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# Notes on *Thelephora atrocitrina* and *T. cuticularis*

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Summary. *Thelephora atrocitrina* and *T. cuticularis* are described and illustrated with microscopical drawings on the basis of fresh material from recent collections. *T. atrocitrina* is easily distinguished from other members of the genus by its characteristic V-shaped, dark gray to blackish basidiomes remarkably contrasted by a white margin with yellow tinges. The microscopically diagnostic character of *T. cuticularis* is the absence of clamp connections.

Zusammenfassung. *Thelephora atrocitrina* und *T. cuticularis* werden anhand von frischem Material aus neueren Funden beschrieben und ihre mikroskopischen Merkmale abgebildet. Typisch für *T. atrocitrina* sind das V-förmige, dunkelgraue bis schwärzliche Basidiom in auffallendem Kontrast zum weissen Rand mit gelben Tönen. Das mikroskopisch diagnostische Merkmal für *T. cuticularis* ist die Abwesenheit von Schnallen an allen Septen.

Riassunto. *Thelephora atrocitrina* e *T. cuticularis* vengono descritte e documentate con illustrazioni degli elementi microscopici sulla base di materiale fresco proveniente da raccolte recenti. Un carattere distintivo di *T. atrocitrina* sono i basidiomi a forma di V, con un colore dal grigio scuro al nerastro, in vistoso contrasto con il margine bianco dalle tipiche sfumature gialle. *T. cuticularis* si riconosce facilmente al microscopio per l'assenza di unioni a fibbia.

## Methods

Microscopic observation was performed on hand-cut slices that were soaked for 5 minutes in 95% ethanol and air-dried. To record the natural color of the cells and any color changes occurring in alkaline milieu the sections were

mounted in water, 2% KOH or a solution consisting of 80% (v/v) of an alkaline buffer (0.1 M glycine, 0.1 M KCl, adjusted to pH = 11.0 with KOH, abbreviated Gly/KOH) and 20% glycerol. Microscopical elements were drawn using either hand-cut slices or squash mounts in 2% KOH stained with aqueous phloxine to enhance contrast. Dimensions of microscopical elements are given as the mean  $\pm$  standard deviation and the range (min–max) of n measurements from S specimens (n/S).

*Thelephora cuticularis* Berk., Lond. J. Bot. 1847, 6: 324

Specimen examined, on which descriptions are based. *Thelephora cuticularis* Berk.: Italy, Province Pordenone, Fanna, 250 m above sea level, on chalky concrete among various residues of construction materials left along a country track, 9. VIII. 1987, leg. G. Zecchin, Herb. A. Baici 2322. Although an association with buried wood or roots via mycorrhiza seems plausible, this could not be ascertained due to the compact nature of the unusual substratum.

Basidiomes solitary or gregarious, individually separable or conrescent, with tapering base, cuneiform, spatulate, flabelliform or irregularly cylindrical, soft coriaceous. Individual basidiomes up to 20 mm tall and 10 mm wide. Abhymenial surface roughly radially rugose, looking innately fibrillose by appressed hyphae, black when fresh, pale brown upon drying. Hymenial surface even, black to purplish black when fresh and wet, fading to pale gray-brown after drying, fertile down to the base. Herbarium material reassumes the original black color upon rehydration. Margin sterile, pubescent, white in the wet state and pale brown after drying. Context up to 1 mm thick, though, pale brown when dry. In the exsiccatum the basidiomes become hard, but not brittle and develop a strong, persisting, more or less unpleasant smell.

Hyphal system monomitic, all hyphae with simple septa (Figs. 1, 2). Context hyphae pale yellowish brown when examined individually, much darker when seen in mass, with unchanging color in both water and alkaline media, slightly thick-walled (about 0.5  $\mu$ m),  $4.7 \pm 0.5 \mu$ m in diameter (range 4.0–5.0  $\mu$ m, n/S = 15/1) with occasional secondary septa, branched frequently in the proximity of a primary septum. Hyphae at the abhymenial surface morphologically similar to context hyphae, but agglutinated, more compact and darker (brownish). Subhymenial hyphae derive from context hyphae that bend at 90 degrees towards the hymenial surface, subhyaline to pale yellowish brown in water, immediately cyanescent in 2% KOH and Gly/KOH, thin-walled,  $4.4 \pm 0.5 \mu$ m wide (range 4.0–5.0  $\mu$ m, n/S = 15/1). Hymenial layer without cystidia, but with sparse short-celled paraphysoid hyphae extending from the subhymenium. Basidioles forming the bulk of the hymenial palisade and eventually developing to basidia, hyaline to pale yellowish brown, not or rarely cyanescent in alkaline solution,  $53 \pm 10 \mu$ m long (range 34–75  $\mu$ m) and  $11 \pm 1.8 \mu$ m wide (range 9.0–16.0  $\mu$ m, n/S = 15/1).

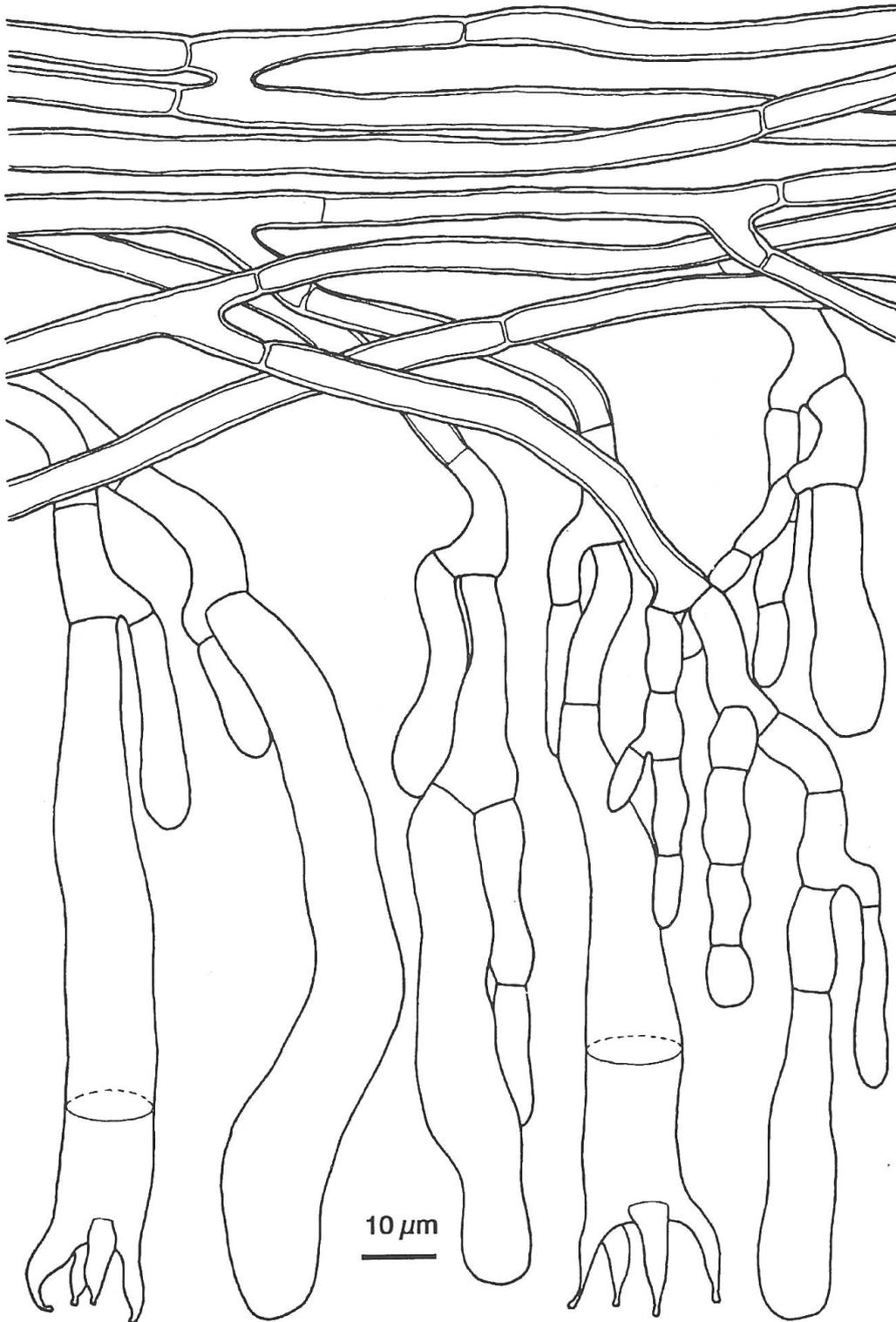


Figure 1. *Thelephora cuticularis* (AB-2322): context and subhymenial hyphae, mature basidia and basidioles.

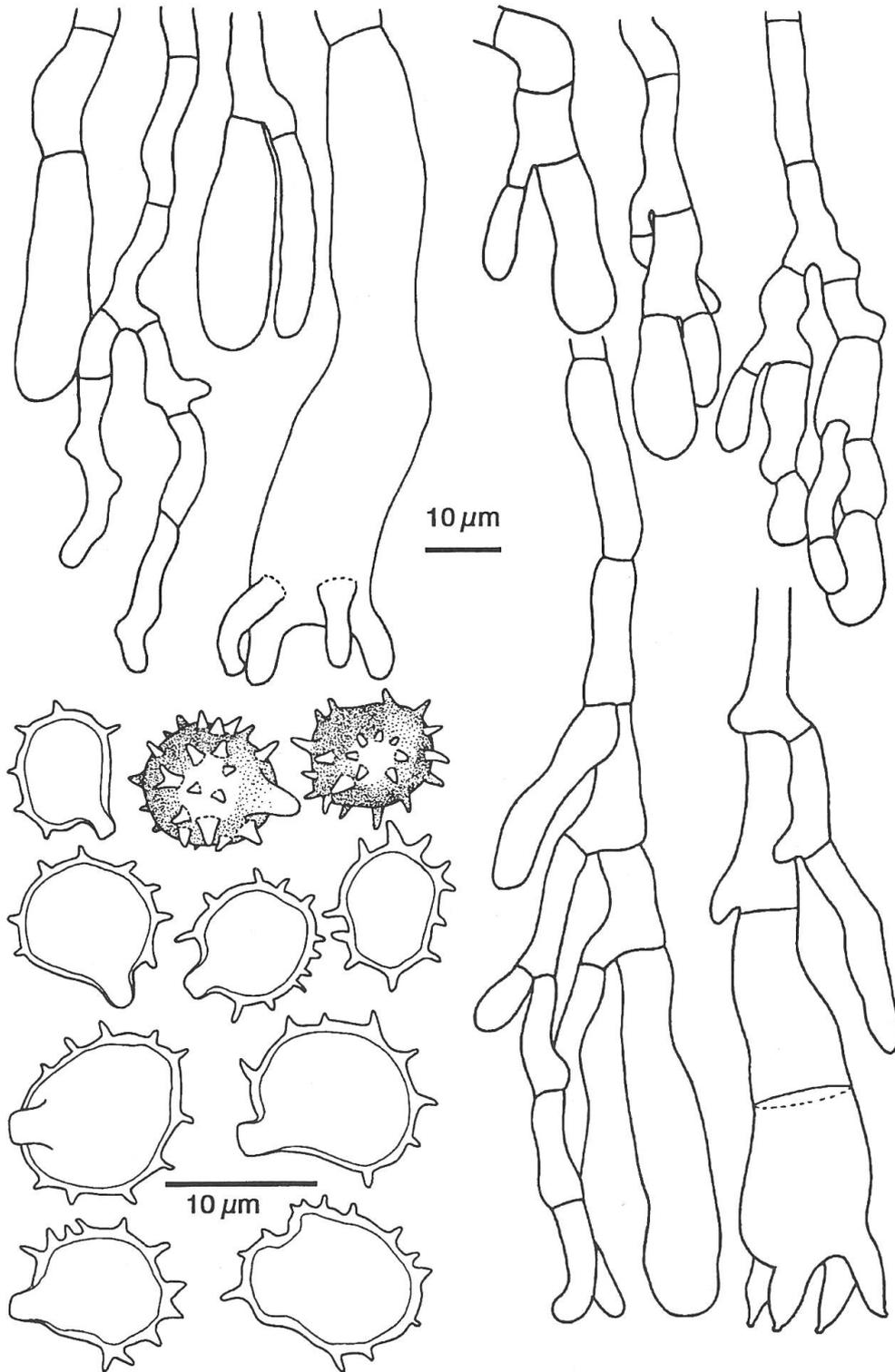


Figure 2. *Thelephora cuticularis* (AB-2322): basidia, paraphysoid elements, subhymenial hyphae and spores.

Basidia clavate, hyaline or pale yellowish brown, not cyanescent in alkaline solution, with a simple septum at the base,  $78 \pm 11 \mu\text{m}$  long (range 60–95  $\mu\text{m}$ ) and  $13.9 \pm 1.4 \mu\text{m}$  wide (range 12.0–16.0  $\mu\text{m}$ , n/S=15/1), with normally four sterigmata that are up to 15  $\mu\text{m}$  long. Many basidia bear a secondary septum at approximately  $\frac{2}{3}$  of the length (measured from the basal septum) and sometimes two such septa divide the cell in three segments of roughly the same length (Figs. 1, 2).

Spores yellowish brown or light brown in both water and alkaline solutions, mostly regular in outline, broadly ellipsoid or rarely subglobose, a few looking somewhat irregular, echinulate with simple (common) or bifurcate spines (less frequent),  $9.9 \pm 1.3 \mu\text{m}$  long (range 8.0–12.0  $\mu\text{m}$ ) and  $8.3 \pm 0.8 \mu\text{m}$  wide, (range 6.0–10.0  $\mu\text{m}$ , n/S=36/1, measurements without spines, Fig. 2).

*Thelephora atrocitrina* Quél., Mem. Soc. Émul. Montbéliard, 2<sup>e</sup> Sér., complément du 5<sup>e</sup> Vol. (1875) 443, tab. II, fig. 8, [= Champ. Jura et Vosges III (1875)15].

Specimens examined, on which descriptions are based. *Thelephora atrocitrina* Quél.: (1) Italy, Province Pordenone, Fanna, 250 m above sea level, on the ground, associated with angiosperms, 2. VIII. 1989, leg. G. Zecchin, Herb. A. Baici 2724; (2) Italy, Province Treviso, Gorgo al Monticano, Bosco Cavalier, 15 m above sea level, on the ground, associated with angiosperms, 28. VI. 1992, leg. G. Ricci, Herb. A. Baici 2757, part of this collection was deposited at the herbarium of the Museo Civico di Venezia (MCVE) under the number MCVE 416; (3) Switzerland, Canton Jura, Foret de Bassecourt, on the ground, 16. IX. 1992, leg. E. Chetelat, det. J. Keller, Herb. J. Keller 5092. These collections represent mature basidiomes and just AB-2724 had a collapsed hymenium, but numerous spores.

Basidiomes solitary or caespitose forming a rosette with separable individuals, typically obconical (sagittal section V-shaped) or pseudo-stalked. The pseudostipe, if present, may be slightly radicate. Individual basidiomes up to 50 mm tall and 35 mm wide. Margin undulate, thick, sharply differentiated from the remaining of the basidiome by its white to vividly citrine color when fresh and pale yellowish to dirty white when old, sterile. Hymenial surface smooth, dark gray to almost black when fresh, except at the paler margin, becoming grayish brown, reddish brown or vinaceous when dry, fertile down to the base. Context 1–2 mm thick, pale brown with dark, shiny zones. The whole basidiome soft coriaceous, loosening much water upon drying and becoming very light and brittle.

Hyphal system monomitic with clamp connections (Figs. 3–5). Context hyphae (Fig. 4) hyaline to pale yellow, of the same color in water and alkaline media, thin-walled or wall slightly thickening, running more or less parallel to the hymenial surface, with frequent branches, clamped at almost all septa,

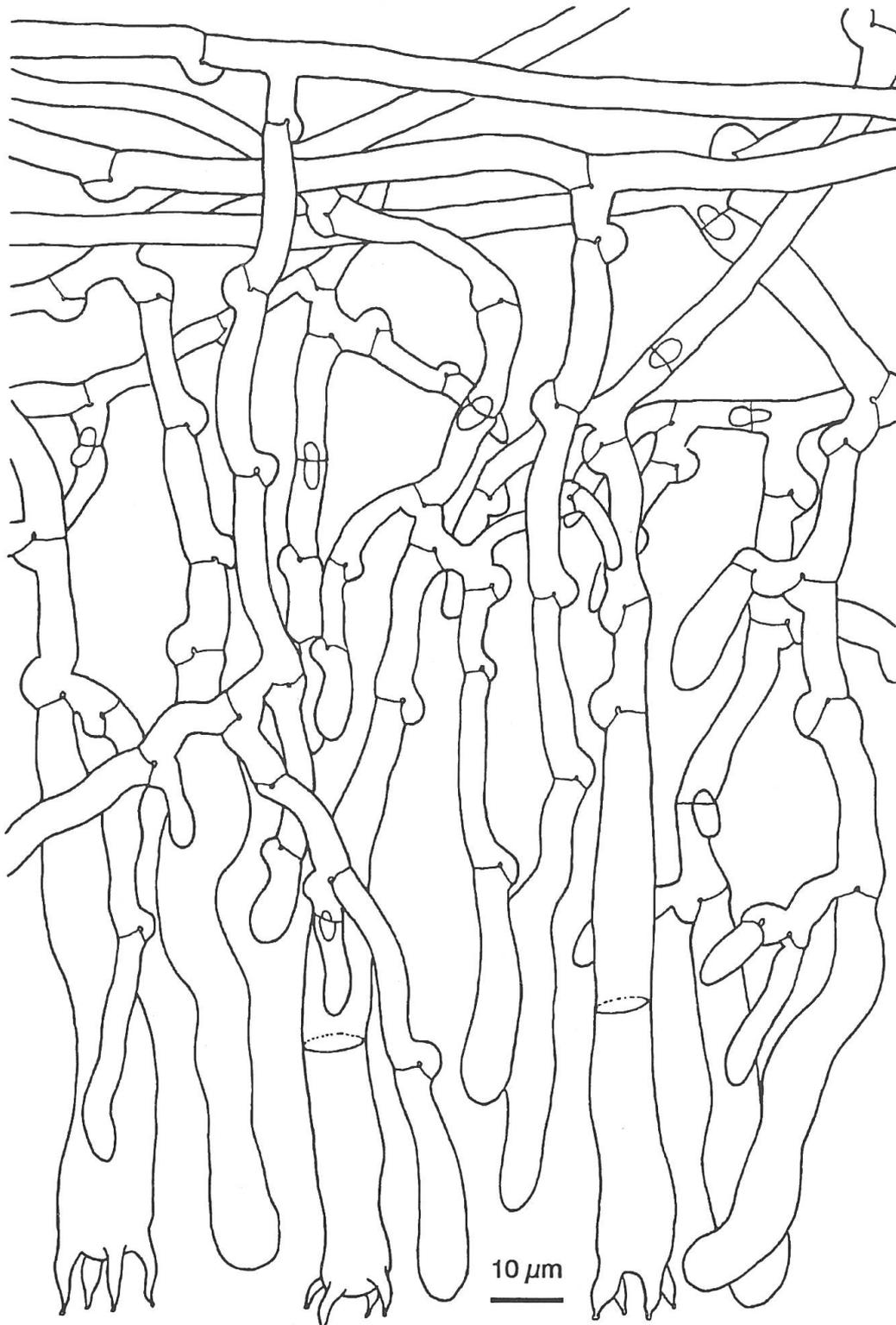


Figure 3. *Thelephora atrocitrina* (AB-2757): subhymenial hyphae, basidia and basidioles.

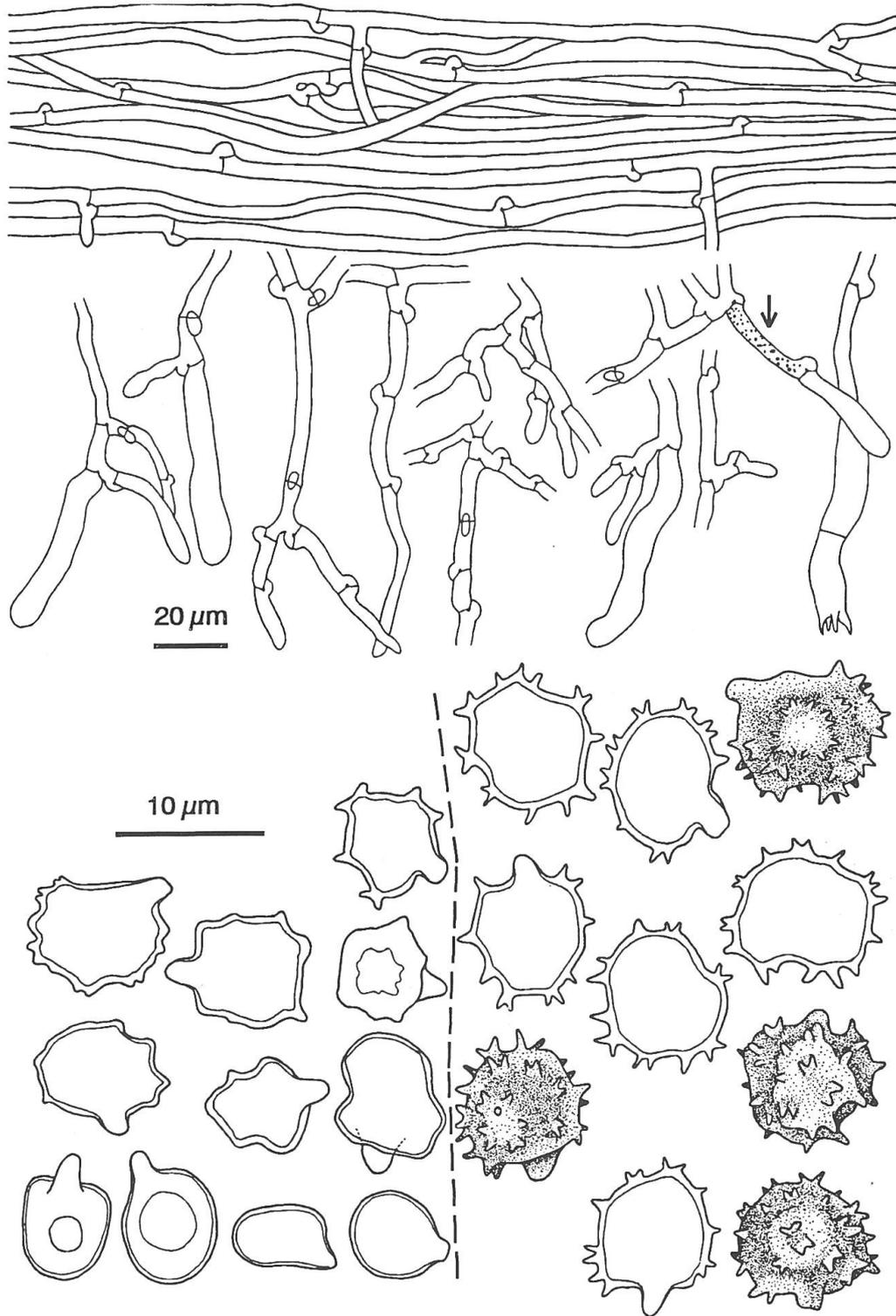


Figure 4. *Thelephora atrocitrina* (AB-2757): context hyphae, subhymenial hyphae and hymenial elements (upper half). Pigments of subhymenial hyphae remain granular in Gly/KOH (arrow). Immature (lower half, left) and mature spores (right).

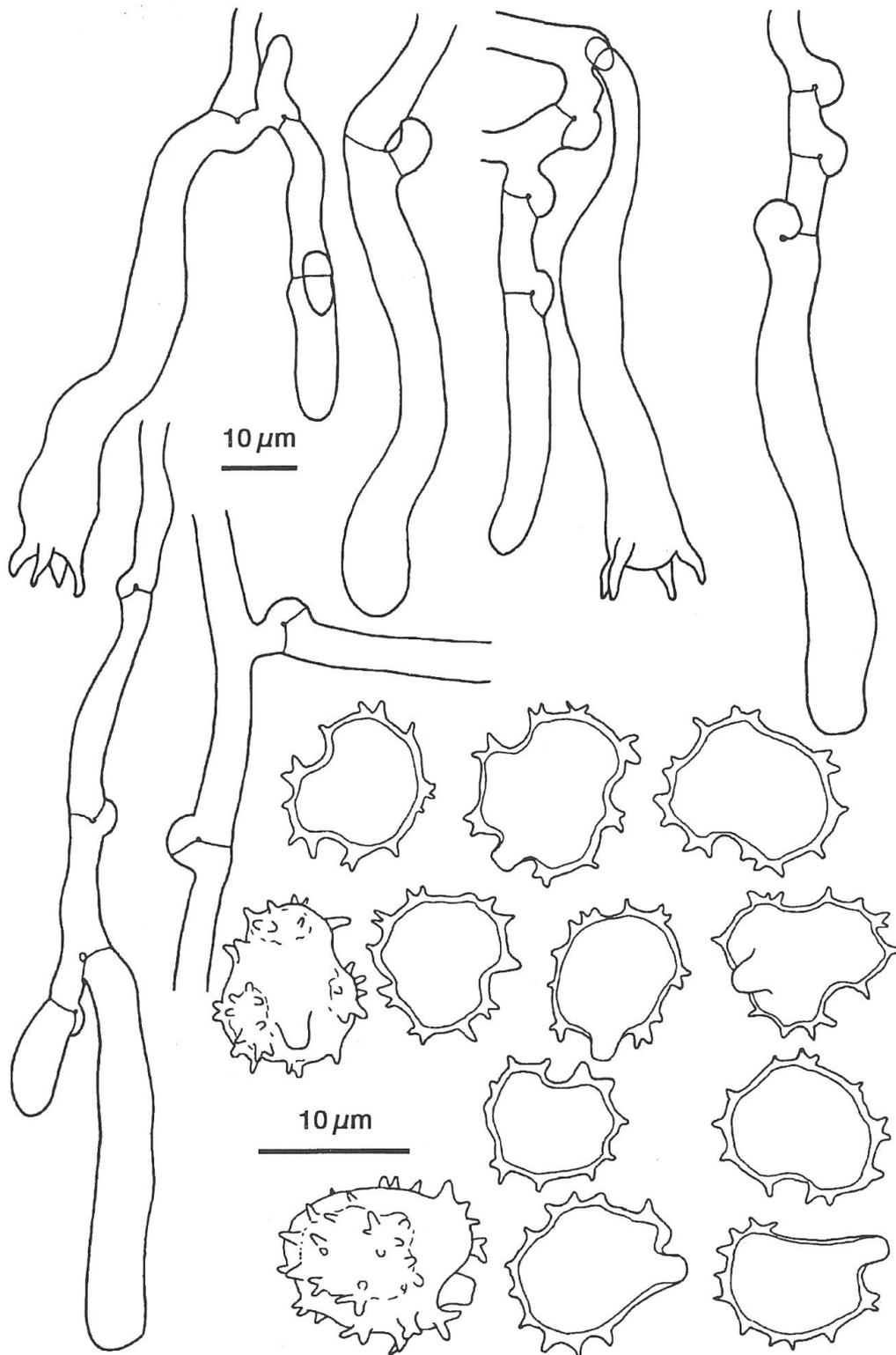


Figure 5. *Thelephora atrocitrina* (J. Keller 5092): basidia, basidioles and spores.

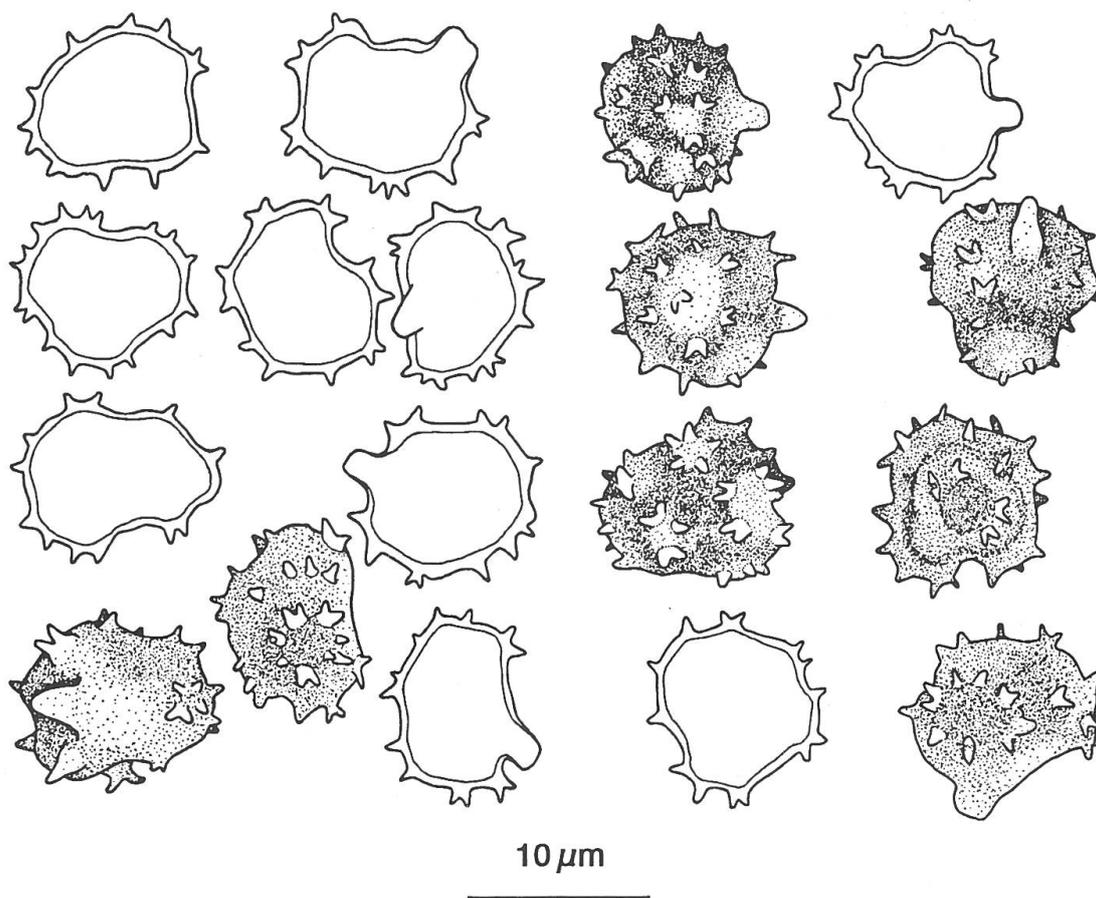


Figure 6. *Thelephora atrocitrina* (AB-2724): spores.

$4.8 \pm 0.7 \mu\text{m}$  wide (range  $3.5\text{--}6.0 \mu\text{m}$ ,  $n/S=45/3$ ). Subhymenial hyphae arising from context hyphae by bending at right angles towards the hymenium, short-celled, clamped at all septa, hyaline in water mounts, typically cyanescent in alkaline solution,  $18 \pm 5.6 \mu\text{m}$  long (range  $9.0\text{--}29.0 \mu\text{m}$ ) and  $4.1 \pm 0.6 \mu\text{m}$  wide (range  $3.0\text{--}5.0 \mu\text{m}$ ,  $n/S=45/3$ ), thin-walled. Pigments of subhymenium remain granular (Fig. 4), deep green in Gly/KOH and slowly dissolve, producing a cyanescent diffusate, in 2% KOH. Hymenial layer without cystidia, the bulk of the cells being formed by basidioles  $48 \pm 10 \mu\text{m}$  long (range  $30\text{--}70 \mu\text{m}$ ) and  $8.1 \pm 1.0 \mu\text{m}$  wide (range  $7.0\text{--}10.0 \mu\text{m}$ ,  $n/S=36/2$ ), with some hyploid cells intermingled between them.

Basidia clavate, hyaline, not cyanescent in alkaline solution, clamped at the base,  $70 \pm 9 \mu\text{m}$  long (range  $50\text{--}90 \mu\text{m}$ ),  $10.2 \pm 1.4 \mu\text{m}$  wide (range  $7.0\text{--}13.0 \mu\text{m}$ ,  $n/S=29/2$ ), usually bearing 4 sterigmata that are up to  $10 \mu\text{m}$  long. Some basidia are secondarily septate (Figs. 3–5).

Spores pale brown, irregularly elliptical or rarely irregularly subglobose,

echinulate with commonly bifurcate or, less frequently, single spines,  $9.0 \pm 0.7 \mu\text{m}$  long (range 7.0–10.5  $\mu\text{m}$ ) and  $7.9 \pm 0.7 \mu\text{m}$  wide (range 6.0–10.0  $\mu\text{m}$ ,  $n/S=108/3$ , measurements without spines, Figs. 4–6). Immature spores elliptical with big apiculus, deprived of spines and lobes, becoming irregular in outline after developing a few lobes and the spines (Fig. 4).

### Discussion

The descriptions of *Thelephora atrocitrina* and *T. cuticularis* in this paper, based solely on the collections mentioned above, agree with those of the literature. *T. cuticularis*, originally described from Ohio (Berkeley, 1847), has been reported from many North American States (Burt, 1914; Coker, 1921; Lentz, 1942). This species is poorly known in Europe and, to the best of our knowledge, was known up to date only from Somerset, England (Corner, 1968). Besides being rare, this interesting species has also been scarcely illustrated microscopically. Our sample AB-2322 from North Italy may represent the first documented collection from this country. The Italian specimen, consisting of many basidiomes in various stages of development, was in good condition, with fertile hymeniums and numerous spores. Judging from the photographs by Burt (1914, plate 5, fig. 14) and Lentz (1942, fig. 2) the morphology of the basidiome depends very much on the substratum, e.g. mossy bark or twigs encrusted by ascending basidiomes that develop reflexed lobes. In this respect, our collection was very particular: The real substratum was likely wood buried in a compact mass of chalky concrete or roots to which the specimen was associated in mycorrhizal connection and this unusual host undoubtedly determined its macroscopical appearance. The diagnostic character of *T. cuticularis* is the absence of clamps from all septa, a unique property within the genus *Thelephora*. A further distinguishing character is given by the quite regular, non-lobate spores, corresponding to the “regular spiny type” recently described by Stalpers (1993). Our specimen retains in the exsiccatum the strong odor described by Berkeley (1847) and Burt (1914), but not noted by Lentz (1942).

*T. atrocitrina* is also a rare species described from some European countries, for instance France (Quélet, l.c.; Bourdot et Galzin, 1927), Germany (Corner, 1968), Austria (Huber, 1941), Netherlands (Maas Geesteranus, 1955), Slovenia (cited by Novacký, 1956), Moravia (Novacký, 1956) and Norway (cited by Julich, 1984). Italy and Switzerland are now added to this list on the basis of the collections described above. Distinguishing characters of *T. atrocitrina* are the obconical basidiomes with undulate margin which is citric-yellow in young and mature specimens and fades out to pale yellowish to dirty white with age. The spores belong to the “irregular spiny type” of Stalpers (1993).

### Acknowledgment

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