Lei Cai · Kevin D. Hyde

New species of *Clohiesia* and *Paraniesslia* collected from freshwater habitats in China

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Abstract Two fungi collected from submerged woody debris were found to represent hitherto undescribed species of the ascomycete genera *Clohiesia* and *Paraniesslia*. They are described as Clohiesia curvispora sp. nov. and Paraniesslia aquatica sp. nov. based on morphological characters. Clohiesia curvispora is characterized by immersed ascomata under a clypeus, and unitunicate, cylindrical asci containing one-celled, curved, elongate-fusiform ascospores. Paraniesslia aquatica is characterized by small, superficial, setose ascomata, and unitunicate, clavate asci containing verrucose, brown ascospores. Each species is illustrated with light micrographs and compared with similar taxa in this article.

Key words Ascomycetes · Freshwater fungi · Lignicolous fungi · Taxonomy

Submerged woody substrata are essential components of freshwater ecosystems (Jacobson et al. 1999). Diverse taxonomic groups of fungi colonize and grow on submerged wood (Tsui and Hyde 2003). Wood-inhabiting fungi in the freshwater environment have received less attention in mainland China, as compared to those in terrestrial environment. Our study of fungal biodiversity in streams in Yunnan of mainland China (Cai et al. 2005, 2006; Zhu et al. 2005) yielded two interesting teleomorphic ascomycetes. Examination of their morphological characteristics showed that they are new species of Clohiesia K.D. Hyde and Paraniesslia K.M. Tsui, K.D. Hyde & Hodgkiss. They are therefore, described, illustrated, and compared to similar taxa.

e-mail: mrcailei@gmail.com

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In this study, samples were processed and examined following the methods described in Cai et al. (2006). Single spore isolations were made on cornmeal agar (CMA) (Choi et al. 1999). Cultures are deposited in HKUCC. Type specimens are deposited in HKU (M). Observations and photographs were made from materials mounted in water. The range between minimum and maximum values for microscopic measurements is given. Mean values are in brackets, with "n" being the number of items measured.

Clohiesia curvispora L. Cai & K.D. Hyde sp. nov. Figs. 1–7

Ascomata 400-600 µm diametro, 400-500 µm alta, globosa vel subglobosa, superficialia vel subimmersa, carbonacea, atro-brunnea vel nigra, papillata, ostiolata, clypeata, gregaria. Peridium 30-50µm crassum, brunneum vel nigrum. Paraphyses 3–4 µm crassae, filamentosae, numerosae, septatae, ramosae, hyalinae. Asci $125-175 \times 8.5-10.5 \,\mu\text{m}$, octospori, unitunicati, cylindrici, pedicellati, tenuitunicati, apice rotundati, apparatu apicali praediti. Ascosporae $32.5-39 \times$ 3.2-5 µm, 1- vel 2-seriatae, unicellulares, hyalinae, guttulatae, curvatim fusiformes, tunico gelatinoso praeditae.

Etymology: Referring to the curved ascospores.

Ascomata 400-600 µm in diameter, 400-500 µm high, perithecioid, globose to subglobose, superficial to partly immersed, carbonaceous, dark brown to black, papillate, ostiolate, clypeate, gregarious. Clypeus light-colored, disclike, thin, surrounding the ostiole, comprising host cells filled with irregular brown hyphae. Ostiole central, short, brown, composed of dark brown pseudoparenchyma of textura angularis, periphysate. Peridium 30-50 µm thick, brown-black, composed of light brown oblong cells. Paraphyses 3–4 µm wide at base, longer than asci, hypha-like, filamentous, numerous, septate, branched, hyaline, tapering distally, embedded in a gelatinous matrix. Asci 125–175 \times $8.5-10.5 \,\mu m \,(\bar{x} = 152 \times 9.6 \,\mu m, n = 10), 8$ -spored, unitunicate, cylindrical, pedicellate, thin-walled, apically rounded, with a nonamyloid, refractive, apical apparatus (ca. 1.5µm long, 2.5–3µm in diameter). Ascospores $32.5-39 \times 3.2-5$ µm $(\bar{x} = 35.5 \times 4 \mu m, n = 25)$, overlapping uniseriate to biseriate,

L. Cai $(\boxtimes)^1 \cdot K.D.$ Hyde

Centre for Research in Fungal Diversity, Department of Ecology and Biodiversity, The University of Hong Kong, Hong Kong SAR, China

¹Present address: Novozymes China, No. 14, XinXi Road, ShangDi Zone, HaiDian District, Beijing 100085, China Tel. +86-10-6298-7888, ext. 306; Fax +86-10-6298-0085

Figs. 1–7. *Clohiesia curvispora*, from holotype. **1** Section of ascoma. **2** Section of the peridium. **3** Paraphyses. **4**, **5** Asci. Note the apical ring. **6**, **7** Ascospores. *Bars* **1** 30μm; **2** 15μm; **3–7** 10μm



Figs. 8–11. Paraniesslia aquatica, from holotype. 8 Section of ascoma. 9 Section of peridium.
10 Asci. 11 Ascospores. Bars 8 30μm; 9 25μm; 10, 11 10μm



Colonies on CMA slow growing, up to 2 cm diameter after 2 months at room temperature (22°–25°C), superficial, woolly, with a few aerial hyphae, pale brown from above, reverse dark brown in the center, white toward the margin, growing in concentric rings (cultures studied in HKUCC 9182).

Habitat: Saprobic on submerged decaying wood.

Anamorph: Unknown.

Material examined: China, Yunnan, Jinghong, on submerged wood in a small forest stream, Sept. 15, 2002, L. Cai, CAI-9BNA38 (holotype: HKU(M) 10854).

Notes: *Clohiesia* K.D. Hyde was established to accommodate the freshwater taxon *C. corticola* from tropical Australia (Hyde 1995). *Clohiesia* had been placed in Annulatascaceae (Sordariales) owing to its relatively massive J-apical ring (Wong et al. 1998). Recent molecular studies, however, have revealed that the family Annulatascaceae is polyphyletic (Campbell and Shearer 2004; Huhndorf et al. 2004; Vijaykrishna et al. 2005). In addition, *Clohiesia* was shown to have phylogenetic affinity with Lasiosphaeriaceae and Sordariaceae instead of other annulatascaceous species (Raja et al. 2003; Duong et al. 2004). Currently, *Clohiesia* is placed in Sordariales incertae sedis (Kirk et al. 2001).

Our species should be classified in the genus Clohiesia because it has immersed ascomata under a clypeus, cylindrical-clavate asci with a refractive apical ring, and fusiform, hyaline, aseptate ascospores, characteristics of Clohiesia (Hyde 1995). There are currently two species in *Clohiesia*, C. corticola K.D. Hyde and C. lignicola K.M. Tsui, K.D. Hyde & Hodgkiss (Hyde 1995; Tsui et al. 1998). Clohiesia curvispora is different from C. corticola, the type species of the genus, in its larger ascospores $(32.5-39 \times 3.2-5 \mu m v s)$. $18.5-26.5 \times 3.7-5 \mu m$), which are distinctly curved. Moreover, C. curvispora differs from C. corticola in its larger ascomata and asci and the thicker peridium. Clohiesia lignicola, however, differs from C. curvispora in the smaller ascospores $(14-28 \times 4.5-6 \mu m)$, which are fusiform-ellipsoid rather than the curved elongate-fusiform ascospores in C. curvispora (Tsui et al. 1998).

Paraniesslia aquatica L. Cai & K.D. Hye, sp. nov.

Figs. 8-11

Ascomata 160–200 μ m diametro, 190–240 μ m alta, superficialia, pyriformia vel subglobosa, membranacea, brunnea vel nigra, papillata, ostiolata, setosa, solitaria vel gregaria. Setae acutae, rectae, non ramosae, septatae, nigrae. Peridium 15–30 μ m, a externe visus *textura epidermoidea*, in sectione longitudinali cellulis 3–4-stratpsis *textura angularis* compositum. Paraphyses septatae, tenuitunicatae, deliquescentes. Asci 68–95 × 9–16 μ m, octospori, clavati, pedicellati, unitunicati, tenuitunicati, apice truncati, apparatu apicali praediti. Ascosporae 15–19 × 6–7.5 μ m, imbricato-uniseriatae vel biseriatae, ellipsoideae, 1-septatae, ad septum constrictae, verrucosae, subhyalinae, tunico gelatinoso praeditae.

Etymology: Referring to its aquatic habitat.

Ascomata 160–200 μ m wide, 190–240 μ m high, perithecioid, superficial, pyriform to subglobose, membranous, brown or black, papillate, ostiolate, solitary to gregarious, with setae. Setae acute, straight, unbranched, septate, black. Peridium 15–30 μ m, textura epidermoidea in surface view, textura angularis in longitudinal section, composed of 3–4 layers of compressed polygonal cells. *Paraphyses* septate, thin-walled, deliquescent. Asci 68–95 × 9–16 μ m ($\bar{x} = 78 \times 12 \mu$ m, n = 15), 8-spored, clavate, pedicellate, unitunicate, thin-walled, apex truncate, with a nonamyloid discoid refractive apical ring. Ascospores 15–19 × 6–7.5 μ m ($\bar{x} = 17.5 \times 7 \mu$ m, n = 25), overlapping uniseriate to biseriate, ellipsoidal, 1-septate, slightly constricted at the septum, verrucose, subhyaline, with a thin mucilaginous sheath. No cultures obtained.

Habitat: Saprobic on wood submerged in freshwater. Anamorph: Unknown.

Material examined: China, Yunnan, Kunming, Qinglongxia, submerged wood in a small stream, Nov. 10, 2002, L. Cai, CAI-11QL34 (holotype: HKU(M) 10856). Paratype, ibid, HKU(M) 10855.

Notes: Paraniesslia (Niessliaceae, Hypocreales) K.M. Tsui, K.D. Hyde & Hodgkiss was recently established to accommodate a fungus collected from Hong Kong freshwater habitats (Tsui et al. 2001). The genus is characterized by small, setose, perithecioid ascomata, unitunicate asci with nonamyloid, discoid, refractive apical ring, and uniseptate, verrucose, brown ascospores. Paraniesslia is also distinct in having deliquescing interascal filaments instead of true paraphyses, characteristics of Hypocreales. Our species is classified in Paraniesslia because it fits well with the generic concept of Paraniesslia (Tsui et al. 2001). There is currently only one species, P. tuberculata K.M. Tsui, K.D. Hyde & Hodgkiss, in this genus. Paraniesslia aquatica can be distinguished from the type species in having larger ascomata $(160-200 \times 190-240 \,\mu m \text{ vs. } 100-120 \times 100-130 \,\mu m)$, asci $(68-95 \times 9-16 \mu m \text{ vs. } 50-75 \times 7-14 \mu m)$, and ascospores $(15-19 \times 6-7.5 \mu m \text{ vs. } 11-14 \times 4-6.5 \mu m)$, and ascospores that are subhyaline at maturity rather than dark brown in P. tuberculate (Tsui et al. 2001).

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References

- Cai L, Zhang KQ, Hyde KD (2005) Ascoyunnania aquatica gen. et sp. nov., a freshwater fungus collected from China and its microcylic conidiation. Fungal Divers 18:1–8
- Cai L, Ji KF, Hyde KD (2006) Variation between freshwater and terrestrial fungal communities on decaying bamboo culms. Antonie Leeuwenhoek 89:293–301
- Campbell J, Shearer CA (2004) Annulusmagnus and Ascitendus, two new genera in the Annulatascaceae. Mycologia 96:822–833
- Choi YW, Hyde KD, Ho WH (1999) Single spore isolation of fungi. Fungal Divers 3:29–38
- Duong LM, Lumyong S, Hyde KD, Jeewon R (2004) Emarcea castanopsidicola gen. et sp. nov. from Thailand, a new xylariaceous taxon based on morphology and DNA sequences. Stud Mycol 50:253–260

- Huhndorf SM, Miller AN, Fernandez FA (2004) Molecular systematics of the Sordariales: the order and the family Lasiosphaeriaceae redefined. Mycologia 96:368–387
- Hyde KD (1995) Tropical Australian freshwater fungi. VII. New genera and species of Ascomycetes. Nova Hedwigia 61:119–140
- Jacobson PJ, Jacobson KM, Angermeier PL, Cherry DS (1999) Transport, retention, and ecological significance of woody debris within a large ephemeral river. J N Am Benthol Soc 18:429–444
- Kirk PM, Cannon PF, David JC, Stalpers JA (2001) Ainsworth & Bisby's dictionary of the Fungi, 9th edn. CAB International, Wallingford, UK
- Raja HA, Campbell J, Shearer CA (2003) Freshwater ascomycetes: *Cyanoannulus petersenii*, a new genus and species from submerged wood. Mycotaxon 88:1–17
- Tsui CKM, Hyde KD (2003) Freshwater mycology. Fungal diversity research series 10. Fungal Diversity Press, The University of Hong Kong, Hong Kong

- Tsui CKM, Hyde KD, Hodgkiss IJ (1998) A new species of *Clohiesia* from Hong Kong. Mycoscience 39:257–259
- Tsui CKM, Hyde KD, Hodgkiss IJ (2001) *Paraniesslia tuberculata* gen. et sp. nov., and new records or species of *Clypeosphaeria*, *Lepto-sphaeria* and *Astrosphaeriella* in Hong Kong freshwater habitats. Mycologia 93:1002–1009
- Vijaykrishna D, Jeewon R, Hyde KD (2005) *Fusoidispora aquatica*: a new freshwater ascomycete from Hong Kong based on morphology and phylogeny inferred from rDNA gene sequences. Sydowia 57:267–280
- Wong SW, Hyde KD, Jones EBG (1998) Annulatascaceae, a new ascomycete family from the tropics. Syst Ascomycetum 16:17–25
- Zhu H, Cai L, Hyde KD, Zhang KQ (2005) A new species of *Acrogenospora* from submerged bamboo in Yunnan, China. Mycotaxon 92:383–386