



## *Trichaptum* (Basidiomycota) in tropical America: a sequence study

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### Abstract

A comprehensive study on *Trichaptum* was performed on the basis of collections from Europe, USA and Central America. The phylogenetic analysis, including 33 newly generated nuclear DNA ITS sequences revealed four new lineages. Combined with morphological evidence, three new lineages are described and illustrated as new species: *Trichaptum agricola*, *T. confertum* and *T. resacarium*; one new lineage represented by a single collection is treated as unidentified taxon. *Nigrofomes melanoporus* ITS sequence is shown to merge into the *Trichaptum* clade.

**Key words** – Hymenochaetales – taxonomy – Costa Rica

### Introduction

The genus *Trichaptum* Murrill 1904 contains mostly pileate polypores with tomentose to hirsute upper surface, blackish-brown, gray, dirty white and more or less purplish on growing tubes, a dimitic/trimitic hyphal system, mostly apically encrusted cystidia, hyaline, thin-walled, mostly cylindrical basidiospores, and causing a white rot (Gilbertson & Ryvarden 1987). Because of tough tissue and trametoid appearance, many species were treated in *Trametes* Fr. 1836 (e.g. Pilát 1936) until Donk (1933) established genus *Hirschioporus* Donk for temperate zone species. Ryvarden & Johansen (1980) concluded that *Hirschioporus* spp. were not essentially different from tropical species with more fleshy and villose pilei, typified by *Trichaptum perrottetii* (Lév.) Ryvarden. Since then it is generally acknowledged that *Trichaptum* is a cosmopolitan, well delimited genus embracing small but very distinct group of polypores, which show many unique features. Except for purplish colors and characteristic cystidia there are also distinctive generative hyphae with small clamps and with imperforate dolipore septa (Traquiar & McKeen 1978), which indicates relationship to *Hymenochaetales* Oberw., not *Polyporales* Gäum. This was confirmed by molecular phylogeny that places species of *Trichaptum* in the so called Hymenochaetoid clade, close to *Hyphodontia/Schizopora* (Hibbet & Binder 2002).

Eight species of *Trichaptum* occur in the temperate zone: *T. abietinum* (Dicks.) Ryvarden, *T. fuscoviolaceum* (Ehrenb.) Ryvarden, *T. laricinum* (P. Karst.) Ryvarden, *T. subchartaceum* (Murrill) Ryvarden (in North America, Zhou et al. 2016), *Trichaptum polycystidiatum* (Pilát) Y.C. Dai, *Trichaptum perenne* Y.C. Dai & H.S. Yuan and *Trichaptum montanum* T. Hatt. in Asia (Dai et al. 2009, Dai 2012), and *T. bifforme* (Fr.) Ryvarden, the last one being reportedly cosmopolitan, growing also in tropical regions. They belong to the most common wood-decaying fungi of great

economic importance. All these species have rather similar spores and cystidia but macroscopic markers, especially juvenile pores size and shape can be used for reliable determination.

Subtropical and tropical regions of South America host more *Trichaptum* species which indicates possible determination problems, because species delimitation is also based mostly on macro-morphology (Ryvarden 2016). Determination supported by DNA-based phylogenetic analysis cannot be applied because few sequences are available in GenBank: two specimens of *T. byssogenum* (Jungh.) Ryvarden, both from East Asia, and two specimens of *T. sector* (Ehrenb.) Kreisel from Brazil (Apr 2017).

In the last few years, we have collected and sequenced many *Trichaptum* specimens in Texas, Florida, Caribbean Region and Costa Rica and encountered problems with species determination. Our aim is to use ITS region sequencing for improving taxonomy of *Trichaptum* occurring in tropical America.

## Materials & Methods

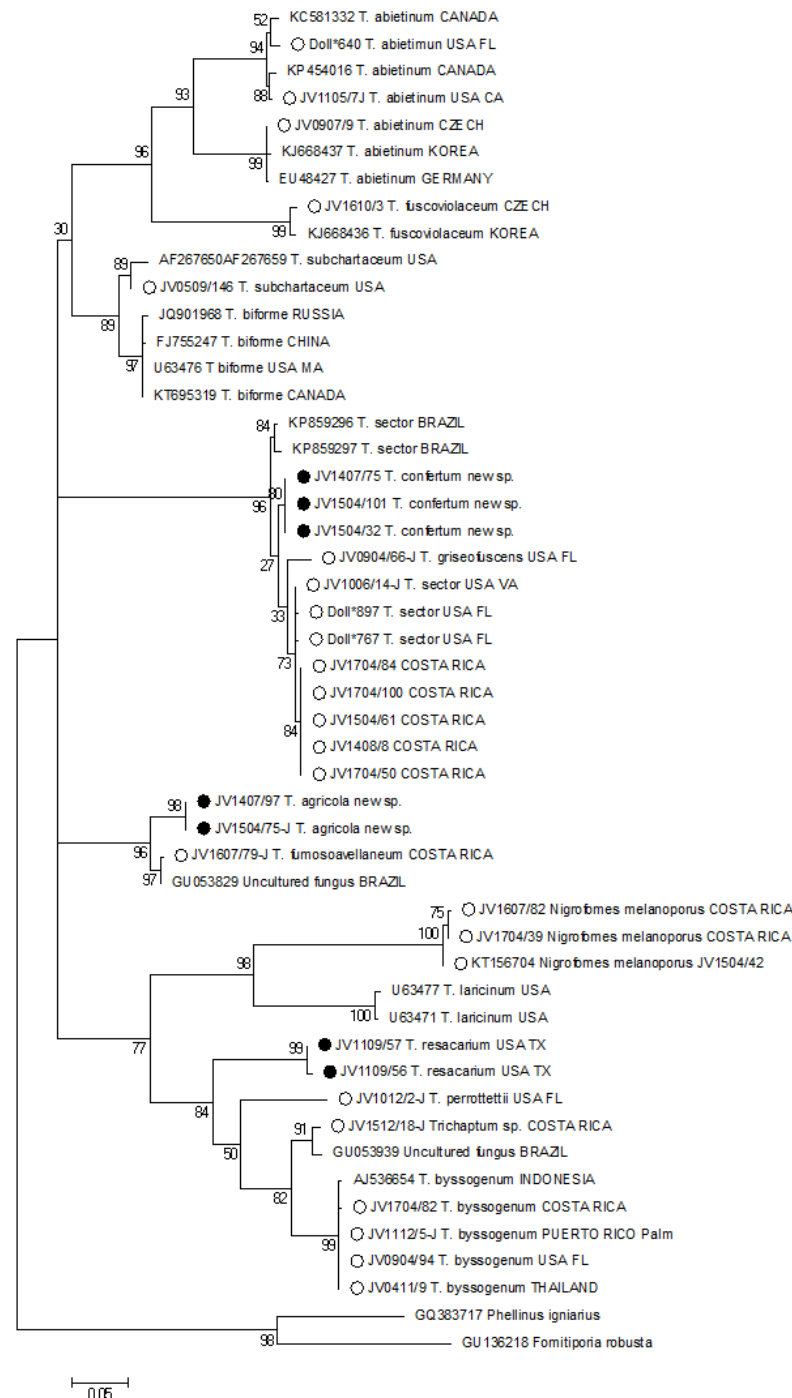
*Trichaptum* specimens were collected during 2005–2017 on various localities in the USA, US Virgin Islands, Puerto Rico, and Costa Rica; standard *T. abietinum* and *T. fuscoviolaceum* also in the Czech Republic, Europe. The specimens were dried and microscopically inspected in Melzer's reagent (IKI) and 5 % KOH. Collected specimens are deposited in the herbarium of the first author, type specimens also in National Museum of the Czech Republic (PRM).

DNA isolation and PCR procedure follow Vlasák & Kout (2011). The ITS region was amplified with primers ITS5 and ITS4 (White et al. 1990) and the sequences were aligned with Clustal W using very low values for gap penalty to accommodate long insertions in *Trichaptum abietinum* sequences: gap opening penalty was set at 4 and gap extension penalty was set at 0. Proper alignment of conserved regions was manually checked. Maximum likelihood analyses were conducted in MEGA6 using the Tamura 3-parameter model with gamma distribution with 5 invariant sites, which was determined as the best-fit model with "Model testing for estimation of distances" option (Tamura et al. 2013).

## Results

30 ITS rDNA region sequences of twelve *Trichaptum* species, six of which were sequenced for the first time, and also three sequences of *Nigrofomes melanoporus* (Mont.) Murrill were generated for this study. In the phylogenetic analysis, another 18 sequences were retrieved from GenBank. Clustal W program produced an alignment of 1335 characters, 350 of which were parsimony informative. In positions where *T. fuscoviolaceum* and *T. abietinum* specimens contain long inserts in ITS1 (Ko & Jung 2002), the alignment produced up to 210 bp long gaps in sequences of other species. Except for the rather structured *T. abietinum*/*T. fuscoviolaceum* clade (Ko & Jung 2002), which is not our object of interest, phylogenetic analysis resolved the taxonomical structure of *Trichaptum* in a more or less expected way (Fig. 1). Sequences of clearly-defined species such as *T. bifforme*, *T. byssogenum*, *T. fumosoavellaneum* (Romell) Rajchenb. & Bianchin., *T. fuscoviolaceum*, *T. laricinum*, *T. perrottettii* and *T. subchartaceum* produced strongly supported terminal clades. *T. byssogenum* sequences from East Asia (type locality) are identical with those from America. USA collections of *T. sector* show sequences slightly different from Costa Rica and Brazil collections. The differences are small, however, and more collections analyzed by multigenic phylogeny are needed to decide, if more species are involved. Somewhat surprisingly, specimens of *Nigrofomes melanoporus* created a sister clade to *T. laricinum*, and through a deeper node they are related also to *T. perrottettii*, a type species of *Trichaptum*. We do not intend to make taxonomical conclusions on generic level based on single-gene sequence. It should be noted, however that *Nigrofomes* has the same blackish-violet colors as most *Trichaptum* species, similar skeletal hyphae, and also a kind of cystidia, even if very rarely occurring. On the other hand, distinct crust on pileus surface and lack of clamps on generative hyphae separate *Nigrofomes* from *Trichaptum*.

Specimens of three *Trichaptum* species which we were not able identify with described species, produced sequences that also make strongly supported terminal clades and are described below as new species.



**Figure 1** – Phylogenetic relationships of 45 *Trichaptum* and 3 *Nigrofomes* specimens inferred with ITS rDNA. *Phellinus igniarius* and *Fomitiporia robusta* were used to root the tree. Topology from maximum likelihood (ML) analysis. Support values along branches from ML bootstrap (500 replicates). The tree is drawn to scale, with branch lengths measured in the number of substitutions per site. Circles indicate sequences generated in this study, black circles specimens of new species. GenBank numbers indicate sequences retrieved from GenBank; two sequences marked as “Uncultured fungus” come from filter-collected spores - see Discussion.

*Trichaptum agricola* Vlasák & Vlasák Jr., sp. nov.

Mycobank number: MB 821863

Etymology – named for its resemblance to *C. theobromae*.

Figs 2

Differs from other *Trichaptum* species by about 2 cm thick basidiocarps, lack of violet colors, large, isodiametric, entire pores 1–3 per mm and by ITS rDNA sequence. Thick-walled and apically encrusted cystidia abundantly present, basidiospores ellipsoid, thin-walled, IKI–,  $4\text{--}5 \times 2.5\text{--}3 \mu\text{m}$ .

Type – Costa Rica, Puntarenas Prov.: La Ensenada Lodge, hardwood prop, 22.IV.2015 Vlasák Jr. 1504/75-J (PRM 945505, JV, GenBank MF380995)

Basidiomes – Annual to biannual, sessile, broadly attached, up to  $10 \times 5 \times 2 \text{ cm}$ , rarely with tube layer somewhat effused on the substrate below the pilei. Upper surface tomentose, velvety on touch, later glabrous, with age becoming warted or with irregular outgrowths, azonate or sometimes slightly concentrically sulcate near the margin, light brown, later with a silvery shine, margin obtuse to sharp, concolorous. Pore surface brown, pores angular, 1–3 per mm, regular, or in parts somewhat radially elongated or labyrinthine, with moderately thin, entire dissepiment, context up



**Figure 2** – A. Microscopical features of *Trichaptum Agricola*. B. spec. 1407/97 in situ. C. holotype in herbarium.

to 10 mm thick, tough, brown, with several distinct growth zones about 2 mm thick, indistinctly delimited towards the pilei cover; tube layer up to 10 mm thick, indistinctly stratified.

Hyphal system – Trimitic, generative hyphae hyaline, thin-walled,  $2\text{--}3 \mu\text{m}$  in diam., with abundant, small clamps, skeletal hyphae brownish, thick-walled,  $2\text{--}4 \mu\text{m}$  in diam., binding hyphae very rare, hyaline,  $2\text{--}3 \mu\text{m}$ , with short branches. Basidia clavate, 4-sterigmate,  $12\text{--}15 \times 5\text{--}6 \mu\text{m}$ . Cystidia very abundant,  $10\text{--}20 \times 4\text{--}7 \mu\text{m}$ , ventricose to fusoid, slightly thick-walled and apically encrusted. Basidiospores ellipsoid, hyaline, smooth, thin-walled, IKI–,  $4\text{--}5 \times 2.5\text{--}3 \mu\text{m}$ .

Distribution – Collected in two dry farmland localities and in both cases the fungus was common on fence props. In one of these localities, it was found also in nearby woods on a dead stag.

Notes – *Trichaptum agricola* looks very much like *Trametes gibbosa* (Pers.) Fr., because of thick basidiocarps with uneven surface, but it is brown in all parts and has large, isodiametric pores. The violet colors, typical for *Trichaptum*, were not noted in our collections, which were however represented by rather old pilei.

***Trichaptum confertum*** Vlasák & Vlasák Jr., sp. nov.

Figs 3

Mycobank number: MB 821866

Etymology – *Confertum* (Lat.): indicating densely arranged pilei.

Distinguished by small, only 0.5–2 cm wide and 1 mm thick pilei, densely imbricate and rather dark, greyish-black with purplish to bluish tints when fresh. Pores and spores smaller than in similar species: pores 6–8 per mm, basidiospores  $4\text{--}4.5 \times 1.8\text{--}2.2 \mu\text{m}$ .

Type – Costa Rica. Puntarenas Prov.: Monteverde, Santa Elena, Cerro Amigos Road, hardwood, 29.VII.2014 Vlasák 1407/75 (JV, PRM 945506, GenBank MF380988, MF380989).

Basidiomes – Annual, pileate, dimidiate to flabelliform, or broadly attached but mostly effused-reflexed, pilei 5–20 mm long and wide and only 1 mm thick, densely imbricate. Upper surface dark gray to black with purplish to bluish tints, fading to brown after drying, adpressed velutinate to tomentose, zonate, margin sharp and wavy, concolorous. Pore surface dark purplish when fresh, greyish brown after drying, pores angular, 6–8 per mm, with thick, entire dissepiments,

somewhat irregular or slightly lacerate in old specimens, context about 0.7 mm thick, tubes concolorous, up to 0.5 mm deep; context duplex, lower part dense and of the same dark brown color as the tube layer, upper layer white and cottony, only about 0.2 mm thick.



**Figure 3** – A. Microscopical features of *Trichaptum confertum*. B. holotype in situ. C. in herbarium.

Hyphal system – Trimitic, generative hyphae hyaline, thin-walled, 2–3 µm in diam., with abundant, small clamps, skeletal hyphae brownish, thick-walled, mostly narrow, 2–3 µm in diam., with rounded ends, binding hyphae rare, but typically developed, 2–3 µm, with many short branches. Basidia clavate, 4-sterigmate, 10–13 × 4–5 µm. Cystidia 10–15 × 3–5 µm, ventricose, thin- to slightly thick-walled and apically encrusted. Basidiospores ellipsoid, hyaline, smooth, thin-walled, IKI–, 4–4.5 (5) × 1.8–2.2 µm, often collapsed to rugged ellipsoid structures.

Distribution – Collected in two localities and noted also in other places in montane cloud forests, always covering large areas on dead logs. Seems to be quite common.

Notes – The species is closely related to *T. sector* but differs by smaller pilei, pores and spores and by bluish or purplish colors when fresh. *T. sector* never develops purplish tinge and the pilei are up to 5 cm wide, with pores 3–5 per mm and spores subcylindrical, 6–7 µm long.

***Trichaptum resacarium*** Vlasák & Vlasák Jr., sp. nov.

Figs 4

Mycobank number: MB 821866

Etymology – Indicating growth around Rio Grande old channels called *resaca* (Spanish).

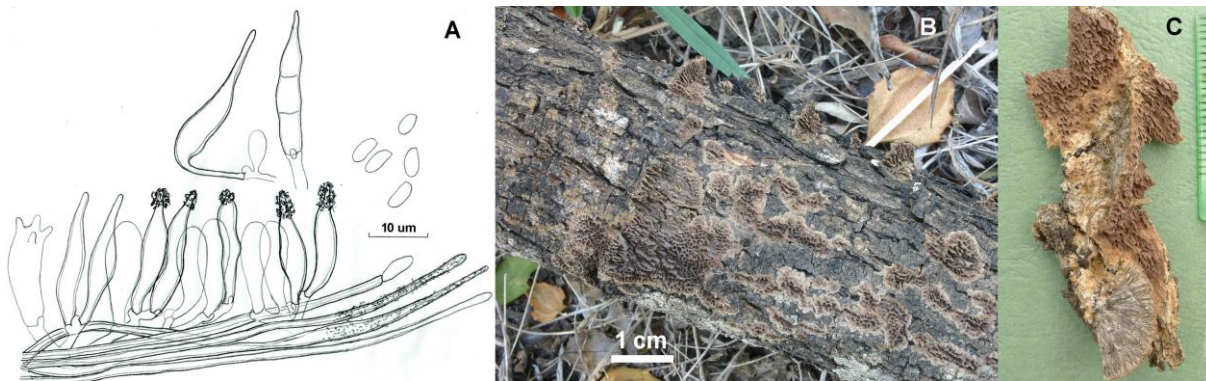
Similar to *T. byssogenum* but distinguished by poorly developed pilei, dark hymenial surface, blackish hairs on pileal surface making a loose net, small spores 3.5–4 × 2 µm, neckige cystidia and heavily incrustated hyphal ends. ITS RNA sequence is also different.

Type – USA. Texas: Cameron County, Pharr, hardwood, 27.IX.2011 Vlasák 1109/57 (JV, PRM 945510, GenBank MF381017, MF381018)

Basidiomes – Annual, resupinate, rarely with small reflexed pilei 0.5–2 × 0.5–1 cm, upper surface greyish tomentose to hispid and radially striate with coarse, blackish hairs making a prominent, loose net, margin sharp, concolorous. Pore surface dark fuliginous with purplish tint, pores angular, (0.5)1–2 per mm, with rather thick dissepiments, irregular or somewhat irpicoid in old specimens, sometimes developed as flat teeth 1–2 × 0.4 × 4 mm, tubes concolorous, up to 4 mm deep; context about 1 mm thick, duplex, lower part dense and of the same dark brown color as the tube layer, upper layer cottony, with blackish hairs.

Hyphal system – Dimitic, generative hyphae hyaline, thin-walled, 2–3 µm in diam., with abundant, small clamps, skeletal hyphae brownish, thick-walled, 2–3 µm in diam., with rounded ends, which in hymenium or on pore-edges often covered with a layer of crystals reminding of *Skeletocutis* incrustation. Basidia clavate, 4-sterigmate, 12–15 × 5–6 µm. Cystidia very abundant, fusoid, 15–30 × 3–5 µm, thin- to slightly thick-walled, mostly with long narrow necks up to 5–10 × 1.5 µm apically encrusted or smooth, light brown. , the hymenial layer very dark under microscope because of abundance of brownish cystidia. Basidiospores sparse, ellipsoid, hyaline, smooth, thin-walled, IKI–, 3.5–4 × 2 µm.

Distribution – Collected twice but noted several times around Rio Grande dead channels on hardwood twigs lying on the ground.



**Figure 4** – A. Microscopical features of *Trichaptum resacarium*. B. holotype in situ. C. spec. 1109/56 herbarium.

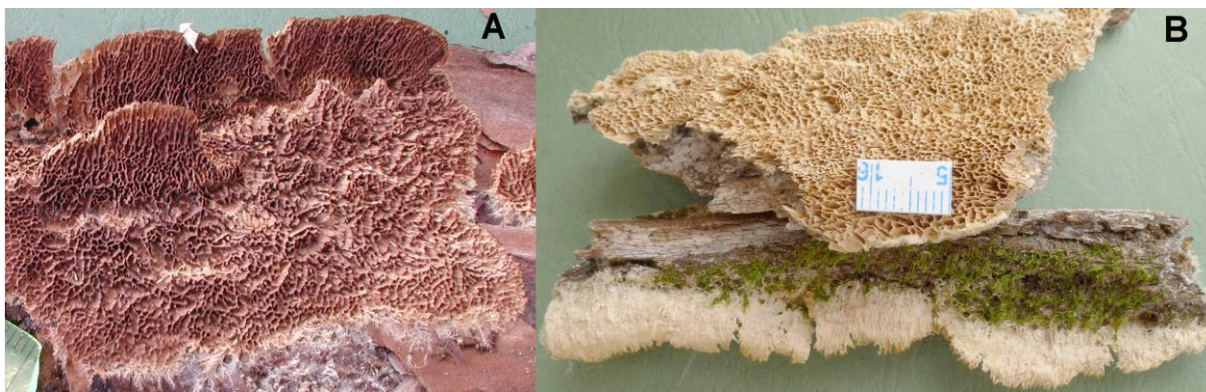
Notes – *Trichaptum cladotrichum* (Berk. & M.A. Curtis 1868) Murrill 1904 collected in Cuba, which was later synonymized with *T. byssogenum* (Ryvarden 1984), differs in having pilei 5–7 cm broad, with “spongy strigose coating without prominent fibers”. Also, dark hymenial surface and small spores are not mentioned in the original or later descriptions.

Notes on some other species from tropical America:

***Trichaptum biforme*** (Fr.) Ryvarden

*T. biforme* is reported as the only cosmopolitan *Trichaptum* species (Ryvarden 2016). It is very common in the eastern USA, Europe and Asia and it is similar to other *Trichaptum* species in that region, showing mostly flabelliform, thin pilei with greyish, zonate surface and violaceous, angular, later irpicoid pores. It should be noted that we were not able to find it in Central America though we have looked for it. There are about 20 sequences of *T. biforme* in GenBank but none of them comes from tropical regions.

***Trichaptum byssogenum*** (Jungh.) Ryvarden



**Figure 5** – A. *Trichaptum byssogenum* spec. 0904/95. B. *Trichaptum* sp. spec. 1512/18-J.

Effused-reflexed basidiomes show characteristic, coarsely-strigose upper surface, greyish tan to whitish, and large angular pores 1–2 per mm, purplish when fresh, dull purplish brown on drying, which become lacerate and partly lamellate towards the margin (Fig. 5A). The basidiospores are cylindrical,  $5.5\text{--}8 \times 2\text{--}2.5 \mu\text{m}$  (Ryvarden 2016), in our measurements somewhat broader, subcylindrical,  $5.5\text{--}7 \times 2.2\text{--}3 \mu\text{m}$ . *T. byssogenum* was described from Java but it is a pantropical species. It is quite common on pines in Florida, but rather rare on hardwoods in Costa Rica and the Caribbean Region.

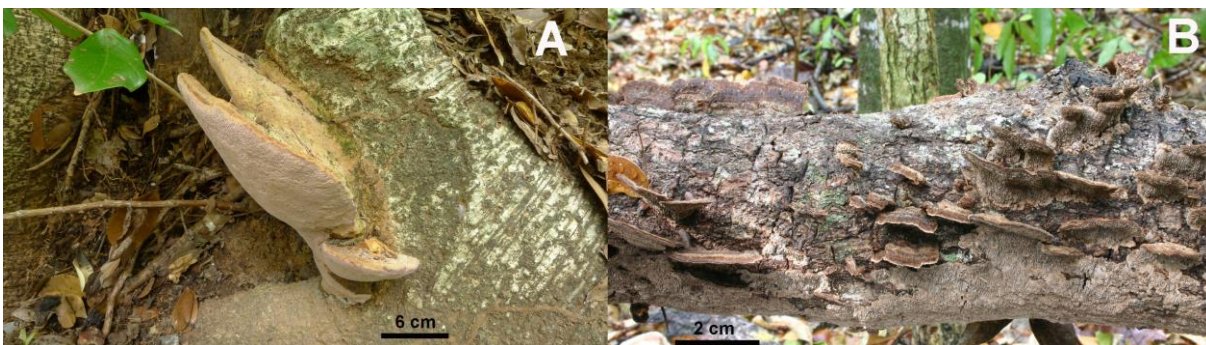
***Trichaptum* sp. 1512/18-J**

This species is similar to *T. byssogenum* but can be distinguished by extremely thin or almost missing context, lack of purplish colors on pores, which are also somewhat larger, 0.5–2 per mm,

isodiametric and without tendency to become lamellate towards the margin (Fig. 5B). Spores are broadly ellipsoid,  $6.5\text{--}8 \times 3.3\text{--}4 \mu\text{m}$ , which is unique in *Trichaptum*. The ITS sequence is related to *T. byssogenum*, but distinctly different. We have found this species only once in Costa Rica; the same sequence, however, is present several times in GenBank as “Uncultured fungus”- see Discussion. Ryvar den and Johansen (1980) show a picture of *Trametes versatilis* Berk 1842 features (synonym of *Trichaptum byssogenum*, collected in the USA) where the spores perfectly correspond to our *Trichaptum* sp. spores. We have not seen the type deposited in Kew and so we cannot decide now if our *Trichaptum* sp. is identical with *Trametes versatilis* or not.

***Trichaptum fumosoavellaneum* (Romell) Raichenb.**

Largest of all *Trichaptum* species, showing massive pilei up to 40 cm in diam. and 10 cm thick (Fig. 6A). Thick context, isodiametric, entire pores, similar spores and ITS sequence indicate relationship with *T. agricola* described above. Nevertheless, except for its size, *T. fumosoavellaneum* is distinguished by smaller pores 3–4 per mm, and usually distinct violet colors.



**Figure 6** – A. *Trichaptum fumosoavellaneum* spec. 1607/79-J. B. *T. griseofuscens* 0904/66-J, both in situ.

***Trichaptum griseofuscens* (Mont.) Ryvar den & Iturr.**

This species is similar and related to *T. sector* but with a hydroid hymenophore from the very beginning (Fig. 6B), more brownish pilei, and narrower spores.

***Trichaptum perrottetii* (Lév.) Ryvar den**

The basidiocarps are pileate, broadly-attached and not decurrent on substrate, 5–10 cm wide and up to 2 cm thick, with unique, 0.5 cm thick, blackish mat of entangled strigose and forked hairs on pileal surface, reminding of *Hexagonia hydroides* pileal cover. Pores violet, on drying becoming brown, 2–3 per mm, later with incised dissepiments. Unmistakable species but rather rare, according to our experience.

***Trichaptum sector* (Ehrenb.) Kreisel**

*T. sector* is a common species of south-eastern USA, easily recognized in the field because of subshiny, white to pale buff pilei and dark greyish pore surface without a purplish tinge. In Costa Rica, however, a different type is present which shows some bluish tints on pores when fresh, somewhat shorter spores and slightly different sequence. It is widely distributed and common in both mountain and lowland wet tropical forests. Two sequences of *T. sector* from Brazil in GenBank may represent still another species, even if closely related. It is possible that *T. sector* in classical sense represents a complex of closely related species that should be studied by more sophisticated methods. Unfortunately, we have not inspected any collection from Brazil (type locality of *T. sector*).

**Discussion**

*Trichaptum* is one of few polypore genera where microscopical routine is of little use for species determination. Macroscopic features are much more significant but sometimes too variable

and not sufficiently discriminating. In such cases, sequence-based phylogeny is of the most importance. ITS sequences provide a robust tool for the discrimination of *Trichaptum* species. For instance, the rather similar *T. biforme* and *T. sector* differ in 54 positions of ITS1 and in 62 positions of ITS2. In contrast, in the case of many other polypores, e.g. *Antrodia* P. Karst., similar species may differ only in 3–4 positions (Spirin et al. 2016, 2017). Analysis of a broad set of newly prepared sequences enabled us to confirm three new species described above and also to open unlooked-for issues of *Trametes versatilis* and *Trichaptum sector* identity and of *Nigrofomes* vs. *Trichaptum* relationship. These questions aim however over the scope of our paper and would require multigenic phylogenetic analyses.

Because our *Trichaptum* sampling did not cover South America we have analyzed aerosol sequencing results of Frohlich-Nowoiski et al. (2012) that originated from sampling performed in Rondonia, Brazil, in the south-western part of the Amazon Basin. In this method, fungal spores are collected for several days on glass fiber filters, bulk DNA is isolated and amplified, cloned, and clones are then individually sequenced. 15 sequences published by Frohlich-Nowoiski et al. (2012) represent according to our analysis *Trichaptum* species and all can be assigned to species studied by us. Six (D01, D08, F06, F07, H09, and H10) belong to *T. byssogenum*, another six (D03, D05, D09, E09, G11 and F03-shown in Fig. 1) correspond with *Trichaptum* sp. 1512/18-J (*Trametes versatilis*?), one (E11, Fig 1) is *T. fumosoavellaneum* and two (E05 and E06) are surprisingly *T. abietinum* - a species which we would not expect in South America. On the other hand, *T. perrottettii*, *T. sector*, and *T. biforme* are not represented, as well as our three newly described species.

Ryvarden (2016) lists 12 *Trichaptum* species in South America. Of them, however, *T. sprucei* belongs to *Phaeodaedalea* Lloyd and *T. durum* (Jungh.) Corner to *Nigroporus* Murrill, according to “*Index fungorum*”. Eight other species which we have studied are treated above. *T. strigosum* Corner seems to be restricted to Amazonia and we have not seen it as well as three other species that were described relatively recently from type localities only (*T. bulbocystidiatum* Ryvarden, *T. deviatum* Ryvarden and *T. variabilis* Ryvarden and Iturriaga). To get more current overview, we have adopted *Trichaptum* key of Ryvarden (2016) for the changes suggested above and inclusion of newly described species.

### Specimens examined

*Nigrofomes melanoporus*. Costa Rica. Puntarenas Prov.: Piedras Blancas, hardwood, 20.IV.2015 Vlasák 1504/38, 1504/42 (JV, GenBank KT156704), Golfito, Playa Nicuesa, hardwood, 18.IV.2017 Vlasák 1704/39, 1704/47 (JV, GenBank MF381028). Alajuela Prov.: Bijagua, hardwood, 28.VII.2016, Vlasák 1607/82 (JV, GenBank MF381027).

*Trichaptum abietinum*. Czech Republic. Hluboka: Karvanice PR, *Picea abies*, 25.VII.2009 Vlasák 0907/9 (JV, GenBank MF381023). USA. California: Purissima Creek, Half Moon Bay, douglas fir, V.2011 Vlasák Jr. 1105/7-J (JV, GenBank MF381024, MF381025). Florida: Lakewood Ranch, *Pinus*, 3.II.2016 Dollinger 640 (JV, GenBank MF381922)

*Trichaptum agricola*. Costa Rica. Guanacaste Prov.: Ahogados, hardwood fence prop, 31.VII. 2014 Vlasák 1407/97, 1407/98 (JV, GenBank MF380993, MF380994), hardwood dead stag Vlasák 1407/99 (JV), Puntarenas Prov: La Ensenada Lodge, hardwood, 22.IV.2015 Vlasák Jr. 1504/75-J (JV, PRM 945505, GenBank MF380995).

*Trichaptum byssogenum*. Costa Rica. Puntarenas Prov.: Dominical, Hacienda Barú, hardwood, 21.IV.2017 Vlasák 1704/82 (JV, GenBank MF381014). Puerto Rico. San Juan, palm tree, XII.2011 Vlasák Jr. 1112/5-J (JV, GenBank MF381013). Thailand. Thaleban Nat. Park, hardwood, 25.XI.2004 J.Kout 0411/9 (JV, GenBank MF381015, MF381016). USA. Florida: Everglades, Long Pine Key, *Pinus*, 22.IV.2009 Vlasák 0904/94 (JV, GenBank MF381012), Sarasota, *Pinus*,



30.XII.2015 *Dollinger 632* (JV). US Virgin Islands: St. John, hardwood, 4.IX.2004 *Vlasák Jr. 0409/22-J* (JV).

*Trichaptum confertum*. Costa Rica. Puntarenas Prov.: Monteverde, Santa Elena, Cerro Amigos Road, hardwood, 29.VII.2014 *Vlasák 1407/75* (JV, PRM 945506, GenBank MF380988, MF380989), El Establo Hotel trails, hardwood, 24.IV.2015 *Vlasák 1504/101* (JV, GenBank MF380992), Golfito, Playa Cacao, hardwood, 19.IV.2015 *Vlasák 1504/32* (JV, GenBank MF380990, MF380991).

*Trichaptum fumosoavellaneum*. Costa Rica. Guanacaste Prov.: Lomas Bardubal, hardwood, 27.VII.2016 *Vlasák Jr. 1607/79-J* (JV, GenBank MF381021).

*Trichaptum fuscoviolaceum*. Czech Republic. Hluboka: Bezdrev, Jaderny, *Pinus sylvatica*, 30.X.2016 *Vlasák 1610/3* (JV, GenBank MF381026).

*Trichaptum griseofuscens*. USA. Florida: Key Largo, John Pennekamp Coral Reefs State Park, hardwood, 21.IV.2009 *Vlasák Jr. 0904/66-J* (JV, GenBank MF381009).

*Trichaptum perrottettii*. USA. Florida: Long Key Natural Area, SW 130th ave, hardwood, XII.2010 *Vlasák Jr. 1012/2-J* (JV, GenBank MF381011).

*Trichaptum resacarium*. USA. Texas: Cameron County, Pharr, hardwood, 27.IX.2011 *Vlasák Jr. 1109/57* (JV, PRM 945510, GenBank MF381017, MF381018), *Vlasák 1109/56* (JV, GenBank MF381019, MF381020).

*Trichaptum sector*. USA. Virginia: Virginia Beach, hardwood, VI.2010 *Vlasák Jr. 1006/14-J* (JV, GenBank MF380997). Florida: Royal Palm St. Park, *Swietenia mahagoni*, 29.VIII.2010 *Vlasák Jr. 1008/70-J* (JV); Sarasota, hardwood, 8.V.2016 *Dollinger 767* (JV, GenBank MF380999); Parrish, Rye Preserve, hardwood, 16.VIII.2016 *Dollinger 897* (JV, GenBank MF380998). Costa Rica. Guanacaste Prov.: Rincon de la Vieja, hardwood, 1.VIII.2014 *Vlasák 1408/8* (JV, GenBank MF381000, MF381001). Puntarenas Prov.: Tarcoles, Laguna Meandrica, hardwood, 22.IV.2015 *Vlasák 1504/61* (JV, GenBank MF381002, MF381003); Golfito, Playa Nicuesa, hardwood, 18.IV.2017 *Vlasák 1704/50* (JV, GenBank MF381004); Dominical, Hacienda Barú, hardwood, 21.IV.2017 *Vlasák 1704/84* (JV, GenBank MF381005, MF381006); Tarcoles, Carara Nat. Park, hardwood, 22.IV.2017 *Vlasák 1704/100* (JV, GenBank MF381007, MF381008).

*Trichaptum* sp. Costa Rica. Guanacaste Prov.: Santa Cruz, hardwood, 28.XII. 2015 *Vlasák Jr. 1512/18-J* (JV, PRM 945507, GenBank MF380996).

*Trichaptum subchartaceum*. USA. New York: Adirondack Park, Fish Creek Pond, *Populus* sp., 18.IX.2005 *Vlasák 0509/146* (JV, GenBank MF381010).

### Key to South-American *Trichaptum* species

1. Basidiocarps pileate ..... 2
1. Basidiocarps resupinate ..... 13
2. Pilei glabrous to adpressed tomentose mixed with glabrous zones ..... 3
2. Pilei strongly strigose to hispid..... 9
3. Hymenophore distinctly hydroid ..... *T. griseofuscens*
3. Hymenophore poroid ..... 4
4. Basidiocarps < 3 mm thick, flexible, often in clusters, pileal surface velutinate to hirsute ..... 5
4. Basidiocarps >10 mm thick, dense and hard, usually single, pileal surface glabrous ..... 7
5. Pore surface grey to black, pores entire, upper surface often grey to whitish ..... *T. sector*

5. Pore surface with violet tinge when fresh, fading to pale or deep brown ..... 6
6. Upper surface light grey to deep beige, pore surface fading to beige, pores 3–5 per mm, later often lacerate or irpicoid..... T. biforme
6. Upper surface blackish-grey, pore surface fading dark grey, pores 6–8 per mm, regular..... T. confertum
7. Pores irregular at least in parts, pore surface split with age, basidiocarp brown..... T. variabilis
7. Pores regular and round, pore surface even, basidiocarps greyish to clay colored ..... 8
8. Pores 3–4 per mm, basidiocarps clay colored or brown with purplish tinge, up to 14 cm thick in massive specimens..... T. fumosoavellaneum
8. Pores 1–3 per mm, basidiocarps brown and without purplish tints, up to 3 cm thick..... T. agricola
9. Pilei with a dense, 5 mm thick mat of dark brown strigose to hispid hairs, basidiocarps sessile..... T. perrottetii
9. Pilei with light gray to whitish hairs, basidiocarps resupinate, effused-reflexed or sessile ..... 10
10. Spores > 3.3 µm in width, pore surface without purplish tinge, pores isodiametric ..... Trichaptum sp. 1512/18-J
10. Spores < 3.3 µm in width, pores with violet tinge when fresh, radially elongated ..... 11
11. Pores 1–2 per mm ..... 12
11. Pores 2–4 per mm, spores ellipsoid, 4.5–6 × 2.5–3 µm..... T. strigosum
12. Spores cylindrical 5.5–8 × 2–2.5 µm, pore surface purplish or purplish brown ..... T. byssogenum
12. Spores ellipsoid 3.5–4 × 2 µm, pore surface dark fuliginous with purplish tint..... T. resacarium
13. Bulbous cystidia present, pores 2–4 per mm ..... T. bulbocystidium
13. Bulbous cystidia absent, pores 5–7 per mm ..... T. deviatum

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