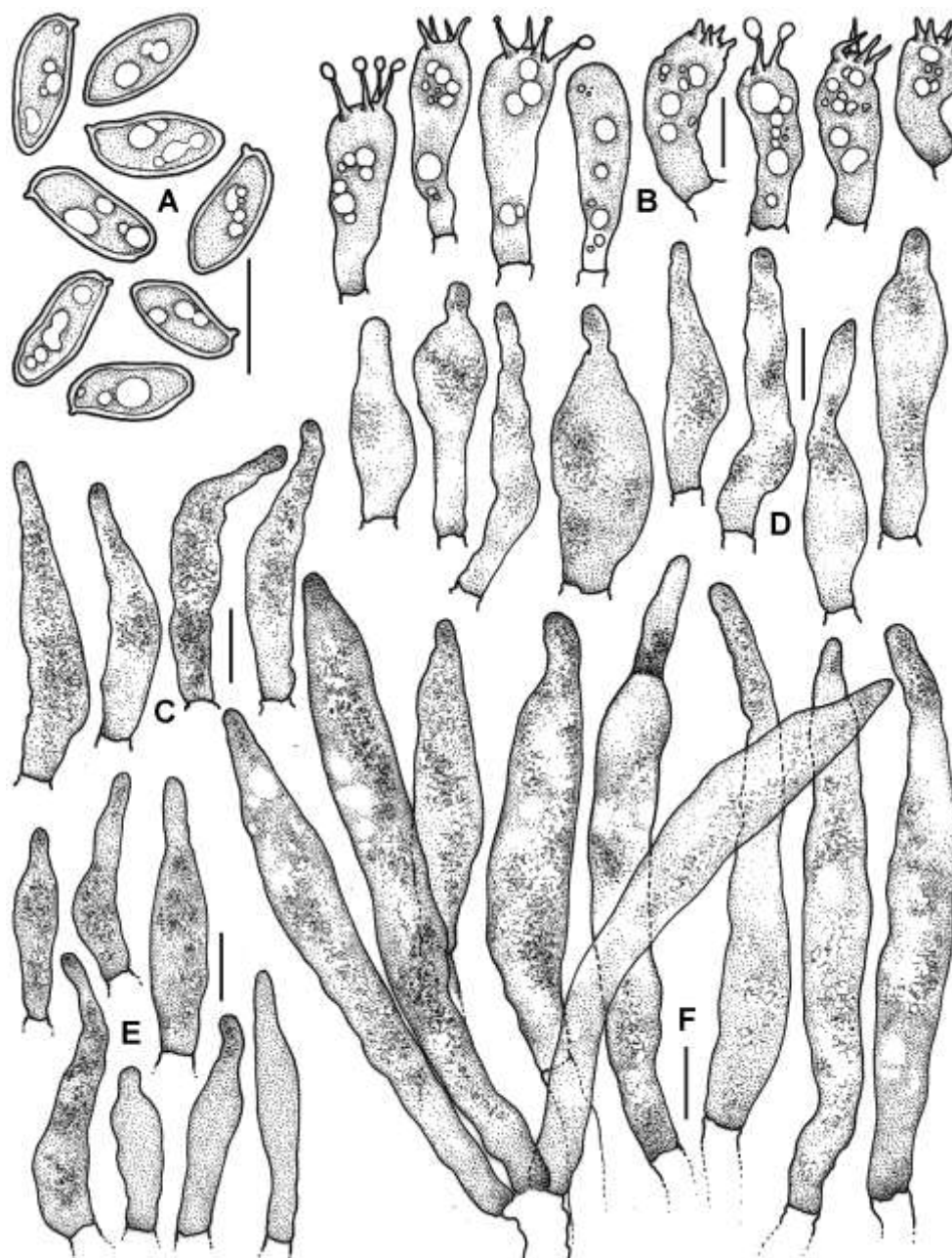


Figure 3: Micromorphological features of *Boletus sharmae* K. Das & D. Chakraborty (KD 13-013). A: Basidiospores. B: Basidia. C: Cheilocystidia. D: Caulocystidia. E: Pleurocystidia. F: Terminal cystidioid cells of pileipellis. Scale bars: A-F = 10 μm .

Research Article

Boletus rubroflammeus A.H. Sm. & Thiers, *Boletus frostii* J.L. Russell, *Boletus vinaceobasis* A.H. Sm. & Thiers, *Boletus luridus* Schaeff. (all reported from North America), *Boletus rhodopurpureus* Smotl. (reported from Europe), and *Boletus erythropus* Pers. (also reported from India as *B. luridus* var. *erythropus* Fr.) resemble *Boletus sharmae*. But, *Boletus luridus* has different coloration of stipe i.e. either yellow overall or in different combinations with red which becomes blue or violaceous with maturity and longer spores ($12-17 \times 5.5-7 \mu\text{m}$) (Smith and Thiers, 1971). *B. frostii* shows distinct and coarse reticulation spreading overall on stipe surface and longer spores: $11-15 (18) \times 4-5 \mu\text{m}$ (Smith & Thiers 1971). *B. rubroflammeus* differs macromorphologically by its persistent deep vinaceous red/ wine red coloured and nonviscid pileus and micromorphologically by tightly interwoven appressed pattern (never trichoderm) of pileipellis (Smith and Thiers, 1971; Bessette *et al.*, 1991) whereas, *B. vinaceobasis* always has a persistent vinaceous colour of the stipe surface spreading up to the base (Smith and Thiers, 1971). Similarly, European species *B. rhodopurpureus* is distinct in the field by its larger (60–150 mm) pileus and coloration of the stipe surface: purple red reticulation on yellow background (Breitenbach and Kränzlin, 1991). *Boletus erythropus* (reported from India in Lakhanpal, 1996) can easily be differentiated from *B. sharmae* by distinctly pruinose (never reticulate) stipe-surface with orange yellow apex, strigose stipe-base, and interwoven nature of hyphal arrangement in pileipellis.

Finally, this Indian collection differs from all the allied taxa (Indian and extralimital) by showing the combination of characters i.e. stipe surface with distinct red reticulation on red background, the stipe-base with persistent yellow coloration and its association with the coniferous trees in subalpine Himalaya.

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Research Article

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