A new species and new records of *Lactarius* (subgenus *Russularia*) in a subtropical cloud forest from eastern Mexico

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Three species of *Lactarius*, subgenus *Russularia*, occurring in a subtropical cloud forest from eastern Mexico (Gulf of Mexico area) were studied. *Lactarius strigosipes* is described here as a new species close to *L. subserifluus*, and *L. areolatus* and *L. minusculus* are new to the catalogue of *Lactarius* from the country. Fresh basidiomes of the three taxa were monitored throughout 2003-2006 in a study plot and in aleatory samplings in a protected forest near Xalapa, Veracruz. The morphological variation, phenological distribution and ecological association of the species with *Carpinus caroliniana* were documented. Descriptions, illustrations of macro- and micromorphological features and discussions are presented.

Key words: *Carpinus*, ectomycorrhizal fungi, *Russulaceae*

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Introduction

Preliminary results of a long term project to study the macromycota of the subtropical cloud forest from Veracruz State where *Lactarius* is one focus of attention, and monitoring of this genus in other areas of Mexico, reveal that the subtropical cloud forest community sustains a rich ectomycorrhizal fungal biota, embracing more than 50% of the *Lactarius* species currently recorded from the country. This is related to the rich vegetational composition of the forest, containing a canopy with a mixture of ectotrophic hosts (*Quercus* spp., *Alnus* spp., *Carpinus caroliniana* Walter, and in some cases, *Fagus* or conifers). These habitats are rich in ectomycorrhizal fungi. However, large areas of this kind of forest have been disturbed and exploited by humans for a long time with the consequent fragmentation of habitat and loss of biodiversity.

This paper deals with three species of *Lactarius* subgenus *Russularia*, studied in a protected subtropical cloud forest near Xalapa, Veracruz; two of them represent new records from Mexico and one is proposed as a new species.

Materials and methods

The study is based on collections in a subtropical cloud (mesophytic) forest, in Santuario del Bosque de Niebla, protected by Instituto de Ecología, A.C. at Xalapa, Veracruz. The site was monitored weekly during 2003-2006, through aleatory samplings and in two delimited plots. Macroscopical descriptions are based on fresh basidiomes, colour references in brackets correspond to Kornerup and Wanscher (1967) and Munsell (1994) (in bold). Measurements and colours of the micromorphological structures were observed in slides mounted in 3% KOH, except for the basidiospores, which were studied in Melzer´s reagent. The methods used to calculate spore ranges and to produce SEM images of their ornamentation are those used by Montoya and Bandala (2003). The $\bar{x}$ corresponds to the range of means of basidiospore length and width, and **Q** to the range of

the mean of Q (length/width ratio) of n collections. Line drawings were made with the aid of a drawing tube. The acronyms for Herbaria are according to Holmgren *et al.* (1990).

Results

Lactarius areolatus Hesler and A.H. Sm., N. Am. Sp. of *Lactarius*, p. 515, (1979).

(Figs 1-3)

Pileus 3-45 mm diam, somewhat campanulate with acute apex when young, becoming convex, to plano-convex or depressed or concave to infundibuliform, with a more or less acute papilla (which is more conspicuous in young stages), in other cases, mainly with age it is more obtuse and sometimes the papilla becomes indistinguishable; surface finely rugulose, slightly venous around the papilla (mainly when young), dry, when expanded or in old specimens the pileipellis becomes cracked and then appears rimose-areolate, pale orange with dark vinaceous-brown (7D7) tinges in young specimens, orange-brown to brick colour (7C7, **5YR 5/6**), reddish-brown (7D6-8) dull brownish-orange (6C5) in some areas or orange-beige, dull pinkish-brown to pale vinaceous-brown (**5YR 6/4**, 7C6) when faded, with darker reddish-brown colours (7EF7) or vinaceous (8DF8) towards the centre; margin decurved and smooth when young, straight to uplifted, wavy and finely striate or sulcate in mature specimens, edge frequently crenate, lobed or irregular, undulate with age, orange-brown (6C6) or grayish-brown (6D6). *Lamellae* more or less close when young to subdistant in mature specimens, decurrent, faintly arcuate, thick, broad 1.5-4 (-5) mm diam, at times dichotomic or with interparietal veins, pinkish-cream-tan or pinkish-orange (7-6A2) with dark orange-brown stains, pale orange (**5YR 8/4**, 5A2), grayish-orange-brown, with pinkish-vinaceous or pale vinaceous tinges (7B5, 7C6) to flesh colour (6B5), grayish-brown to brown (6C5, D6), grayish-vinaceous **(5YR 5/4)** to vinaceous (7D6-7), darker with age, with different length series of lamellulae, margin stained orange-brown when bruised, edge smooth. *Stipe* 12-60 × 1-6 mm, cylindrical to slightly wider towards the base, frequently sinuous, apex commonly striate because of the lamellae prolongation, hygrophanous, not viscid, dull and canescent, the base irregularly covered with fine mycelium when young, hygrophanous, grayish-vinaceous (7C3), dull brown (6D6), orange-vinaceous, grayish-vinaceous or dull vinaceous (7C6-4), vinaceous (8C5, 8D6-7) to vinaceous-brown (7D6), finally brownish-vinaceous (**5YR 5/3-5/4**, 7DE5), chocolate-brown towards the base, at times caespitose, darkening when bruising; base glabrous or with fine or sometimes whitish or yellowish mycelium downwards, whitish or concolour. *Context* slightly hygrophanous, pinkish-orange, spotted brownish-vinaceous, pileus context thin. *Odour* of geranium in fresh condition, spicy when dried (more or less nutty), taste mild. *Latex* watery to serous, unchanging, not staining white paper.

Basidiospores 6.5-8 (-8.5) × 5.5-7.5 µm, $\bar{x}$ = 7-7.7 × 6-6.8 µm, **Q** = 1.1-1.2, subglobose to broadly ellipsoid, cristate, verrucose, with continuous bands or ridges which at times form subreticulate areas, ornamentation 0.8 µm high; under SEM seen to have irregular verrucae, at times with a pronounced base and in some cases joined forming subreticulate areas, also some with continuous isolate ridges with irregular lobed margin. *Basidia* 3-60 × 8-12 µm, clavate, tetrasporic. *Pleurocystidia* absent. *Marginal cells* of the lamellae 9-17 × 4.5-10.8 µm, broadly clavate, subcylindrical, more or less similar to the terminal elements of the pileipellis but also basidiole-like. *Pseudocystidia* 3-5 µm diam, subcylindrical, contorted, originated from the trama. *Pileipellis* pseudoparenchymatous, composed of irregular subglobose or inflated elements 11-33 µm diam, and some intermixed hyphae 3-4 µm diam, terminal cells 15.6-33 × 6-11 µm, subcylindrical, broadly inflated or pyriform, yellowish in KOH, with thin irregular hyphal patches which cover mounds of pseudoparenchymatous tissue, hyphae 2-3 µm diam. *Context* hyphae 3-6.2 µm diam, laticifers 5-16 µm diam, abundant, sphaerocytes 8-45.6 µm diam, in rosettes. *Hymenophoral* trama hyphae 3.5-10 µm diam, laticifers 3-12 µm diam, sphaerocytes 8-18 µm diam, scarce.

Habitat: Gregarious, rarely solitary. In subtropical cloud (mesophytic) forest, near *Carpinus caroliniana,* at 1300 m alt.

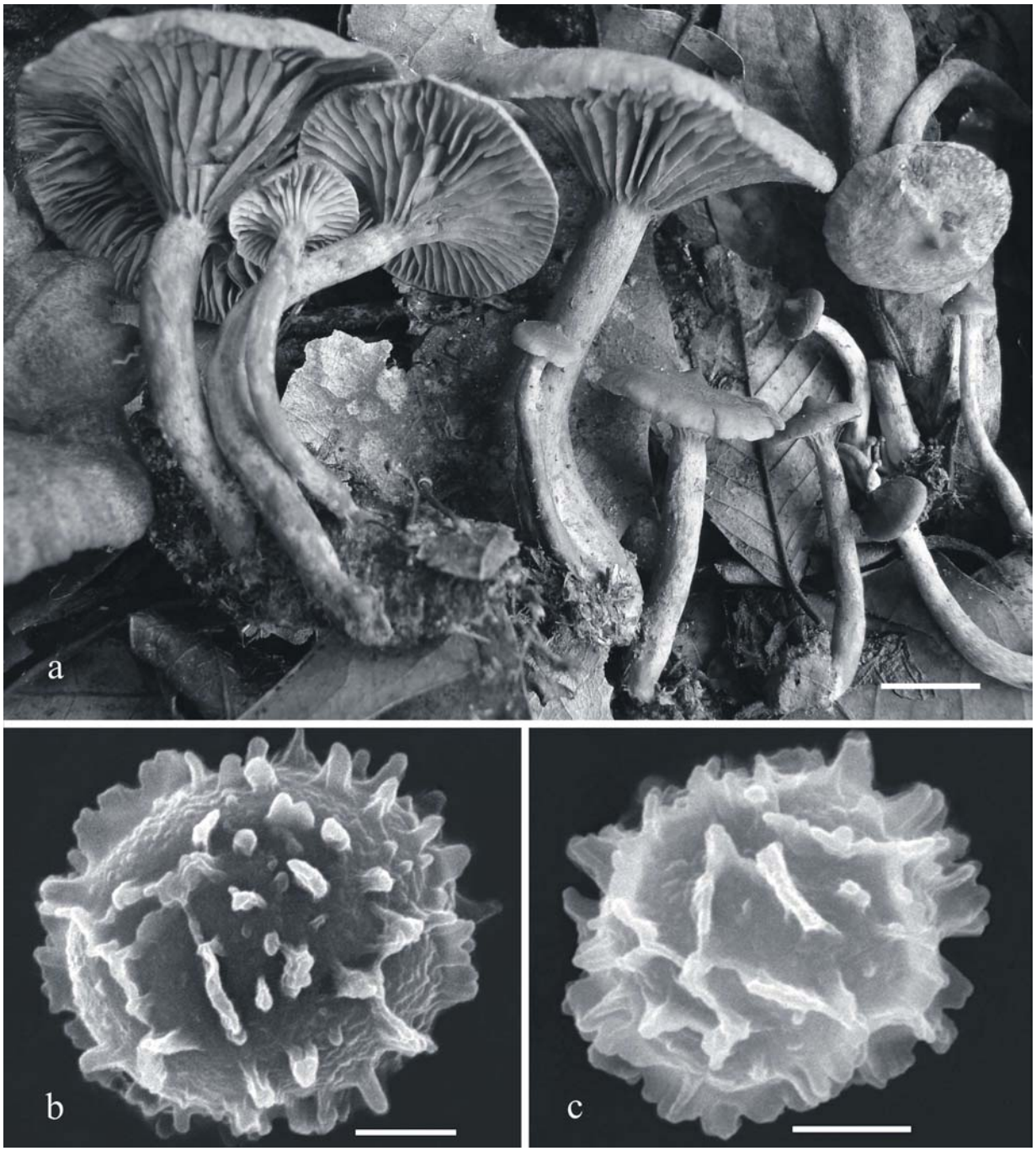

Fig. 1. *Lactarius areolatus* (Montoya 4447). **a.** Basidiomes. **b-c.** Basidiospores under SEM. Bars: a = 10 mm, b-c = 2 μm.

Known distribution: U.S.A., Mexico.

Material examined: MEXICO. Veracruz, Mpio. Xalapa, Instituto de Ecología, Santuario del Bosque de Niebla, 26 June 2003, *Corona* 260; September 25 2003, *Corona 337*; 9 July 2004, *Montoya, 4137*; 10 August 2004, *Montoya 4163*; 22 September 2004, *Montoya 4190*; 6 October 2004, *Montoya 4204*; 19 October 2004, *Montoya 4227*; 3 November 2004, *Montoya 4239*; 9 March 2005, *Jarvio 2022*; 27 May 2005, *Jarvio 2076;* 30 May 2006, *Montoya 4447*; 7 June 2006, *Montoya 4484* (all at XAL).

Other material examined: U.S.A. Michigan, Washtenaw Co., Gorman Lake Woods, Chelsea, 19 August 1972, *A.H. Smith 81607* (**Holotype** of *Lactarius areolatus* MICH).

Notes: Field observations show that basidiomes of *Lactarius areolatus* have a wide

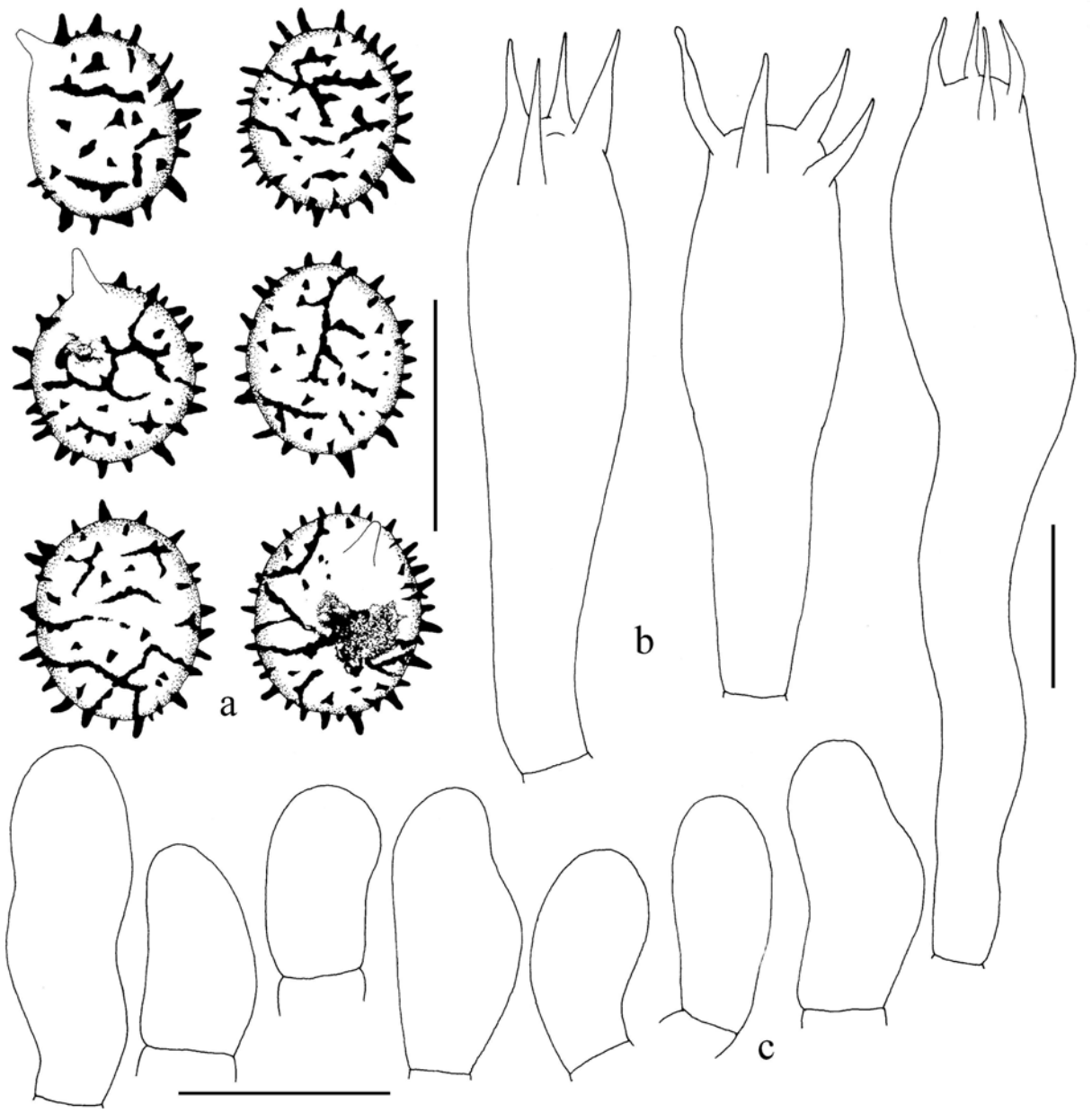

Fig. 2. *Lactarius areolatus* (Montoya 4227). **a.** Basidiospores. **b.** Basidia. **c.** Marginal cells of lamellae. Bars = 10 μm.

colour variation, with a range of orange-brown, brick colour, reddish-brown or dull brownish-orange, with somewhat paler lamellae darkening with age. The combination represented by colour, lamellae disposition (close to subdistant) and a papillate pileus helps to recognize this fungus. The pileipellis frequently becomes cracked with age but this is not always constant. This species produces aromatic basidiomes, and when dried, the odour is even more concentrated. Microscopically the basidiospore ornamentation is characteristic, composed of isolated warts and some bands or ridges forming subreticulate areas. The Mexican specimens share a similar basidiospore range and form to that shown by the holotype ($\overline{x}$ = 7.3 × 6 μm, **Q** = 1.2). Likewise, the basidiospore ornamentation and pileipellis features are also similar to the type.

Among the North American *Lactarius* (Hesler and Smith, 1979) of Sect. *Olentes*, *L. rimosellus* is macroscopically similar to *L. areolatus*. According to our observations (Montoya and Bandala, 2005) and in agreement

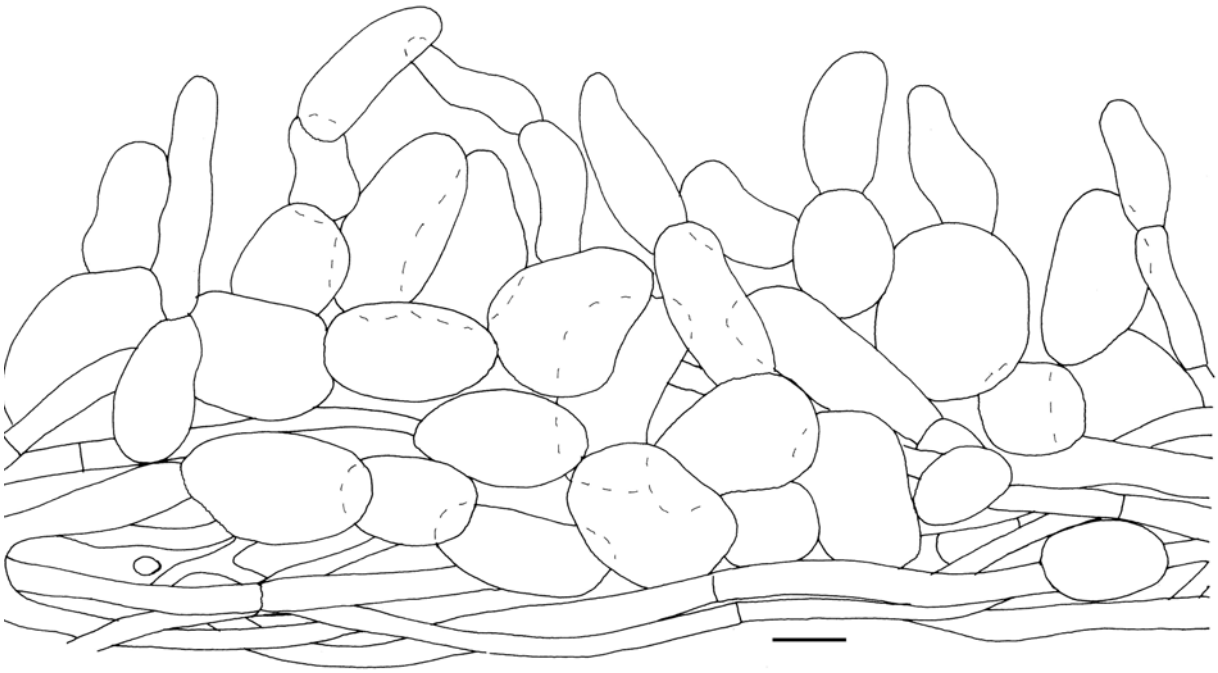

Fig. 3. *Lactarius areolatus* (Montoya 4227). Pileipellis. Bar = 10 μm.

with Hesler and Smith (1979), both show a consistent difference with regard to the basidiospore ornamentation. In *L. rimosellus*, the basidiospores are distinctly baculate to verrucose, not forming reticulations or continuous ridges as in *L. areolatus*. We have only found a few specimens of *L. rimosellus* that let us know part of the macroscopic variation range, they however, show certain difference with regard to lamellae arrangement, being closer in this species than in *L. areolatus*. New collections of *L. rimosellus* will help to recognize if these differences are consistent, or if there are additional features that help to recognize them.

In the study area during 2003-2006, basidiomes of *L. areolatus* proved to be very common and constant, being dominant in time, space and in production (number/weight), in comparison with other macrofungi recorded in the area of study. *Lactarius areolatus* basidiomes appear through all seasons of the year in proximity to *Carpinus caroliniana* but also at times in areas covered with a mixture of *C. caroliniana* and *Quercus* spp.

Lactarius strigosipes Montoya & Bandala **sp. nov.** (Figs 4-6)

MycoBank: 510757

Etymology: *strigosipes* (Lat.) refers to the presence of strigose structures at stipe base.

Pileus 8-42 mm latus, applanatus vel plano-depressus, siccus, aurantiacus, rubiginosus vel aurantio-lateritius, margine undulato. *Lamellae* densae, angustae, 1-2 mm latae, subdecurrentes, pallide aurantiaceae, griseo-aurantiaceae ad brunneo-vinaceae. *Stipes* 18-80 × 1.5-6 mm, gracilis, siccus, pileo concolor vel pallide, fistulosis, glaber; basim villosostrigosa. *Odor* in sicco condimento similis. *Gustu* nullus. *Latex* hyalinus vel translucido-albus. *Sporae* 6-7.5 × 6-7 (-7.5) μm, globosae ad subglobosae, subreticulatae vel reticulatae, ornamentum usque ad 1-2 μm altis, macula suprahilaris leviter amyloidea. *Cystidia* absentia. *Contextus* filamentosus, cellulis globosis presentia. *Pileipellis* pseudoparenchymata, cellulis subglobosis vel irregularis instructa, 7.2-30 μm latis, interdum cum hyphis cylindricis vel cellulis pyriformibus ad superficies.

Holotype: MEXICO. Veracruz, Mpio. Xalapa, Instituto de Ecología, Santuario del Bosque de Niebla, 8 June 2006, *Montoya 4521* (XAL).

Pileus 8-42 mm diam, convex in young specimens, plano-depressed in age, expanding at maturity, young specimens rarely with papilla, more frequently with an obtuse umbo, nonviscid, rugose-venose in the centre, hygro-

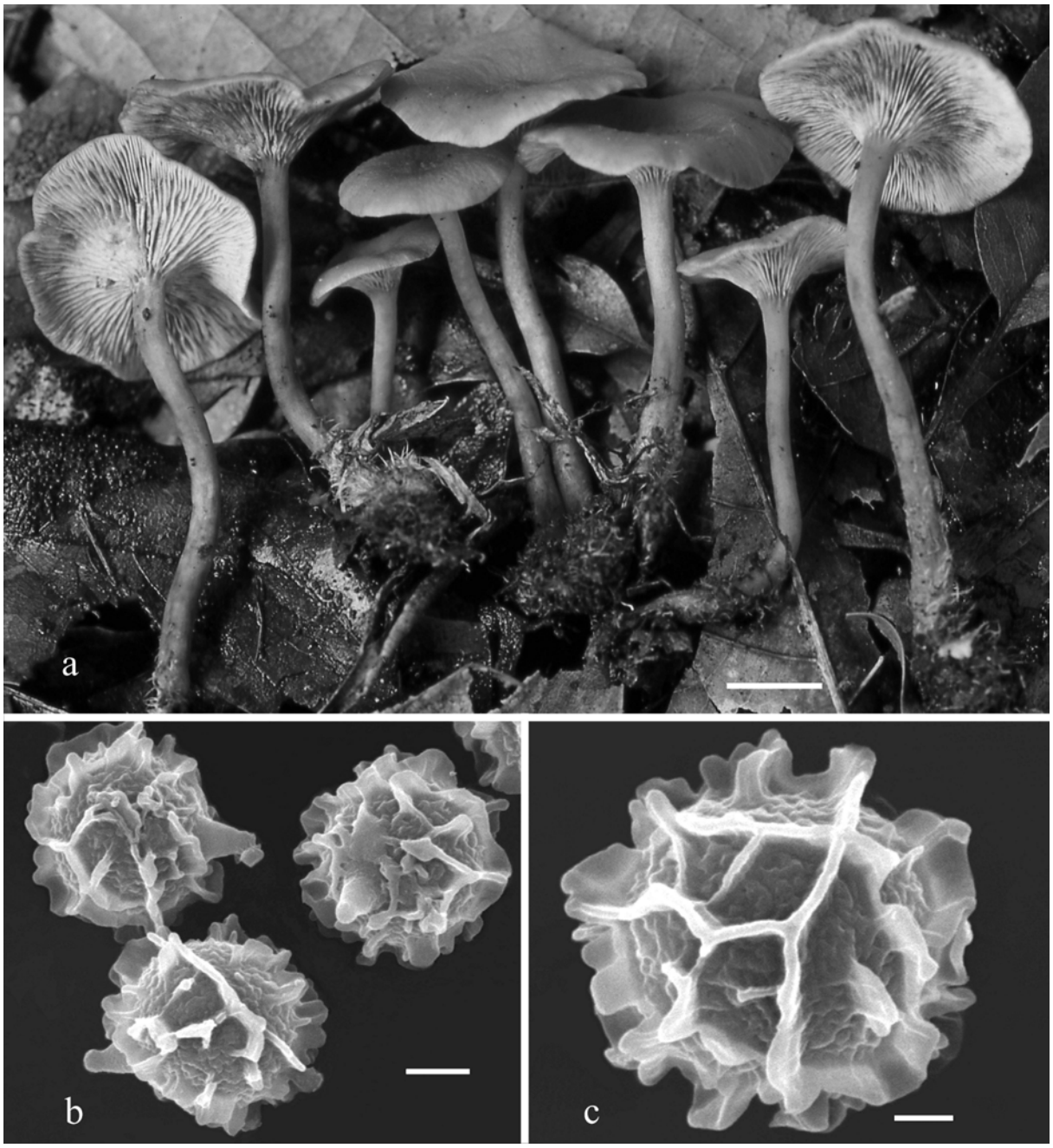

Fig. 4. *Lactarius strigosipes*. **a.** Basidiomes (Corona 268). **b-c.** Basidiospores under SEM (**from holotype**). Bars: a = 10 mm, b = 2 μm, c = 1 μm.

phanous, in young specimens they are orange-brown, honey-orange (6CD8-7D8), then orange to orange-brown (6B6, 6C5, 6-7C7, 6D6-8), rusty to reddish-brown (7DE6) or wet brick (7C8), more intense towards the centre (7C8, 7DE6) to bright orange-brown (6D7-8), reddish-brown to vinaceous (7DE7, 8C6) old specimens can reach darker brown (6E7, 7F6) tinges in the centre, the rest of the surface remaining orange colour, faded when dehydrated mainly towards the margin; edge involute, whitish and finely pubescent in young specimens, becoming decurved and smooth, at times lobed or irregularly undulate. *Lamellae* crowded inclusive in adult specimens, adnate to decurrent, arcuate, dichotomic towards the stipe area, narrow 1-2 mm broad, thin, with different length series of lamellulae, beige-

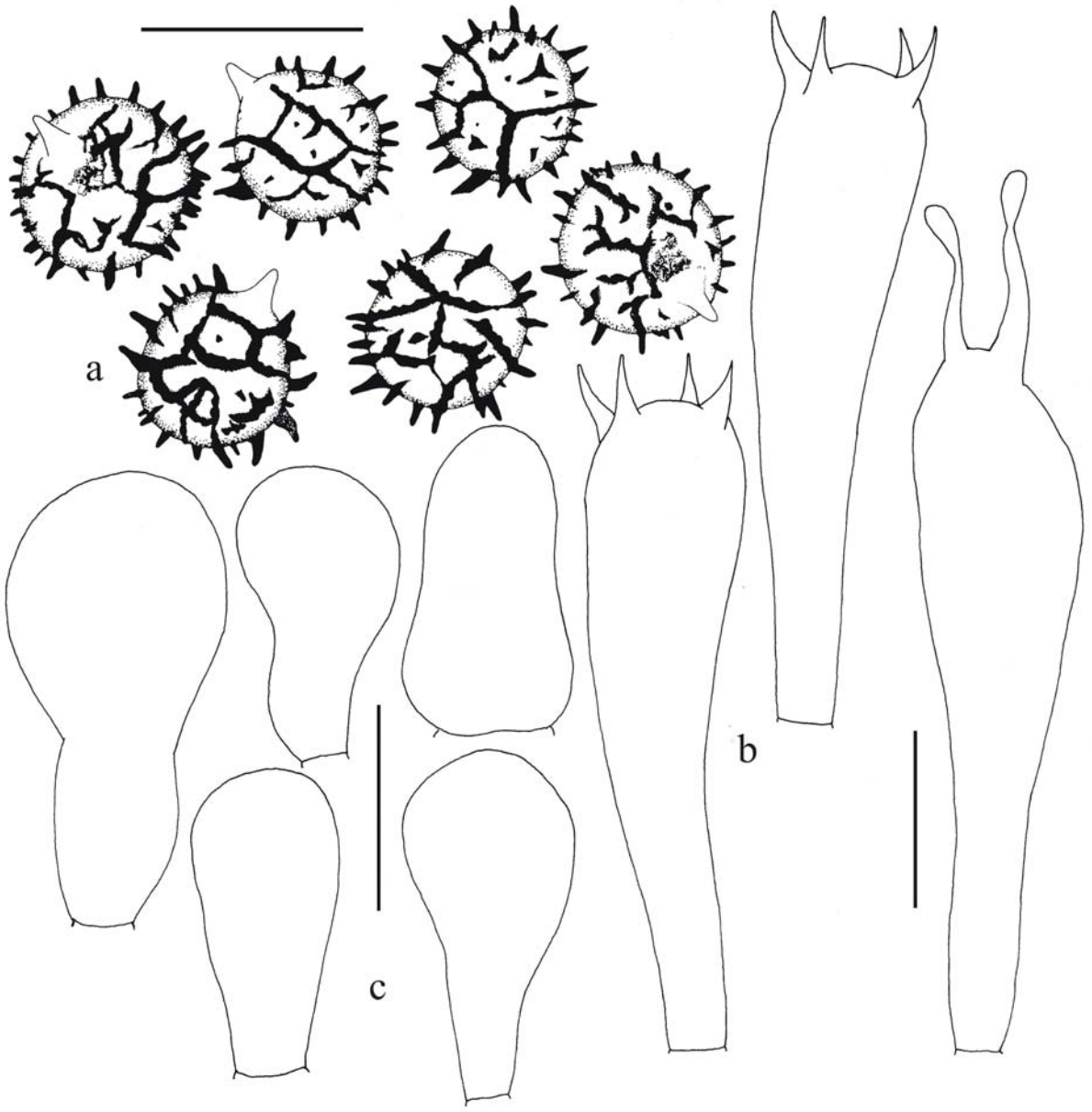

Fig. 5. *Lactarius strigosipes* (**from holotype**). **a.** Basidiospores. **b.** Basidia. **c.** Marginal cells of lamellae. Bars = 10 µm.

orange (6B5), grayish-vinaceous or dark vinaceous-brown (7C5, 7D7), orange-brown (6CB6) in mature specimens, beige-orange to grayish-orange (5B5, 6B4-5) when seen in group, or pale orange (5A4-3), beige-orange (5B4, 6B5) on side view, not yellow, gradually stained grayish-orange or brown-orange (6B5, 6C6) in old specimens, edge irregular, at times bifurcate. *Stipe* 18–80 × 1.5–6 mm, subcylindrical, sinuous, paler than pileus, smooth, apex somewhat striate, hygrophanous, fibrous, showing longitudinal fibers when cut, bright-orange to ochraceous-orange (6C7, 5B5) upwards, bright orange-brown when young (6C8), then orange-brown (6E8, 6D7-8) when old, not reddish, slightly paler at apex with reddish tinges and at times up to 2/3 downwards the stipe vinaceous-brown (7D7-8, 7E8, 8D8), apex slightly tomentose, hollow, at times growing in fascicles; base strigose, covered with numerous thick and hairy structures, beige-pinkish or orange-brown. *Context* of pileus thin, hygrophanous, fibrous in the stipe, whitish with grayish-orange tinges

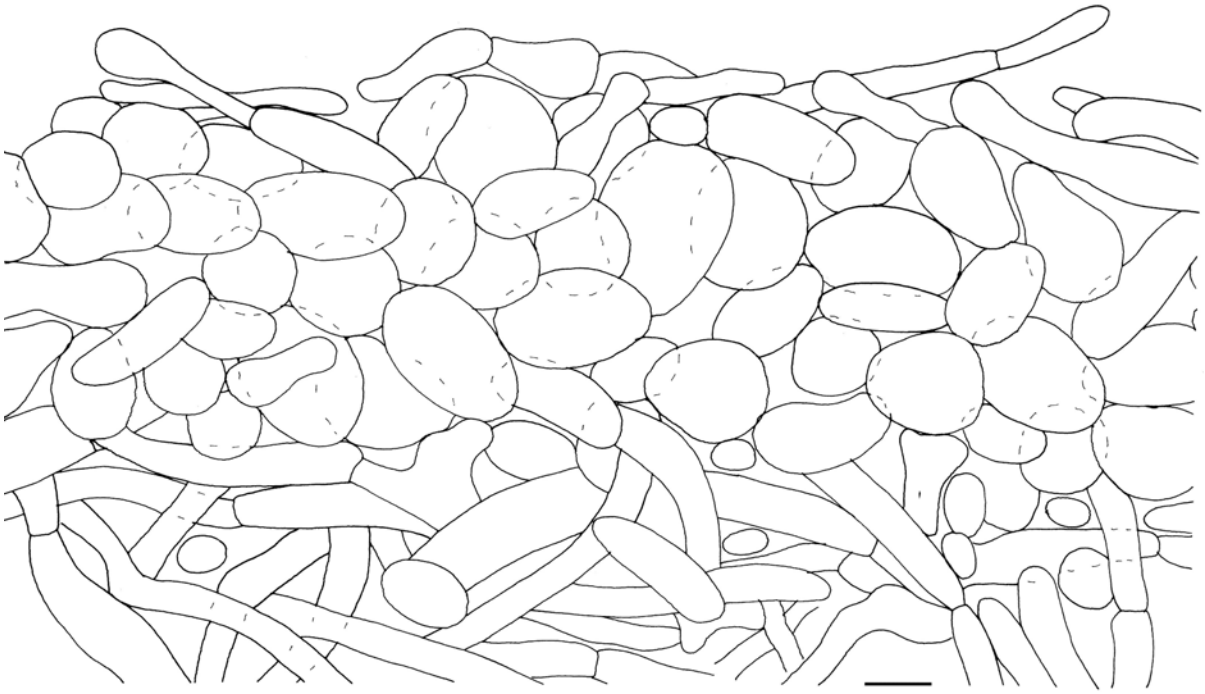

Fig. 6. *Lactarius strigosipes* (**from holotype**). Pileipellis. Bar = 10 μm.

towards the stipe. *Odour* agreeable, intense, somewhat of geranium, strongly spicy when dried (more or less nutty). *Taste* mild. *Latex* serous, watery-white, unchanging, not staining white paper. *Spore print* cream-yellowish.

Basidiospores 6-7.5 × 6-7 (-7.5) μm, $\bar{x}$ = 6.7-7.2 × 6.4-6.5 μm, **Q** = 1-1.13, globose to subglobose, subreticulate, with isolated ridges and warts, ornamentation 1-2 μm high, suprahilar plage at times with faint amyloid dots. Under SEM the ornamentation appears composed of short and wide ridges joined forming reticulations or free and with isolated warts, the ridges have a more or less continuous or irregularly lobed edge, some basidiospores look more reticulated than other. *Basidia* 30-51 × 8.4-10.8 μm, clavate, subcapitate, at times subcylindrical, bi- or tetrasporic, with granular refringent contents. *Cystidia* lacking. *Pseudocystidia* 2.4-3.6 μm diam, subcylindrical, contorted, apex rounded or somewhat slender, yellowish. *Marginal cells* clavate, subcylindrical or somewhat capitate, 14.4-22 × 7.2-10 μm, yellowish, thin walled. *Pileipellis* pseudoparenchymatous, a thin layer 48-96 μm depth, composed of broadly inflated cells, other subisodiametric (7.2-30 μm diam), thin walled, yellowish, some areas with patches of cylindrical cells, 3.6-6 μm diam, other with terminal subclavate or at times pyriform cells, 20.4-31.2 × 6-10.8 μm, thin walled, hyaline. *Context hyphae* 3.6-15.6 μm diam, laticifers 3.6-16.8 μm diam, sphaerocytes 18-36 μm diam, in rosettes, thin walled. *Hymenophoral trama* narrow, 38.4-60 μm broad, hyphae 3.6-6 μm diam, laticifers 2.4-7.2 μm.

Habitat: Gregarious, in small troops, near *Carpinus caroliniana*, in subtropical cloud (mesophytic) forest, at 1300 m alt.

Material examined: MEXICO. Veracruz, Mpio. Xalapa, Instituto de Ecología, Santuario del Bosque de Niebla, 22 January 2003, *Corona 192*; 26 June 2003, *Corona 262*; 2 July 2003, *Corona 268*; 17 September 2003, *Corona 331*; 7 July 2004, *Montoya 4122*; 9 July 2004, *Montoya 4144*; 11 August 2004, *Montoya 4157*; 8 June 2006, *Montoya 4521* (**holotype**); 6 July 2006, *Montoya 4601*. Mpio. San Andrés Tlalnehuayocan, San Antonio, 10 September 2001, *Montoya 3689* (all at XAL).

Other material examined: U.S.A. Michigan**,** near Leslie, 24 July 1900, *Longyear* (**Holotype** of *Lactarius subserifluus* NYS).

Notes: *Lactarius strigosipes* is a slender, orange-brown to reddish-brown fungus, with a characteristic odour in fresh and dry condition, having narrow, thin and crowded lamellae, strigose stipe base, thin pileus context with sphaerocytes presence, subreticulate basidiospores, and pseudoparenchymatous pileipellis.

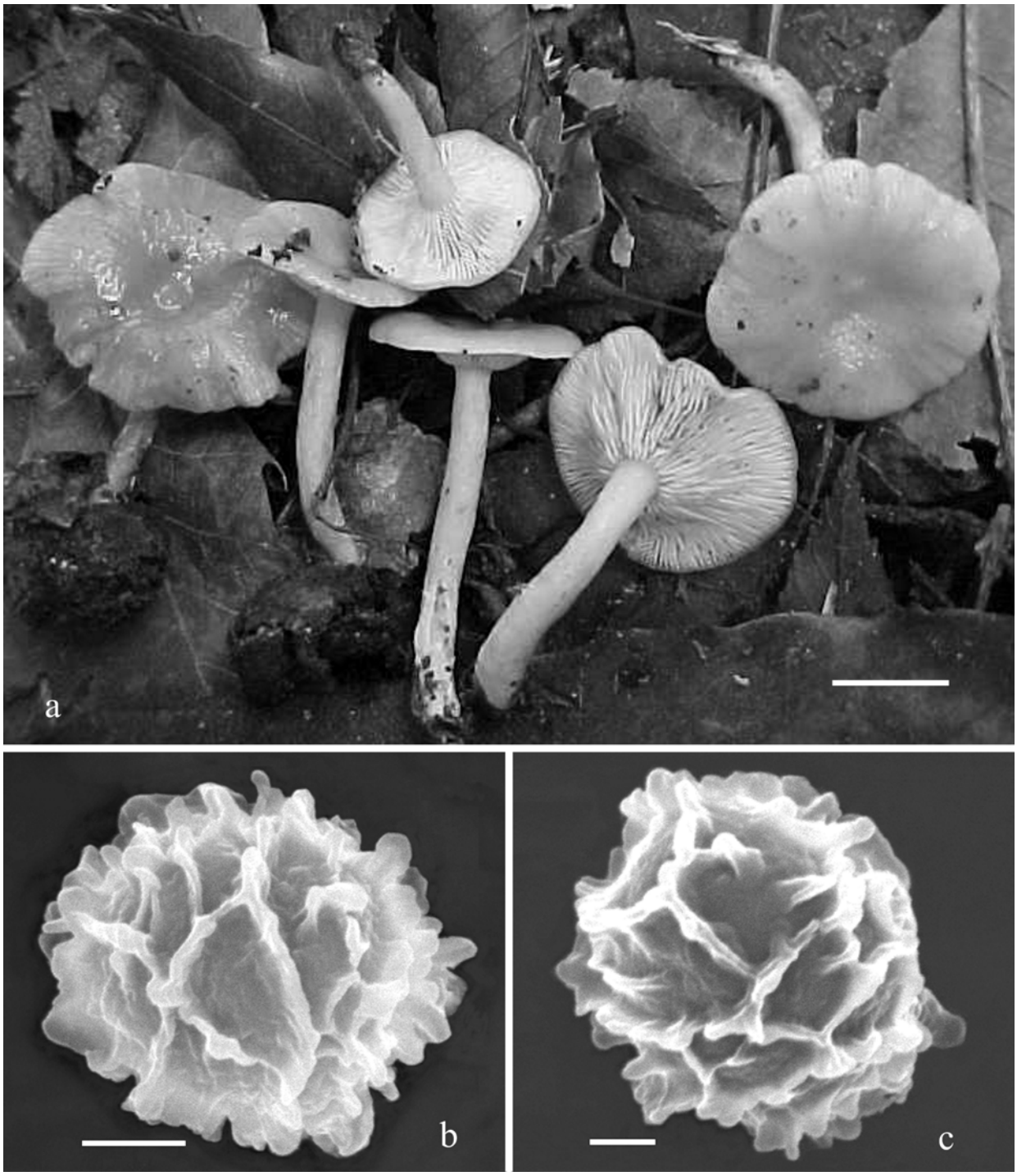

Fig. 7. *Lactarius minusculus* (Montoya 4009). **a.** Basidiomes. **b-c.** Basidiospores under SEM. Bars: a = 10 mm, b = 2 μm, c = 1 μm.

This species could be confused with the American *Lactarius subserifluus* Longyear (another member of Sect. *Olentes*), which is superficially similar. *Lactarius subserifluus* differs however, by the distinctly thicker, more distant and broader lamellae, as showed in the type specimen and depicted also in the basidiomes illustrated by Longyear (1901). The discrepancies are also evident when comparing stipes in both taxa. According to Longyear (1901) in *L. subserifluus* the stipe base is pruinose (observed also in the type specimen),

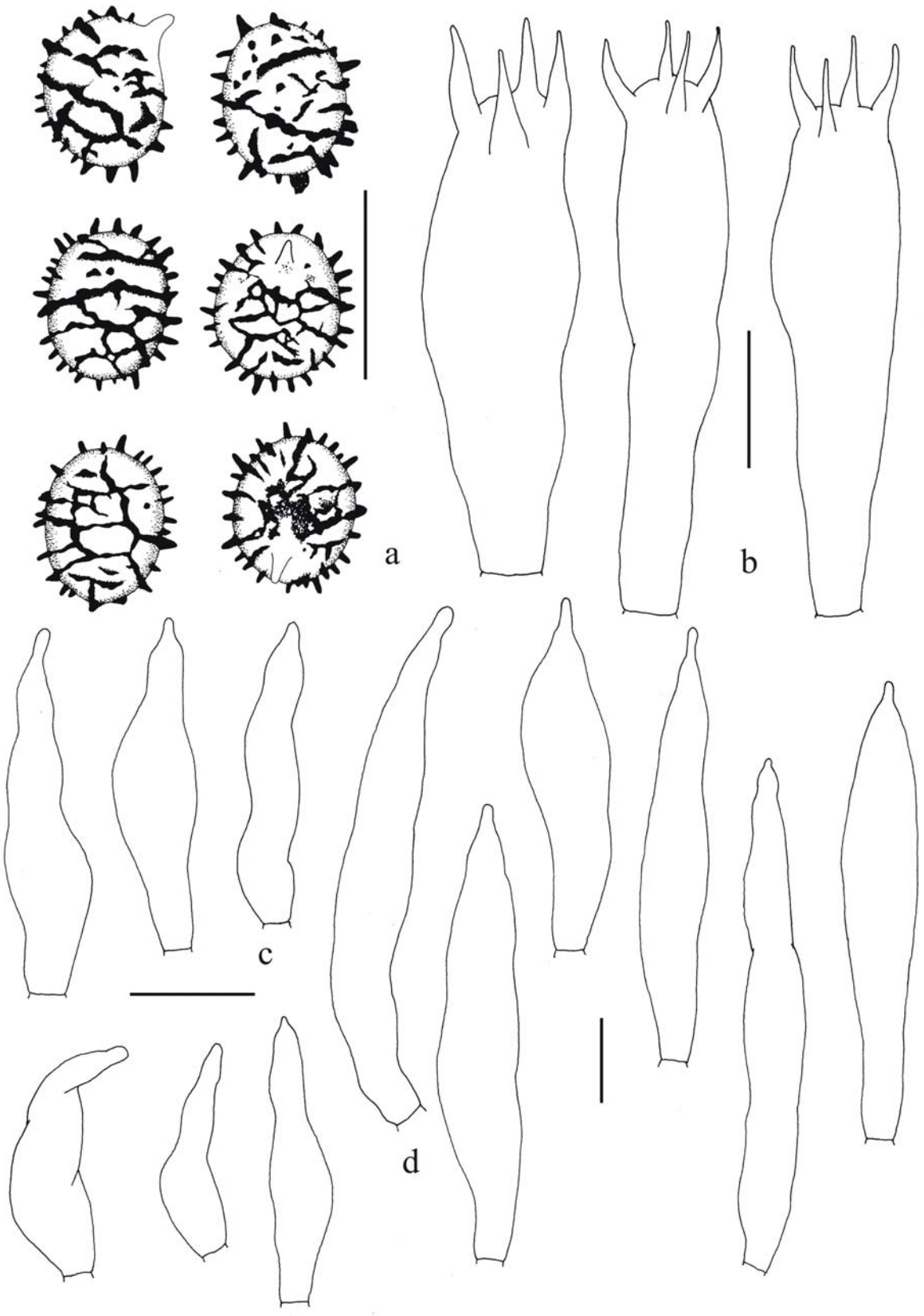

Fig. 8. *Lactarius minusculus* (Montoya 4121). **a.** Basidiospores. **b.** Basidia. **c.** Cheilocystidia. **d.** Pleurocystidia. Bars = 10 μm.

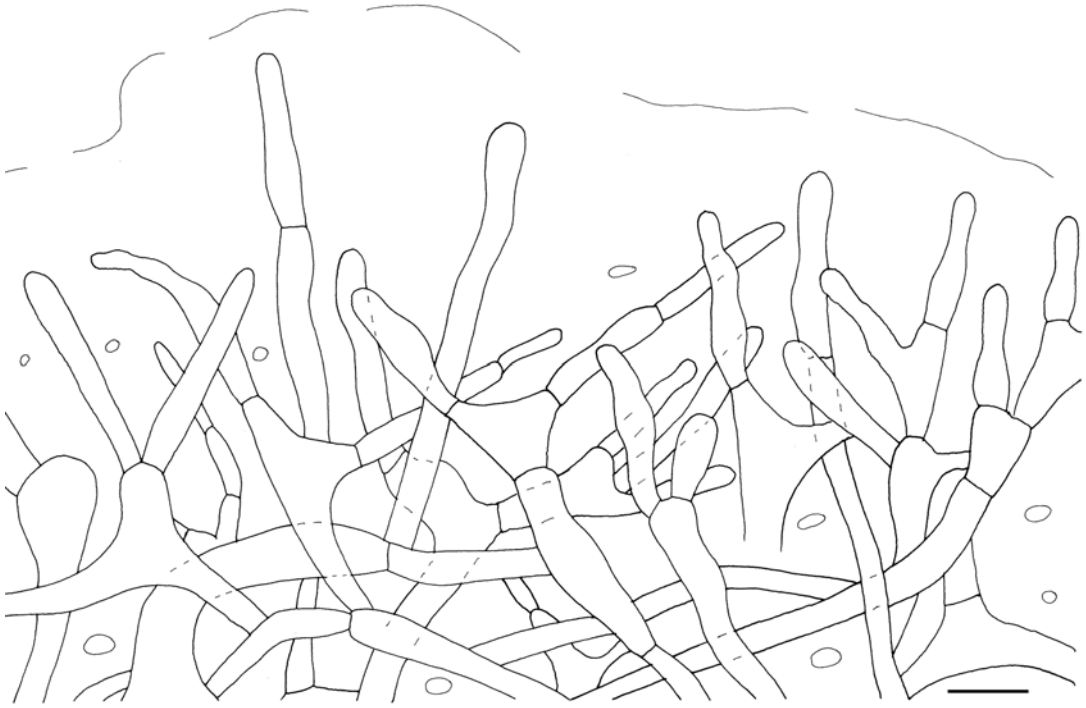

Fig. 9. *Lactarius minusculus* (Montoya 4095). Pileipellis. Bar = 10 μm.

while the Mexican taxon produces conspicuous, evident strigose structures. In addition, *Lactarius subserifluus* was described as odorless (Longyear, 1901), while *L. strigosipes* emits a distinct smell, either in fresh or in dry condition. Moreover, the type specimen of *L. subserifluus* lacks rosettes of sphaerocytes as also Hesler and Smith (1979) observed in collections from U.S.A. Description and illustration provided by Hesler and Smith (1979) of *L. subserifluus* also shows that it is a different species. They additionally recorded a faintly peppery taste and mentioned that the base of the stipe is strigose; we did not see this feature in the type.

In the area of study, *Lactarius strigosipes* is sympatric with *L. areolatus* (see above) but the basidiomes of the first one appear mostly in short periods, mainly through the summer-autumn seasons, when it becomes relatively common in the area.

Lactarius minusculus Burl., Bull. Torrey Bot. Club 34, p. 88 (1907). (Figs 7-9)

Pileus 5-30 mm diam, convex, plano-convex to depressed in the centre with age, concave to uplifted, at times with a small acute papilla, yellow-orange, grayish-orange to flesh colour (5A6, 5B5-8, 5C5-6), honey colour (**7.5 YR 6/8)**, orange with brown tinges (6C6), centre pale orange (5A4-6) to brighter orange (6A6-5, B6-7, 6C6), orange-brown (6C8-7, 6D7), margin paler, pruinose at edge when young, viscid to subviscid, at times lubricous, hygrophanous; margin finely striate to crenate, decurved when young to moderately incurved and undulate, finely translucent striate, pale yellow (4A3-5), thinner and irregular towards the edge. *Lamellae* crowded to close, adnate to decurrent, broad, 1.5-3 mm width, frequently bifurcate, arcuate; pale orange (5A2-4) or yellowish, cream-yellow (3A2, 4A2-3) to pale salmon (6A3) or grayish-orange (6B4), with different length series of lamellulae. *Stipe* 11-53 × 1.5-6 mm, subcylindrical to slightly wider towards the base, solid, yellowish-cream (4A2-3), pale salmon (5A2-4, 5B4) to grayish-orange (6B4-6, 6C6), paler towards the apex to cream-orange, dry to slightly oily, glabrous, smooth, base with rests of mycelium or at times somewhat strigose. *Context* slightly hygrophanous, margin concolorous with pileus

surface, pale orange (5A2) towards the centre. *Odour* mild to agreeable. *Taste* mild to slightly acrid. *Latex* white to serous, unchanging, taste slightly bitter then acrid, colour invariable in white paper.

Basidiospores 7-8 (-8.5) × 6-6.5 µm; $\bar{x}$ = 7.6 × 6.2 µm; **Q** = 1.21; broadly ellipsoid, subreticulate or with an incomplete reticulum, ornamentation up to 1.25 µm high, suprahilar plage at times with a faint irregular amyloid tinge; under SEM seen to be composed of irregular ridges, joined or at times disarticulated and also isolated warts present, the ridges edge is variable from more or less continuous to irregularly lobed. *Basidia* 41-45 × 8–13.5 µm, tetrasporic, clavate, yellowish, thin walled. *Pleurocystidia* 23.5-75 × 6-11 µm, subfusoid, subventricose, apex acute or at times mucronate or contorted. *Pseudocystidia* 2-4 µm diam, subcylindrical, contorted, yellowish. *Cheilocystidia* 16.5-29 × 3.5-7 µm, subfusoid, subventricose, at times mucronate. *Pileipellis* an ixotrichoderm of 90-140 µm broad, gelatinous matrix thick or at times thin, with more or less erect, filamentous, subcylindrical, sinuous cells, thin walled, yellowish, terminal elements 2-5 µm diam, other more inflated of 6-10 µm diam, with presence of isolated laticifers with dense contents. *Context* hypae 4-10 µm diam, laticifers 4-12 µm diam, sphaerocytes 18-35 µm diam. *Hymenophoral trama* hyphae 4-5 µm diam, laticifers 2-6 µm diam.

Habitat: Gregarious, in small troops, frequently among moss. In subtropical cloud (mesophytic) forest, under *Carpinus caroliniana* or in other areas between *Quercus* cf. *germana*-*C. caroliniana*., at 1300 m alt.

Known distribution: U.S.A., Mexico

Material examined: MEXICO. Veracruz, Mpio. Xalapa, Instituto de Ecología, Santuario del Bosque de Niebla, 4 July 2003 *Montoya 4009*, 16 June 2004, *Montoya 4095*; 7 July 2004, *Montoya 4121*; 9 July 2004, *Montoya 4134*; 17 August 2004, *Montoya 4173*; 9 November 2004, *Montoya 4241*; 3 March 2005, *Montoya 4244*; 31 May 2005, *Jarvio 2116*; 12 July 2005, *Jarvio 2191*; 21 November 2005, *Jarvio 2440*; 11 November 2005, *Montoya 4422*.
(all at XAL).

Other material examined: U.S.A. New York, Vermont, *Burlingham*, 21, 23 July 1906 (**Holotype** of *Lactarius minusculus,* NY).

Notes: *Lactarius minusculus* has been placed in sect. *Russularia* (Hesler and Smith, 1979). It can be recognized by the combination of its short size, colour variation, viscid pileus, close and broad lamellae, somewhat acrid taste, the basidiospore form and ornamentation. *Lactarius minusculus* is one of the common species found among moss at the study site and at times some basidiomes were recorded growing on *Quercus* acorns or wood remains. Hesler and Smith (1979) cited it growing in moss or on decayed wood too. According to the monitoring in the area, *L. minusculus* produces scarce fructifications, however, it appears practically all the year, from spring to autumn, even a couple of times it was observed growing in January.

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