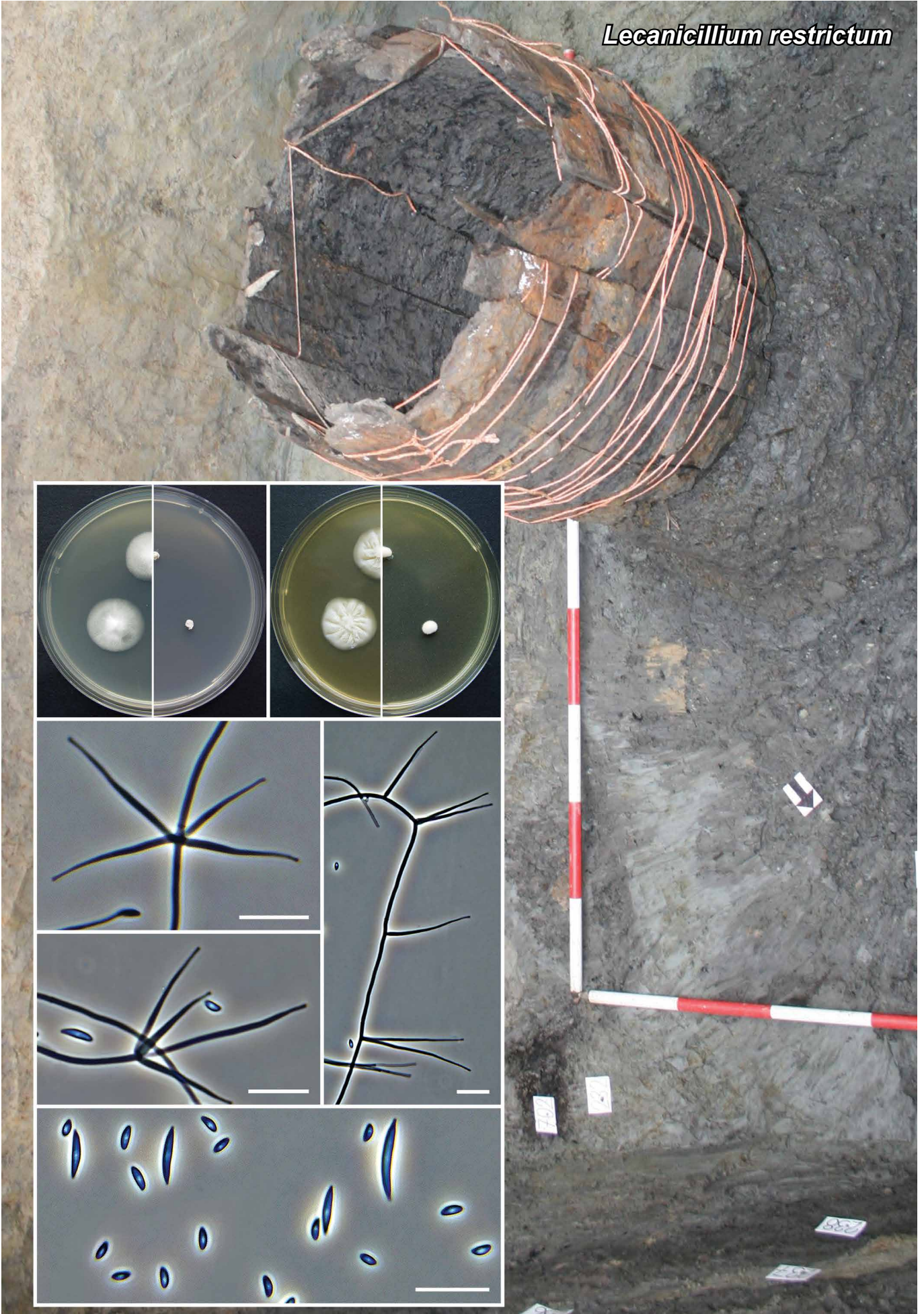


*Lecanicillium restrictum*



Fungal Planet 734 – 13 July 2018

***Lecanicillium restrictum*** Hubka, Kubátová, Nonaka, Čmoková & Řehulka, *sp. nov.*

*Etymology.* *restrictum* (res.tric'tum. L. neut. part. adj.); limited, restricted, referring to the slow growth at room temperature (25 °C).

Classification — *Cordycipitaceae*, *Hypocreales*, *Sordariomycetes*.

On PCA: *Phialides* produced on aerial hyphae, solitary or aggregated in whorls of 2–5 phialides, tapering toward the tip, (12–)17–30(–36) µm long (mean ± standard deviation; 22.4 ± 4.8), basal part 0.5–1.5 (1.1 ± 0.2) µm wide, 0.3–0.5 µm wide on the tip. *Conidia* dimorphic, macroconidia with pointed ends, fusiform or slightly falcate, smooth-walled, 1-celled, (5–)6–10(–12) × 1–1.5 µm (7.5 ± 1.3 × 1.1 ± 0.1), microconidia usually without sharply pointed ends, ovate, ellipsoidal, obovate or fusoid, frequently slightly curved, smooth-walled, 1-celled, 2.5–3 × 1–1.5 µm (3 ± 0.4 × 1.1 ± 0.1). No microscopic crystals observed.

Culture characteristics — (in the dark, at 20 °C after 14 d): Colonies on PCA 20–23 mm diam (10–12 mm after 7 d), white, cottony, centrally raised, margin entire, no exudate and soluble pigments, reverse yellowish white (4A2; Kornerup & Wanscher 1967). Colonies on MEA 19–22 mm diam (10–12 mm after 7 d), yellowish white (4A2), waxy, delicately funiculose, umbonate, radially wrinkled, margin entire, no exudate and soluble pigments, reverse pale yellow (4A3). Colonies on PDA 21–25 mm diam (11–13 mm after 7 d), yellowish white (4A2), floccose to delicately funiculose, umbonate, radially wrinkled, margin entire, no exudate and soluble pigments, reverse yellowish white (4A2) to pale yellow (4A3). Growth rates at 15 °C on PCA/MEA/PDA: 8–10/8–10/9 mm after 7 d and 17–21/17–20/18–21 mm after 14 d, respectively. Growth rates at 25 °C on PCA/MEA/PDA: 1–3/2–4/2–4 mm after 7 d and 2–4/4–5/3–6 mm after 14 d, respectively. No growth to microcolonies on PCA and MEA at 27 °C; no growth at 30 °C.

*Typus.* CZECH REPUBLIC, Starý Bohumín, surface of the wooden barrel found during archaeological excavations, 3 Mar. 2014, coll. M. Kiecoň & P. Malík, isol. J. Řehulka (holotype PRM 946543, isotype PRM 946544, culture ex-type CCF 5252 = CBS 143072; SSU-ITS-LSU, *tef1-α* and *tub2* sequences GenBank LT548279, LT626943 and LT989952, MycoBank MB824887).

*Colour illustrations.* Wooden barrel found during archaeological excavations, Starý Bohumín, Czech Republic; 14-d-old colonies of *L. restrictum* on PCA (left) and MEA (right), left half of Petri dish: colony at 20 °C, right half: 25 °C; whorls of phialides and solitary phialides; micro- and macroconidia. Scale bars = 10 µm.

Notes — BLAST analysis with the ITS rDNA region sequence gave closest hits to *L. testudineum* CCF 5201<sup>T</sup> (99 %, 497/499 bp, GenBank LT548278), *L. kalimantanense* NBRC 105406<sup>T</sup> (94 %, 465/494 bp, GenBank AB360356), *L. wallacei* CBS 101237<sup>T</sup> (93 %, 448/484 bp, GenBank EF641891) and *Verticillium indonesiacum* BTCC-F36<sup>T</sup> (93 %, 462/495 bp, GenBank AB378516). LSU rDNA showed 99 % similarity to *L. testudineum* (99 %, 589/592 bp, GenBank LT548278) and *L. wallacei* (541/548 bp, GenBank AY184967), and 98 % similarity to *L. kalimantanense* (580/589 bp, GenBank AB360356) and *V. indonesiacum* (580/589 bp, GenBank AB378516). The *tub2* sequence showed 91 % similarity to *L. testudineum* (1 225/1 348 bp, GenBank LT548284) and the *tef1-α* sequence 94 % similarity to *L. testudineum* (936/992 bp, GenBank LT626942).

*Lecanicillium restrictum* is characteristic by having slow growth at 25 °C, optimum temperature for growth around 20 °C and the production of dimorphic conidia. *Lecanicillium testudineum* has an optimum temperature for growth around 25 °C and smaller macroconidia than *L. restrictum*. Microconidia of *L. restrictum* are smaller than conidia produced by *L. kalimantanense* (3.5–12 × 1–2 µm) (Sukarno et al. 2009). Phialides of *V. indonesiacum* are most frequently produced in a single whorl at the end of erect hyphae (Sukarno et al. 2009). *Lecanicillium wallacei* grows more rapidly than *L. restrictum* on PCA at 20 and 25 °C (Zare & Gams 2001).

The best scoring maximum likelihood tree calculated from ITS rDNA and *tef1-α* sequences shows the species relationships within the genus *Lecanicillium*. The optimal partitioning scheme (PartitionFinder v. 1.1.1; Lanfear et al. 2012) divided the dataset into four partitions with the following substitution models: the GTR+G substitution model was used for ITS1 and ITS2 regions, JC+I model for the 5.8S nrDNA region and the 2nd codon positions of *tef1-α*, F81+I+G model for the 1st codon positions of *tef1-α*, and HKY+G model for the 3rd codon positions of *tef1-α*. The tree was constructed with IQ-TREE v. 1.4.0 (Nguyen et al. 2015). The dataset contained 30 taxa and a total of 1 583 characters of which 478 were variable and 357 parsimony-informative. Bootstrap support values at branches were obtained by generating 1 000 bootstrap replicates. Only bootstrap support values ≥ 70 % are shown; ex-type strains are indicated by a superscript <sup>T</sup>. The tree is rooted with *Simplicillium lanosoniveum* CBS 704.86 and *S. obclavatum* CBS 311.74<sup>T</sup>.

For phylogenetic tree see Fungal Planet 735.

Vit Hubka, Department of Botany, Faculty of Science, Charles University, Benátská 2, 128 01 Prague 2, Czech Republic; Laboratory of Fungal Genetics and Metabolism, Institute of Microbiology of the CAS, v.v.i, Vídeňská 1083, 142 20 Prague 4, Czech Republic; e-mail: hubka@biomed.cas.cz  
Alena Kubátová, Department of Botany, Faculty of Science, Charles University, Benátská 2, 128 01 Prague 2, Czech Republic; e-mail: alena.kubatova@natur.cuni.cz  
Kenichi Nonaka, Kitasato Institute for Life Sciences, Kitasato University, 5-9-1 Shirokane, Minato-ku, Tokyo 108-8641, Japan; e-mail: ken@lisci.kitasato-u.ac.jp  
Adéla Čmoková, Laboratory of Fungal Genetics and Metabolism, Institute of Microbiology of the CAS, v.v.i, Vídeňská 1083, 142 20 Prague 4, Czech Republic; e-mail: cmokova@gmail.com  
Jiří Řehulka, Department of Zoology, Silesian Museum, Nádražní okruh 31, 746 01 Opava, Czech Republic; e-mail: rehulka@szm.cz