



# A re-evaluation of Neotropical *Junghuhnia* s.lat. (*Polyporales*, *Basidiomycota*) based on morphological and multigene analyses

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## Key words

Mycodiversity  
phylogeny  
Steccherinaceae  
taxonomy

**Abstract** *Junghuhnia* is a genus of polypores traditionally characterised by a dimitic hyphal system with clamped generative hyphae and presence of encrusted skeletocystidia. However, recent molecular studies revealed that *Junghuhnia* is polyphyletic and most of the species cluster with *Steccherinum*, a morphologically similar genus separated only by a hydroid hymenophore. In the Neotropics, very little is known about the evolutionary relationships of *Junghuhnia* s.lat. taxa and very few species have been included in molecular studies. In order to test the proper phylogenetic placement of Neotropical species of this group, morphological and molecular analyses were carried out. Specimens were collected in Brazil and used for DNA sequence analyses of the internal transcribed spacer and the large subunit of the nuclear ribosomal RNA gene, the translation elongation factor 1- $\alpha$  gene, and the second largest subunit of RNA polymerase II gene. Herbarium collections, including type specimens, were studied for morphological comparison and to confirm the identity of collections. The molecular data obtained revealed that the studied species are placed in three different genera. Specimens of *Junghuhnia carneola* represent two distinct species that group in a lineage within the phlebioid clade, separated from *Junghuhnia* and *Steccherinum*, which belong to the residual polyporoid clade. Therefore, the new genus *Geesterania* is proposed including two species, *G. carneola* comb. nov. and *G. davidii* sp. nov. Neotropical specimens identified as *Junghuhnia nitida* represent a different lineage from the European species and are described as *Steccherinum neonitidum* sp. nov. In addition, the new combinations *Steccherinum meridionale*, *Steccherinum polycystidiferum* and *Steccherinum undigerum*, as well as the new name *Flaviporus tenuis*, are proposed.

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## INTRODUCTION

The genus *Junghuhnia* (Basidiomycota, Polyporales) is characterised by a dimitic hyphal system, clamped generative hyphae, small basidiospores with variable shape and presence of large encrusted skeletocystidia. Traditionally, it has been separated from *Steccherinum* only by the hymenophore configuration (poroid in *Junghuhnia* and hydroid in *Steccherinum*), but microscopically both genera present almost identical features. *Antrodiella* is also related but distinguished by the lack of cystidia (Ryvarden 1991). However, recent studies including molecular data (Miettinen et al. 2012) showed that the morphological segregation of *Junghuhnia* and *Steccherinum* is not supported by molecular phylogeny. Many species treated as *Junghuhnia* in the literature group with species with hydroid hymenophore in the *Steccherinum* clade, separated from *J. crustacea*, the type of *Junghuhnia*. Furthermore, there are no microscopic characters enabling delimitation between *Junghuhnia* s.str. (including *J. crustacea*) and *Junghuhnia* s.lat. (species included in *Steccherinum* clade) due to convergent micromorphology. In the Neotropics, 11 names were previously reported in *Junghuhnia*: *J. carneola*, *J. chlamydospora*, *J. globospora*, *J. meridionalis*, *J. minuta*, *J. neotropica*, *J. nitida*, *J. polycystidifera*, *J. semisupiniformis*, *J. subundata* and *J. undigera*

(Ryvarden 1985, 2007, 2015, Lindblad & Ryvarden 1999, Westphalen et al. 2012). However, molecular data of all of them are unavailable and their phylogenetic position is unknown. Therefore, the current study addresses a knowledge gap, using molecular and morphological data to elucidate the systematics of Neotropical *Junghuhnia* and related genera.

## MATERIAL AND METHODS

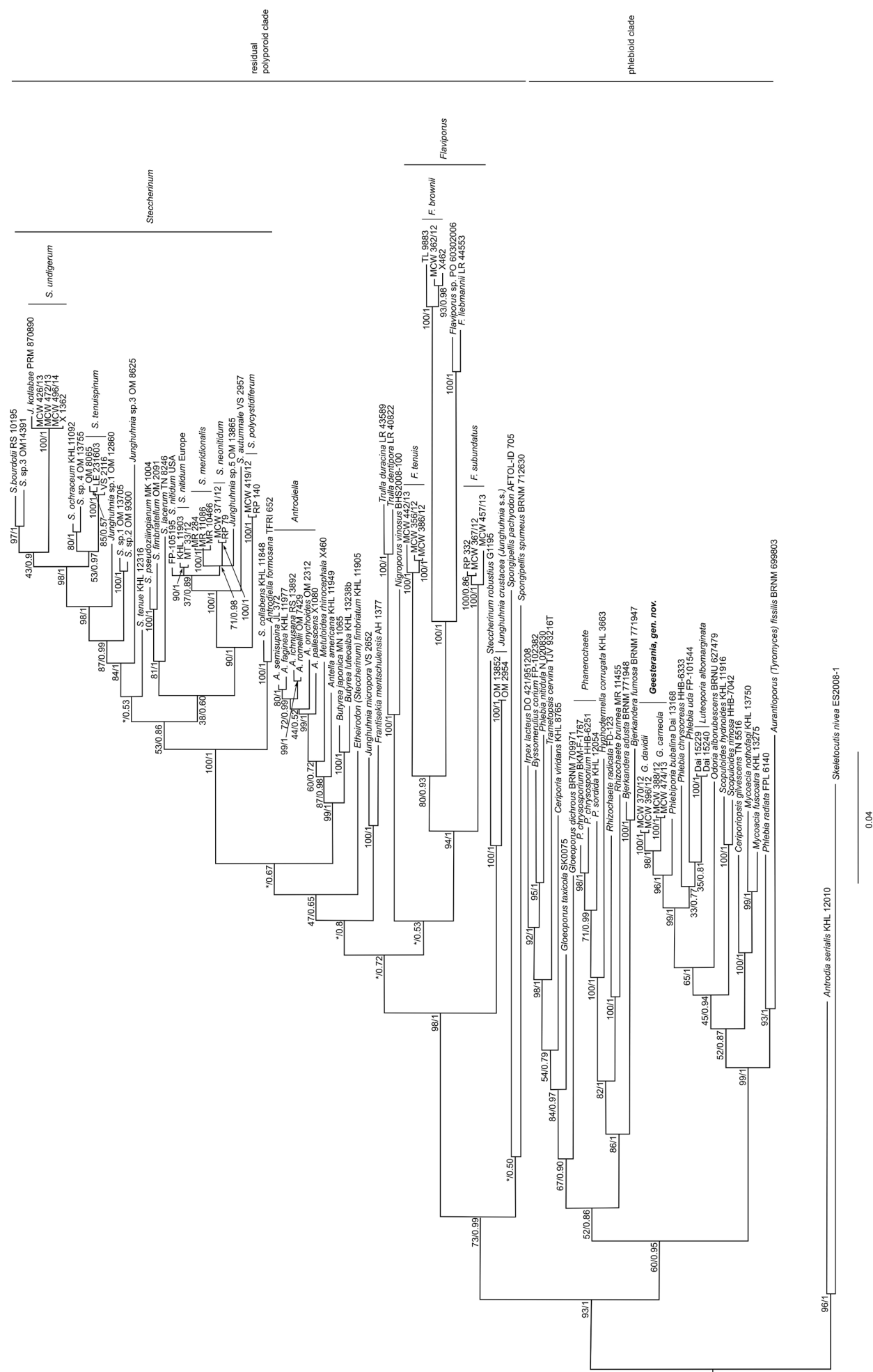
### Morphological analysis

Specimens were collected in southern Brazil in Santa Catarina, São Paulo and Rio Grande do Sul States. Additional specimens of Neotropical *Junghuhnia* s.lat. from BPI, FLOR, ICN, O, PRM and S herbaria (Thiers 2016) were studied for morphological revision. For micromorphology observations, hand-cut sections of the basidiomes were prepared on microscope slides with 3 % KOH solution and stained with 1 % aqueous phloxine. When necessary, small pieces of the basidiomes were kept in NaOH 3 % solution under 60 °C for about 12 h in order to obtain a better separation of the hyphae and interpret the hyphal system. The samples were then used for preparing microscope slides. All microscopic structures observed were measured with aid of an eyepiece micrometer and, when possible, a minimum of twenty measurements of each structure were taken. The abbreviations and codes used for the measurements are: Lm  $\times$  Wm = means of length and width, Q = range of length/width ratios, Qm = length/width mean and n = x/y (x = number of measurements from a given number; y of specimens). Drawings of the microstructures were made with the aid of a drawing tube. Cresyl violet was used for observation on metachromatic reactions.

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**g. 1** Phylogenetic tree of ITS-LSU regions conducted by Bayesian analysis. Numbers at branches indicate maximum likelihood bootstrap proportion and Bayesian posterior probability values. The asterisk (\*) marks different topologies in both analyses. The bar indicates number of expected substitutions per position.

**Table 1** List of sequences used in this study. \* Sequences obtained in this study.

Specimen	GenBank No.			
	ITS	LSU	TEF1- $\alpha$	RPB2
<i>Antella americana</i> (KHL 11949)	JN710509	JN710509	JN710711	–
<i>Anrodia serialis</i> (KHL 12010)	JX109844	JX109844	JX109898	JX109870
<i>Anrodiella faginea</i> (KHL 11977)	JN710514	JN710514	JN710712	–
<i>Anrodiella ichnusana</i> (RS 13892)	JN710516	JN710516	–	–
<i>Anrodiella onychoides</i> (OM 2312)	JN710517	JN710517	–	–
<i>Anrodiella pallescens</i> (X1080)	JN710518	JN710518	–	–
<i>Anrodiella romellii</i> (OM 7429)	JN710520	JN710520	–	–
<i>Anrodiella semisupina</i> (JL 372)	JN710521	JN710521	–	–
<i>Aurantioporus fissilis</i> (BRNM 699803)	HQ728292	HQ729002	–	–
<i>Bjerkandera adusta</i> (BRNM 771948)	KT305935	KT305935	KT305938	–
<i>Bjerkandera fumosa</i> (BRNM 771947)	KT305937	KT305937	–	–
<i>Butyrea japonica</i> (MN 1065)	JN710556	JN710556	JN710718	–
<i>Butyrea luteoalba</i> (KHL 13238b)	JN710558	JN710558	JN710719	–
<i>Byssomerulius corium</i> (FP-102382)	KP135007	KP135230	KP134921	–
<i>Ceriporia viridans</i> (KHL 8765)	AF347109	AF347109	–	–
<i>Ceriporiopsis gilvescens</i> (TN 5516)	HQ659222	HQ659222	–	–
<i>Flaviporus brownii</i> (TL 9883)	JN710537	JN710537	–	–
<i>Flaviporus brownii</i> (X462)	JN710538	JN710538	–	–
<i>Flaviporus brownii</i> * (MCW 362/12)	KY175008	KY175008	KY175022	–
<i>Flaviporus liebmanni</i> (LR 44553)	JN710540	JN710540	–	–
<i>Flaviporus minutus</i> * (MCW 442/13)	KY175001	KY175001	–	–
<i>Flaviporus minutus</i> * (MCW 356/12)	KY175002	KY175002	–	–
<i>Flaviporus minutus</i> * (MCW 386/12)	KY175003	KY175003	–	–
<i>Flaviporus</i> sp. (PO 60302006)	JN710542	JN710542	–	–
<i>Flaviporus subundatus</i> * (MCW 367/12)	KY175004	KY175004	–	–
<i>Flaviporus subundatus</i> * (MCW 457/13)	KY175005	KY175005	–	–
<i>Flaviporus subundatus</i> * (RP 332)	KY175006	KY175006	–	–
<i>Frantisekia mentschulensis</i> (AH 1377)	JN710544	JN710544	–	–
<i>Geesteriana carneola</i> * (MCW 388/12)	KY174999	KY174999	KY175013	KY175011
<i>Geesteriana carneola</i> * (MCW 474/13)	KY175000	KY175000	KY175014	–
<i>Geesteriana davidii</i> * (MCW 370/12)	KY174997	KY174997	KY175015	–
<i>Geesteriana davidii</i> * (MCW 396/12)	KY174998	KY174998	KY175016	KY175012
<i>Gloeoporus dichrous</i> (BRNM 709971)	EU546097	FJ496709	–	–
<i>Gloeoporus taxicola</i> (SK 0075)	JX109847	JX109847	JX109901	JX109873
<i>Hyphodermella corrugata</i> (KHL 3663)	EU118630	EU118630	–	–
<i>Irpex lacteus</i> (DO 421/951208)	JX109852	JX109852	JX109911	JX109882
<i>Junghuhnia crustacea</i> (OM 13852)	JN710554	JN710554	–	–
<i>Junghuhnia crustacea</i> (OM 2954)	JN710553	JN710553	–	–
<i>Junghuhnia kotlabae</i> * (PRM 870890)	KY175007	KY175007	–	–
<i>Junghuhnia micropora</i> (VS 2652)	JN710559	JN710559	JN710720	–
<i>Junghuhnia</i> sp1 (OM12860)	JN710563	JN710563	JN710723	–
<i>Junghuhnia</i> sp3 (OM 8625)	JN710564	JN710564	JN710724	–
<i>Junghuhnia</i> sp4 (X1362)	JN710565	JN710565	JN710725	–
<i>Junghuhnia</i> sp5 (OM 13865)	JN710566	JN710566	JN710726	–
<i>Luteoporia albomarginata</i> (Dai 15229)	KU598873	KU598878	–	–
<i>Luteoporia albomarginata</i> (Dai 15240)	KU598874	KU598879	–	–
<i>Metuloides rhinocephala</i> (X460)	JN710562	JN710562	–	–
<i>Mycoacia fuscoatra</i> (KHL13275)	JN649352	JN649352	JX109908	JX109879
<i>Mycoacia nothofagi</i> (KHL13750)	GU480000	GU480000	–	–
<i>Nigroporus vinosus</i> (BHS2008-100)	JX109857	JX109857	–	–
<i>Odoria alborubescens</i> (BRNU 627479)	JQ821319	JQ821318	–	–
<i>Phanerochaete chrysosporium</i> (BKM-F-1767)	HQ188436	GQ470643	HQ188379	–
<i>Phanerochaete chrysosporium</i> (HHB-6251)	KP135094	KP135246	–	KP134954
<i>Phanerochaete sordida</i> (KHL 12054)	EU118653	EU118653	–	–
<i>Phlebia chrysocreas</i> (HHB-6333)	KP135358	KP135263	–	KP134908
<i>Phlebia uda</i> (FP-101544)	KP135361	KP135232	–	KP134909
<i>Phlebia nitidula</i> (N 020830)	EU118655	EU118655	–	–
<i>Phlebia radiata</i> (FPL 6140)	AY854087	AF287885	AY885156	AY218502
<i>Phlebioporia bubalina</i> (Dai 13168)	KC782526	KC782528	–	–
<i>Rhizochaete brunnea</i> (MR 11455)	AY219389	AY219389	–	–
<i>Rhizochaete radicata</i> (FD-123)	KP135407	KP135279	–	KP134937
<i>Scopuloides hydroides</i> (KHL 11916)	EU118665	EU118665	–	–
<i>Scopuloides rimosa</i> (HHB-7042)	KP135350	KP135282	–	KP134903
<i>Skeletocutis nivea</i> (ES2008-1)	JX109858	JX109858	–	–
<i>Spongipellis pachyodon</i> (AFTOL-ID 705)	DQ249277	AY629322	DQ028599	DQ408123
<i>Spongipellis spumeus</i> (BRNM 712630)	HQ728288	HQ728288	–	–
<i>Steccherinum autumnale</i> (VS 2957)	JN710549	JN710549	JN710716	–
<i>Steccherinum bourdotii</i> (RS 10195)	JN710584	JN710584	–	–
<i>Steccherinum collabens</i> (KHL 11848)	JN710552	JN710552	JN710717	–
<i>Steccherinum fimbriatellum</i> (OM 2091)	JN710555	JN710555	–	–
<i>Steccherinum fimbriatum</i> (KHL 11905)	JN710530	JN710530	–	–
<i>Steccherinum formosanum</i> (TFRI 652)	EU232184	EU232268	–	–
<i>Steccherinum lacerum</i> (TN 8246)	JN710557	JN710557	–	–
<i>Steccherinum meridionalis</i> * (MR 284)	KY174992	KY174992	KY175019	KY175009
<i>Steccherinum meridionalis</i> * (MR 11086)	KY174993	KY174993	–	–
<i>Steccherinum meridionalis</i> * (MR 10466)	KY174994	KY174994	–	–
<i>Steccherinum neonitidum</i> * (MCW 371/12)	KY174990	KY174990	KY175017	KY175010

Table 1 (cont.)

Specimen	GenBank No.			
	ITS	LSU	TEF1-α	RPB2
<i>Steccherinum neonitidum</i> * (RP 79)	KY174991	KY174991	KY175018	–
<i>Steccherinum nitidum</i> (FP-105195)	KP135323	KP135227	–	KP134964
<i>Steccherinum nitidum</i> (KHL 11903)	JN710560	JN710560	JN710721	–
<i>Steccherinum nitidum</i> * (MT 33/12)	KY174989	KY174989	–	–
<i>Steccherinum ochraceum</i> (KHL 11902)	JN710590	JN710590	JN710730	JN710738
<i>Steccherinum polycystidiferum</i> * (MCW 419/12)	KY174995	KY174995	KY175021	–
<i>Steccherinum polycystidiferum</i> * (RP 140)	KY174996	KY174996	–	–
<i>Steccherinum pseudozilingianum</i> (MK 1004)	JN710561	JN710561	JN710722	–
<i>Steccherinum robustius</i> (G1195)	JN710591	JN710591	–	–
<i>Steccherinum</i> sp1 (OM 13705)	JN710592	JN710592	JN710731	–
<i>Steccherinum</i> sp2 (OM 9300)	JN710593	JN710593	–	–
<i>Steccherinum</i> sp3 (OM 14391)	JN710594	JN710594	JN710732	–
<i>Steccherinum</i> sp4 (OM 13755)	JN710596	JN710596	–	–
<i>Steccherinum tenue</i> (KHL 12316)	JN710598	JN710598	JN710733	–
<i>Steccherinum tenuispinum</i> (OM 8065)	JN710599	JN710599	–	–
<i>Steccherinum tenuispinum</i> (LE231603)	KM411452	KM411452	–	–
<i>Steccherinum tenuispinum</i> (VS 2116)	JN710600	JN710600	–	–
<i>Steccherinum undigerum</i> * (MCW 426/13)	KY174986	KY174986	KY175020	–
<i>Steccherinum undigerum</i> * (MCW 472/13)	KY174987	KY174987	–	–
<i>Steccherinum undigerum</i> * (MCW 496/14)	KY174988	KY174988	–	–
<i>Trametopsis cervina</i> (TJV 93216T)	JN165020	JN164796	JN164882	JN164877
<i>Trulla dentipora</i> (LR 40822)	JN710512	JN710512	–	–
<i>Trulla duracina</i> (LR 43589)	JN710513	JN710513	–	–

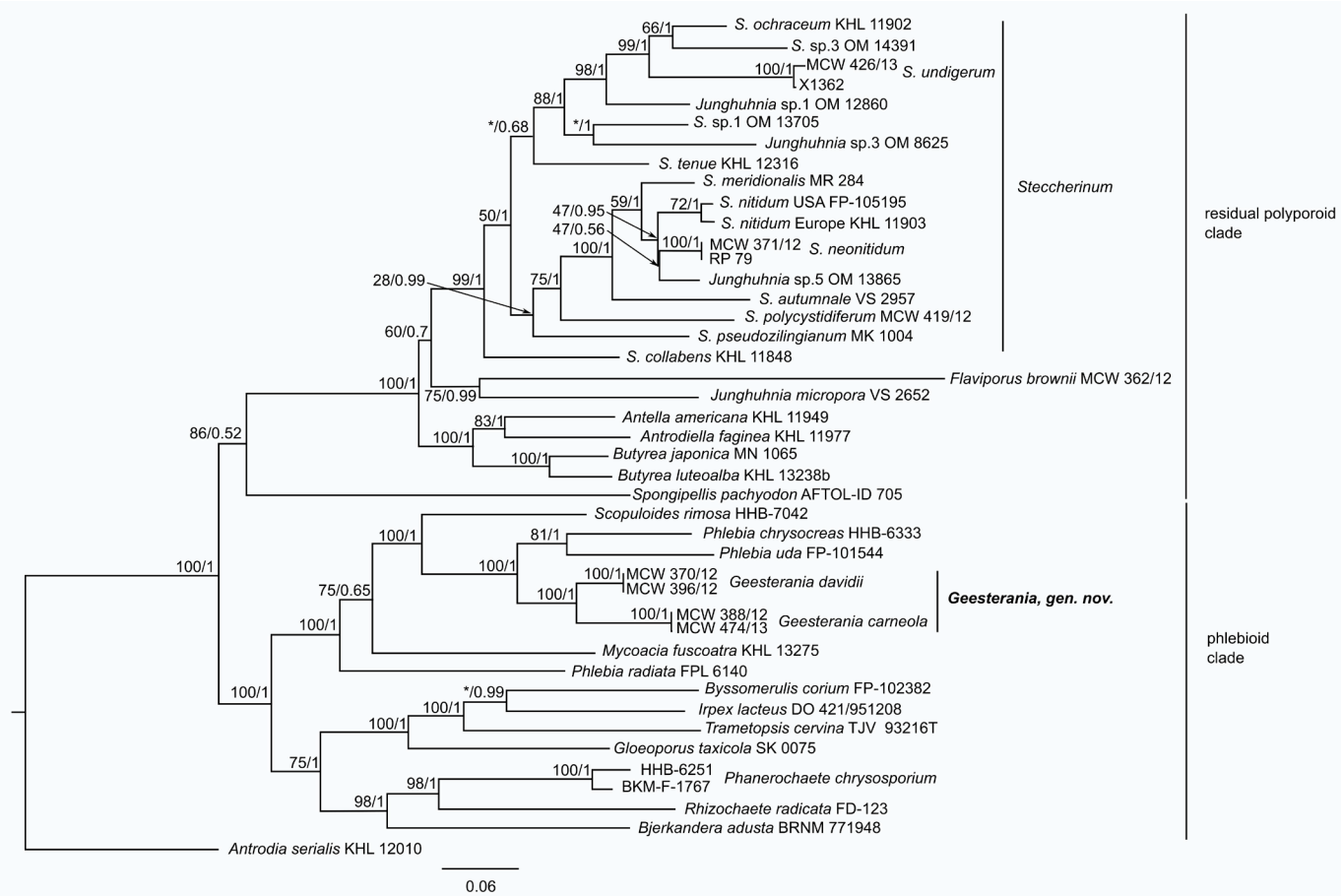


Fig. 2 The phylogenetic tree of ITS, LSU, TEF1-α, RPB2 regions conducted by Bayesian analysis. Numbers at branches indicate maximum likelihood bootstrap proportion and Bayesian posterior probability values. The asterisk (\*) marks different topologies in both analyses. The bar indicates number of expected substitutions per position.

### Culturing, mating tests and nuclear staining

Sporeprints were obtained from fresh basidiomes and used to prepare monosporic and polysporic cultures. Cultures were grown and kept in malt extract agar (MEA) or potato dextrose agar (PDA) at 25 °C. In order to clarify the mating system of the species in question, mating tests were performed according to Hallenberg (1984). At least nine monosporic cultures of each species were used for mating tests. For nuclear behaviour verification, Giemsa stain was used for nuclear staining technique following Boidin (1958). After staining, the mycelia were observed under microscope and nuclear behaviour was classified as Normal, Astatocoenocytic, Heterocytic or Holocoenocytic (Boidin 1971). Cultures obtained were kept at the Botany Institute of São Paulo Culture Collection and Culture Collection of Basidiomycetes (Institute of Microbiology, Czech Academy of Sciences, Prague). Additional cultures from CIEFAP culture collection (CIEFAPcc) were also used in this study.

### DNA extraction and amplification

DNeasy Plant Mini Kit (QIAGEN) and Power Soil DNA Isolation Kit (MO BIO) were used for DNA extraction from cultures and basidiomes, respectively. In cases where basidiome extraction was not successful, the 10 first steps of DNeasy Plant Mini Kit were followed and DNA extraction of the samples proceeded using magnetic-bead technology of MagNA Pure compact system (Roche). When necessary, DNA was purified using DNA Clean & Concentration Kit (Zymo Research). DNA amplification of the internal transcribed spacer (ITS) and large subunit (LSU) regions of ribosomal RNA gene were performed using ITS5/ITS4-Basidio (Nikolcheva & Bärlocher 2004) primer combination for ITS and LR0R/LR6 for LSU. The PCR regimes followed Tomšovský et al. (2010a). For translation elongation factor 1- $\alpha$  gene (TEF1- $\alpha$ ; primers: 983F/2218R), touchdown PCR with gradually reduced annealing temperature (60–50 °C) was performed (Rehner & Buckley 2005). For the second largest subunit of RNA polymerase II gene (RPB2; primers fRPB25F/bRPB271R), a touchdown protocol modified from Rehner & Buckley (2005) EF- $\alpha$  protocol was performed. In some samples where PCR did not present good results, a nested PCR was performed according to Tomšovský et al. (2010b). Amplification products were sent for purification and sequencing in MacroGen Ltd. (Korea).

### Phylogenetic analysis

All the sequences obtained were initially checked in Chromas 2.6 (<http://technelysium.com.au/wp/chromas/>) and BioEdit (Hall 1999) software and, when necessary, adjusted manually to correct minor sequencing errors. Two combined dataset of DNA sequences were prepared, one using ITS-LSU and one with four molecular markers (ITS, LSU, TEF1- $\alpha$ , RPB2). Reference sequences for both datasets were chosen based on studies of Miettinen et al. (2012), Floudas & Hibbett (2015), Binder et al. (2013) and through BLAST searches in the NCBI (National Center for Biotechnology <http://www.ncbi.nlm.nih.gov>), and are summarised in Table 1. The datasets were aligned using MAFFT version 7 (<http://mafft.cbrc.jp/alignment/server/>). In order to remove highly variable regions with low homology in the 4 gene dataset, Gblocks curation was performed on ITS sequences and intron regions of TEF1- $\alpha$  and RPB2 sequences were deleted manually. The evolutionary models for each gene and for combined datasets were inferred with the jModelTest 2.c1.4 (Darriba et al. 2012) using the Akaike information criterion. The best fit models selected were TrN+I+G for TEF1- $\alpha$  region, TIM2+I+G for RPB2 and GTR+I+G for ITS, LSU, and for both combined datasets. Bayesian trees and posterior probabilities were estimated with the MrBayes 3.2.6 software (Ronquist et al. 2012). The analysis was run for 10 million generations,

sampling every 1 000 generations. Burnin was set to 10 % of the trees. Maximum Likelihood analyses were conducted in RAXML-HP v. 8 (Stamatakis 2014) with a GTRCAT model of evolution. Number of bootstrap replicates was halted automatically (autoMRE) and 252 replicates were used for the ITS-LSU dataset and the 204 for four marker dataset. All analyses were conducted in CIPRES Science Gateway (Miller et al. 2010).

## RESULTS

Molecular data obtained show that the studied Neotropical *Junghuhnia* spp. are polyphyletic and occur in three different genera (Fig. 1, 2): *Steccherinum* (supported in ML and BA of both ITS-LSU and combined dataset), *Flaviporus* (unsupported in ML in ITS-LSU dataset and supported in ML and BA of both ITS-LSU and combined dataset) and a new genus in the phlebioid clade (supported in ML and BA of both datasets). None of the studied species is related to the type of the genus, *J. crustacea*. The ITS-LSU dataset includes 1952 positions including 1070 conserved, 825 variable and 136 singleton. The four gene dataset includes 3482 positions including 2191 conserved, 1287 variable and 317 singleton.

The description of a new polypore genus in the phlebioid clade, *Geesterania*, including *J. carneola*, is supported by both the phylogenetic data and morphological characters. Moreover, *J. carneola* is in fact a complex of two species. Our data also show that the Neotropical *J. nitida* represents a different species separated from the European specimens (the species was described from Europe) and here is described as a new species, *Steccherinum neonitidum*. In addition, three new combinations: *Steccherinum meridionale*, *Steccherinum polycystidiferum* and *Steccherinum undigerum*, and one new name: *Flaviporus tenuis*, are proposed and the position of *J. subundata* in *Flaviporus* (Ginns 1980) is confirmed. Full descriptions and illustrations of the new genus and the two new species are presented below, as well as comments on the new combinations and name proposed. Identification keys for the genera and species of Neotropical *Junghuhnia* s.lat. are also provided. Further discussion on the four Neotropical species not included in this study (*J. chlamydospora*, *J. globospora*, *J. neotropica* and *J. semisupiniformis*) is presented.

## TAXONOMY

***Geesterania* Westphalen, Tomšovský & Rajchenb., gen. nov.**  
— MycoBank MB822658; Fig. 3, 4

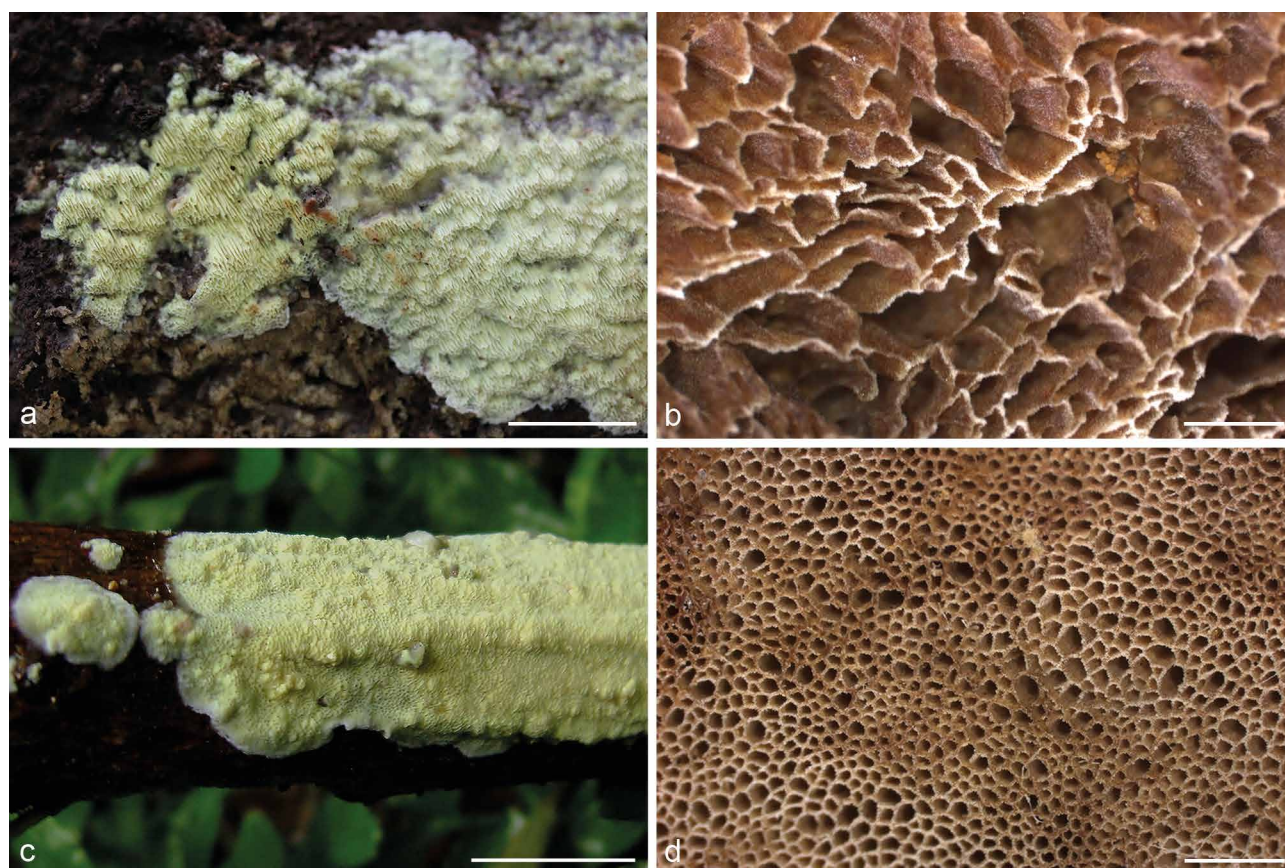
*Type.* *Geesterania carneola* (Bres.) Westphalen & Rajchenb.

*Etymology.* Named in honour of Rudolph Arnold Maas Geesteranus (The Hague 1911–Oegstgeest 2003) for his extensive contribution to the mycology, including studies of the genus *Steccherinum* and of several phlebioid species.

*Diagnosis.* *Geesterania* is characterised by the basidiomes becoming reddish when bruised, the abundant, long, thin- to thick-walled, finely encrusted skeletocystidia, the metachromatic skeletal hyphae and cystidia, a bipolar mating system and astatocoenocytic nuclear behaviour.

Basidiomes resupinate, adnate, becoming reddish when bruised and/or upon drying, soft to fleshy when fresh, becoming harder and somewhat waxy when dried. Hymenophore poroid; pores regular to irregular, round to angular. Hyphal system dimitic, generative hyphae clamped, skeletal hyphae metachromatic, hyphae IKI-. Skeletocystidia present in the trama and dissepiments, cylindrical to clavate, encrusted at the apex with thin crystals, usually very long and appearing as skeletal hyphae ends, metachromatic, IKI-. Basidiospores ellipsoid to subcylindrical, hyaline, smooth, IKI-.





**Fig. 3** a, b. *Geesterania carneola*; c, d. *G. davidii*. — a, c. Fresh basidiomes; b, d. detail of pore surface in dried basidiomes. — Scale bars: a, c = 2 cm; b, d = 1 mm. — Photos: a. D.L. Komura; b, d. M.C. Westphalen; c. M.A. Reck.

**Notes** — *Geesterania* is characterised by resupinate, somewhat fleshy basidiomes typically becoming reddish when bruised or dried. The colour change is variable, and in some basidiomes only some reddish spots appear while others become pale reddish brown to flesh coloured throughout. Microscopically, the very long finely encrusted (skeleto)cystidia are typical. In the trama, the cystidia are usually markedly thick-walled and long, arising from skeletal hyphae that slowly increase in diameter. For that reason, the distinction between skeletal hyphae and cystidia sometimes is not clear and the cystidia may be interpreted as wide encrusted skeletal hyphae ends. Here we choose to use the term cystidia as these structures can be very conspicuous in the dissepiments and in the trama, sometimes having many crystals covering a widened apex. In addition, the skeletal hyphae and cystidia are metachromatic, while the generative hyphae, hymenium and basidiospores are non metachromatic in Cresyl Violet. *Geesterania* differs from other polypore genera exactly by this combination of characteristics, which is unique and unknown in other poroid fungi. Even though the type species had been placed in *Junghuhnia*, morphological, biological and molecular data support it as a different genus. *Junghuhnia* s.str. and *Steccherinum* present long encrusted skeletocystidia. However, in both genera the cystidia are encrusted with large crystals and can be easily distinguished from the skeletal hyphae, as they present a more abrupt widening and more evident encrustation. In addition, the skeletal hyphae in *Junghuhnia* and *Steccherinum* are non metachromatic in Cresyl Violet and only the cystidia present a variable metachromatic reaction, while in *Geesterania* the cystidia and skeletal hyphae are metachromatic throughout. Furthermore, culture studies had already shown that *G. carneola* is bipolar and astatocoenocytic, while species of *Junghuhnia* s.str. and *Steccherinum* s.lat. are tetrapolar and have a normal nuclear behaviour (David & Rajchenberg 1985, Rajchenberg

2011). The combination of bipolarity and astatocoenocyt is characteristic in the phlebioid clade (Rajchenberg 2011), and our molecular data confirm the position of *Geesterania* there (Fig. 1). Phylogenetically, the closest related polypore species known is *Phlebioporia bubalina* (Chen & Cui 2014) but, morphologically, it substantially differs by presenting a monomitic hyphal system with simple septate dextrinoid hyphae and lack of cystidia. The recently described genus and species *Luteoporia albomarginata* (Wu et al. 2016) is also phylogenetically related to *Geesterania*, presenting similar resupinate basidiomes with presence of cystidia-like structures. However, *Luteoporia* differs in being monomitic, turning red in contact with KOH, and having differently shaped thin-walled cystidia.

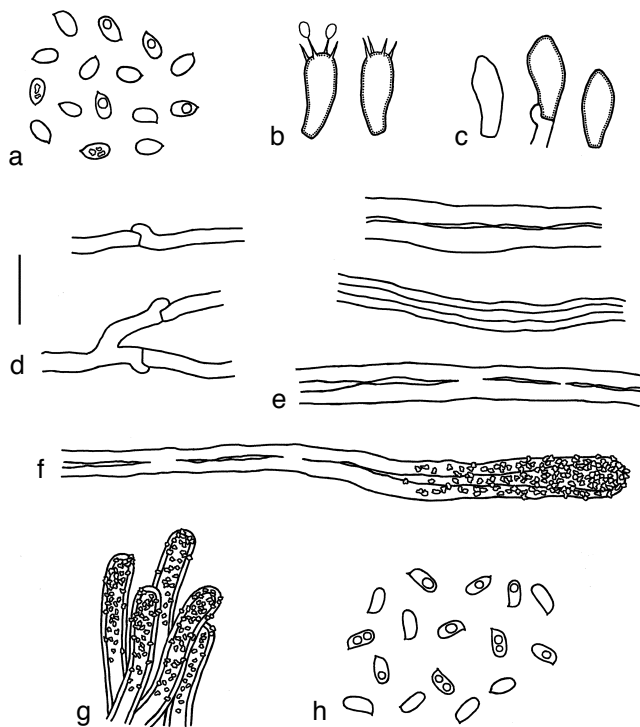
***Geesterania carneola* (Bres.) Westphalen & Rajchenb., comb. nov.** — MycoBank MB822659; Fig. 3a–b, 4a–g

≡ *Poria carneola* Bres., Hedwigia 35: 282. 1896, basionym.

≡ *Junghuhnia carneola* (Bres.) Rajchenb., Revista Invest. Agropecu., Ser. 5, 19: 45. 1984.

**Specimens examined.** BRAZIL, Paraná, Paranaguá, Prox. a Rodovia PR 508, 2 Sept. 2013, M.C. Westphalen 458/13 (SP 446225); Rio Grande do Sul, São Francisco de Paula, FLONA, 30 Apr. 2012, M.C. Westphalen 388/12 (SP 446186); Santa Catarina, Blumenau, Moller (S F159730), syntype; ibid., Moller 284 (S F15834), syntype; ibid., Moller 465 (S F15836, syntype); Itapoá, RPPN Volta Velha, 17 Nov. 2012, M.C. Westphalen 412/12 (FLOR); Santo Amaro de Imperatriz, Parque Estadual da Serra do Tabuleiro, Hotel Plaza Caldas da Imperatriz, 24 Jan. 2014, M.C. Westphalen 480/14 (SP 446238); São Paulo, Santo André, Reserva Biológica do Alto da Serra de Paranapiacaba, 5 Dec. 2013, M.C. Westphalen 474/13 (SP 446234); São Luiz do Paraitinga, P.E. da Serra do Mar, Núcleo Sta. Virgínea, 17 Apr. 2013, R.M. Pires RP2 (SP 446259).

**Notes** — This species was described from southern Brazil (Santa Catarina State) by Bresadola (1896) and since then



**Fig. 4** a–g. *Geesterania davidii*; h. *G. carneola*. — a. Basidiospores; b. basidia; c. cystidioles; d. generative hyphae; e. skeletal hyphae; f. cystidium from the trama; g. cystidia from the dissepiments; h. basidiospores. — Scale bar = 10 µm. — Drawn by M.C. Westphalen.

registered from different tropical and subtropical regions in South and Central America (Lowe 1966, Carranza-Velásquez & Ruiz-Boyer 2005, Robledo & Rajchenberg 2007, Westphalen et al. 2012). It is characterised by its yellowish basidiomes that become reddish when bruised and sometimes after drying and the large and very irregular pores (3–5 per mm) that are often split and lacerate, forming a daedaloid surface. Microscopically it presents a dimitic hyphal system, thin to thick-walled cystidia encrusted with fine crystals and narrowly ellipsoid basidiospores ( $4.0\text{--}5.0 \times 2.0\text{--}2.5$  µm,  $Lm \times Wm = 4.38 \times 2.38$ ,  $Q = 1.60\text{--}2.25$ ,  $Qm = 1.86$ ,  $n = 30/2$ ).

***Geesterania davidii*** Westphalen & Rajchenb., *sp. nov.* — MycoBank MB822660; Fig. 3c–d, 4h

**Holotype.** BRAZIL, Rio Grande do Sul, Morrinhos do Sul, Perdida, 14 May 2012, M.C. Westphalen 396/12 (SP 446193).

**Etymology.** Named in honour of Alix David (Lyon, France), for her extensive work on the biology of fungi, especially her numerous investigations on mating systems and nuclear behaviour of polypores.

Basidiomes annual, resupinate, thin, soft and somewhat fleshy when fresh, becoming hard and slightly waxy when dried, margin narrow to absent, regular, white to cream-coloured; pore surface pale yellow to pale greenish yellow when fresh, becoming reddish when bruised and straw-coloured to pinkish brown after drying; pores regular, round to angular, with thin and often dentate dissepiments, sometimes partially split, 5–7(–8) per mm, tubes concolour with the pore surface, up to 2 mm deep; subiculum very thin, up to 0.2 mm thick, cream-coloured when fresh, becoming ochraceous when dried. Hyphal system dimitic; generative hyphae with clamps, branched or more rarely unbranched, hyaline and thin-walled, 2.0–4.0 µm wide; skeletal hyphae hyaline to slightly yellowish, thick-walled to almost solid, unbranched, 3.0–5.0 µm wide. Cystidia appearing as thickened skeletal hyphal ends, in the dissepiments slightly thick-walled, in the trama forming a continuous with the skeletal hyphae with

very thick walls to almost solid, but usually with a wider lumen at the apex, finely encrusted, 5.0–8.0 µm diam. Cystidioles fusoid to ventricose,  $9\text{--}11 \times 4\text{--}4.5$  µm. Basidia clavate, 4-sterigmate,  $10\text{--}12 \times 4\text{--}5$  µm; basidiospores ellipsoid, hyaline, smooth, thin-walled, usually with one or two oil drops,  $3.5\text{--}4.5 \times 2.0\text{--}3.0$  µm,  $Lm \times Wm = 4.02 \times 2.60$ ,  $Q = 1.33\text{--}1.80$ ,  $Qm = 1.55$ ,  $n = 30/1$ .

**Distribution** — Known only from southern Brazil, in Rio Grande do Sul State.

**Substratum** — Growing on dead branches and logs of unidentified angiosperms.

**Additional specimens examined.** BRAZIL, Rio Grande do Sul, São Francisco de Paula, CPCN Pró-Mata, 14 Nov. 2009, M.C. Westphalen 289/09 (ICN 154468); *ibid.*, 18 Apr. 2012, M.C. Westphalen 370/12 (SP 446173).

**Notes** — *Geesterania davidii* is characterised by yellowish basidiomes that become reddish when bruised and after drying, the medium to small pores and the small ellipsoid basidiospores. It differs from *G. carneola* in smaller and more regular pores, sometimes only partially split (Fig. 3c–d). In addition, *G. carneola* presents slightly longer and thinner, narrowly ellipsoid spores. Even though there are cases of overlapping in basidiospore size between the two species and only a detailed examination can reveal the differences, the values of  $Q$  and  $Qm$  show the difference in spore shape.

***Flaviporus tenuis*** Westphalen, Rajchenb. & Tomšovský, *nom. nov.* — MycoBank MB822514

≡ *Junghuhnia minuta* I. Lindblad & Ryvarden, Mycotaxon 71: 346. 1999, basionym.

**Etymology.** *tenuis* refers to the small and fragile pilei characteristic of this species.

**Specimens examined.** BRAZIL, Paraná, Piraquara, Morro do Canal, 4 Sept. 2013, M.C. Westphalen 470/13 (SP 446231); Santa Catarina, Florianópolis, UCAD, 12 Mar. 2012, M.C. Westphalen 356/12 (SP 446164); Rio Grande do Sul, São Francisco de Paula, FLONA, 30 Apr. 2012, M.C. Westphalen 386/12 (SP 446184); *ibid.*, 15 Apr. 2013, M.C. Westphalen 427/13 (SP 446207) and 430/13 (SP 446210); *ibid.*, CPCN Pró-Mata, 19 Apr. 2013, M.C. Westphalen 442/13 (SP 446218). — ECUADOR, Orellana, Yasuni National park, Yasuni Scientific Research Station, 9–12 Mar. 2002, L. Ryvarden 44687 (O).

**Notes** — Due to the presence of strongly agglutinated hyphae, *Junghuhnia minuta* morphologically fits the concept of *Flaviporus*, which is supported by our molecular data (Fig. 1). Since the name *Flaviporus minutus* is already taken by a different species (Wu et al. 2017), we propose the new name *Flaviporus tenuis*. This species can easily be recognised by the very small and brittle basidiomes growing in clusters, that become resinous-hard upon drying. Microscopically, it is very similar to *F. brownii*, presenting hyphal pegs and thick-walled encrusted cystidia. Macroscopically, *F. tenuis* is similar to *F. liebmanni*, presenting whitish basidiomes with darkening of the pileus surface upon drying. However, *F. liebmanni* differs in solitary to imbricate larger basidiomes and microscopically it lacks encrusted thick-walled cystidia. For a full description see Westphalen et al. (2012; as *Junghuhnia minuta*).

***Steccherinum meridionale*** (Rajchenb.) Westphalen, Tomšovský & Rajchenb., *comb. nov.* — MycoBank MB822515

≡ *Junghuhnia collabens* var. *meridionalis* Rajchenb., Sydowia 40: 236. 1987, basionym.

≡ *Junghuhnia meridionalis* (Rajchenb.) Rajchenb., Austral. Syst. Bot. 16 (4): 477. 2003.

**Specimens examined.** ARGENTINA, Chubut, P.N. Los Alerces, Río Arrayanes y Menéndez, 8 May 1991, M. Rajchenberg 11086 (CIEFAPcc 54); Neuquén, P.N. Lanín, Lago Lácar, Cascada Chachín, 19 May 1999, M. Rajchenberg 11924 (CIEFAPcc 284). — BRAZIL, Paraná, Morretes, Serra da Graciosa, 13 Oct. 2009, M.A. Reck 251/09 (ICN 154709); Rio Grande do Sul, São



Francisco de Paula, FLONA, 22 June 2009, M.C. Westphalen 238/09 (ICN 154290); *ibid.*, 26 Mar. 2010, M.C. Westphalen 295/10 (ICN 154659); *ibid.*, 26 Mar. 2010, M.C. Westphalen 303/10 (ICN 154660); Derrubadas, Parque Estadual do Turvo, 15 Sept. 2009, M.A. Reck 198/09 (ICN 154340); Santa Catarina, Santo Amaro da Imperatriz, Parque Estadual Serra do Tabuleiro, 18 Sept. 2010, M.A. Reck 559/10 (ICN 154719). — CHILE, Palena, Chaitén, ruta a Caleta Gonzalo, km 20, 6 Apr. 1996, M. Rajchenberg 10466 (CIEFAPcc 55); Valdivia, Corral, Thaxter, Aug. 1905 (BAFC 31012), holotype.

**Notes** — *Steccherinum meridionale* is characterised by cinnamon to brick red resupinate basidiomes with small pores (7–9 per mm) and waxy soft consistency when fresh, becoming hard upon drying. Microscopically, it presents abundant skeletocystidia projecting above the hymenium and sub-cylindrical basidiospores ( $3.0\text{--}4.0 \times 1.5\text{--}2.0\ \mu\text{m}$ ). This species was first described as a variety of *Junghuhnia collabens* (Rajchenberg 1987a) but later, using cultural features and intercompatibility tests, Rajchenberg (2003) verified it as an autonomous taxon. Our data shows that *S. meridionale* and *J. collabens* are not phylogenetically related (Fig. 1) and that the former groups close to the Neotropical species *S. neonitidum*. *Junghuhnia meridionalis* was described from the Patagonian Andes forests in southern Argentina and Chile and later registered from Neotropical regions (Westphalen et al. 2010, 2012). Unfortunately, the DNA amplification and sequencing of Brazilian specimens was unsuccessful, but we decided to keep the Neotropical specimens under the name *S. meridionale* and include it in the identification key. Further work can clarify if the Neotropical and Patagonian specimens belong to the same widely distributed species or if the Neotropical collections represent a morphologically similar but molecularly separated species. For a full description see Westphalen et al. (2012; as *J. meridionalis*).

***Steccherinum neonitidum* Westphalen & Tomšovský, sp. nov.**  
— MycoBank MB822516; Fig. 5, 6

**Holotype.** BRAZIL, Rio Grande do Sul, São Francisco de Paula, CPCN Pró-Mata, 18 Apr. 2012, M.C. Westphalen 371/12 (SP 446174).

**Etymology.** *Neo-* means new in Latin. The name refers to morphological similarity of the species to *Steccherinum nitidum* (Pers.) Vesterh. (syn. *Junghuhnia nitida*).

Basidiomes annual, resupinate, very thin, easily separable from the substrate and often detaching upon drying, soft when fresh, becoming corky and somewhat brittle when dried; margin irregular, white, narrow to wide, very thin, cottony and fimbriate, up to 5 mm wide; pore surface pinkish cream to very pale pinkish cinnamon; pores regular, round to angular, sometimes partially lacerate, 8–10 per mm; tubes up to 1.0 mm deep, concolour with the pore surface; subiculum white to cream-coloured, up to 0.5 mm



Fig. 5 *Steccherinum neonitidum*. — Scale bar = 2 cm. — Photo: M.A. Reck.

thick. Hyphal system dimitic; generative hyphae clamped, thin to slightly thick-walled, hyaline,  $2.0\text{--}3.5\ \mu\text{m}$  diam; skeletal hyphae unbranched to branched, thick-walled to solid, hyaline to yellowish,  $2.0\text{--}5.0\ \mu\text{m}$  diam. Skeletocystidia, abundant, clavate, thick-walled and heavily encrusted, metachromatic, immersed in the trama or projecting above the hymenium,  $6.0\text{--}10.0\ \mu\text{m}$  diam. Basidia clavate, 4-sterigmate; basidiospores ovoid to ellipsoid, smooth and hyaline,  $3.0\text{--}4.0 \times 2.0\text{--}3.0\ \mu\text{m}$ ,  $Lm \times Wm = 3.35 \times 2.53\ \mu\text{m}$ ,  $Q = 1.2\text{--}1.5$ ,  $Qm = 1.34$ ,  $n = 40/2$ .

**Mating system** — Tetrapolar. Ten monosporic cultures (SP 446174) were confronted and the mating types distributed as follows:  $A_1B_1$ : 1, 2, 4;  $A_2B_2$ : 6, 10;  $A_1B_2$ : 3, 5, 8, 9;  $A_2B_1$ : 7.

**Nuclear behaviour** — Normal (monosporic culture monokaryotic, polysporic culture dikaryotic).

**Distribution** — Southern and south-eastern Brazil.

**Substratum** — Growing on dead branches of unidentified angiosperms.

**Additional specimens examined.** BRAZIL, Rio Grande do Sul, Cambará do Sul, PARNA da Serra Geral, Itaimbezinho, R.M. Silveira & R.T. Guerrero 048, 19 Nov. 1987 (ICN); *ibid.*, Fortaleza, 29 Apr. 2012, M.C. Westphalen 380/12 (SP 446179); São Francisco de Paula, CPCN Pró-Mata, 30 May 2009, M.C. Westphalen 222/09 (ICN 154296); *ibid.*, FLONA, 22 June 2009, M.C. Westphalen 236/09 (ICN 154294) and M.C. Westphalen 241/09 (ICN 154295); São Paulo, São Luiz do Paraitinga, P.E. da Serra do Mar, Núcleo Sta. Virgínea, 12 June 2013, R.M. Pires & C.M. Ishida RP79 (SP 445975).

**Notes** — *Steccherinum neonitidum* is characterised by pinkish, thin resupinate basidiomes with small pores and fimbriate white margins. Microscopically, it can be recognised by the small ovoid to ellipsoid basidiospores and the abundant encrusted skeletocystidia. This species has been traditionally identified in Brazil as *J. nitida* due to its morphological similarities. However, *Steccherinum nitidum* (= *Junghuhnia nitida*) can be distinguished by larger pores (4–7 per mm; in margins of basidiomes pores can split to 3 per mm) and the somewhat longer basidiospores ( $3.7\text{--}4.5 \times 2.0\text{--}2.7\ \mu\text{m}$ ,  $Lm \times Wm = 4.26 \times 2.41\ \mu\text{m}$ ,  $Q = 1.65\text{--}1.88$ ,  $Qm = 1.77$ ,  $n = 187/9$ ). A careful analysis of herbarium specimens identified as *J. nitida* in other

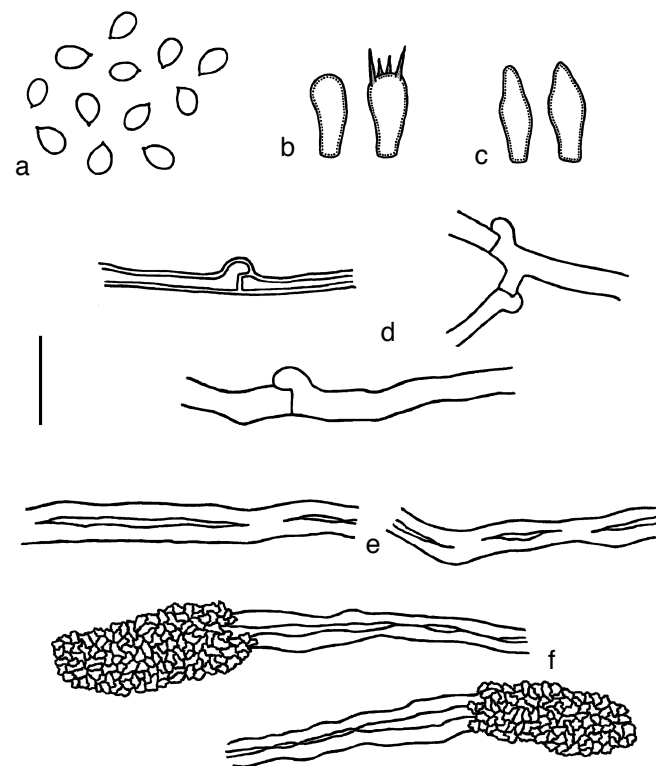


Fig. 6 *Steccherinum neonitidum*. a. Basidiospores; b. basidia; c. cystidiales; d. generative hyphae; e. skeletal hyphae; f. skeletocystidia. — Scale bar = 10  $\mu\text{m}$ . — Drawn by M.C. Westphalen.



Neotropical areas may greatly increase the distribution of this species and confirm if *S. nitidum* is restricted to temperate regions in the Northern Hemisphere, while *S. neonitidum* is distributed throughout Neotropical regions. *Steccherinum meridionale* is also morphologically similar, but can be separated by the darker, cinnamon to brick-red basidiomes and the sub-cylindrical basidiospores.

***Steccherinum polycystidiferum* (Rick) Westphalen, Tomšovský & Rajchenb., comb. nov.** — MycoBank MB822517

≡ *Poria polycystidifera* Rick, Iheringia, Bot. 7: 281. 1960, basionym.  
≡ *Junghuhnia polycystidifera* (Rick) Rajchenb., Nordic J. Bot. 7 (5): 566. 1987.

**Specimens examined.** BRAZIL, Rio Grande do Sul, Caxias do Sul, Terceira Léguas, 10 Dec. 2012, *M.C. Westphalen* 419/12 (SP 446201); São Salvador, 4 Apr. 1944, *J. Rick s.n.* (PACA FR 18603), holotype; Porto Alegre, Morro Santana, 14 Dec. 2007, *M.C. Westphalen* 070/07 (ICN 154122); Santa Catarina, Blumenau, Parque Nacional da Serra do Itajaí, 12 June 2012, *M.A.B. Silva et al.* 253 (FLOR 49252); São Paulo, São Luiz do Paraitinga, P.E. da Serra do Mar, Núcleo Sta. Virgínea, 29 Oct. 2013, *R.M. Pires* 140 (SP 446271).

**Notes** — *Steccherinum polycystidiferum* is characterised by cream-coloured to beige resupinate basidiomes, becoming slightly darker upon drying, small pores (6–8 per mm) and ellipsoid to sub-cylindrical small basidiospores (3.0–4.0 × 1.5–2.0 µm). *Steccherinum meridionale* presents very similar micromorphology and can be distinguished only by the basidiomes of cinnamon to brick-red colour. Despite the morphological similarities, *S. polycystidiferum* presents a phylogenetic isolate position, but close to the *S. nitidum* group. This seems to be a rare species, first described from northern Argentina as *Junghuhnia microspora* (Rajchenberg 1983), and later synonymised with *J. polycystidifera* (Rajchenberg 1987b), a species originally described from southern Brazil as *Poria polycystidifera* (Rick 1960).

***Steccherinum undigerum* (Berk & M.A. Curtis) Westphalen & Tomšovský, comb. nov.** — MycoBank MB822518

≡ *Polyporus undigerus* Berk. & M.A. Curtis, Bot. J. Linn. Soc. 10: 317. 1869, basionym.  
≡ *Junghuhnia undigera* (Berk. & M.A. Curtis) Ryvarden, Mycotaxon 20 (2): 359. 1984.  
= *Junghuhnia kotlabae* Pouzar, Czech Mycol. 55 (1–2): 2. 2003.  
= *Junghuhnia complicata* Blumenf. & J.E. Wright, Mycotaxon 19: 472. 1984.

**Mating system** — Tetrapolar. Two specimens were studied, each with nine monosporic cultures confronted, and the mating types distributed as follows:

(SP 446233)  $A_1B_1$ : 1, 2, 5, 8;  $A_2B_2$ : 4, 9;  $A_1B_2$ : 3, 6;  $A_2B_1$ : 7.  
(SP 446206)  $A_1B_1$ : 1, 2, 8;  $A_2B_2$ : 3;  $A_1B_2$ : 4, 6;  $A_2B_1$ : 5, 7, 9.

**Nuclear behaviour** — Normal (monosporic culture monokaryotic, polysporic culture dikaryotic).

**Specimens examined.** ARGENTINA, Misiones, Puerto Libertad, *ad culturam arborum coniferarum Alto Paraná*, S. Blumenfeld, 25 Mar. 1981, *ad Pinus taeda vivo* (BAFC 28947), holotype of *J. complicata*. — BRAZIL, Paraná, Curitiba, Parque Barigui, 26 Jan. 2014, *M.C. Westphalen* 485/14 (SP 446242); Rio Grande do Sul, São Francisco de Paula, CPCN Pró-Mata, 18 Apr. 2012, *M.C. Westphalen* 374/12 (SP 446176); *ibid.*, 19 Apr. 2013, *M.C. Westphalen* 443/13 (SP 446219); FLONA, 30 Apr. 2012, *M.C. Westphalen* 387/12 (SP 446185); Dom Pedro de Alcântara, Mata da Cova Funda, 14 May 2012, *M.C. Westphalen* 392/12 (SP 446189); Morrinhos do Sul, Perdida, 14 May 2012, *M.C. Westphalen* 395/12 (SP 446192); Caxias do Sul, Terceira Léguas, 10 Dec. 2012, *M.C. Westphalen* 418/12 (SP 446200); São Paulo, Santo André, P.E. do Alto da Serra de Paranapiacaba, 5 Dec. 2013, *M.C. Westphalen* 472/13 (SP 446233); São Luiz do Paraitinga, PE da Serra do Mar, Núcleo Santa Virgínea, Trilha Ipiranga, 7 Nov. 2012, *M.C. Westphalen* 406/12 (SP 446199); *ibid.*, 26 Apr. 2014, *M.C. Westphalen* & *R.M. Pires* 496/14 (SP 446249); *ibid.*, 27 Apr. 2014, *M.C. Westphalen* & *R.M. Pires* 501/14 (SP

446254); P.E. Fontes do Ipiranga, 20 May 2013, *M.C. Westphalen* 426/13 (SP 446206); *ibid.*, P.E. da Cantareira, 20 June 2013, *M.C. Westphalen* 446/13 (SP 446221). — CUBA, *sine loc.*, C. Wright 457 (K 181077), holotype; Pinar del Rio, Soroa near San Cristóbal, 13 Jan. 1967, *F. Kotlaba* (PRM 878650), holotype of *J. kotlabae*; Pinar del Rio, Soroa, 3 Feb. 1967, *F. Kotlaba* (PRM 870890), paratype of *J. kotlabae*.

**Notes** — *Steccherinum undigerum* is characterised by pileate, thin and flexible effused-reflexed or more rarely sessile basidiomes that become corky and somewhat brittle upon drying. Even though it presents a poroid hymenophore, the pores (4–6 per mm) present dentate dissepiments and are often split and somewhat irregular. Features of the pore surface is what distinguishes this species from others in the *Steccherinum* clade. Microscopically, it presents typical skeletocystidia of *Steccherinum*, more easily observed in the dissepiments edges but also found in the trama, a dimitic hyphal system and ellipsoid to subglobose basidiospores (4–5 × 3.5–4.5 µm), larger than other species in the group. For a full description see Westphalen et al. (2012; as *J. undigera*). Phylogenetically, *S. undigerum* is the only studied species that groups very close to *S. ochraceum* (Fig. 1, 2), type of the genus (for further comments see Discussion). Our materials present identical ITS and LSU sequences to a specimen from Cuba, type locality of this species, available at GenBank as *Junghuhnia* sp. 4. (Miettinen et al. 2012). *Steccherinum undigerum* is a very common species in southern Brazil and is often found in different areas of the Atlantic Rainforest.

*Junghuhnia kotlabae* is another species described from Cuba (Pouzar 2003) that presents morphological features almost identical to *J. undigera* and in its original description was only separated by the presence of thin-walled cystidia (absent in *J. undigera*). The basidiospore dimensions are 3.7–5.5 × 3–4.2 µm (Pouzar 2003) or 3.7–4.9 × 3–3.9 µm (our observation). In addition, the ITS sequence we obtained from the paratype of *J. kotlabae* presents only two different bases from specimens of *J. undigera* from Brazil. Since both the morphological and molecular similarities are high and cystidia present variation within a species, we consider *J. kotlabae* a synonym of *J. undigera*.

**Additional specimens examined.** *Flaviporus subundatus* — BRAZIL, Paraná, Paranaguá, Prox. a Rodovia PR 508, 2 Sept. 2013, *M.C. Westphalen* 457/13 (SP 446224); Curitiba, Parque Barigui, 26 Jan. 2014, *M.C. Westphalen* 483/14 (SP 446240); Rio Grande do Sul, São Francisco de Paula, FLONA, 16 Apr. 2012, *M.C. Westphalen* 367/12 (SP 446171); *ibid.*, 30 Apr. 2012, *M.C. Westphalen* 381/12 (SP 446180); *ibid.*, 15 Apr. 2013, *M.C. Westphalen* 445/13 (SP 446220); Santa Catarina, Itapoá, RPPN Volta Velha, 17 Nov. 2012, *M.C. Westphalen* 411/12 (FLOR); São Paulo, São Paulo, Parque Estadual da Cantareira, 27 June 2012, *M.C. Westphalen* 398/12 (SP 446195); P.E. da Serra do Mar, Núcleo Santa Virgínea, 27 Apr. 2014, *R.M. Pires* RP332 (SP 446276); Peruíbe, Estação Ecológica Juréia-Itatins, 25 Mar. 2014, *M.C. Westphalen* 494/14 (SP 446247). — CUBA, Guantánamo, Baracoa, El Yunque Mountain, Mar. 1903, *L.M. Underwood & F.S. Earle* 1168 (BPI 0243504), isotype. *Steccherinum nitidum* — AUSTRIA, Styria, Graz, Grazer Bergland, Schöckel Mountain, 31 Sept. 1975, *P. Döb- beler & J. Poelt* (PRM 801862). — CZECH REPUBLIC, Brno, Žebětín, 30 Oct. 2015, *M. Tomšovský* 52/2015 (BRNM 781249); Znojmo, Vranov nad Dyjí, Braitava nature monument, 29 Aug. 2012, *M. Tomšovský* 33/2012 (BRNM 781248). — GERMANY, Rhineland, Bonn, Bad Godesberg, Alnus glutinosa, 8 Feb. 1968, *H. Gorcholt* (PRM 690499). — HUNGARY, Tapolca, Lesence valley, *Carpinus betulus*, 15 Sept. 1978, *V. Holubová* (PRM 818395); Mátra Mts, Kékes Mt, *Fagus sylvatica*, 22 Sept. 1978, *J. Klán* (PRM 818365). — SLOVAKIA, Zvolen, Hriňová, *Fagus sylvatica*, 28 Aug. 1982, *F. Kotlaba*, (PRM 828459), Hajnáčka, Pohanský hrad nature reserve, *Quercus* sp., 22 July 1993, *J. Holec* (PRM 886250), Humenné, Svetlice, *Fagus sylvatica*, 23 Oct. 1987, *F. Kotlaba* (PRM 854505). *Junghuhnia chlamydospora* — BELIZE, Stan Creek Distr., Corkscomb basin wildlife sanctuary, 16 Nov. 2001, leg. *L. Ryvarden* 44241 (O), holotype. *Junghuhnia globospora* — VENEZUELA, Merida Prov., Monte Zerpa by Merida 2000 m, 29 Jan. 2001, *L. Ryvarden* 45503 (O), holotype. *Junghuhnia semisupiniformis* — MEXICO, Japala, 1909, *Murrill* (O), holotype. — PUERTO RICO, Rio Grande, Big tree trail, 18 June 1996, *L. Ryvarden* 38921 (O 911195).

**Key to Neotropical *Junghuhnia* s.lat. genera**

1. Basidiomes becoming reddish or blackish when bruised and/or after drying, cystidia appearing as elongated finely encrusted skeletal hyphae ends and sometimes hard to differentiate from them, skeletal hyphae metachromatic . . . . . *Geesterania*
1. Basidiomes not changing colour when bruised, sometimes darker after drying, cystidia easily distinguished from the skeletal hyphae, smooth or covered with large crystals, skeletal hyphae non metachromatic . . . . . 2
2. Basidiomes strongly shrinking when dried, resinous-hard, cystidia variable, thin to thick-walled, smooth or heavily encrusted, hyphae strongly agglutinated, hyphal system pseudo-dimitic . . . . . *Flaviporus*
2. Basidiomes with no remarkable shrinking when dried, corky to waxy, cystidia thick-walled and heavily encrusted, hyphae not agglutinated, hyphal system strictly dimitic . . . . . *Steccherinum*

**Key to Neotropical *Junghuhnia* s.lat. species**

1. Basidiomes strictly resupinate . . . . . 2
1. Basidiomes pileate, effused-reflexed to sessile . . . . . 9
2. Basidiomes changing colour when bruised or dried, cystidia finely encrusted, skeletal hyphae metachromatic . . . . . 3
2. Basidiomes with no remarkable colour change when bruised or dried, cystidia smooth or heavily encrusted . . . . . 5
3. Basidiomes becoming blackish in parts when dried, dextrinoid chlamydospores present . . . . . *J. chlamydospora*
3. Basidiomes becoming reddish when bruised and often when dried, dextrinoid chlamydospores absent . . . . . 4
4. Pore surface irregular, pores 3–5 per mm, strongly split, basidiospores narrowly ellipsoid . . . . . *G. carneola*
4. Pore surface regular, pores 5–8 per mm, entire or only slightly split, basidiospores ellipsoid . . . . . *G. davidii*
5. Generative hyphae simple-septate, hyphae strongly agglutinated, cystidia hard to observe and sometimes only large crystals seen in the trama . . . . . *F. subundatus*
5. Generative hyphae clamped, hyphae not agglutinated, cystidia easily observed . . . . . 6
6. Basidiomes cinnamon to brick-red or cream to straw coloured, tough and somewhat waxy, basidiospores sub-cylindrical . . . . . 7
6. Basidiomes white to cream or pinkish, thin, papery to corky, basidiospores globose to widely-ellipsoid . . . . . 8
7. Pore surface brick-red, pale cinnamon or dull red . . . . . *S. meridionale*
7. Pore surface cream to beige . . . . . *S. polycystidiferum*
8. Basidiomes pale pinkish cream to pale orange-pink, basidiospores broadly ellipsoid to ovoid ( $3.0\text{--}4.0 \times 2.0\text{--}3.0 \mu\text{m}$ ), cystidia heavily encrusted . . . . . *S. neonitidum*
8. Basidiomes white to cream, basidiospores globose to subglobose, cystidia smooth . . . . . *J. globospora*
9. Basidiomes resinous-hard when dried, hyphae strongly agglutinated and hard to observe . . . . . 10
9. Basidiomes corky to tough, hyphae not agglutinated and easily observed . . . . . 11
10. Basidiomes growing in clusters, usually dorsally attached, cystidia clavate, heavily encrusted . . . . . *F. tenuis*
10. Basidiomes solitary, cystidia elongated with a ventricose apex, smooth . . . . . *J. neotropica*

11. Basidiomes flexible when fresh becoming papery to corky upon drying, pores 5–6 per mm, basidiospores  $4.5\text{--}5.0 \times 3.5\text{--}4.5 \mu\text{m}$  . . . . . *S. undigerum*
11. Basidiomes tougher, somewhat dense when dried, pores 8–10 per mm, basidiospores  $2.5\text{--}3.5 \times 2.0\text{--}3.0 \mu\text{m}$  . . . . . *J. semisupiniformis*

**DISCUSSION**

Our results reveal that none of the studied Neotropical species previously placed in *Junghuhnia* are related to the type of the genus, *J. crustacea*. Instead, they are placed in different clades representing three genera:

***Steccherinum* clade**

Type. *Steccherinum ochraceum*.

*Steccherinum meridionale*, *S. neonitidum*, *S. polycystidiferum* and *S. undigerum* are part of this group. Even though *Steccherinum* is well-supported within the residual polyporoid clade (Miettinen et al. 2012, Binder et al. 2013), the *Steccherinum* clade is not yet resolved and more data are needed to better understand the evolutionary relations of its species. Under current knowledge, we decided to consider *Steccherinum* in a wide sense and to transfer the studied species into this genus, agreeing with Miettinen & Ryvarden (2016). The current wide delimitation of *Steccherinum* has a high phylogenetic support and can be defined morphologically by dimitic species with encrusted skeletocystidia and small subglobose to subcylindrical spores. Currently there are no morphological characters that would support the segregation of *Steccherinum* in different genera. However, further studies including more molecular and morphological data may elucidate if *Steccherinum* is a wide genus or if it encompasses several smaller genera. If the genus is further split, the Neotropical species could be transferred to *Chaetoporus*, a genus name available with *C. nitidus* (= *Junghuhnia nitida*) as type (Donk 1967, Miettinen et al. 2012). The only exception is *S. undigerum*, which groups close to *S. ochraceum*, type of *Steccherinum*.

***Flaviporus***

Type. *Flaviporus brownii*.

Our molecular data shows that *Junghuhnia minuta* and *J. subundata* group with *Flaviporus*, which is also supported by morphology. Species in *Flaviporus* are separated phylogenetically by rather long branches (Fig. 1), which may indicate that there are missing data that could clarify the relations among species of this group. Morphologically, *Flaviporus* species present basidiomes that become very hard and somewhat resinous after drying, cystidia with variable shape and strongly agglutinated hyphae, which support the genus as monophyletic. The hyphal system in *Flaviporus* is difficult to interpret due to the hyphae agglutination, especially in dried specimens. Our observations of *F. tenuis* and *F. brownii* treated in NaOH solution showed that many thick-walled hyphae present clamps, but some long aseptate segments can also be observed. In addition, there is no clear distinction between the hyphae present, so we classify the hyphal system as monomitic to pseudo-dimitic instead of strictly dimitic, as defined by Ginns (1980). *Flaviporus subundatus* differs by lacking clamp connections in the generative hyphae, but our molecular data support keeping this species under *Flaviporus*. Future studies including a wider sampling may clarify if the presence of clamp connections is variable in the genus or if *F. subundatus* belongs to a separate lineage. For full descriptions of *F. subundatus* see Lowe (1966), Ginns (1980) and Westphalen & Silveira (2012).

## Geesterania

Type. *Geesterania carneola*.

This third group recognised in this study comprises two species previously identified as *Junghuhnia carneola* that belong to the phlebiod clade. The phylogenetic position of these species is supported by biological data, since *J. carneola* was characterised by having a bipolar mating system and an astatoenocytic nuclear behaviour (David & Rajchenberg 1985), a combination typical of the phlebiod species, while species of *Steccherinum* are tetrapolar and present a normal nuclear behaviour (Rajchenberg 2011). Morphological and molecular data obtained support the description of a new genus and a new species. *Geesterania* is unique among polypores by the combination of basidiomes changing colour when bruised or after drying, the presence of long, finely encrusted cystidia and by the dimitic hyphal system with clamped generative hyphae and metachromatic skeletal hyphae. The two species identified in the genus are very similar but can be distinguished by the pore surface (larger, irregular and strongly split pores in *G. carneola* and smaller regular pores in *G. davidii*) and by a small difference in spore shape (Fig. 4).

The data presented in this study expand the knowledge of *Junghuhnia* s.lat. in the Neotropics and are in agreement with Miettinen et al. (2012), showing that the genus is highly polyphyletic and currently only *J. crustacea* has been confirmed as *Junghuhnia* s.str. During a survey of polypores in southern Brazil, seven species considered morphologically to belong to *Junghuhnia* s.lat. were collected and studied. The data obtained support the description of the new genus *Geesterania*, including one new species, as well as a new *Steccherinum* species and five new combinations of species previously treated in *Junghuhnia*. We present here full descriptions and illustrations of the new species, comments on the newly combined species and identification keys. However, more studies are required to clarify the evolutionary relationships of species previously treated as *Junghuhnia*, especially the ones included in the *Steccherinum* clade. Furthermore, the phylogenetic positions of *J. semisupiniformis*, *J. chlamydospora*, *J. globospora* and *J. neotropica* are still unknown. Here we segregated these four species by morphological groups and presented them in the identification key, but molecular data are needed to verify their proper generic position. Ryvarden (2015) also included *J. sobria* in his study of Neotropical *Junghuhnia* taxa. *Junghuhnia sobria* was originally characterised by the presence of gloecystidia (Lowe 1977), a character absent in all other species of the group, therefore we consider to exclude it from *Junghuhnia* s.lat.

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## REFERENCES

- Binder M, Justo A, Riley R, et al. 2013. Phylogenetic and phylogenomic overview of the Polyporales. *Mycologia* 105: 1350–1373. doi: <https://doi.org/10.3852/13-003>.
- Boidin J. 1958. Essai biotaxonomique sur les Hydnés résupinés et les Corticiés. *Revue de Mycologie. Mémoire Hors Série* 6: 1–387.
- Boidin J. 1971. Nuclear behavior in the mycelium and the evolution of Basidiomycetes. In: Petersen RH (ed), *Evolution in the higher Basidiomycetes*: 129–148. Knoxville, The University of Tennessee Press.
- Bresadola G. 1896. *Fungi Brasilienses lecti a cl. Dr. Alfredo Möller*. *Hedwigia* 35: 276–302.
- Carranza-Velásquez J, Ruiz-Boyer A. 2005. Checklist of polypores of Costa Rica. *Revista Mexicana de Micología* 20: 45–52.
- Chen JJ, Cui BK. 2014. *Phlebiporia bubalina* gen. et. sp. nov. (Meruliaceae, Polyporales) from Southwest China with a preliminary phylogeny based on rDNA sequences. *Mycological Progress* 13: 563–573. doi: <https://doi.org/10.1007/s11557-013-0940-4>.
- David A, Rajchenberg M. 1985. Pore fungi from French Antilles and Guiana. *Mycotaxon* 22: 285–325.
- Darriba D, Taboada GL, Doallo R, et al. 2012. jModelTest 2: more models, new heuristics and parallel computing. *Nature Methods* 9 (8): 772. doi: <https://doi.org/10.1038/nmeth.2109>.
- Donk MA. 1967. Notes on European polypores – II. *Persoonia* 5 (1): 47–130.
- Floudas D, Hibbett DS. 2015. Revisiting the taxonomy of Phanerochaete (Polyporales, Basidiomycota) using a four gene dataset and extensive ITS sampling. *Fungal Biology* 119: 679–719. doi: <https://doi.org/10.1016/j.funbio.2015.04.003>.
- Ginns J. 1980. The genus *Flaviporus* Murrill (Polyporaceae). *Canadian Journal of Botany* 58: 1578–1590.
- Hall TA. 1999. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series* 41: 95–98.
- Hallenberg N. 1984. Compatibility between species of Corticiaceae s.l. (Basidiomycetes) from Europe and North America. *Mycotaxon* 21: 335–388.
- Lindblad I, Ryvarden L. 1999. Studies in neotropical polypores. 3. New and interesting Basidiomycetes (Poriales) from Costa Rica. *Mycotaxon* 71: 335–359.
- Lowe JL. 1966. Polyporaceae of North America. The genus *Poria*. State University College of Forestry at Syracuse University, Technical Publication n° 90.
- Lowe JL. 1977. On *Polyporus sobrius*. *Kew Bulletin* 31 (3): 753–754.
- Miettinen O, Larsson E, Sjökvist E, et al. 2012. Comprehensive taxon sampling reveals unaccounted diversity and morphological plasticity in a group of dimitic polypores (Basidiomycota, Polyporales). *Cladistics* 28: 251–270. doi: <https://doi.org/10.1111/j.1096-0031.2011.00380.x>.
- Miettinen O, Ryvarden L. 2016. Polypore genera *Antella*, *Austeria*, *Butyrea*, *Citripora*, *Metuloidea* and *Trulla* (Steccheriaceae, Polyporales). *Annales Botanici Fennici* 53: 157–172.
- Miller MA, Pfeiffer W, Schwartz T. 2010. Creating the CIPRES Science Gateway for inference of large phylogenetic trees. *Proceedings of the Gateway Computing Environments Workshop (GCE)*, 14 Nov. 2010, New Orleans, LA: 1–8.
- Nikolcheva LG, Bärlocher F. 2004. Taxon-specific primers reveal unexpectedly high diversity during leaf decomposition in a stream. *Mycological Progress* 3: 41–49. doi: <https://doi.org/10.1007/s11557-006-0075-y>.
- Pouzar Z. 2003. A new polypore from Cuba: *Junghuhnia kotlabae*. *Czech Mycology* 55 (1–2): 1–6.
- Rajchenberg M. 1983. New South American resupinate polypores. *Mycotaxon* 16: 500–506.
- Rajchenberg M. 1987a. Xylophilous Aphyllophorales (Basidiomycetes) from the southern Andean forests. Additions and corrections. II. *Sydowia* 40: 235–249.
- Rajchenberg M. 1987b. Type studies of Polyporaceae (Aphyllophorales) described by J. Rick. *Nordic Journal of Botany* 7: 553–568.
- Rajchenberg M. 2003. Taxonomic studies on selected Austral polypores. *Australian Systematic Botany* 16: 473–485.
- Rajchenberg M. 2011. Nuclear behavior of the mycelium and the phylogeny of Polypores (Basidiomycota). *Mycologia* 103 (4): 677–702. doi: <https://doi.org/10.3852/10-310>.
- Rehner SA, Buckley EP. 2005. A *Beauveria* phylogeny inferred from nuclear ITS and EF1- $\alpha$  sequences: evidence for cryptic diversification and links to *Cordyceps* teleomorphs. *Mycologia* 97: 84–98.
- Rick J. 1960. Basidiomycetes Eubasidii in Rio Grande do Sul – Brasília 4. Meruliaceae, Polyporaceae e Boletaceae. *Iheringia Série Botânica* 7: 193–295.
- Robledo GL, Rajchenberg M. 2007. South American polypores: first annotated checklist from Argentinean Yungas. *Mycotaxon* 100: 5–9.
- Ronquist F, Teslenko M, Van der Mark P, et al. 2012. MrBayes 3.3: efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology* 61: 539–542.
- Ryvarden L. 1985. Type studies in the Polyporaceae 17. Species described by W.A. Murrill. *Mycotaxon* 23: 169–198.
- Ryvarden L. 1991. Genera of polypores: Nomenclature and taxonomy. *Synopsis Fungorum* 5: 1–363.
- Ryvarden L. 2007. Studies in Neotropical polypores 23. New and interesting wood-inhabiting fungi from Belize. *Synopsis Fungorum* 23: 32–50.
- Ryvarden L. 2015. Neotropical polypores Part 2, Polyporaceae, Abortiporus – Nigroporus. *Synopsis Fungorum* 34: 232–443.



- Stamatakis A. 2014. RAxML Version 8: A tool for Phylogenetic Analysis and Post-Analysis of Large Phylogenies. *Bioinformatics* 30 (9): 1312–1313. doi: <https://doi.org/10.1093/bioinformatics/btu033>.
- Thiers B. 2016. Index Herbariorum: a global directory of public herbaria and associated staff. New York Botanical Garden's Virtual Herbarium. <http://sweetgum.nybg.org/ih/>.
- Tomšovský M, Menkis A, Vasaitis R. 2010a. Phylogenetic relationships in European Ceriporiopsis species inferred from nuclear and mitochondrial ribosomal DNA sequences. *Fungal Biology* 114: 350–358. doi: <https://doi.org/10.1016/j.funbio.2010.02.004>.
- Tomšovský M, Sedlák P, Jankovský L. 2010b. Species recognition and phylogenetic relationships of European Porodaedalea (Basidiomycota, Hymenochaetales). *Mycological Progress* 9 (2): 225–233. doi: <https://doi.org/10.1007/s11557-009-0628-y>.
- Westphalen MC, Reck MA, Silveira RMB. 2010. Ganoderma chalceum and Junghuhnia meridionalis: new records from Brazil. *Mycotaxon* 111: 11–18.
- Westphalen MC, Reck MA, Silveira RMB. 2012. The genus Junghuhnia in Brazil. *Nova Hedwigia* 94: 209–220. doi: <https://doi.org/10.1127/0029-5035/2012/0094-0209>.
- Westphalen MC, Silveira RMB. 2012. Resupinate polypores from mixed ombrophilous forests in southern Brazil. *Mycotaxon* 122: 111–122.
- Wu F, Chen JJ, Ji XH, et al. 2017. Phylogeny and diversity of the morphologically similar polypore genera Rigidoporus, Physisporinus, Oxyporus and Leucophellinus. *Mycologia* 109 (5): 749–765. doi: <https://doi.org/10.1080/00275514.2017.1405215>.
- Wu F, Yuan Y, Chen JJ, et al. 2016. Luteoporia albomarginata gen. et sp. nov. (Meruliaceae, Basidiomycota) from tropical China. *Phytotaxa* 263 (1): 31–41. doi: <https://doi.org/10.11646/phytotaxa.263.1.3>.