



Research article

Lactarius indohirtipes and *L. sharmai* (Russulales, Basidiomycota): two novel species from Jammu and Kashmir, India

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Abstract. Exploration of macrofungi in the Jammu and Kashmir regions, India, led to the discovery of two novel species, described herein as *Lactarius indohirtipes* sp. nov. and *L. sharmai* sp. nov. This paper presents detailed morphological descriptions and illustrations, as well as a phylogenetic analysis based on nrITS sequences. A comparative analysis with related species is also included.

Keywords. Macrofungi, nrITS, phylogeny, Russulaceae, taxonomy.

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Introduction

The Union Territory of Jammu and Kashmir, situated in the northernmost part of India, is characterised by species-rich vegetation consisting of broadleaf, coniferous, and mixed temperate forests, providing suitable habitats for ectomycorrhizal fungi. Recent mycosystematic studies in this Himalayan region (Kumar *et al.* 2021; Altaf *et al.* 2022; Verma *et al.* 2022) have underscored this fact. However, despite these studies, the region remains relatively underexplored in terms of wild mushroom diversity.

Lactarius Pers. (Russulales, Basidiomycota) is a globally distributed genus in the family Russulaceae Lotsy, encompassing over 640 recognised species (Wisitrassameewong *et al.* 2016; Paloi *et al.* 2019; Kalichman *et al.* 2020; Verma *et al.* 2021, 2022, 2023; Bera & Das 2023). India alone has reported approximately 100 taxa (Atri *et al.* 2016; Das *et al.* 2020; Verma *et al.* 2021, 2022, 2023; Bera & Das 2023). All species of this genus form ectomycorrhizal (ECM) associations with diverse angiospermic and gymnospermic trees in tropical and temperate forests (Das *et al.* 2020). The genus *Lactarius* is subdivided into three subgenera: *L.* subg. *Lactarius*, *L.* subg. *Russularia* (Fr.) Kauffman and *L.* subg.

Plinthogalus (Burl.) Hesler & A.H.Sm. (Buyck *et al.* 2010; Wisitrassameewong *et al.* 2016; De Crop *et al.* 2017; Das *et al.* 2020; Verma *et al.* 2022).

Species within *Lactarius* subg. *Russularia* are characterised by a dry pileus surface, with predominantly orange, reddish brown or brown colours, and typically white, unchanging latex, although exceptions exist with yellow latex or colour changes (Verbeken *et al.* 2001; Paloi *et al.* 2019). The pileipellis structure varies greatly, ranging from an epithelium to hyphoepithelium or (tricho-)palisade to a cutis (Heilmann-Clausen *et al.* 1998; Das & Sharma 2005; Wisitrassameewong *et al.* 2014; Paloi *et al.* 2019). White Species of *L.* subg. *Plinthogalus* are characterised by their brownish to grey or blackish basidiomata with a dry, velutinous, rugose to velvety pileus; pale yellow to darker spore print; pinkish or reddish context discolouration; the presence of white to yellow latex; ornamented basidiospores with high ridges forming a reticulate to zebroid pattern; mostly lacking macrocystidia and most commonly a palisade type of pileipellis, though sometimes it may be a trichoderm to trichopalisade or an ixohymeniderm. (Heilmann-Clausen *et al.* 1998; Le *et al.* 2007; Das & Chakraborty 2014; Das *et al.* 2017; Uniyal *et al.* 2019).

To explore the macrofungal diversity of the region, systematic surveys were conducted across different parts of the north-western Himalayas, particularly in the Jammu and Kashmir regions. These surveys led to the discovery of two novel *Lactarius* species, one belonging to subg. *Russularia* and the other to subg. *Plinthogalus*. Subsequent micromorphological observations and a nrITS-based molecular phylogenetic analysis confirmed the novelty of these taxa. This paper describes these two new species, providing detailed descriptions and illustrations.

Material and methods

Macro- and micromorphology

Fresh basidiomata were collected in the field and photographed using a Nikon D5300. Macromorphological descriptions, information about the habitat and associated hosts were recorded from fresh specimens in the field. Terminology of macro-morphology was used in accordance with Vellinga (1988) and Heilmann-Clausen *et al.* (1998). Colour codes were followed according to Kornerup & Wanscher (1978). Various chemical spot tests (10% KOH, FeSO₄ and guaiacol) for colour reactions on fresh specimens were performed on the pileus surface, stipe surface and context. All anatomical details were observed from dried samples by making freehand sections which were mounted in 5% KOH, 1% Phloxine and 1% Congo Red (Largent *et al.* 1977) and examined under an Olympus CH20i compound microscope. Micromorphological elements were drawn with a Camera lucida at 2000× magnification. Microphotographs of the various elements were captured with a digital camera attached to an Olympus CX33 compound microscope. Basidium length excludes the length of the sterigmata. Basidiospores were stained with Melzer's Reagent and 40 basidiospores were measured from each sample. Basidiospore measurements are reported as minimum–mean–maximum length × minimum–mean–maximum width and Quotient (Q) = length/width ratio with Qm the average Q of all basidiospores.

Scanning electron microscope (SEM) images of basidiospores were obtained from dried specimens that were directly mounted on a double-sided adhesive tape pasted on a metallic magnification in high vacuum mode (20 KV) to observe patterns of spore ornamentation. SEM studies were carried out with a JOEL JSM-IT-300 model installed at the CSIR-Indian Institute of Integrative Medicine (CSIR-IIIM), Jammu. Holotypes of the two species were deposited in the Central National Herbarium (CNH), Howrah, West Bengal (acronym CAL), while paratypes were deposited in the Herbarium of Department of Botany, University of Jammu (acronym HBJU).

DNA extraction, PCR amplification and sequencing

The Plant II Kit (Macherey-Nagel) was used to isolate nuclear genomic DNA from 100 mg of dried basidiocarps. The ITS regions of the nuclear ribosomal DNA were amplified using the primers ITS1 and ITS4 (White *et al.* 1990). PCR amplification reactions were carried out in a 20 µl reaction volume which contained 1× Phire PCR buffer (contains 1.5 mM MgCl₂), 0.2 mM each dNTPs, 1 µl DNA, 0.2 µl Phire Hotstart II DNA polymerase enzyme, 0.1 mg ml⁻¹ BSA, and 3% DMSO, 0.5 M Betaine and 5 pM of forward and reverse primers. PCR amplification was carried out in a PCR thermal cycler (Gene Amp PCR System 9700, Applied Biosystems) programmed for 2 min at 96°C, followed by 30 cycles of 30 s at 96°C, 40 s at 50°C, and a final stage of 4 min at 60°C. The PCR products were purified with QIAquick Gel Extraction Kit (QIAGEN, Germany) and then subjected to Sanger sequencing in an automated DNA sequencer (ABI3730xl DNA Analyzer, Applied Biosystems, USA) using the same primers as for amplification. All generated nrITS sequences were deposited in GenBank under accession numbers (OP185366, OP185367 for *Lactarius indohirtipes* sp. nov. and OP185255, PP273500 for *L. sharmai* sp. nov.).

Phylogenetic analysis

A phylogenetic analysis based on nrITS sequences was carried out to establish the phylogenetic placement of the novel species. The dataset consists of nrITS sequences of collection and reference sequences acquired from nBLAST search against GenBank (<https://www.ncbi.nlm.nih.gov/genbank>; Altschul *et al.* 1997; Clark *et al.* 2016) and relevant published phylogenies (Stubbe & Verbeken 2012; Lee *et al.* 2015; Wisitrassameewong *et al.* 2015, 2016; Das *et al.* 2017, 2020; Uniyal *et al.* 2018, 2019; Paloi *et al.* 2019; Verma *et al.* 2021, 2022, 2023). For analysing the phylogenetic placement of the novel species, two separate datasets were prepared. For *Lactarius indohirtipes* sp. nov., 49 nrITS sequences (including our newly generated sequences) of *Lactarius* were used and for *L. sharmai* sp. nov., 49 nrITS sequences (including our newly generated sequences) of *Lactarius* and *Lactifluus* (Pers.) Roussel were used. *Lactarius pterosporus* Romagn. (AY331013) and *L. friabilis* H.T.Le & D.Stubbe (EF560667) were used as outgroups for *L. indohirtipes*. For *L. sharmai*, species of the sister genus *Lactifluus* (*Lactifluus bertillonii* (Neuhoff ex Z.Schaeff.) Verbeken (MH125230) and *Lactifluus deceptivus* (Peck) Kuntze (AY854089)) were used as outgroups. Sequences were aligned using MAFFT ver. 7 (Katoh & Standley 2013) and manually edited in Bioedit ver. 7.2.5 (Hall 1999). A phylogenetic analysis of nrITS sequences was undertaken based on Maximum Likelihood (ML) criteria computed in RAxML GUI 2.0 (Edler *et al.* 2021). 1000 bootstrap replicates were analysed to obtain nodal support values.

Results

Phylogenetic inferences

Inferences drawn from the phylogenetic analysis confirmed the novelty of *Lactarius indohirtipes* (GenBank OP185366, OP185367) and its placement in the *L. hirtipes* clade, formed of *L. aurantiobrunneus* X.H.Wang (MK351925, MK351923), *L. fulvihirtipes* X.H.Wang (GenBank MK253491), *L. hirtipes* J.Z.Ying (MG589767, MG589766, MW075673, MW075668) and *L. alpinihirtipes* X.H.Wang (MG589774, MG589782, MG589785) and is also confirmed by macro- and microscopic characters of the taxa. Within its clade, it is well segregated with 68% nodal support value (Fig. 1). *Lactarius sharmai* sp. nov. (GenBank OP185255, PP273500) clustered within a clade formed by an Indian species, *L. pleuromacrocytidiatus* Uniyal, K.Das & R.P.Bhatt (GenBank MF791871, MF405081) reported from Garhwal Himalayas of Uttarakhand, but segregated with a nodal support of 100% (Fig. 2).

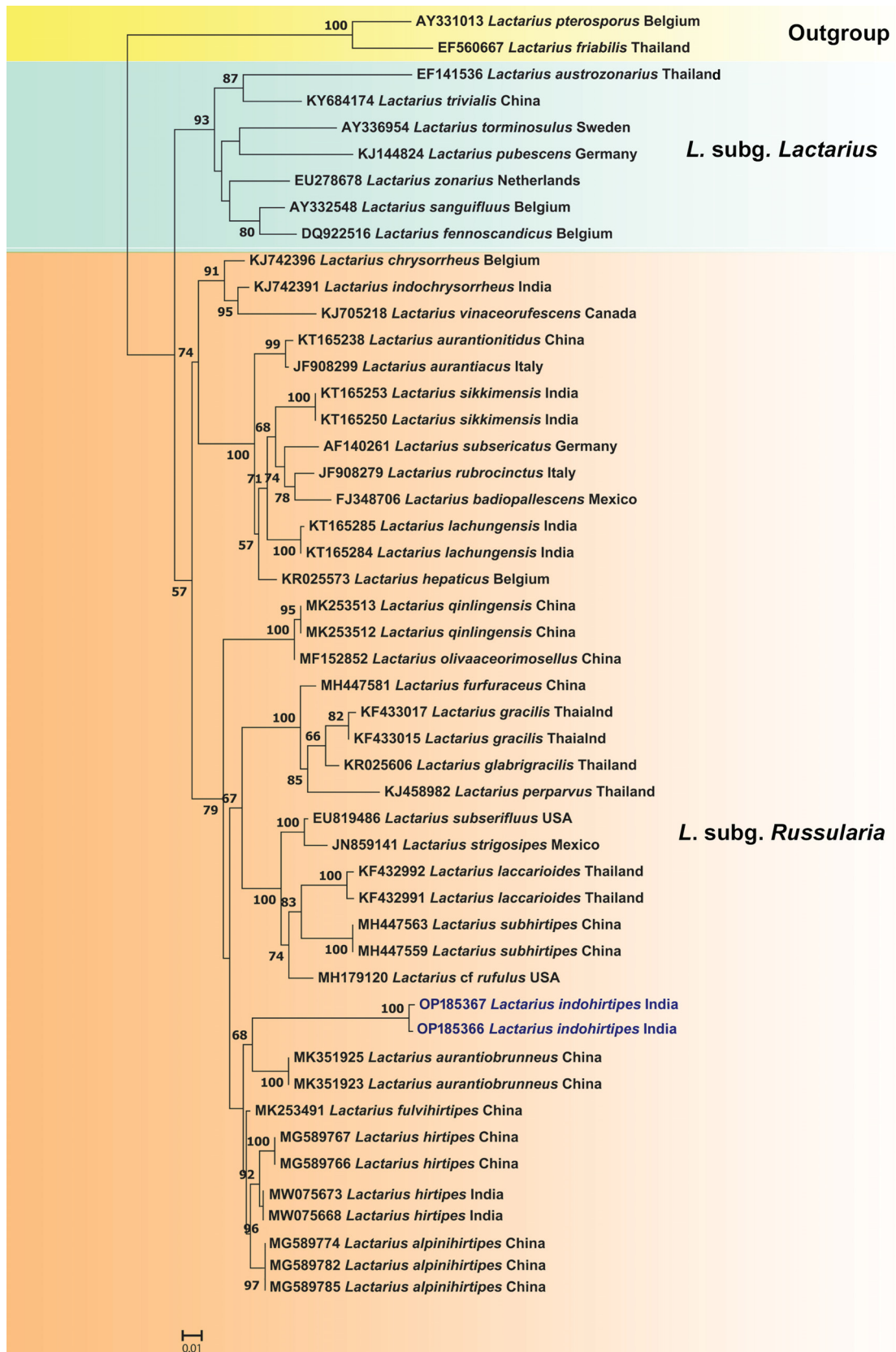


Fig. 1. ML phylogram (RAxML) inferred from the nrITS alignment. *Lactarius indohirtipes* K.Verma, Uniyal & Mehmood sp. nov. is highlighted in bold blue.

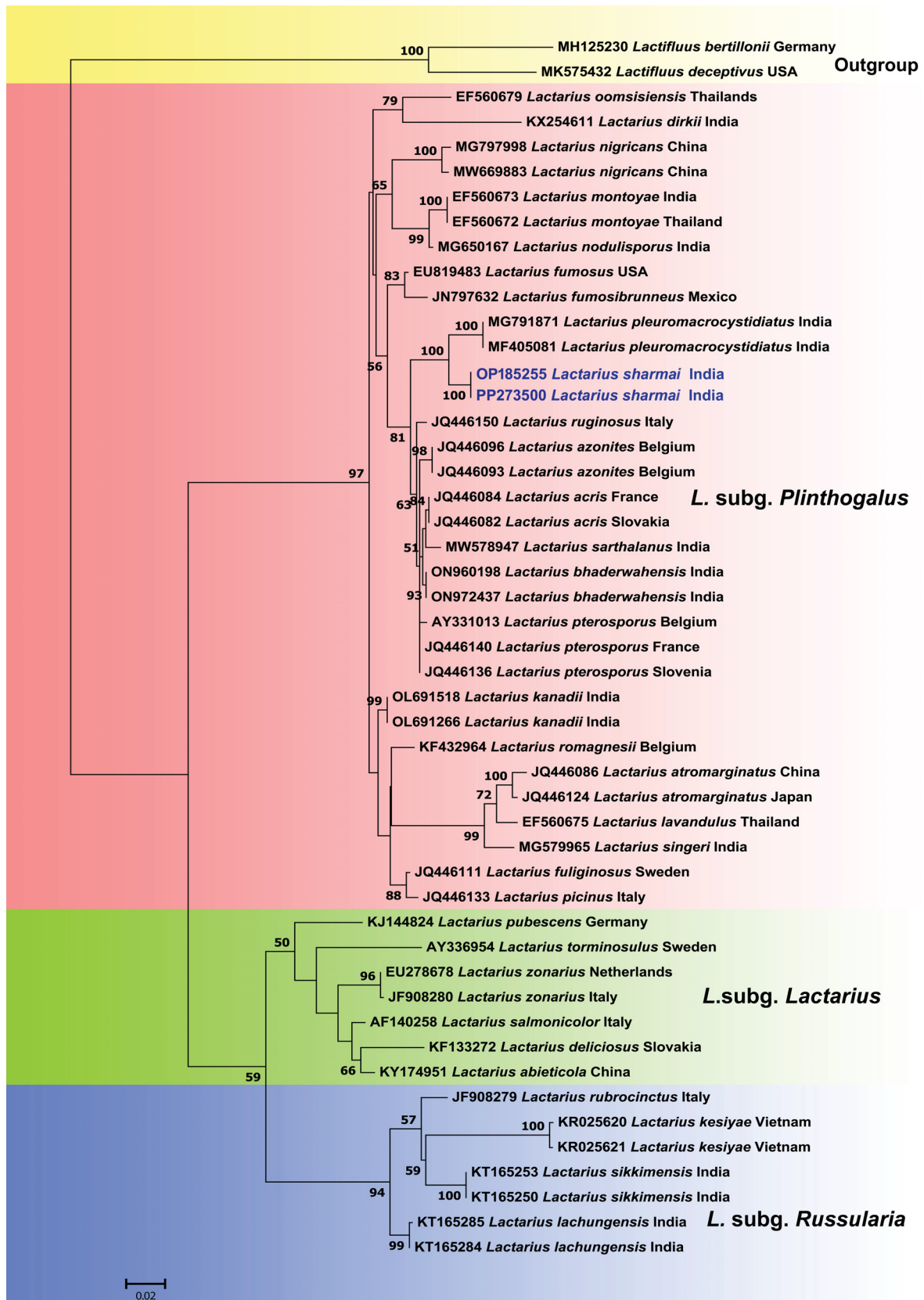


Fig. 2. ML phylogram (RAxML) inferred from the ITS-rDNA alignment. *Lactarius sharmai* K.Verma, Uniyal & Mehmood sp. nov. is highlighted in bold blue.

Taxonomic treatment

Phylum Basidiomycota R.T.Moore
Class Agaricomycetes Doweld
Order Russulales Kreisel ex P.M.Kirk, P.F.Cannon & J.C.David
Family Russulaceae Lotsy
Genus *Lactarius* Pers.

Lactarius indohirtipes K.Verma, Uniyal & Mehmood sp. nov.
MycoBank: MB847561
Figs 3–4

Diagnosis

Lactarius indohirtipes sp. nov. is characterized by the presence of a brownish orange to light orange, umbonate, viscid pileus, a stipe with reddish brown hairs at the base, white unchanging latex, subglobose to broadly ellipsoid basidiospores with ornamentations composed of ridges and isolated warts forming partial to almost complete reticulum, the absence of cheilocystidia, an ixocutis type of pileipellis and its association with *Quercus* sp.

Etymology

The name ‘*indohirtipes*’ refers to the occurrence of the species in India and its morphological and phylogenetic similarities to species of the *Lactarius hirtipes* complex.

Type material

Holotype

INDIA • Jammu and Kashmir, Doda, Bhaderwah, Chatergala; 32°53'13.50" N, 75°47'28.70" E; 3046 m a.s.l.; 10 Jul. 2018; *K. Verma LK-02*; under *Quercus* sp. in temperate broad-leaved forest; GenBank no.: OP185366 (nrITS); CAL 1918.

Paratype

INDIA • Jammu and Kashmir, Doda, Bhaderwah, Padri; 32°54.485' N, 75°48.643' E; 3103 m a.s.l.; 10 Aug. 2021; *K. Verma LK-03*; under *Quercus* sp. in temperate broad-leaved forest; GenBank no.: OP185367 (nrITS); HBJU/M/51.

Description

Basidiomata small to medium sized. Pileus 15–45 mm diam., convex with incurved margin, a central umbo when young, gradually becoming planoconvex at maturity, with slight depression when mature, umbo may or may not persist; surface smooth, viscid; pilear surface light to brownish orange (5A5–5C6), darker at the center, gradually fading on maturity to brownish orange (5C6), light orange (5A4–5A5) at the margin; cuticle not peeling easily; margin incurved when young, gradually becomes decurved, entire. Lamellae 2–3 mm wide, subdecurrent, highly crowded (20 L+I/cm at pilear margin); lamellulae in 5 series, pale orange (5A3) but turning slightly brownish on bruising, edge entire. Stipe 35–80 × 5–8 mm, central; cylindrical, stuffed, surface dry, orange to brownish orange (6B7–6C7) gradually darker at base often with reddish brown hairs at the base. Context moderately thin at pileus in comparison to the stipe, pale orange to light orange (5A3–5), unchanged with 10% KOH, greenish grey (1C2–1C3) with FeSO₄, becoming brownish with guaiac. Latex abundant, white, unchanging on exposure. Taste mild. Odour fruity. Spore print not obtained.



Fig. 3. *Lactarius indohirtipes* K.Verma, Uniyal & Mehmood sp. nov. (CAL 1918). **A.** Basidiomata in the field. **B.** Basidiomata at the base camp. **C.** Lamellae showing latex. **D.** Pleuroseuodocystidia. **E, F.** Basidia. **G.** Pleuromacrocytidia. **H.** Transverse section through pileipellis. **I.** Lamellae edge. **J.** Basidiospores under light microscope. **K–M.** Basidiospores under SEM. Scale bars: A, C = 20 mm; D–J = 10 µm; K–M = 2 µm.

Basidiospores $8.5\text{--}9.5\text{--}11.2 \times 6.8\text{--}7.9\text{--}9.3 \mu\text{m}$, ($n = 40$), $Q = 1.06\text{--}1.20\text{--}1.40$, usually subglobose to broadly ellipsoid; ornamentations up to $0.7 \mu\text{m}$ high, composed of ridges and isolated warts, ridges sometimes connected to form partially to almost complete reticulum; suprahilar spot inamyloid. Basidia $35\text{--}42 \times 8\text{--}13 \mu\text{m}$, subclavate, 4-spored; sterigmata $3\text{--}6 \times 1.5\text{--}1.8 \mu\text{m}$. Pleuromacrocystidia $35\text{--}48.5 \times 5\text{--}8.5 \mu\text{m}$, moderate to abundant, emergent up to $15 \mu\text{m}$, cylindric to subcylindric with acute, fusoid to subfusoid apices, thin-walled; content dense, granular to crystalline. Pleuropseudocystidia $3\text{--}5 \mu\text{m}$ wide, scarce to moderate, slightly emergent mostly non-emergent, cylindrical to slightly tortuous, with rounded apex. Lamellae edge fertile with basidia and basidioles. Cheilomacrocystidia not found.

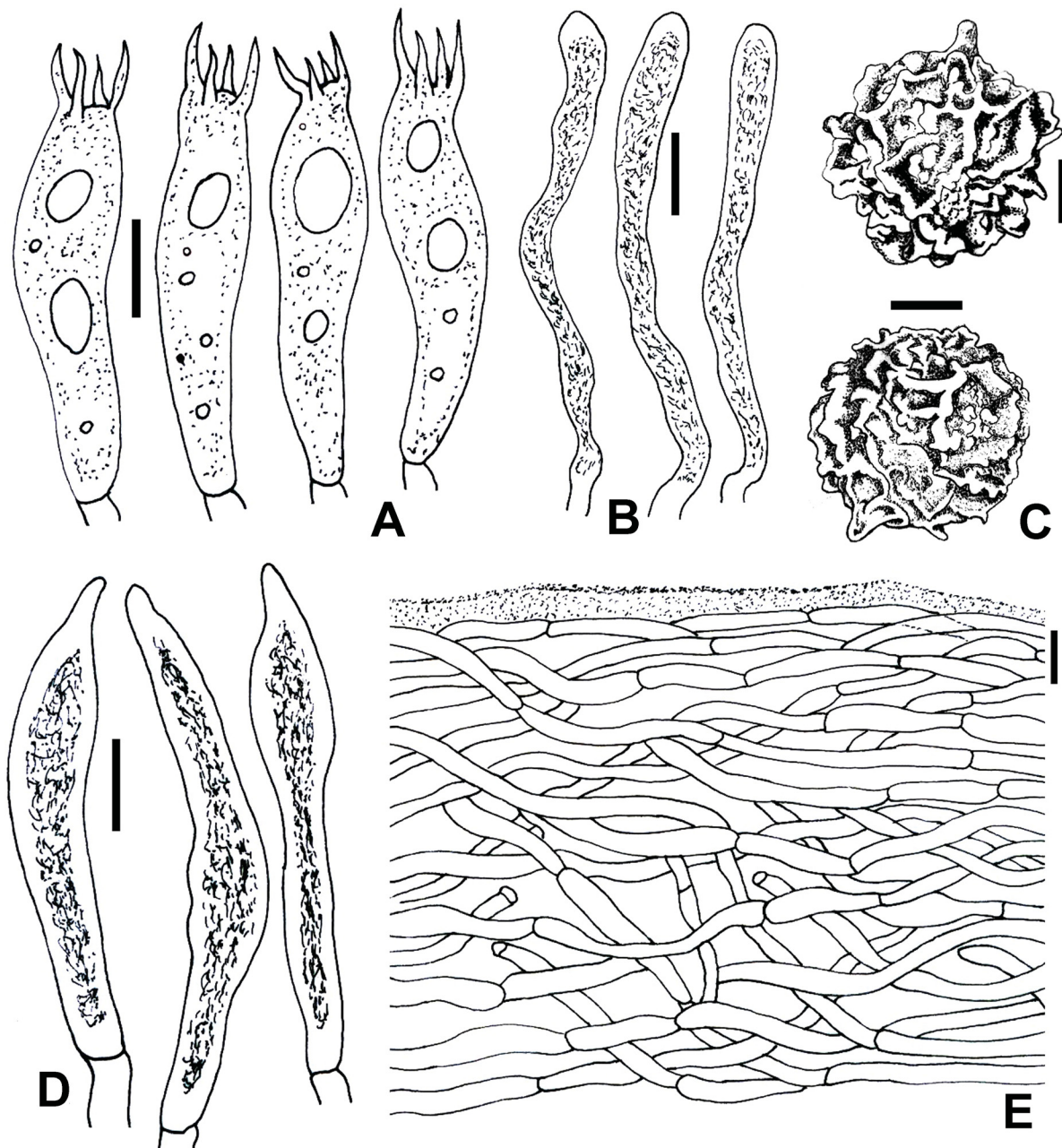


Fig. 4. Line drawings of *Lactarius indohirtipes* K.Verma, Uniyal & Mehmood sp. nov. (CAL 1918). **A.** Basidia. **B.** Pleuropseudocystidia. **C.** Basidiospores. **D.** Pleuromacrocystidia. **E.** Transverse section through pileipellis. Scale bars: A–B, D–E = $10 \mu\text{m}$; C = $2 \mu\text{m}$.

Subhymenium up to 15 µm thick, cellular. Hymenophoral trama composed of lactifers and few nests of sphaerocytes connected with connecting hyphae. Pileipellis 45–65 µm thick, an ixocutis, a thin layer of gluten up to 10 µm thick. Suprapellis 25 µm thick, composed of parallelly arranged hyphae. Subpellis is cellular to sub-cellular 45µm thick, hyphae 4.5µm thick. Stipitipellis a cutis, composed of compactly arranged septate hyphae.

Lactarius sharmai K.Verma, Uniyal & Mehmood sp. nov.

Mycobank: MB847562

Figs 5–6

Diagnosis

Lactarius sharmai sp. nov. can be identified by its yellowish white to greyish yellow, brownish grey, dry, velvety, radially rugulose pileus surface, greyish orange to pale yellow lamellae turning deep orange when damaged; extensive forkations, white abundant latex that changes into light orange to yellowish orange or pastel yellow on lamellae, globose to broadly ellipsoid basidiospores with high ridges (≤ 1.6 µm), the absence of pleuromacrocystidia, abundant cheiloleptocystidia and a trichopalisade type of pileipellis.

Etymology

The epithet '*sharmai*' refers to Prof. Yash Pal Sharma to honour his valuable contributions to the macrofungal flora of Jammu and Kashmir, and Ladakh.

Type material

Holotype

INDIA • Jammu and Kashmir, Udampur, Majalta, Samanabanj; 32°42.996' N, 75°25.931' E; 1749 m a.s.l.; 8 Aug. 2020; *K. Verma & T. Mehmood LK-012*; GenBank no.: OP185255 (nrITS); CAL 1919.

Paratype

INDIA • Jammu and Kashmir, Kathua, Machedi; 32°40.047' N, 75°37.231' E; 1807 m a.s.l.; 4 Aug. 2021; *K. Verma & T. Mehmood LK-120*; under *Quercus* sp. and *Pinus roxburghii* in subtemperate mixed forest; GenBank no.: PP273500 (nrITS); HBJU/M/52.

Description

Basidiomata medium sized. Pileus 50–68 mm diam., convex when young, plano-convex to applanate with depressed centre when mature; surface dry, velvety, radially rugulose, yellowish white (4A2) to greyish yellow (4B2) when young, blond (5C4) to brownish grey (4D2) when mature, centre olive brown (4D3) to brownish grey (4D2); margin undulate, decurved to plane. Lamellae 4–6 mm broad, adnate to subdecurrent, greyish orange (5B4), pale yellow (4A3) towards edge, staining light brown (5C5) to brownish orange (5D5) after 30 minutes, crowded (15–18L+l/cm), edges concolorous, staining yellowish brown (5D8) when damaged; lamellulae plentiful, unevenly distributed, 10–12 lengths, forked in 5–6 ranks. Stipe 45–80 × 8–10 mm, cylindric, slightly tapered towards base, surface smooth, longitudinally rugose; yellowish white (3A2–3A3), greyish yellow (4B4) to blond (4C4). Context 3–4 mm thick in pileus, brittle, white to cream white (1B2), staining light orange (6A4–6A5) to greyish yellow (4B4–4B5) on exposure, stuffed in stipe; not staining with FeSO₄ and KOH, brownish red (8D7–8D8) with guaiacol. Latex abundant, white, changing to light orange (6A4–6A5) to yellowish orange (6A5–6A6), pastel yellow to light yellow (3A4–3A6) on lamellae. Taste mild. Odour pleasant. Spore deposit not obtained.

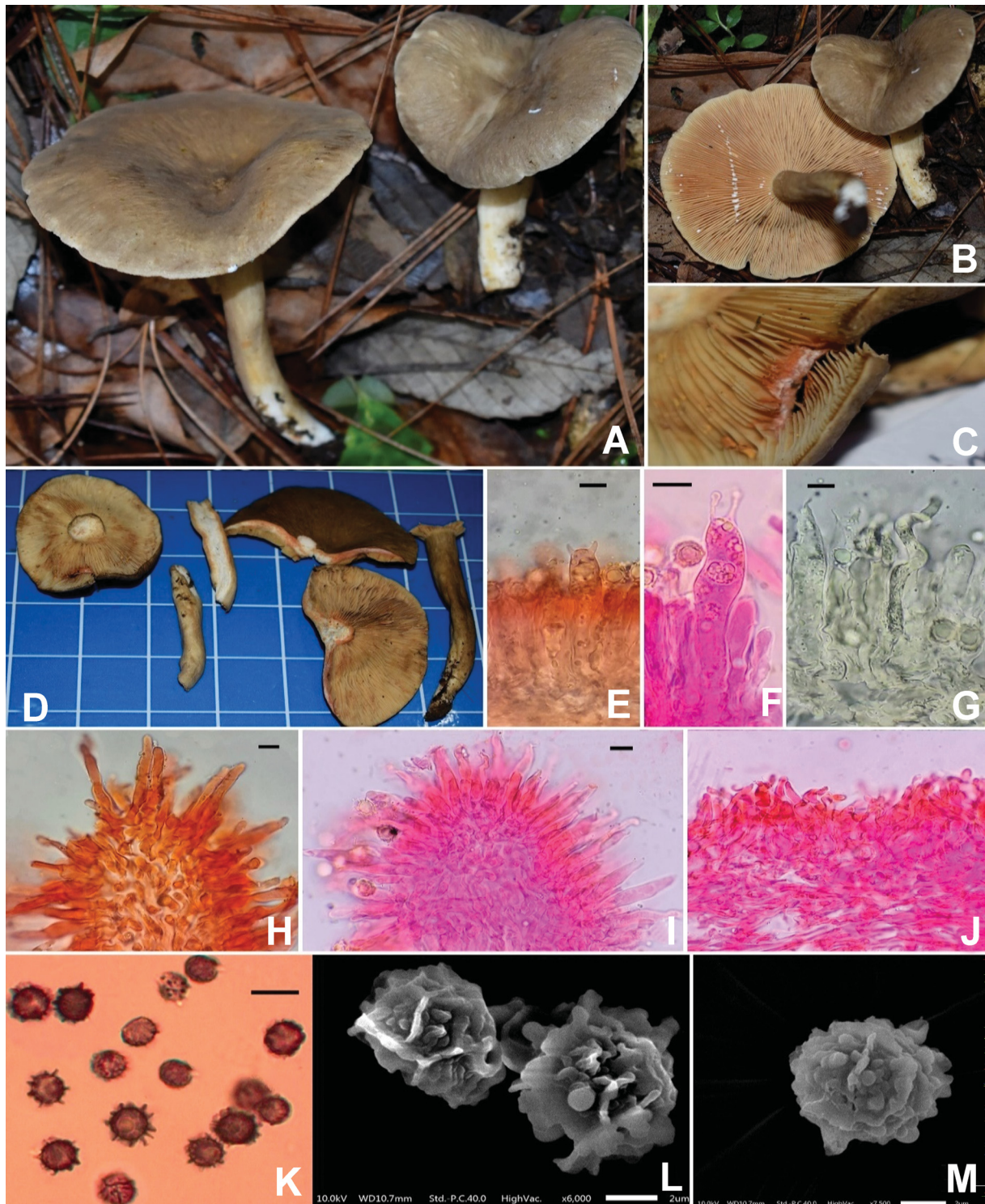


Fig. 5. *Lactarius sharmai* K.Verma, Uniyal & Mehmood sp. nov. (CAL 1919). **A.** Fresh basidiomata in the field. **B.** Lamellae showing latex. **C.** Bruised context. **D.** Basidiomata at the base camp. **E–F.** Basidia. **G.** Pseudocystidia. **H–I.** Cheiloleptocystidia. **J.** Transverse section through pileipellis. **K.** Basidiospores under light microscope. **L–M.** Basidiospores under SEM. Scale bars: E–K = 10 µm; L–M = 2 µm.

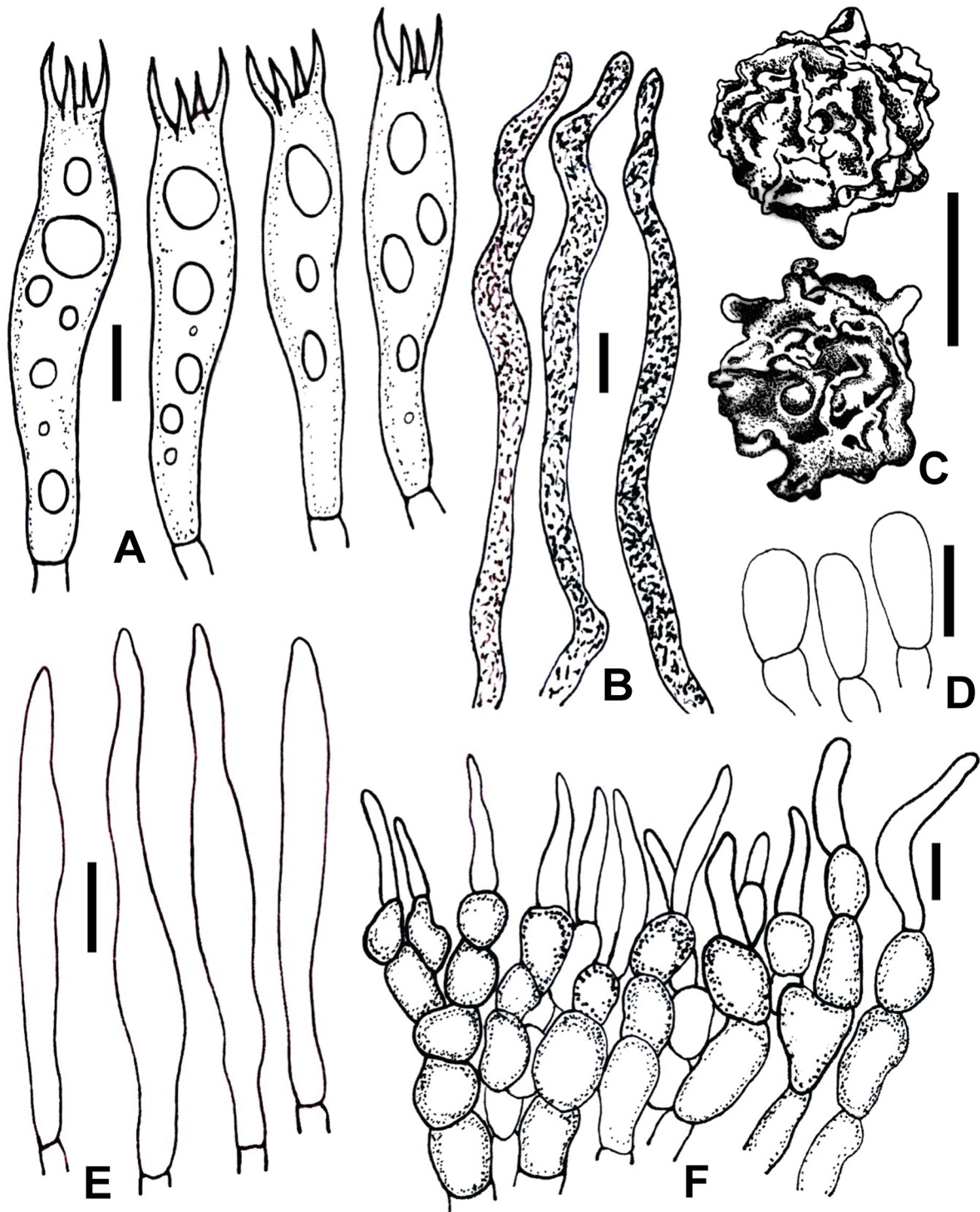


Fig. 6. Line drawings of *Lactarius sharmai* K.Verma, Uniyal & Mehmood sp. nov. (CAL 1919). A. Basidia. B. Pleuropseudocystidia. C. Basidiospores. D. Marginal cells. E. Cheiloleptocystidia. F. Transverse section through pileipellis. Scale bars: A–B, D–F = 10 µm; C = 5 µm.

Basidiospores $8.0\text{--}9.13\text{--}11 \times 7.0\text{--}8.4\text{--}10.0 \mu\text{m}$, ($n = 40$), $Q = 1.00\text{--}1.09\text{--}1.29$, globose to broadly ellipsoid, ornamentations composed of ridges ($0.8\text{--}1.6 \mu\text{m}$ high) with entire to wavy edges, often with rounded isolated warts, some shorter and irregular ridges are present in between main ridges, never forming a reticulum; plage distally amyloid. Basidia $39\text{--}43 \times 11\text{--}17 \mu\text{m}$, subclavate, 2- or 4-spored; sterigmata $5\text{--}7 \times 0.3\text{--}0.7 \mu\text{m}$. Pleuromacrocystidia absent. Pseudocystidia abundant, $5\text{--}10 \mu\text{m}$ diam., emergent up to $10\text{--}15 \mu\text{m}$, tortuous at base, unbranched. Cheilomacrocystidia absent. lamellar edges sterile. Cheiloleptocystidia abundant, $30\text{--}54 \times 5\text{--}6 \mu\text{m}$, emergent up to $20\text{--}25 \mu\text{m}$, cylindric to clavate, apices round, with one to two cells at the base. Marginal cells $12\text{--}14 \times 6\text{--}7 \mu\text{m}$, cylindric to subclavate, often multiseptate, thin-walled, with brown intracellular pigmentation. Hymenophoral trama composed of abundant lactifers, up to $4\text{--}6 \mu\text{m}$ diam. Pileipellis a trichopalisade, $70\text{--}100 \mu\text{m}$ thick; suprapellis $40\text{--}65 \mu\text{m}$ deep, composed of cylindric to subclavate cells with brown intracellular pigmentation; subpellis $45\text{--}70 \mu\text{m}$ thick, composed of cylindrical to almost rounded cells. Stipitipellis a palisade to a trichopalisade, $60\text{--}75 \mu\text{m}$ thick; hyphae $4\text{--}5 \mu\text{m}$ diam., terminal cells with intracellular brown pigment.

Discussion

The combination of characteristics such as pileus with brownish orange colour, white and unchanging latex, stipe with hairs at base, subglobose to broadly ellipsoid basidiospores with ornamentation forming partial to complete reticulum, and ixocutis type of pileipellis clearly categorizes *Lactarius indohirtipes* sp. nov. within *Lactarius* subg. *Russularia* sect. *Russularia* (Heilmann-Clausen *et al.* 1998; Paloi *et al.* 2019). *Lactarius indohirtipes* is an addition to the *L. hirtipes* species complex due to its close phylogenetical and morphological resemblance to some Chinese species within this complex, including *L. fulvihirtipes*, *L. hirtipes*, *L. alpinihirtipes*, *L. subhirtipes* X.H.Wang and *L. aurantiobrunneus*. The species within the *L. hirtipes* complex can be easily confused with *L. indohirtipes* in the field due to their similar morphology. These share several characteristics such as slender fruiting bodies, basidiospores with reticulate ornamentation, a hyphoepithelium pileipellis, and absence of cheilomacrocystidia (Wang 2018). However, *L. fulvihirtipes* can be distinguished by its brownish yellow-coloured pileus, distant lamellae, white watery latex, sterile lamellar edge and smaller ($7.5\text{--}8.6\text{--}9.5 \times 7.0\text{--}7.7\text{--}8.5 \mu\text{m}$) broadly ellipsoid basidiospores and ixo-epithelium to hyphoepithelium type of pileipellis (Wang 2018). *Lactarius hirtipes*, which is also close to *L. indohirtipes*, can be differentiated by its white latex changing to whey white, slightly smaller ($5.7\text{--}7.0\text{--}7.9 \times 5.2\text{--}6.6\text{--}7.3 \mu\text{m}$) basidiospores, epithelium type of pileipellis, and association with *Castanopsis* sp. or *Picea* sp. and *Rhododendron* sp. (Wang & Liu 2002; Bera & Das 2021). *Lactarius subhirtipes* can be distinguished from *L. indohirtipes* by slender brownish orange basidiocarps, polished pileus with a papilla, watery latex, small globose spores with relatively isolated and acute ornamentation and absence of macrocystidia. *Lactarius alpinihirtipes* can be differentiated from *L. indohirtipes* by its irregularly striate margin, rugose pileus surface, smaller ($7.5\text{--}10.0 \times 6.5\text{--}8.0 \mu\text{m}$), broadly ellipsoid to ellipsoid basidiospores, a hyphoepithelium type of pileipellis and its occurrence under *Pinus densata* Mast., *Picea* sp. and *Quercus* sp. (Wang 2017). *Lactarius aurantiobrunneus* can be separated from *L. indohirtipes* by a more orange-coloured pileus, a dry and strongly rugose pileus surface, a crenulate margin, the medium distant lamellae, the watery or cream-watery latex, slightly smaller basidiospores ($7.0\text{--}9.0 \times 6.5\text{--}8.0 \mu\text{m}$, $Q = 1.12 \pm 0.05$); high ornamentation ($1.0\text{--}2.0 \mu\text{m}$), never as reticulate and epithelium type of pileipellis (Wang *et al.* 2019). Another Indian species, namely *L. atrii* Van de Putte & K.Das, is similar to *L. indohirtipes*. However, *L. atrii* can be distinguished from *L. indohirtipes* by its light brown basidiocarp with a very strigose base, obtuse isolated warts in the basidiospore ornamentation, the large and protruding macrocystidia and by its occurrence in mixed coniferous forest of *Picea*, *Tsuga* and *Juniperus*.

The presence of a greyish-yellow to blond-brownish-grey pileus with a radially rugulose, dry and velvety surface, the absence of macrocystidia, the presence of cheiloleptocystidia, a trichopalisade type of pileipellis with dark brown pigmented hyphal elements certainly placed *Lactarius sharmai*

sp. nov. in subg. *Plinthogalus* (Heilmann-Clausen *et al.* 1998; Stubbe & Verbeken 2012). *Lactarius pleuromacrocytidiatus* (an Indian species) shares a slight morphological and phylogenetic resemblance with *L. sharmai*, showing a 98.5% BLAST similarity. However, both species were easily segregated in the phylogenetic tree. In addition, the presence of a radially rugose pileus, white latex changing to pale greyish orange on the lamellae and to yellow on the context; smaller basidiospores ($6.5\text{--}8 \times 6\text{--}7 \mu\text{m}$) with moderately high ornamentations (up to $1.7 \mu\text{m}$ high); and the presence of hyaline pleuromacrocytidia delineates *L. pleuromacrocytidiatus* from *L. sharmai* (Uniyal *et al.* 2019). Some European taxa, namely *L. azonites* (Bull.) Fr., *L. ruginosus* Romagn., *L. pterosporus* Romagn., and *L. acris* (Bolton) Gray are also phylogenetically close to *L. sharmai* and can also be mistaken in the field due to their similar morphology (shape, colours of pileus and lamellae, occurrence under broad-leaved trees). However, *L. azonites*, can be differentiated from *L. sharmai* by its pale ochraceous to greyish brown pileus adnate, moderately distant, whitish chrome lamellae, white context becoming bright pink to orange-pink and hyphoepithelium to a trichoepithelium type of pileipellis. *Lactarius ruginosus* differs from *L. sharmai* by the greyish brown to olive pileus surface, the crenate pileus margin, the context white turning salmon pink, adnate, distant lamellae, basidiospores with a spiral pattern of ridges and higher ornamentations (up to $2 \mu\text{m}$) which never form a reticulum, a trichoepithelium type of pileipellis, and by its association with *Fagus* sp. and *Carpinus* sp. *Lactarius pterosporus* can be differentiated from *L. sharmai* by its olivaceous buff to dark greyish colour pileus, less abundant latex drying to pale greyish-orange on the lamellae and smaller ($7.7\text{--}8.0 \times 6.4\text{--}7.1 \mu\text{m}$), subglobose to ellipsoid basidiospores with a spiral pattern of ridges forming an incomplete reticulum. Similarly, *L. acris* can be differentiated from *L. sharmai* by its creamy white to dark pinkish pileus, the crenulate pileus margin, the viscid, sticky, pileus surface, the context white turning into pink to brownish orange, and an ixo-oedotrichoderm to ixotrichopalisade type of pileipellis with capitate terminal elements (Heilmann-Clausen *et al.* 1998; Stubbe & Verbeken 2012). Furthermore, another Indian species, namely *L. sarthalanus* K.Verma, Uniyal, Y.P.Sharma & Mehmood, also shows some resemblance with *L. sharmai*, but can be separated from the new species by its light brown to dark brown pileus, its lamellae bruising to deep orange when injured, the white latex that turns into pastel red on exposure, and by its occurrence in coniferous forests under *Cedrus deodara* (Roxb.) G.Don (Verma *et al.* 2021).

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