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Two new keratinophilic fungal species

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Abstract

Two new keratinophilic fungal species, *Chrysosporium jingzhouense* and *C. clavisporum*, were isolated from farmland soil of Jingzhou City, Hubei Province, and from plant root soil of Guigang City, Guangxi Province, China, respectively, using child hair as bait. Molecular and morphological characters were used to identify taxonomic status. The phylogeny shows *C. jingzhouense* is clustered with *C. articulatum* and *C. keratinophilum*; while *C. clavisporum* appears to be related to *C. qinghaiense*, *C. indicum* and *C. linfenense*. These previously described *Chrysosporium* species can be easily distinguished from our proposed new species based on morphology alone. The holotypes, ex-types and ex-isotypes have been deposited in the Institute of Fungus Resources (GZAC).

Keywords: filamentous fungi, morphological character, phylogeny, identification

Introduction

Keratinophilic fungal species are distributed worldwide, and include many natural colonizers of keratinic substrates, particularly where human and animal populations exert strong selective pressure on the environment (Marchisio *et al*. 1991). These fungi secrete many useful enzymes, including keratinase, cellulase and amylase, and also produce active compounds with anti-cancer, anti-fungal and anti-parasitic properties (Marchisio 2000). Moreover, these fungi show a wide tolerance toward several ecological and environmental factors, including pH, temperature, and elevation (McAleer 1980). Subsequently, keratinophilic fungi have been used in biotechnology for the disposal of refuse, and for the production of animal foods, fertilizers, glues and rare amino acids, from poultry farm and tannery wastes (Marchisio 2000). Keratinophilic fungi are also involved in recycling carbon, nitrogen, and sulfur from α-keratins in natural environments, and play an important role in ecological systems (Marchisio 2000). However, keratinophiles have also been considered potential pathogens, and have attracted the attention of dermatologists and mycologists due to associations with human and animal mycoses (De Hoog *et al.* 2000, Gan *et al.* 2002, Anstead *et al.* 2012, Sigler *et al.* 2013). Pathogenic keratinophilic fungi isolated have predominantly been *Microsporum* Gruby, *Trichophyton* Malmsten, *Aphanoascus* Zukal, *Chrysosporium* Corda, *Geomyces* Traaen and *Myceliophthora* Costantin genera (Kamath *et al*. 2015, Suankratay *et al*. 2015, Anbu *et al*. 2004).

Keratinophilic fungi have received increased attention in recent years; in particular their distribution in soil has been investigated at many sites around the world (Anbu *et al*. 2004, Zarrin & Haghgoo 2011, Shadzi *et al*. 2002). Hair, wool and/or nails are usually used as bait for the keratinophilic fungi or in keratinolysis assays (Marchisio 2000). Several mycologists began investigating keratinophilic fungi in China in 2010 from soil, sludge and feather samples, using child hair as bait, obtaining and reporting several previous and novel strains: *Geomyces guiyangensis* Y.F. Han, Y. Luo & Z.Q. Liang (Luo *et al*. 2016), *Chrysosporium qinghaiense* Y.F. Han *et al*. (Han *et al*. 2013), *C. sanyaense* Y.W. Zhang *et al*. (Zhang *et al*. 2013), *C. hubeiense* Y.W. Zhang, Y.F. Han & Z.Q. Liang (Zhang *et al*. 2016a), *C. guizhouense* Y.W. Zhang, Y.F. Han & Z.Q. Liang (Zhang *et al*. 2016a) and *C. shanxiense* Y.W. Zhang *et al*. Our current survey isolated keratinophilic fungi from Hubei and Guangxi Provinces, and identified two new species based on molecular and morphological analyses. This paper provides a phylogenetic tree, descriptions, and illustrations of the novel species.

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Materials and Methods

Sample collection and strain isolation

Two strains (EB1301M and EB1303M) were isolated from farmland soils of Jingzhou City in Hubei Province (N 30°21′27.37″, E 112°19′58.37″), and two others (G80.1 and G80.2) were isolated from plant root soils of Guigang City in Guangxi Province (N 23°18′54.75″, E 109°48′17.78″). Soil samples were added to sterilized hair fragment about 10 mm in length and kept moist at 26 °C for approximately one month. When fungal growth was observed, the hair powder was mixed with sterilized water in an Erlenmeyer flask, and 1 mL suspensions were evenly spread on Martin's medium and incubated at 26 °C. Pure cultures were then isolated and transferred to potato dextrose agar (PDA) slants stored at −70 °C. The holotypes and ex-isotypes were deposited at the Institute of Fungus Resources, Guizhou University (GZAC) (Zhang *et al*. 2016a, b).

Morphological identification

Isolates were transferred to PDA and Czapek agar, incubated at 26 °C for 14 days, and subjected to macroscopic examination. Fungal micro-characteristics were examined with a Motic microscope (Guangzhou, Motic Co., China) and photographed. Diagnostic features were then defined based on these observations. Finally, the fungi were morphologically identified according to colony characteristics and conidiogenous structures (van Oorschot 1980, Han *et al*. 2013, Zhang *et al*. 2013, 2016a, b).

DNA extraction, PCR amplification and nucleotide sequencing

Total genomic DNA was extracted from fresh sporulating cultures after seven days at 26 °C using a Fungal DNA Mini Kit (Omega Biotech, Doraville, GA, USA) according to the manufacturer's protocol and then stored at –20 °C. The ITS-5.8S rDNA region was amplified with primers ITS5 (5′-GGTGAGAGATTTCTGTGC-3′) and ITS4 (5′-TCCTCCGCTTAT TGA TATGC-3′) (Zhang *et al*. 2016a). Amplifications were carried out in 25 μL volumes, with 12.5 μm 2× Master Mix, 2 μm template DNA, 1 μm primer ITS4, 1 μm primer ITS5, and 8.5 μm ddH_2O. Amplification conditions were: 5 mins at 94 °C, then 35 cycles of 94 °C for 40 s, 50 °C for 40 s, 72 °C for 1 min, and finally 10 min at 72 °C. The resulting PCR products were sequenced by Sangon Biotech (Shanghai, China) using the same primers. The generated ITS-5.8S rDNA sequences were submitted to GenBank (KY026599 to KY026602).

TABLE 1. Species list for the phylogeny analysis and the information of ITS-5.8S rDNA.

Species names	Strain No.	GenBank No.	Species names	Strain No.	GenBank No.
Chrysosporium articulatum	UAMH 4320	AJ007841	*C. merdarium*	CBS 408.72	AJ390384
C. carmichaelii	CBS 643.79	AJ007842	*C. minutisporosum*	IMI 379912	AJ131689
**C. clavisporum*	G80.1	KY026601	*C. oceanitesii*	MR11771	HG329729
**C. clavisporum*	G80.2	KY026602	*C. pilosum*	IMI 356294	AJ390385
C. europae	UAMH 4587	AJ007843	*C. pseudomerdarium*	CBS 631.79	AJ390386
C. evolceanui	RV26475	AJ005368	*C. qinghaiense*	GZUIFR-11	JX868607
C. filiforme	CBS 187.82	AJ131680	*C. queenslandicum*	IFM 51121	AB219228
C. fluviale	FMR 6005	AJ005367	*C. sanyaense*	GZUIFR-A10222M	JQ809269
C. georgii	CBS 272.66	AJ007844	*C. shanxiense*	GZUIFR-EB1601M.1	KX462168
C. guarroi	CCFVB CH10	EU018451	*C. shanxiense*	GZUIFR-EB1601M.2	KX462169
C. guarroi	CCFVB CH11	EU018452	*C. shanxiense*	GZUIFR-EB1601M.3	KX462130
C. guizhouense	EM14.2002	KT948765	*C. siglerae*	UAMH 6541	AJ131684
C. hubeiense	EM66601	KJ849227	*C. speluncarum*	CCF3761	AM949569
C. indicum	GZUIFR-3-4	HQ685965	*C. speluncarum*	CCF3760	NR_137537
**C. jingzhouense*	EB1301M	KY026599	*C. submersum*	IMI 379911	AJ131686
**C. jingzhouense*	EB1303M	KY026600	*C. sulfureum*	CBS 634.79	AJ390387
C. keratinophilum	IFO 7584	AJ131681	*C. tropicum*	UAMH 691	NR_131267
C. linfenense	GZUIFR-H31	FJ392561	*C. undulatum*	IMI 375884	AJ007845
C. luchnowense	IMI 112798	AJ131682	*C. vallenarense*	CBS 627.83	AJ390389
C. longisporum	UTHSCR4380	HF547873	*C. vespertilium*	RV 27093	AJ007846
C. lobatum	CBS 666.78	AJ131688	*C. zonatum*	IFM 51122	AB219229
C. magnasporum	FMR11770	HG329727	*Myceliophthora thermophila*	H127-1	JX868606
C. mephiticum	CBS 320.86	AJ131683			

Note: * Strains sequenced in the study.

Molecular phylogenetic analysis

ITS-5.8S rDNA sequences of 40 *Chrysosporium* species were downloaded from GenBank. An ITS-5.8S rDNA sequence of *Myceliophthora thermophila* (Apinis) Oorschot was also retrieved to use as an outgroup (Table 1). Alignment of the ITS-5.8S rDNA region of the all sequences in this study, was achieved with MAFFT v. 7.037b (Katoh & Standley 2013), followed by manual adjustment to maximize sequence similarity. Sequence alignment editing was done with BioEdit (Hall 1999).

Phylogenetic trees were constructed using Bayesian analysis (Zhang *et al*. 2015) and maximum likelihood (ML) (Felsenstein 1981). The GTR+I+G nucleotide substitution model was used for Bayesian analysis, as suggested by Modeltest 3.7 (Posada & Crandall 1998). Posterior probabilities were determined by Markov chain Monte Carlo (MCMC) sampling in MrBayes v. 3.2 (Ronquist *et al*. 2012) using the estimated model of evolution. Six simultaneous Markov chains were run for 1,000,000 generations, with trees sampled every 100th generation (resulting in 10,000 total trees). The first 2,000 trees, which represented the burn-in phase of the analysis, were discarded; the remaining 8,000 trees were used to calculate posterior probabilities in the majority rule consensus tree. Each run was examined with the program Tracer v. 1.5 (Drummond & Rambaut 2007) after the analysis finished, to determine burn-in and to confirm that the runs had converged. The ML tree was obtained in MEGA v. 6 (Tamura *et al*. 2013), with gaps treated as missing data, and all other parameters left at default conditions. Bootstrap support for nodes in the resulting trees was assessed using 1,000 replications per analysis. Sequences derived in this study were deposited in GenBank. Sequence alignments were uploaded to TreeBASE (www.treebase.org/treebase/index.html, ID20147), and taxonomic novelties were deposited in MycoBank (www.MycoBank.org).

Results

Molecular phylogenetic analysis

The final aligned ITS-5.8S rDNA sequence dataset from 45 fungi is 477 characters long. Phylogenies obtained using maximum likelihood and Bayesian MCMC analyses are largely congruent (Fig. 1). The majority of branches are strongly supported in both analyses. Most of the strains obviously separated into two clades, with *C. filiforme* Sigler, J.W. Carmich. & H.S. Whitney and *C. pseudomerdarium* Oorschot clustering together. Other strains group together with high support values (1/97) in another clade, of which *C. sanyaense* Y.W. Zhang *et al*., *C. pilosum* Gené, Guarro & Ulfig, *C. longisporum* Stchigel *et al*. and *C. guarroi* J. Cabañes & Abarca are separated into a subclade. Eleven strains, including *C. carmichaelii* Oorschot, assorted into another subclade. Twenty-one strains, including our four isolates (EB1301M, EB1303M, G80.1 and G80.2) assorted into a larger subclade in the genus *Chrysosporium*. Strains G80.1 and G80.2 cluster together with high support values (1/99), and are related to *C. qinghaiense* Y.F. Han, J.D. Liang & Z.Q. Liang, *C. indicum* (H.S. Randhawa & R.S. Sandhu) Garg, and *C. linfenense* Z.Q. Liang, J.D. Liang & Y.F. Han. Strains EB1301M and EB1303M also cluster together with high support values (0.98/94), and appear to be related to *C. articulatum* Scharapov and *C. keratinophilum* D. Frey ex J.W. Carmich, though the support values are low (0.63/-).

Description and taxonomy

Chrysosporium jingzhouense Y.W. Zhang, Y.F. Han & Z.Q. Liang *sp. nov.* (Fig. 2)

MycoBank No.: MB 818911, GenBank: KY026599 KY026600

Type:—CHINA. Hubei Province: Jingzhou City, N 30°21′27.37″, E 112°19′58.37″. Holotype GZUIFR-EB1303M was isolated from farmland soil by Y.R.Wang

Colonies on PDA attaining 25 mm in 14 days at 26 °C, white, fluffy, round, margin regular; reverse light yellow; *hyphae* hyaline, smooth, 1.6–4.3 µm; *racquet hyphae* present, 8.6–15 × 3.2–6.5 µm. *Terminal and lateral conidia* mostly on short protrusions or on side branches, mostly solitary or forming chains, single-celled, occasionally two-celled, smooth-walled, oblong-ovate to oblong-ellipsoidal, 4.3–16.2 × 3.2–8.6 µm ($\bar{x}$ = 7.8 ± 1.1 × 5.6 ± 0.2, n = 50), sometimes clavate, 8.6–25.9 × 3.2–10.8 µm, with broad basal scars (0.8–5.4 µm). *Intercalary conidia* present, solitary, tubby to oblong-clavate, 4.3–32.4 × 2.2–7.6 µm ($\bar{x}$ = 15.8 ± 1.1 × 4.8 ± 0.2, n = 50). *Chlamydospores* absent.

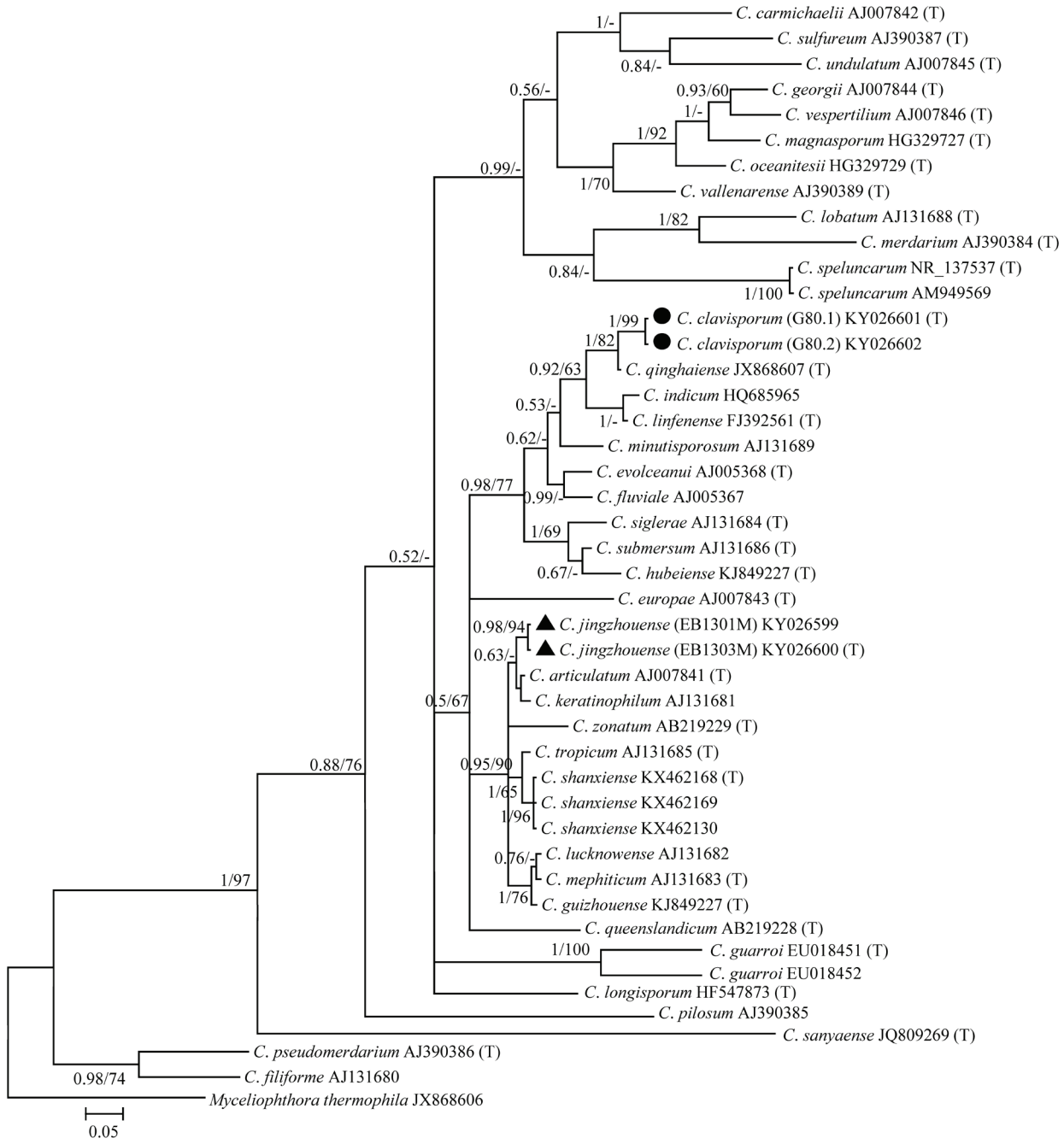

FIGURE 1. Phylogenetic tree of *Chrysosporium* spp. constructed from ITS-5.8S rDNA sequences. Statistical support values (Bayesian posterior probability/maximum likelihood bootstrap percentage) are shown at nodes. The tree was rooted by using *Myceliophthora thermophila* as outgroup.

Etymology:—jingzhouense, referring to Jingzhou City in Hubei Province where the type locality is situated.

Material examined:—The ex-type EB1303M and ex-isotype EB1301M were isolated from farmland soil in Jingzhou City, Hubei Province in March 2012 by Y.R. Wang. Samples were deposited in the Institute of Fungus Resource, Guizhou University (GZAC).

Distribution:—Hubei Province, China.

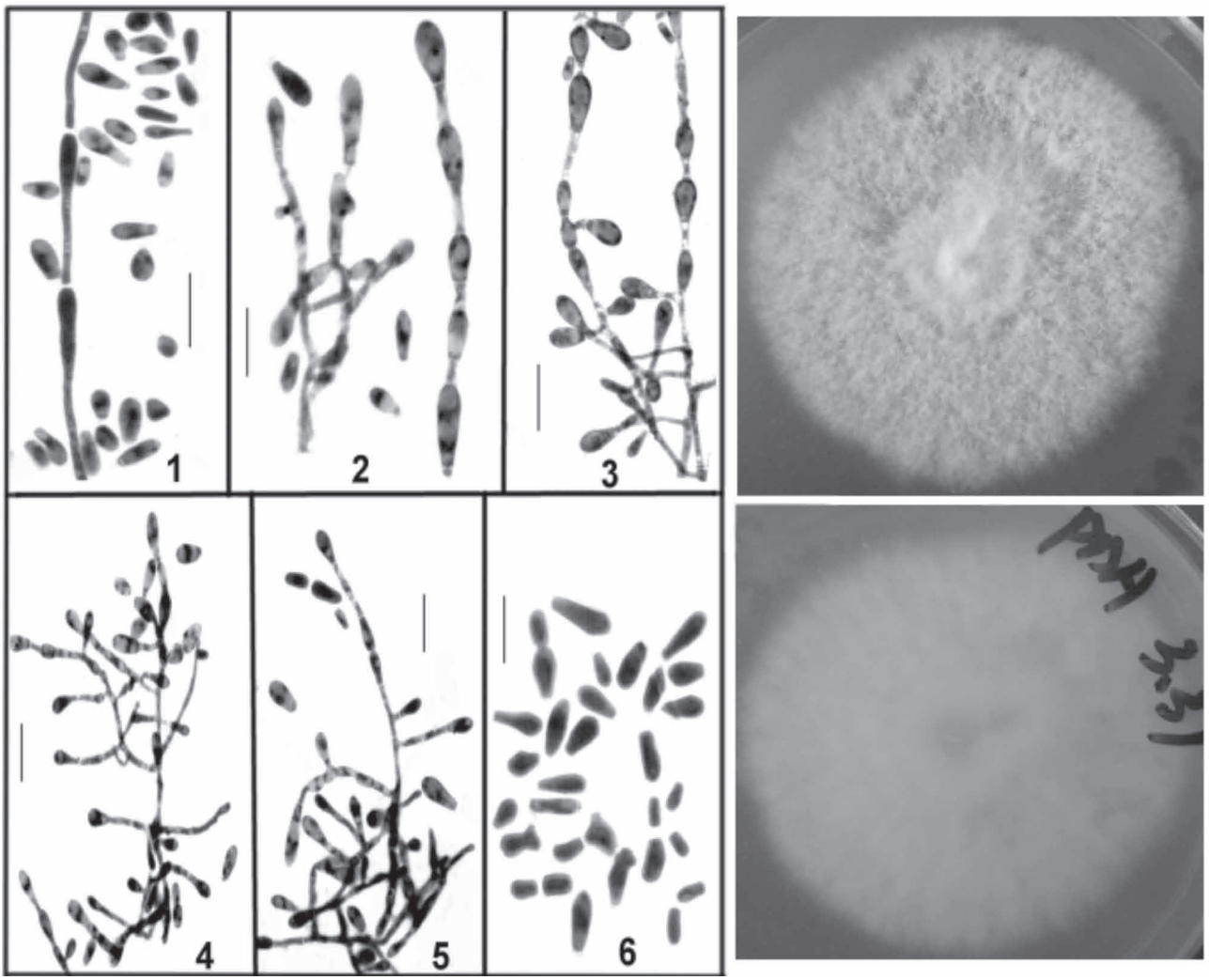

FIGURE 2. Colony and conidiogenous structures of *Chrysosporium jingzhouense* (holotype). **1, 4–5.** Conidiogenous structures. **2.** Racquet hyphae. **3.** Intercalary conidia. **6.** Terminal and lateral conidia. **7–8.** Colony (top and reverse) on PDA. Scale bars 1–6 = 10 μm, 7–8 = 10 mm.

Chrysosporium clavisporum Y.W. Zhang, Y.F. Han & Z.Q. Liang *sp. nov.* (Fig. 3)

MycoBank No.: MB 818912, GenBank: KY026601 KY026602

Type:—CHINA. Guangxi Province: Guigang City, N 23°18′54.75″, E 109°48′17.78″. Holotype GZUIFR-G80.1 was isolated from the plant root soil by Y. Luo.

Colonies on PDA attaining 53 mm in 14 days at 26 °C, white, sparsely fluffy, dense in center and margin loop, round, margin irregular, with deep fissures; reverse red brown in center and light yellow in margin; *hyphae* hyaline, smooth, 1.5–3.5 μm; *racquet hyphae* present, 7.5–15 × 5–7.5 μm. *Terminal and lateral conidia* mostly on short protrusions or on side branches, smooth-walled, mostly solitary, single-celled, clavate to long-ellipsoidal, 5–10 × 2.5–5 μm ($\bar{x}$ = 7.5 ± 1.1 × 3.6 ± 0.1, n = 50); with broad basal scars (2.5–5 μm) and sometimes lightly inflated collar-shaped structures between conidiogenous cells and conidia. *Intercalary conidia* and *chlamydospores* absent.

Etymology:—clavisporum, referring to the shape of conidia.

Material examined:—The ex-type G80.1 and ex-isotype G80.2 were isolated from the tree root soil in Macao River, Guigang City, Guangxi Province on April, 2014 by Y. Luo. Samples were deposited in the Institute of Fungus Resource, Guizhou University (GZAC).

Distribution:—Guangxi Province, China.

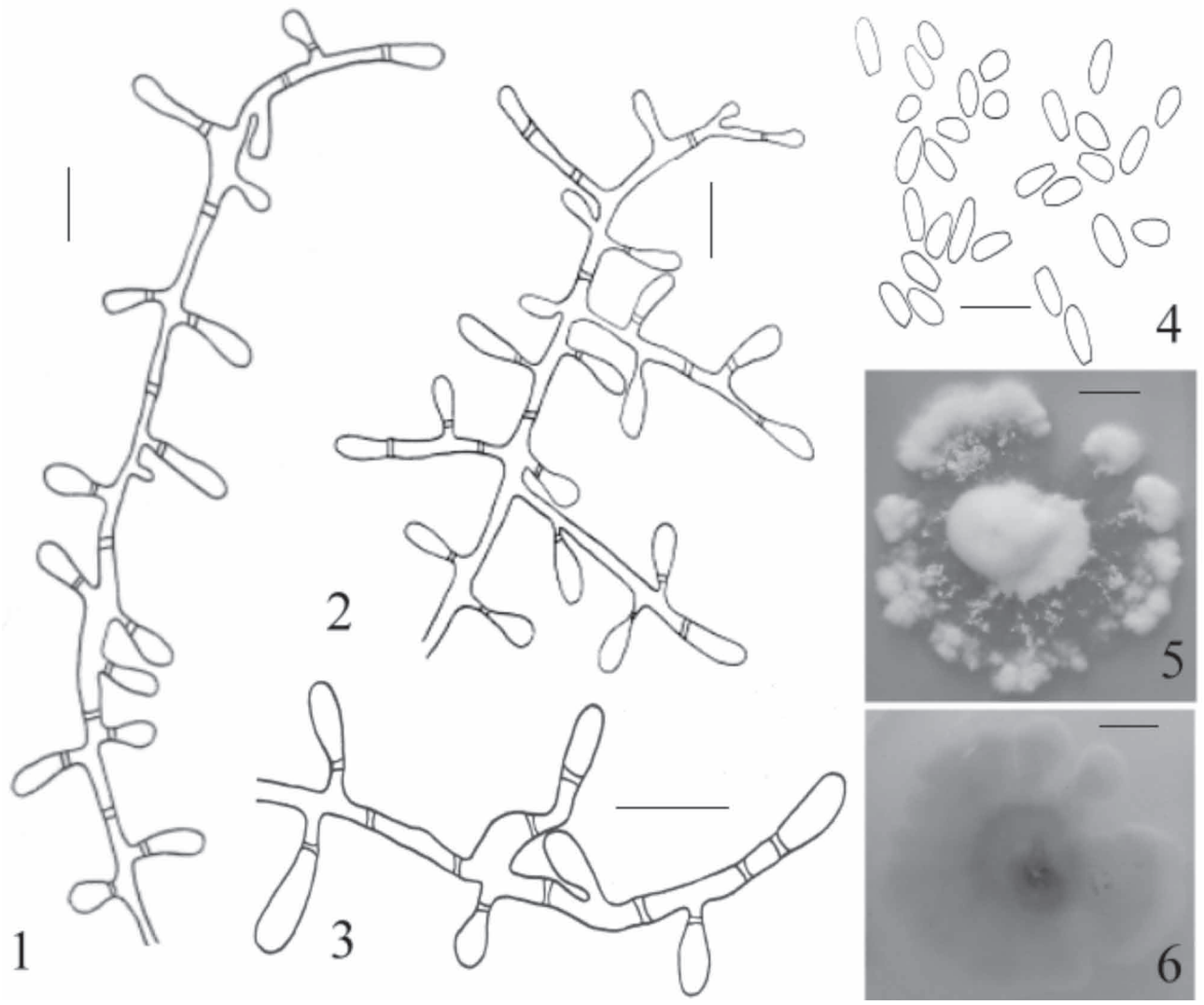

FIGURE 3. Colony and conidiogenous structures of *Chrysosporium clavisporum* (holotype). **1–3.** Conidiogenous structures. **4.** Conidia. **5–6.** Colonies (top and reverse) on PDA. Scale bars 1–4 = 10 μm, 5–6 = 10 mm.

Discussion

Many new fungal species have been reported in recent years based on DNA sequences, and this has dramatically increased the number of known fungi (Dai & Zhuang, 2010; Dai *et al*. 2015). DNA sequences, especially those of ITS-5.8S rDNA, have become an important tool in evolutionary biology. Numerous studies worldwide have demonstrated that ITS-5.8S rDNA can successfully distinguish most fungal species, an important characteristic for fungal identification and recognition (Schoch *et al*. 2012, Kiss *et al*. 2012). Although ITS-5.8S, β-tubulin, and the D1/D2 region of the nuclear 28S subunit (nucLSU) sequences have all been used for phylogenetic reconstruction in the genus *Chrysosporium*, ITS-5.8S rDNA sequences alone were able to successfully distinguish *Chrysosporium* species in two studies (Stchigel *et al*. 2013, Vidal *et al*. 2000). Subsequent phylogenetic analyses in the genus *Chrysosporium* have been predominantly based on ITS-5.8S sequences (Han *et al*. 2013, Vidal *et al*. 2000, Pitt *et al*. 2013).

Chrysosporium jingzhouense clusters with *C. articulatum* and *C. keratinophilum* in our phylogenetic analyses, albeit with low node support values. However, *C. articulatum* has a felty colony, different conidia size (3.5–9.5 × 3–6 μm) and sometimes slightly thick-walled conidia (Oorschot 1980); *C. keratinophilum* has felt-like or powdery colonies and smooth-walled or echinate conidia (Oorschot 1980), which distinguish it from *C. jingzhouense* with its fluffy colonies and smooth-walled conidia. *C. clavisporum* is related to *C. qinghaiense*, *C. indicum*, and *C. linfenense* in our phylogenetic analyses, however, *C. qinghaiense* has powdery colonies and lack racquet hyphae. *C. indicum* has smaller terminal or lateral conidia (3.6–8.1 × 1.8–2.2 μm) and produces intercalary conidia. *C. linfenense* has smaller and

ellipsoidal or fusiform conidia (3.2–5.4 × 1.4–2.2 µm), which differ from *C. clavisporum* with its larger and clavate to long-ellipsoidal conidia (5–10 × 2.5–5 µm), racquet hyphae, sometimes lightly inflated collar-shaped structures between conidiogenous cells and conidia, and no intercalary conidia.

Combining our molecular analyses and classic morphology, the two new species have been described and distinguished from related species, and holotypes designated. The diagnostic characteristics of *C. jingzhouense* are racquet hyphae present, terminal and lateral conidia solitary or on short chains, oblong-ovate to oblong-ellipsoidal, 4.3–16.2 × 3.2–8.6 µm; intercalary conidia solitary, tubby to oblong-clavate, 4.3–32.4 × 2.2–7.6 µm, with broad basal scars (0.8–5.4 µm). *C. clavisporum* can be distinguished by having racquet hyphae, more lateral conidia, clavate to long-ellipsoidal, 5–10 × 2.5–5 µm, with broad basal scars (2.5–5 µm), and sometimes lightly inflated collar-shaped structures between conidiogenous cells and conidia.

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References

Anbu, P., Hilda, A. & Gopinath, S.C.B. (2004) Keratinophilic fungi of poultry farm and feather dumping soil in Tamil Nadu, India. *Mycopathologia* 158: 303–309.
http://dx.doi.org/10.1007/s11046-004-3465-1

Anstead, G.M., Sutton, D.A. & Graybill, J.R. (2012) Adiaspiromycosis causing respiratory failure and a review of human infections due to *Emmonsia* and *Chrysosporium* spp. *Journal of Clinical Microbiology* 50: 1346–1354.
http://dx.doi.org/10.1128/JCM.00226-11

Dai, Y.C. & Zhuang, J.Y. (2010) Numbers of fungal species hitherto known in China. *Mycosystema* 29: 62–628. [in Chinese]

Dai, Y.C., Cui, B.K., Si, J., He, S.H., Hyde, K.D., Yuan, H.S., Liu, X.Y. & Zhou, L.W. (2015) Dynamics of the worldwide number of fungi with emphasis on fungal diversity in China. *Mycological Progress* 14: 62.
http://dx.doi.org/10.1007/s11557-015-1084-5

De Hoog, G.S., Guarro, J. & Gen JéFigueras, M.J. (2000) *Atlas of clinical fungi.* Centraalbureau voor Schimmel-cultures, Utrecht, Netherlands, 1126 pp.

Drummond, A. & Rambaut, A. (2007) BEAST: Bayesian evolutionary analysis by sampling trees. *BMC Evolutionary Biology* 7: 214.
http://dx.doi.org/10.1007/10.1186/1471-2148-7-214

Felsenstein, J. (1981) Evolutionary trees from DNA sequences: a maximum likelihood approach. *Journal of Molecular Evolution* 17 (6): 368–376.
https://doi.org/10.1007/BF01734359

Gan, G.G., Kamamlzaman, A., Gob, K.Y., Ng, K.P., Na, S.L. & Soo-Hoo, T.S. (2002) Non-sporulating *Chrysosporium*: an opportunistic fungal infection in a neutropenic patient. *The Medical Journal of Malaysia* 57: 118–122.

Hall, T.A. (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series* 41: 95–98.

Han, Y.F., Wang, Y.R., Liang, J.D. & Liang, Z.Q. (2013) A new species of the genus *Chrysosporium* from the farmland soil of Qinghai Province. *Mycosystema* 32: 606–611. [in Chinese]

Kamath, P.M., Prasad, V., Shenoy, V.S., Mukundan, A. & Shenoy, S. (2015) *Chrysosporium*: an uncommon fungus in chronic rhinosinusitis. *Journal of Clinical and Diagnostic Research* 9: MD01–MD02.
http://dx.doi.org/10.7860/JCDR/2015/11676.5688

Katoh, K. & Standley, D.M. (2013) Mafft multiple sequence alignment software version 7: improvements in performance and usability. *Molecular Biology and Evolution* 30: 772–780.
http://dx.doi.org/10.1093/molbev/mst010

Kiss, L., Schoch, C.L., Seifert, K.A., Caldeira, K., Myhrvold, N.P., Alvarez, R.A. & Mullins, R.D. (2012) Limits of nuclear ribosomal DNA internal transcribed spacer (ITS) sequences as species barcodes for Fungi. *Proceedings of the National Academy of Sciences*

109: 10741–10742.
http://dx.doi.org/10.1073/pnas.1207143109

Luo, Y., Chen, W.H., Wang, Y., Han, Y.F. & Liang, Z.Q. (2016) A new *Geomyces* species producing melanin in the medium. *Mycosystema* 35: 123–130. [in Chinese]

Marchisio, V.F. (2000) Keratinophilic fungi: their role in nature and degradation of keratinic substrates. *Revista Iberoamericana de Micologia* 17: 86–92.

Marchisio, V.F., Curetti, V., Cassinelli, C. & Bordese, C. (1991) Keratinolytic and keratinophilic fungi in the soil of Papua New Guinea. *Mycopathologia* 115: 113–120.
http://dx.doi.org/10.1007/BF00436799

McAleer, R. (1980) Investigation of keratinophilic fungi from soil in western-Australia: a preliminary survey. *Mycopathologia* 72: 155–166.
http://dx.doi.org/10.1007/BF00572658

Pitt, J.I., Lantz, H., Pettersson, O.V. & Leong, S.L.L. (2013) *Xerochrysium* gen. nov. and *Bettsia*, genera encompassing xerophilic species of *Chrysosporium*. *IMA Fungus* 4: 229–241.
http://dx.doi.org/10.5598/imafungus.2013.04.02.08

Posada, D. & Crandall, K.A. (1998) Modeltest: testing the model of DNA substitution. *Bioinformatics* 14: 817–818.
http://dx.doi.org/10.1093/bioinformatics/14.9.817

Ronquist, F., Teslenko, M., van der Mark, P., Ayres, D.L., Darling, A., Höhna, S., Larget, B., Liu, L., Suchard, M.A. & Huelsenbeck, J.P. (2012) MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology* 61: 539–542.
http://dx.doi.org/10.1093/sysbio/sys029

Schoch, C.L., Seifert, K.A., Huhndorf, S., Robert, V., Spouge, J.L., Levesque, C.A., Chen, W. & Fungal barcoding Consortium (2012) Nuclear ribosomal internal transcribed spacer (ITS) region as a universal DNA barcode marker for fungi. *Proceedings of the National Academy of Sciences* 109: 6241–6246.
http://dx.doi.org/10.1073/pnas.1117018109

Shadzi, S., Chadeganipour, M. & Alimoradi, M. (2002) Isolation of keratinophilic fungi from elementary schools and public parks in Isfahan, Iran. *Mycoses* 45: 496–499.
http://dx.doi.org/10.1046/j.1439-0507.2002.00798.x

Sigler, L., Hambleton, S. & Paréc, J.A. (2013) Molecular characterization of reptile pathogens currently known as members of the *Chrysosporium* anamorph of *Nannizziopsisvriesii* complex and relationship with some human-associated isolates. *Journal of Clinical Microbiology* 51: 3338–3357.
http://dx.doi.org/10.1128/JCM.01465-13

Stchigel, A.M., Sutton, D.A., Cano-Lira, J.F., Cabañes, F.J., Abarca, L., Tintelnot, K., Wickes, B.L., García, D. & Guarro, J. (2013) Phylogeny of c*hrysosporia* infecting reptiles: proposal of the new family *Nannizziopsiaceae* and five new species. *Persoonia* 31: 86–100.
http://dx.doi.org/10.3767/003158513X669698

Suankratay, C., Dhissayakamol, O., Uaprasert, N. & Chindamporn, A. (2015) Invasive pulmonary infection caused by *Chrysosporium articulatum*: the first case report. *Mycoses* 58: 1–3.
http://dx.doi.org/10.1111/myc.12270

Tamura, K., Stecher, G., Peterson, D., Filipski, A. & Kumar, S. (2013) MEGA6: molecular evolutionary genetics analysis version 6.0. *Molecular Biology and Evolution* 30 (12): 2725–2729.
http://dx.doi.org/10.1093/molbev/mst197

van Oorschot, C.A.N. (1980) A revision of *Chrysosporium* and allied genera. *Studies in Mycology* 20: 1–89.

Vidal, P., Vinuesa, M.D.L.A., Sánchez-Puelles, J.M. & Guarro, J. (2000) Phylogeny of the anamorphic genus *Chrysosporium* and related taxa based on rDNA internal transcribed spacer sequences. *Revista Iberoamericana de Micología* 17: 22–29.

Zarrin, M. & Haghgoo, R. (2011) Survey of keratinophilic fungi from soils in Ahvaz, Iran. *Jundishapur Journal of Microbiology* 4: 191–194.

Zhang, Y.W., Han, Y.F., Liang, J.D. & Liang, Z.Q. (2013) A new species of the genus *Chrysosporium* from the rhizosphere soil of China. *Mycosystema* 32: 612–616.

Zhang, Y.W., Chen, W.H., Zeng, G.P., Wang, Y.R., Zou, X., Han, Y.F., Qiu, S.Y. & Liang, Z.Q. (2016a) Two new *Chrysosporium* (Onygenaceae, Onygenales) from China. *Phytotaxa* 270 (3): 210–216.
http://dx.doi.org/10.11646/phytotaxa.270.3.5

Zhang, Y.W., Chen, W.H., Zou, X., Han, Y.F. & Liang Z.Q. (2016b) A new *Chrysosporium* species from sparrow's nest in Shanxi Province. *Mycosystema* 35 (11): 1–8.