

## The dilemma of species recognition in the field when sequence data are not in phase with phenotypic variability

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**Abstract** – This contribution on the genus *Cantharellus* in North America introduces three new look-alikes of already known species in the eastern United States and thereby exposes the problem of species delimitation in *Cantharellus*. The small, reddish pink to orange *C. corallinus* sp. nov. is yet another look-alike of *C. cinnabarinus*, while the new *C. flavolateritius* is proposed as a new southern relative of *C. lateritius*. Both new species are, however, genetically clearly divergent. Such a scenario of “cryptic species” corresponds to a concept that is widely admitted even by field mycologists and therefore easily accepted. In contrast, typical forms of the here newly described *C. velutinus* sp. nov. are nearly indistinguishable from *C. lateritius*, but the phenotypic variability of this new species (as supported by phylogenetic inference of the *TEF-1* gene) is so impressive that it is hard to accept that the various observed phenotypic forms all belong to a single species as indeed suggested by their identical sequence data. The latter scenario, that of lumping easily separable phenotypic forms in the field under a single species epitheton encounters much more criticism from most field mycologists and is only reluctantly accepted.

**Cantharellales / phylogeny / *Cantharellus corallinus* / *Cantharellus flavolateritius* / *Cantharellus velutinus* / North America / *TEF-1* gene/ taxonomy**

## INTRODUCTION

*Cantharellus* appears more and more to be a genus in which species retain a highly variable overall habit in the field, something that will again be demonstrated in the present paper with a quite impressive example from North America. The general color of the fruiting bodies depends most likely on a mixture of different carotenoid pigments that are present in variable quantities and the existence of albino forms has recently been demonstrated for several European chanterelles (Olariaga *et al.* 2015), Petersen (1985) already reported on the existence of bright yellow forms of *C. cinnabarinus* in the Appalachian mountains, whereas the coexistence of pink, bright yellow to bright orange color forms has been illustrated for the Malagasy *R. variabilicolor* Buyck & V. Hofstetter (in Ariyawansa *et al.* 2015). Due to its bright red color and small stature, *C. cinnabarinus* (Schwein.) Schwein. was generally considered to be a very common, widespread and unproblematic taxon among American chanterelles. In 2011, Buyck *et al.* described *C. texensis* Buyck & V. Hofstetter as a perfect look-alike from Texas where the latter species is far more common than *C. cinnabarinus*. In this paper, we describe *C. corallinus* sp.nov., yet another, apparently rare look-alike of *C. cinnabarinus* and compare it to similarly colored American chanterelles in subg. *Cinnabarinus* Buyck & V. Hofstetter. Quite undistinguishable in the field, these species can nevertheless be morphologically distinguished by clear differences in their pileipellis structure; they are equally significantly divergent at the molecular level.

In this paper, we will also describe two other new species. The first is a look-alike of *C. lateritius*; it is distinctly differentiated at the molecular level, but more collections will have to confirm whether or not it is also recognizable in the field. The third new species, *C. velutinus* sp. nov., is molecularly significantly different from other known American chanterelles. However, it is so surprisingly variable in its general stature and overall color that we at first doubted the correctness of the obtained sequences. Our results were also met with disbelief from various colleagues. Depending on the general morph, this new species can be confused with quite a number of other yellow, orange to even pink species in subg. *Cantharellus*.

## MATERIALS AND METHODS

*Morphological data.* – All sequenced collections were gathered by the first author and collaborators in the past few years. All cited specimens are deposited at the mycological herbarium of the Paris' Natural History Museum (PC) unless indicated otherwise. The color notations indicated in the descriptions follow Kornerup and Wanscher (1978). Microscopic features were examined and sketched by B. Buyck using a camera lucida setup. Original drawings for all elements of the hymenium or pellis were made at 2400. All microscopic observations and measurements were made in ammoniacal Congo red, after a short aqueous KOH pretreatment to improve tissue dissociation and matrix dissolution. Measurements of basidiospores cite length, width and length/width ratio (Q) in the following format: (minimum measured-) mean minus stand.dev. – *mean value* – mean plus stand.dev. (-maximum measured); statistics on spore measurements are based on 20 spores/collection. References to infrageneric placements follow the recently published genus phylogeny

(Buyck *et al.* 2014). Coloured illustrations are provided for all newly introduced taxa.

*Molecular data.* – Molecular sampling, sequence data and phylogenetic analyses are identical to those explained in Buyck *et al.* (2016c, this issue).

## RESULTS

### Phylogenetic results

All three newly described species received significant support in the *TEF*-1 analysis.

Both collections here identified as *Cantharellus corallinus* form a highly supported monophyletic clade (MLbs = 99%; BPP = 1). Our phylogeny (Fig. 1) places this species as sister to the European *C. friesii* Quél., although without support. *Cantharellus cinnabarinus* (MLbs = 93%; BPP = 1) is strongly supported as sister to the Chinese *C. phloginus*, while *C. texensis* occupies an unresolved, more basal position with respect to all of the above species.

All three collections of the newly described *C. flavolateritius* (MLbs = 93%; BPP = 1) are resolved as monophyletic and sister with high support (MLbs = 97%; BPP = 1) to *C. lateritius*. Both species constitute the most basal clade of *Cantharellus* subg. *Cantharellus*, although relationships within subg. *Cantharellus* are not fully resolved.

The clade composed of all collections of the newly described *C. velutinus* (MLbs = 96%; BPP = 1) occupies, within the analysed dataset, an isolated position in a strongly supported *Cantharellus* subg. *Cantharellus* (MLbs = 100%; BPP = 0.97).

### Taxonomy

*Cantharellus corallinus* Buyck & V. Hofstetter *sp. nov.*

**Figs 2-4, 11, 13**

*Mycobank*: MB 818376

*Diagnosis*: Differs from *Cantharellus texensis* and *C. cinnabarinus* in obtained sequence data for *TEF*-1 gene.

*Holotypus*: UNITED STATES. **Missouri**: St Louis Co., Forest 44, Conservation area, GIS: 38 31.636/- 090 31.021, 16 July 2011, 1083/Jay Justice MO-Canth-2 (PC 0713846)

**Fruiting bodies** slender and small. **Cap** up to 20-30 mm diam., convex to plane-convex or pitted-uneven with inrolled margin, soon becoming slightly depressed in the center with the outer margin often uplifted, irregular and lobed-sinuuous; surface pinkish red to orange-reddish or bright orange, dull, hygrophanous and rapidly becoming a very pale pinkish when drying out. **Hymenophore** decurrent, not abruptly delimited from the sterile stipe surface, forming rather well-developed gill-folds, mostly strongly veined-anastomosed in between, slightly paler than the cap surface. **Stipe** slender, 20-30 × 3-6 mm, subcylindrical, concolorous to paler than the cap surface, particularly near the base, solid. **Context** whitish, pink beneath cap surface, rather thick in the cap center. **Taste** mild. **Odor** faintly fruity. **Spore print** off-white in light deposit.

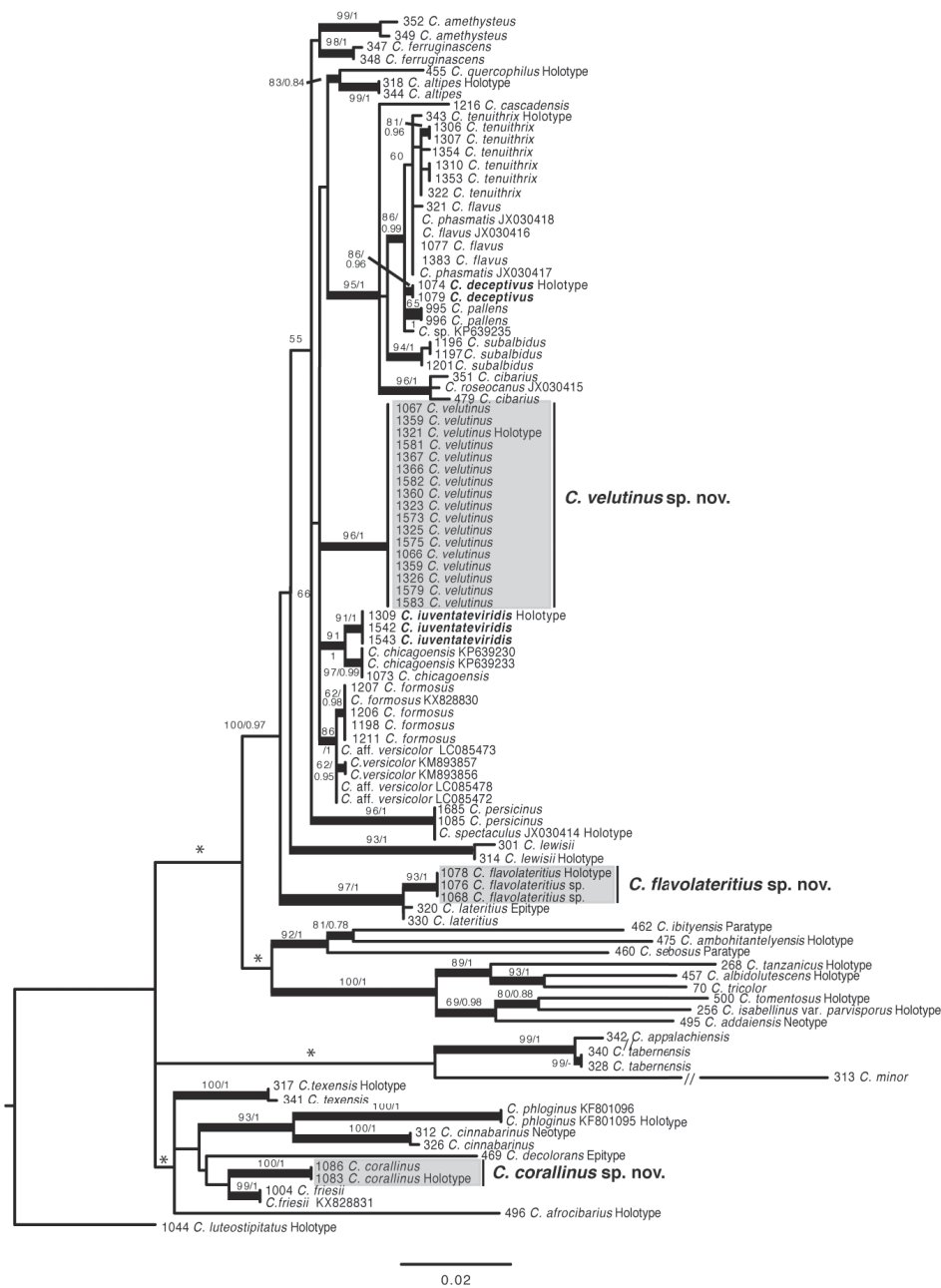
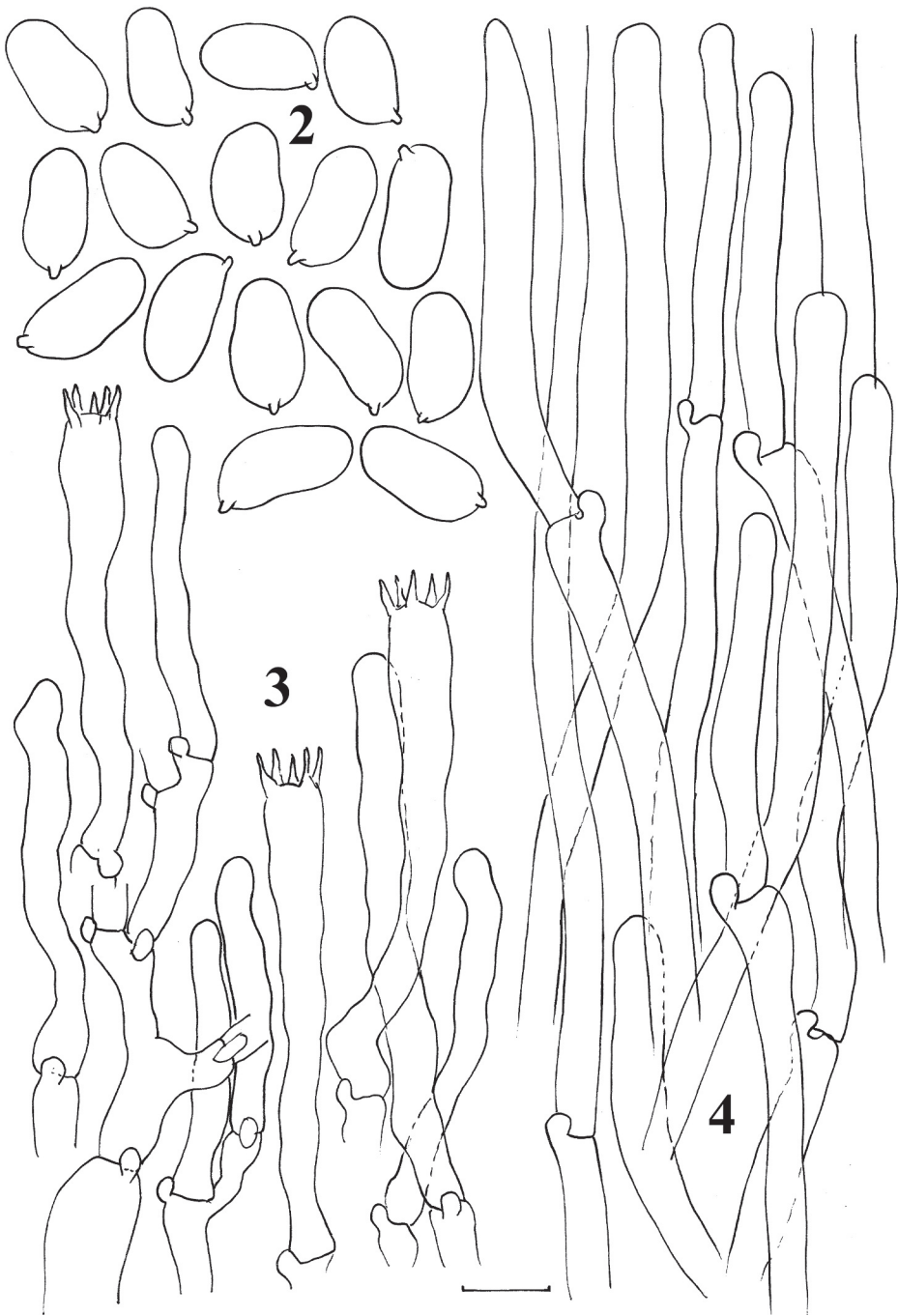


Fig. 1. Most likely tree from the maximum likelihood analysis of the *TEF*-1 region of selected taxa of *Cantharellus*. Branches that received significant support from both maximum likelihood bootstrap (MLbs  $\geq 70\%$ ) and Bayesian posterior probabilities (BPP  $\geq 0.95$ ) are in bold and are reported along the branches. Newly described species in this paper are highlighted. Asterisks (\*) indicate nodes that were implemented as backbone constraints in phylogenetic analyses.



Figs 2-4. *Cantharellus corallinus* (holotype). 2. Spores. 3. Basidia and basidiola. 4. Hyphal extremities of the pileipellis. (Scale 10  $\mu$ m, 5  $\mu$ m for spores). Drawings B. Buyck.

**Spores** ellipsoid to narrowly ellipsoid, rarely more elongate,  $(7.3)8.4\text{--}7.91\text{--}8.4(9.2) \times (3.7)3.9\text{--}4.27\text{--}4.6(5.0) \mu\text{m}$ ,  $Q = (1.5)1.7\text{--}1.86\text{--}2.0(2.2)$ , often inflated in the lower part to peanut-shaped, smooth, thin-walled, inamyloid. **Basidia** relatively short,  $(40)50\text{--}60(-75) \times 7\text{--}8 \mu\text{m}$ , (4)5-6-spored, narrowly clavate to subcylindrical, with multiguttulate contents; basidiola subcylindrical, undulate to repeatedly constricted, often basally somewhat inflated. **Subhymenium** well-developed and deep, strongly ramifying, not filamentous but composed of cells of rapidly increasing diam. toward trama, up to  $15 \mu\text{m}$  diam. **Cystidia** absent. **Pileipellis** composed of thin-walled, often more or less parallel, subcylindrical and relatively narrow hyphae measuring mostly  $4\text{--}8 \mu\text{m}$  diam., with sparse septa and few free endings; terminal cells difficult to find, subcylindrical, mostly exceeding  $50 \mu\text{m}$  in length, rounded-obtuse at the tips, regular in outline, neither tapering nor otherwise differentiated. **Clamp connections** abundant in all parts.

*Additional specimens examined:* UNITED STATES. **Missouri:** St Louis Co., Forest 44, Conservation area, GIS: 38.31.636 /- 090.31.021, 16 July 2011, 1086/Jay Justice MO-Canth-5 (PC 0713849)

*Commentary:* When comparing our figs 11-12 and 13-14, it is clear that his species is a new look-alike of *C. cinnabarinus* (for which it was initially collected). It belongs equally in *C.* subg. *Cinnabarinus* and possesses the typical characteristics of the subgenus: red-pink-orange fruiting bodies having predominantly thin-walled hyphal extremities on the cap surface and clamp connections in all tissues (Buyck *et al.* 2014). Our species also exhibits the deep, intensely ramifying subhymenium of rapidly widening cells typical for some other members of this clade.

Spores of the paratype are more elongate but have identical measurements for spore width  $[(8.3)8.6\text{--}8.92\text{--}9.3(9.6) \times (3.5)3.9\text{--}4.27\text{--}4.6(5.0) \mu\text{m}$ ,  $Q = (1.8)1.9\text{--}2.10\text{--}2.3(2.6)]$ . This second collection clearly demonstrates the variability of the general color as it is clearly bright orange. In form and size of its spores, the paratype is near-identical to *C. texensis*, while the spores of the holotype are closer to those of *C. cinnabarinus* (see Buyck & Hofstetter 2011).

The pileus surface in *C. cinnabarinus* is composed of a very loose superficial tissue that emits dispersed and more or less clavate terminal cells with cell walls of variable thickness, a feature it shares with *C. tabernensis* and *C. appalachiensis* (see Buyck *et al.* 2010). Because of the absence of such thick-walled extremities in the pileipellis, our species is microscopically more similar to *C. texensis*, although the thin-walled hyphae are wider in the latter species. Such subtle differences may disappear when more collections are examined.

*Cantharellus persicinus* (in subg. *Cantharellus*) may also present forms that are very similar in overall habit but it has a pileus surface covered by hyphal extremities in which the 3-5 last cells are distinctly thick-walled with a terminal cell that is rarely clavate (see Buyck *et al.* 2016c, this issue). Furthermore, spores of *C. persicinus* are not only longer but especially much more voluminous than those of any known species in subg. *Cinnabarinus*.

***Cantharellus flavolateritius* Buyck & V. Hofstetter *sp. nov.***

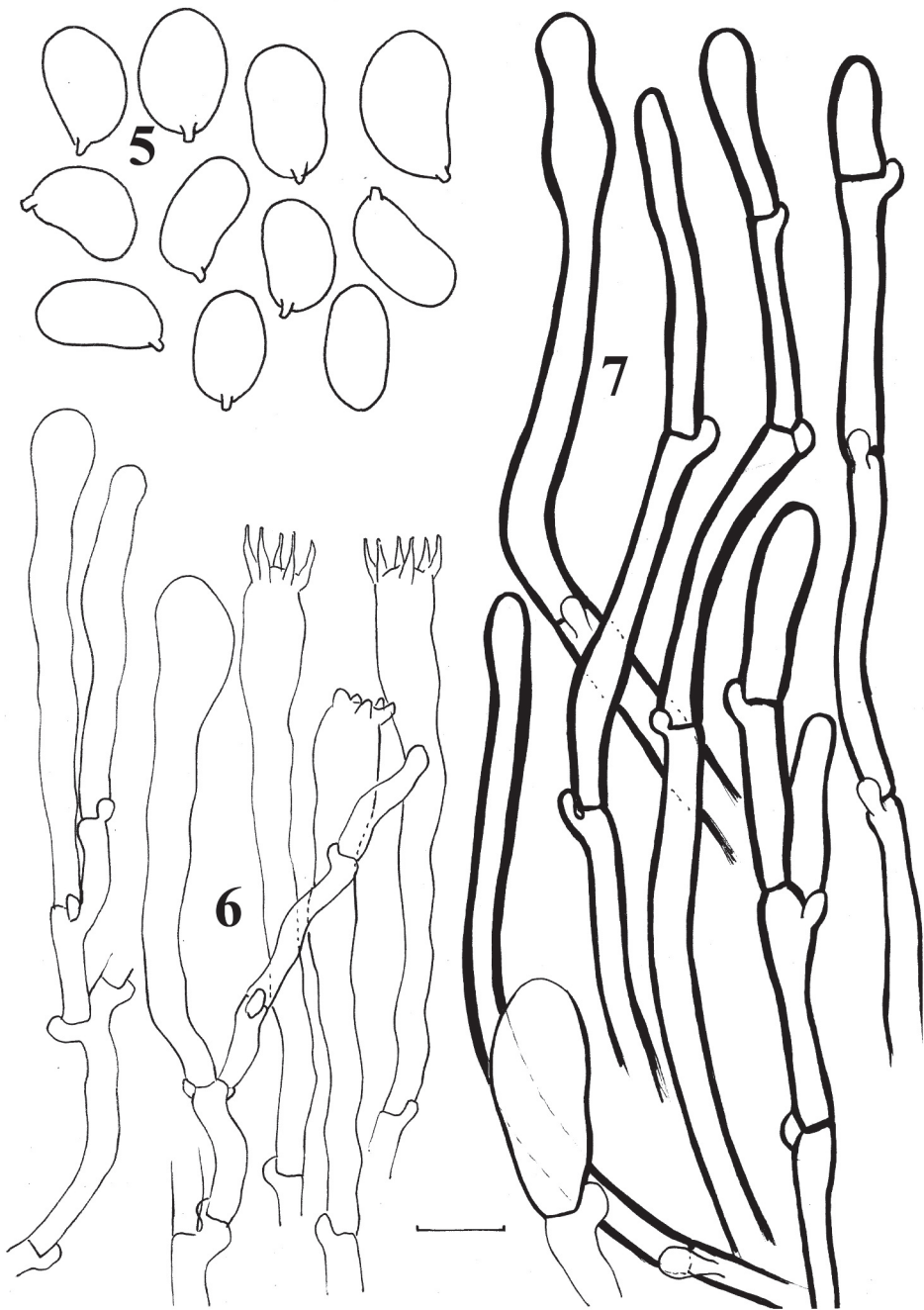
**Figs 5-7, 15-16**

*Mycobank:* MB 818377

*Diagnosis:* Differs slightly from *C. lateritius* in the apically more inflated but less irregularly undulating-wavy outline of the terminal cells of the pileipellis, but is principally supported by sequence data obtained for the *TEF-1* gene.

*Holotypus:* UNITED STATES. **North Carolina.** South of Highlands Biological Station, along Walking Stick Rd, GIS 035 00.497/- 083 09.678, growing on side of dirt road, 21 June 2011, 1078/Jay Justice NC Canth-4 (PC 0713852)





Figs 5-7. *Cantharellus flavolateritus* (holotype) 5. Spores. 6. Basidia and basidiola. 7. Hyphal extremities of the pileipellis. (Scale 10  $\mu$ m, 5  $\mu$ m for spores). Drawings B. Buyck.

**Fruiting bodies** gregarious, firm and stout. **Pileus** soon with a central depression, very firm and fleshy when young, rapidly becoming thinner toward cap margin and very irregularly folded-lobed, finally sometimes funnel-shaped, of a rather bright yellow, smooth or developing a scurfy- frosty appearance in the central part. **Hymenophore** strongly decurrent, composed of radially oriented, low anastomosing veins, locally (almost) smooth, concolorous or paler than the cap or stipe surface, developing pinkish hues with age. **Stipe** stout and firm, yellow to off-white, when yellow frequently pale to off-white at the base, bruising darker when injured, solid. **Context** whitish, firm, only reluctantly (?) yellowing when bruised. **Spore print** white (observed when dry).

**Spores** ellipsoid to narrowly ellipsoid,  $(7.1)7.2-7.88-8.5(10.0) \times (4.0)4.2-4.71-5.2(5.8) \mu\text{m}$ ,  $Q = (1.4)1.5-1.69-1.8(2.1)$ , smooth. **Basidia** long and slender, up to  $85 \mu\text{m}$  long and  $7-9 \mu\text{m}$  wide, mostly 5-spored; basidioles developing frequently a strongly inflated apical part, irregularly undulate-wavy in outline. **Subhymenium** filamentose. **Cystidia** none. **Pileipellis** an aerated, loose tissue with many easily recognizable, free endings of hyphae at the surface; hyphal extremities composed of  $4-7(10) \mu\text{m}$  wide, subcylindrical cells, septa rather distant and having conspicuous clamps, with thickened walls (ca.  $1 \mu\text{m}$ ); terminal cell often rather short, clavulate or apically slightly inflated, rarely ellipsoid, mostly  $20-50(70) \mu\text{m}$  long, sometimes more or less wavy-undulate in outline. **Clamp connections** abundant in all tissues.

*Additional material examined:* UNITED STATES. **North Carolina.** Nantahala national forest, standing Indian campground, 3 Aug. 2010, 1076/Jay Justice NC-CANT-2 (PC 0713851). **Arkansas.** Lake Sylvia Recreational Area, in the campground, 29 May 2010, 1068/Jay Justice AR-CANT-4 (PC 0713850)

*Commentary:* Morphologically speaking, *C. flavolateritius* is difficult to separate from *C. lateritius* as recently epitypified with a specimen that was collected in Texas (Buyck & Hofstetter 2011). The name *C. lateritius* (Berk.) Singer is based on *Craterellus lateritius* Berk., the holotype of which was collected more than a century ago in the south, viz. the state of Alabama (Berkeley 1873). More collections are clearly needed to figure out whether the geographical ranges of both taxa overlap or not. The Arkansas collection produced identical *TEF-1* sequence data, but has a much more orange cap; it was too young to generate reliable spore measurements. Spores of the second collection from North Carolina (Fig. 15) are very similar to those of the holotype collection [JJ NC Cant-2:  $(6.5)7.0-7.38-7.8(7.9) \times (4.2)4.4-4.67-5.0(5.2) \mu\text{m}$ ,  $Q = (1.3)1.4-1.59-1.7(1.9)$ ], but equally near-identical to spores of the *C. lateritius* epitype (Buyck & Hofstetter 2011).

***Cantharellus velutinus* Buyck & V. Hofstetter *sp. nov.***

**Figs 8-10, 21-32**

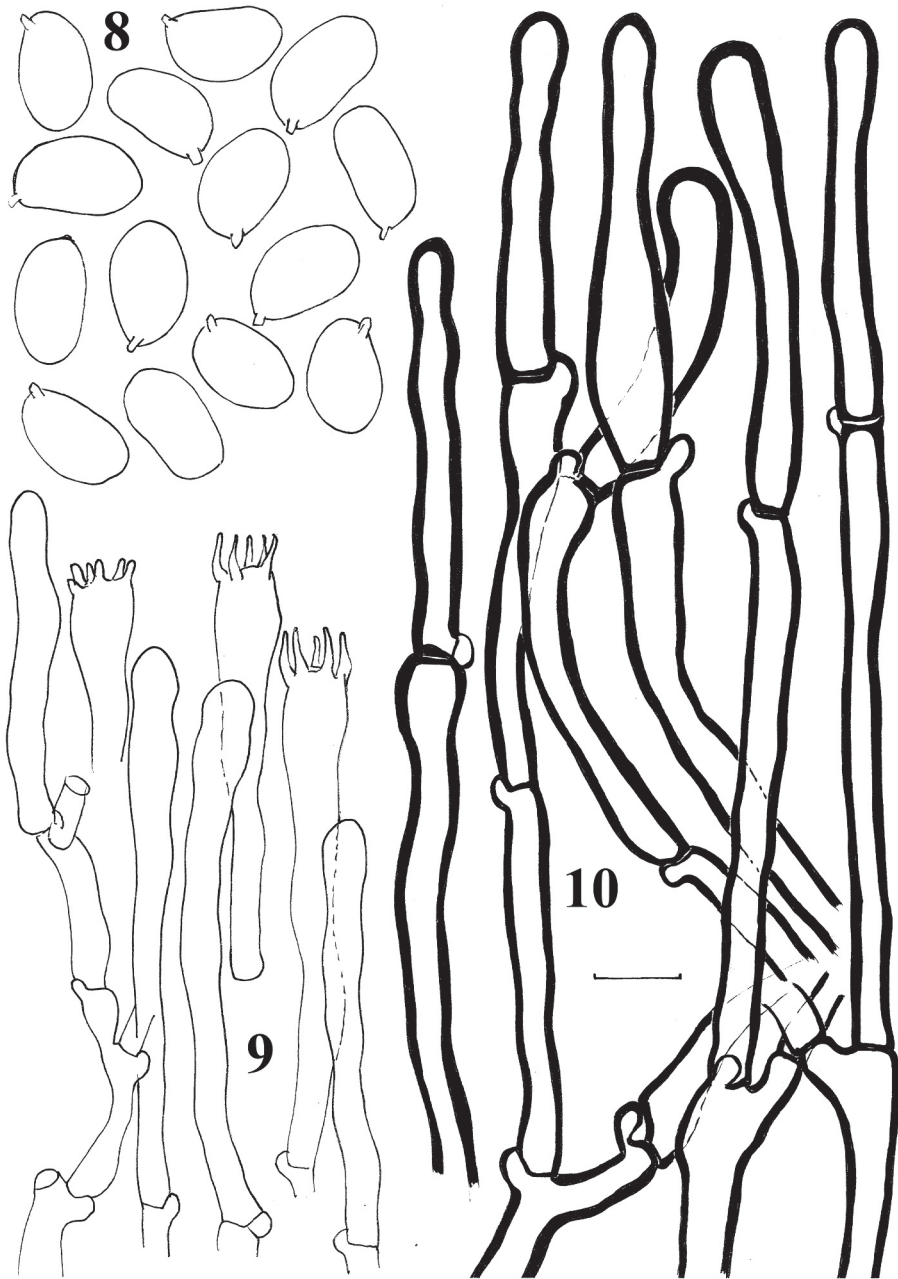
*Mycobank:* MB 818378

*Diagnosis:* *C. velutinus* is extremely similar to *C. lateritius*, although its hymenophore lacks pinkish tinges and the terminal cells in the pileipellis are slightly less undulate-irregular in outline compared to the latter species. It differs most notably from all known chanterelles by sequence data of the *TEF-1* gene.

*Holotype:* United States. **Texas.** Tyler Co., Big Thicket National Preserve, Beech Creek Unit, along path running from the beginning of the trail near parking lot inside the Unit, in Beech-Magnolia-Loblolly pine forest, elev. 55-70 m, N 30.72265-W 094.22493, 25 June 2014, leg. Buyck, Lewis & Hofstetter, in Buyck 1321/BB14.038 (PC 0142227)

*Etymology:* refers to the pubescent-hairy covering of the pileus





Figs 8-10. *Cantharellus velutinus* (holotype): typical, *lateritius*-like form. 8. Spores. 9. Basidia and basidiola. 10. Hyphal extremities of the pileipellis. (Scale 10  $\mu$ m, 5  $\mu$ m for spores). Drawings B. Buyck.



Fig. 11. *Cantharellus corallinus* (paratypus, 1086/Jay Justice MO-Canth-5). Photo. J. Justice.



Fig. 12. *Cantharellus cinnabarinus* (1069/JJ AR cant-5). Photo. J. Justice.



Fig. 13. *Cantharellus corallinus* (1083 /JJ MO cant-2, holotypus). Photo J. Justice.



Fig. 14. *Cantharellus cinnabarinus* (JJ NC-Cant-1). Photo. J. Justice.





Fig. 15. *Cantharellus flavolateritius* (paratype North Carolina). Photo Jay Justice



Fig. 16. *Cantharellus flavolateritius* (holotype, North Carolina). Photo Jay Justice

Figs 17-20. *Cantharellus lateritius* (on the left), for comparison with figs 21-24. *C. velutinus*, typical orange-colored form (on the right). All collections identified by sequencing and gathered in Big Thicket Natural Preserve, TX. Photos B. Buyck







Figs 25-26. *Cantharellus velutinus*. Pink form, specimens (identification not confirmed by sequencing) with similar hymenophore as the specimens of the typical form, i.e. pale and composed of low veins (collected in West Virginia, photos W. Roody).





Figs 27-28. *Cantharellus velutinus*. Pink form. Specimens with hymenophore composed of comparatively well-developed, distinctly pink gill folds (identified by sequencing, both collected in West Virginia; near Carnifax Ferry 28 July 2004 (DEWV5575, top) and Seneca 24 July 2006 (DEWV8498, bottom), photos W. Roody.



Figs 29. *Cantharellus velutinus*. cibarioid form, note the poorly developed hymenophore, Arkansas (photo J. Justice).



Figs 30. *Cantharellus velutinus*. cibarioid form, note the salmon pinkish color variant on the right, Arkansas (photos J. Justice).





Figs 31. *Cantharellus velutinus*, typical form (BB14.198). Detail of the pubescent-hairy cap surface, here particularly accentuated by the poorly hydrated condition of the fruiting body (photo B. Buyck).



Figs 32. *Cantharellus velutinus*, pink form. Detail of the pubescent-hairy cap surface; note also the formation of secondary caps on the adult pileus surface, a phenomenon quite frequently observed in this species (photo W. Roody).



Fig. 33. *Cantharellus velutinus*, typical yellow form (BB14.045; Big Thicket National Preserve, TX; photos B. Buyck).



Fig. 34. *Cantharellus velutinus*, typical yellow form (BB14.198; Shroomdom, near Poplarville, MS; photos B. Buyck).

**Cap** medium-sized, up to 60(80) mm in diam., with cap margin thin-fleshed and long-time inrolled, becoming rapidly depressed to even funnel-shaped with age, usually an egg-yolk orange to light or bright orange (4-5A6-7), but also a vivid clear yellow (2-4A6-8), mat, smooth, greasy-humid when wet but under hand lens distinctly pubescent-hairy all over, becoming almost squamulose and covered by a dense whitish tomentum when dry; cap margin thin, staying inrolled or downward for a long time, mostly irregularly and sometimes strongly lobed-folded, finally uplifted with age. **Stipe** often relatively long and slender, up to 80 mm long and 10(15) mm wide, cylindrical or slightly narrowing downward, often curved in its lower half, smooth to faintly fibrillose-pubescent similar to the cap surface, in drier conditions sometimes distinctly and more or less horizontally cracking into pale velutinous patches, off-white to very pale yellowish, turning slowly to a dirty yellowish brown upon handling, solid. **Hymenophore** strongly decurrent, not abruptly delimited from the sterile stipe surface, composed of low forking veins, sometimes strongly veined-anastomosed in between, off-white to pale cream over pale yellowish with age, without any pinkish hues. **Context** firm, fleshy, whitish, yellow to orange yellow under the cap surface, not energetically staining on handling but very slowly turning yellowish when handled as does the whole mushroom. **Smell** agreeable, of apricot. **Taste** mild. **Spore print** white when fresh, distinctly turning pale yellow when dry.

**Spores** ellipsoid to narrowly ellipsoid, (6.7)7.3-7.84-8.4(9.2)  $\times$  (3.7)4.2-4.61-5.0(5.2)  $\mu\text{m}$ ,  $Q=(1.4)1.5-1.71-1.9(2.2)$ , often somewhat lariform or peanut-shaped, thin-walled, smooth. **Basidia** moderately long, mostly 55–75(85)  $\times$  8–10  $\mu\text{m}$ , (4)5-6-spored. **Subhymenium** distinctly filamentous. **Cystidia** none. **Hyphal extremities of the pileipellis** with conspicuously thickened cell-walls [ca 1(1.5)  $\mu\text{m}$ ], composed of rather long, subcylindrical cells, mostly 5-12  $\mu\text{m}$  diam.; the terminal cell clavulate to subcylindrical, often somewhat undulating, ca 40-60  $\mu\text{m}$  long, obtuse-rounded. **Clamp connections** abundant in all tissues.

#### Additional examined and sequenced material:

##### Typical (lateritius-like) form:

UNITED STATES. **Texas.** Tyler Co., Big Thicket National Preserve, Beech Creek Unit, along path running from the beginning of the trail near parking lot inside the Unit, in Beech-Magnolia-Loblolly pine forest, elev. 55-70 m, N 30.72232/W 094.22395, 25 June 2014, leg. Buyck, Lewis & Hofstetter, BB 14.033 (PC 0142222); *ibid.*, N 30.72265/W 094.22493, BB 14.038 (PC 0142227); *ibid.*, N 30.72265/W 094.22493, BB 14.042 (PC 0142231); *ibid.*, N 30.72256/W 094.22269, BB 14.044 (PC 0142233); *ibid.*, N 30.72232/W 094.22395, BB 14.045 (PC 0142234); Hardin Co., Big Thicket National Preserve, Jack Gore Baygall Unit, in Beech-Magnolia-Loblolly pine forest, elev. 15-30 m, N 30.48126/W 094.15858, 27 June 2014, leg. Buyck, Lewis & Hofstetter in BB 14.078 (PC 0142267), BB 14.095 (PC 0142284); Big Thicket National Preserve, Lance Rosier Unit, along Cotton Rd., in Beech-Magnolia-Loblolly pine forest, elev. 24-32 m, 1 July 2014, N 30.27125/W 094.50455, leg. Buyck, Lewis & Hofstetter in BB 14.104 (PC 0142293); *ibid.*, N 30.27240/W 094.50562, BB 14.105 (PC 0142294), BB 14.106 (PC 0142295), BB 14.111 (PC 0142300), BB 14.115 (PC 0142304); *ibid.*, N 30.26927/W 094.50048, BB 14.110 (PC 0142299), BB 14.112 (PC 0142301). **Mississippi.** Pearl River Co., near Poplarville, Shroomdom property, in bottomland hardwoods, N 30.85495/W 089.63277, elev. 50-70 m, BB 14.198 (PC 0142387).



**Cibarioid form:**

**Arkansas.** Lake Sylvia area, under oak trees in camping ground area, GIS N34,51.906 - W92,49.089, 29 May 2010, 1066/Jay Justice AR-CANT-1 (PC 0142426); *ibid.*, GIS N34,51.911 - W92,49.102, 1067/Jay Justice AR-CANT-2 (PC 0142427)

**Pink form:**

**West-Virginia:** Barbour Co., Teter Creek Lake Wildlife Management Area, on soil under mixed broadleaf forest with *Quercus* spp., *Carya* sp., *Pinus* sp., GIS 597256-4329414, 5 August 2007, William C. Roody 1581/WRWV07-74 (DEWV8944); Clay Co., near Flat Fork Church, on ground under *Fagus grandifolia* and *Quercus* sp. woods, GIS 505904-4262783, 03 July 2013, 1190 ft alt, Donna M. Mitchell 1583/DMWV13-36 (DEWV10727); Fayette Co., Plum Orchard Lake Wildlife Management Area, on ground under *Tsuga canadensis*, *Quercus alba*, *Q. rubra*, *Carya* sp., *Fagus grandifolia*, 19 August 2004, William C. Roody 1573/WRWV04-695 (DEWV5391); Greenbrier Co., Greenbrier State Forest, on ground under *Pinus strobus* and *Quercus* spp., GIS 556392 4176371, 27 July 2009, 1990 ft. alt., William C. Roody 1582/WRWV09-64 (DEWV9938); Nicholas Co., Carnifax Ferry State Park, on ground under *Tsuga canadensis*, *Quercus* sp., *Carya* sp., *Pinus* sp., GIS 505433 4228474, 1640 ft. alt., 27 July 2004, William C. Roody 1575/WRWV04-284 (DEWV5575); Pendleton Co., George Sponaugle's land, on soil under *Quercus* sp., few *Pinus* sp. among ferns, 14 August 2003, excursion Mycological Assoc. of Washington, William C. Roody 1574/MAWV03-818 (DEWV5405); Pocahontas Co., Seneca State Forest, on soil under *Pinus strobus*, *Quercus alba*, 24 July 2006, William C. Roody 1580/WRWV06-587A (DEWV8498); Tucker Co., Clover Run, Monongahela National Forest, on soil under *Fagus grandifolia*, *Quercus* sp., *Tsuga canadensis*, 28 August 2005, William C. Roody 1579/WRWV05-1326 (DEWV7759); Wyoming Co., Twin Falls State Park, Moonshiners Trail, on soil under *Tsuga canadensis*, *Quercus prinus*, *Q. sp.*, *Carya* sp., 29 July 2004, William C. Roody 1576/WRWV04-377 (DEWV6031)

*Commentary:* The description above is entirely based on the typical, i.e. *C. lateritius*-like form of this species which we found to be locally quite common in Beech-Magnolia-Loblolly pine forests of the Big Thicket National Preserve in Texas. It can easily be mistaken for specimens of *C. lateritius*, the arche-type of a smooth chanterelle (Buyck 2014) as both species can be surprisingly similar in overall color, texture and habit (compare left and right columns of figs 17-24). *Cantharellus lateritius*, however, usually has a more pinkish and, at least in certain parts, smoother hymenophore and is usually also growing in large troops, composed of many, sometimes several dozens of specimens, whereas *C. velutinus* typically forms small groups of a few to less than a dozen specimens. Use of a hand lens reveals that both species have a distinct pubescent-fibrillose cap surface, but in both it can appear smooth to almost greasy at the touch, most likely depending on the weather conditions.

There exist two distinct color forms of this "typical" *C. velutinus* as some collections were "bright yellow" whereas others were "bright orange". Fruitings producing bright yellow caps are less reminiscent of *C. lateritius*, but can be confused with several other yellow chanterelles near the "*cibarius*" complex, such as *C. tenuithrix* or *C. flavus*. However, the latter species have a much better developed,



bright yellow to orange-yellow hymenophore that is more or less concolorous to the cap surface, whereas this typical form of our new species has a poorly veined and much paler hymenophore compared to the cap surface, even at maturity. Confusion with young fruiting bodies of *C. phasmatis*, which also has a pale hymenophore, should be more likely.

Under the microscope, the spores of *C. lateritius* and *C. velutinus* are very similar and also other microscopic features show only very minor differences. One can nevertheless observe that, in general, *C. lateritius* has a pileipellis composed of more slender extremities with less thick-walled cells that are more irregular and undulating in outline, and it also has on average longer basidia (see Buyck *et al.* 2011).

Although the first author has been collecting in the Gulf States for nearly 15 years now, this species has rarely been found and one interesting observation is that *C. lateritius* and *C. velutinus* were never found in the same locality, as if they were mutually exclusive. As far as *C. velutinus* is concerned, the presence of *Fagus grandifolia* seems too frequent to be a simple coincidence, but more observations are needed to confirm this relationship. In the Appalachians, it has been recorded mainly in association with oaks and hemlock (Roody 2003).

Quite interestingly, however, there also exists a form that has identical *TEF-1* sequences as the typical form, but which apparently lacks yellow pigments and takes a predominantly pink coloration. This pink form is often also more robust and can have a short stipe (see figs 25-26). This pink form is generally considered by most American mycologists to correspond to *C. persicinus* and was also frequently referred to as such in molecular studies on North American chanterelles (Dunham *et al.* 2003; Montcalvo *et al.* 2006; Foltz *et al.* 2013; Kuo 2015, Leacock *et al.* 2016), but *C. persicinus* is quite unrelated to *C. velutinus* (see Buyck *et al.* 2016c this issue). This pink form is so far only known from the central and southern Appalachian Mountains in eastern North America and possesses identical microscopic features. It shares the pale-coloured, veined or weakly developed gill folds and the moderate yellowing of the surface and context upon handling or bruising.

Our figs 27-28 illustrate also specimens in which the pink color extends to the hymenophore and where the hymenophore is considerably better developed (both sequenced). Both the form with pale and the one with pink hymenophore are interpreted as being conspecific by most North American mycologists and to represent *C. persicinus* (see e.g. Kuo, 2015). Kuo (l.c. ) and also Roody (2003, p. 128) picture "*C. persicinus*" as a strongly staining species with the stipe base turning rapidly and vigorously deep ferruginous, quite different from the typical form of *C. velutinus* where the stipe, as well as the rest of the fruiting body, stains only reluctantly when fresh. In the herbarium, however, dried fruiting bodies of both forms can have partly to completely reddish brown stipe surfaces and also the general color difference between collections of the pink form and those of the typical form disappear and both forms become undistinguishable.

Spores of pink *C. velutinus* are similar in size and form compared to the typical form, i.e. ellipsoid to narrowly ellipsoid and not particularly large, e.g.  $(6.3)7.0-7.43-7.9(8.4) \times (3.8)4.0-4.25-4.6(5.0) \mu\text{m}$ ,  $Q = (1.4)1.6-1.75-1.9(2.1)$  for DEWV 5575 (Fig. 27).

The clade corresponding to *C. velutinus* comprises also two genetically identical specimens from Arkansas, again with a very different general habit that can be considered as a "third form" that is more reminiscent of the *C. cibarius* complex and comprises both yellow and salmon-pinkish fruiting bodies (Figs 29-30).

Although our *tef-1* analysis does not allow to place *C. velutinus* with more precision within subg. *Cantharellus*, a combined analysis of partial ITS-LSU sequences places it as sister to *C. californicus* (see Buyck *et al.* 2016b this issue).

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## REFERENCES

- ARIYAWANSA H.A., HYDE K.D., JAYASIRI S.C., BUYCK B., CHETHANA K.W.T., DAI D.Q., DAI Y.C., DARANAGAMA D.A., JAYAWARDENA R.S., LUCKING R., GHOBAD-NEJHAD M., NISKANEN T., THAMBUGALA K.M., VOIGT K., ZHAO R.L., LI G.J., DOILOM M., BOONMEE S., YANG Z.L., CAI Q., CUI Y.Y., BAHKALI A.H., CHEN J., CUI B.K., CHEN J.J., DAYARATHNE M.C., DISSANAYAKE A.J., EKANAYAKA A.H., HASHIMOTO A., HONGSANAN S., JONES E.B.G., LARSSON E., LI W.J., LI Q.R., LIU J.K., LUO Z.L., MAHARACHCHIKUMBURA S.S.N., MAPOOK A., MCKENZIE E.H.C., NORPHANPHOUN C., KONTA S., PANG K.L., PERERA R.H., PHOOKAMSAK R., PHUKHAMSAKDA C., PINRUAN U., RANDRIANJOHANY E., SINGTRIPOP C., TANAKA K., TIAN C.M., TIBPROMMA S., ABDEL-WAHAB M.A., WANASINGHE D.N., WIJAYAWARDENE N., ZHANG J.F., ZHANG H., ABDEL-AZIZ F.A., WEDIN M., WESTBERG M., AMMIRATI J.F., BULGAKOV T.S., LIMA D.X., CALLAGHAN T.M., CALLAC P., CHANG C.H., COCA L.F., DAL-FORNO M., DOLLHOFFER V., FLIEGEROVA K., GREINER K., GRIFFITH G.W., HO H.M., HOFSTETTER V., JEEWON R., KANG J.C., WEN T.C., KIRK P.M., KYTÖVUORI I., LAWREY J.D., XING J., LI H., LIU Z.Y., LIU X.Z., LIIMATAINEN K., LUMBSCH H.T., MATSUMURA M., MONCADA B., NUANKAEW S., PARNMEN S., DE AZEVEDO SANTIAGO A.L.C.M., SOMMAI S., SONG Y., DE SOUZA C.A.F., DE SOUZA-MOTTA C.M., SU H.Y., SUETRONG S., WANG Y., WEI S.F., WEN T.C., YUAN H.S., ZHOU L.W., RĚBLOVÁ M., FOURNIER J., CAMPORESI E., LUANGSA-ARD J.J., TASANATHAI K., KHONSANIT A., THANAKITPIPATTANA D., SOMRITHIPOL S., DIEDERICH P., MILLANES A.M., COMMON R.S., STADLER M., YAN J.Y., LI X.H., LEE H.W., NGUYEN T.T.T., LEE H.B., BATTISTIN E., MARSICO O., VIZZINI A., VILA J., ERCOLE E., EBERHARDT U., SIMONINI G., WEN H.A. & CHEN X.H., 2015 — Fungal Diversity Notes 111-252: taxonomic and phylogenetic contributions to fungal taxa. *Fungal Diversity* 75: 1-197.
- BUYCK B., 2014 — Exploring the diversity of “smooth chanterelles” (*Cantharellus*, Cantharellales). *Cryptogamie, Mycologie* 35(1), 23-40.
- BUYCK B., LEWIS D.P., EYSSARTIER G. & HOFSTETTER V., 2010 — *Cantharellus quercophilus* sp.nov. and its comparison to other small, yellow or brown American chanterelles. *Cryptogamie, Mycologie* 31(1): 17-33.
- BUYCK B., CRUAUD C., COULOUX A. & HOFSTETTER V., 2011 — *Cantharellus texensis* sp. nov. from Texas, a Southern lookalike of *C. cinnabarinus* revealed by *tef-1* sequence data. *Mycologia* 103: 1037-1046.
- BUYCK B. & HOFSTETTER V., 2011 — The contribution of *tef-1* sequences for species delimitation in the *Cantharellus cibarius* complex in the southeastern USA. *Fungal Diversity* 49: 35-46.
- BUYCK B., KAUFF F., EYSSARTIER G., COULOUX A. & HOFSTETTER V., 2014 — A multilocus phylogeny for worldwide *Cantharellus* (Cantharellales, Agaricomycetidae). *Fungal Diversity* 64(1): 101-121.
- BUYCK B., MOREAU P.-A., COURTECUISSÉ R., KONG LUZ A., ROY M. & HOFSTETTER V., 2016a — *Cantharellus coccolobae* sp. nov. and *Cantharellus garnieri*, two tropical members of *Cantharellus* subg. *Cinnabarinus*. *Cryptogamie, Mycologie* 37(3): 391-403.

- BUYCK B., OLARIAGA I. & HOFSTETTER V., 2016b — Setting the record straight on North American *Cantharellus*. *Cryptogamie, Mycologie* 37(3): 405-417.
- BUYCK B., OLARIAGA I., LOONEY, B., JUSTICE J. & HOFSTETTER V., 2016c — Wisconsin chanterelles revisited and first indications for very wide distributions of *Cantharellus* species in the United States East of the Rocky Mountains. *Cryptogamie, Mycologie* 37(3): 345-366.
- DUNHAM S.M., O'DELL T.E. & MOLINA R., 2003 — Analysis of nrDNA sequences and microsatellite allele frequencies reveal a cryptic chanterelle species *Cantharellus cascadenis* sp. nov. from the American Pacific Northwest. *Mycological Research* 107: 1163-1177.
- FOLTZ M.J., PEREZ K.E. & VOLK T.J., 2013 — Molecular phylogeny and morphology reveal three new species of *Cantharellus* within 20 m of one another in western Wisconsin, USA. *Mycologia* 105(2): 447-461.
- MONCALVO J.M., NILSSON R.H., KOSTER B., DUNHAM S.M., BERNARUER T., MATHENY P.B., MCLENON T., MARGARITescu, WEIß M., GARNICA S., DANELL E., LANGER G., LANGER E., LARSSON E., LARSSON K.-H. & VILGALYS R., 2007 (for 2006) — The cantharelloid clade: dealing with incongruent gene trees and phylogenetic reconstruction methods. *Mycologia* 98(6): 937-948.
- KUO M., 2015 — *Cantharellus persicinus*. Retrieved from the *MushroomExpert.Com* Web site: [http://www.mushroomexpert.com/cantharellus\\_persicinus.html](http://www.mushroomexpert.com/cantharellus_persicinus.html)
- LEACOCK P.R., RIDDELL J., WILSON A.W., ZHANG C. & MUELLER G.M., 2016 — *Cantharellus chicaoensis* sp. nov. is supported by molecular and morphological analysis as a new yellow chanterelle in Midwestern United States. *Mycologia* 108(4): 765-772.
- OLARIAGA I., BUYCK B., ESTEVE-RAVENTOS F., HOFSTETTER V., MANJON J.L., MORENO G. & SALCEDO I., 2015. — Assessing the taxonomic identity of white and orange specimens of *Cantharellus*: occasional colour variants or independent species? *Cryptogamie, Mycologie* 36(3): 287-300.
- OLARIAGA I., MORENO G., MANJÓN J.L., SALCEDO I., HOFSTETTER V., RODRÍGUEZ D. & BUYCK B. 2016 — *Cantharellus* (Cantharellales, Basidiomycota) revisited in Europe through a multigene phylogeny. *Fungal Diversity*. in press.
- PETERSEN R.H., 1985 — Notes on Clavarioid Fungi. XIX. Colored illustrations of selected taxa, with comments on *Cantharellus*. *Nova Hedwigia*. 42: 151-160.
- ROODY W.C., 2003 — *Mushrooms of West Virginia and the Central Appalachians*. The University Press of Kentucky, Lexington, KY.