

Insight into the systematics of microfungi colonizing *Dodonaea viscosa* in Honghe

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Abstract

Members of *Dodonaea* are broadly distributed across subtropical and tropical areas of southwest and southern China. This host provides multiple substrates that can be richly colonized by numerous undescribed fungal species. There is a severe lack of microfungi studies on *Dodonaea* in China, and consequently, the diversity, phylogeny and taxonomy of these microorganisms are all largely unknown. This paper presents two new genera and four new species in three orders of Dothideomycetes gathered from dead twigs of *Dodonaea viscosa* in Honghe, China. All new collections were made within a selected area in Honghe from a single *Dodonaea* sp. This suggests high fungal diversity in the region and the existence of numerous species awaiting discovery. Multiple gene sequences (non-translated loci and protein-coding regions) were analysed with maximum likelihood and Bayesian analyses. Results from the phylogenetic analyses supported placing *Haniomyces dodonaeae* gen. et sp. in the Teratosphaeriaceae family. Analysis of *Rhytidhysterium* sequences resulted in *Rhytidhysterium hongheense* sp. nov., while analysed Lophiostomataceae sequences revealed *Lophiomurispota hongheensis* gen. et sp. nov. Finally, phylogeny based on a combined dataset of pyrenochaeta-like sequences demonstrates strong statistical support for placing *Quixadomyces hongheensis* sp. nov. in Parapyrenochaetaceae. Morphological and updated phylogenetic circumscriptions of the new discoveries are also discussed

Introduction

Fungi are cosmopolitan, featuring a broad geographic distribution and high level of diversity compared to plants and other organisms. Yunnan Province, China, as part of the GMS, is home to an extremely wide variety of ecosystems. Mycologists working in Yunnan have recently focused their attention on abundant "less-researched habitats" for fungal occurrences, including caves, forests, grasslands, lakes, karst landscapes and mountains; accordingly, there is a rich body of literature documenting novel discoveries across the region. Among publications documenting fungal encounters across Yunnan Province, ascomycetes are critically neglected when compared to the amount of research on basidiomycetes. Regrettably, studies on microfungi in Honghe are virtually non-existent.

This study is a part in a series comprising an exhaustive taxonomic effort to document the microfungi of Yunnan Province. In this study, we collected fresh fungal specimens from dead woody twigs of *Dodonaea* species at the Centre for Mountain Futures (CMF), an applied research centre jointly managed by World Agroforestry (ICRAF) and the Kunming Institute of Botany, Chinese Academy of Sciences (CAS), in Honghe County of the Honghe Hani and Yi Autonomous Prefecture. Using morphology and multi-gene phylogenetic evidence retrieved from the gathered ascomycetes, we characterized two new genera and four new species in the orders Capnodiales, Hysteriales and Pleosporales from dead twigs of *Dodonaea viscosa* in Honghe.

Dodonaea viscosa is a perennial evergreen woody shrub belonging to the family Sapindaceae. It is drought- and pollution-resistant as well as capable of growing on poor soils and rocky sites. The plant can also easily inhabit open areas and secondary forests. *Dodonaea viscosa* is effective at performing sand dune fixation and controlling coastal erosion since its roots function as excellent soil binders. It can also be used to reclaim marshes. It is also grown as an ornamental plant owing to its shiny foliage and pink-red winged fruit. Moreover, it is a well-known topic in environmental impact studies to determine the growth and yield of crops based on the presence of *D. viscosa* as well as study its capacity to increase resilience to pollution and drought.

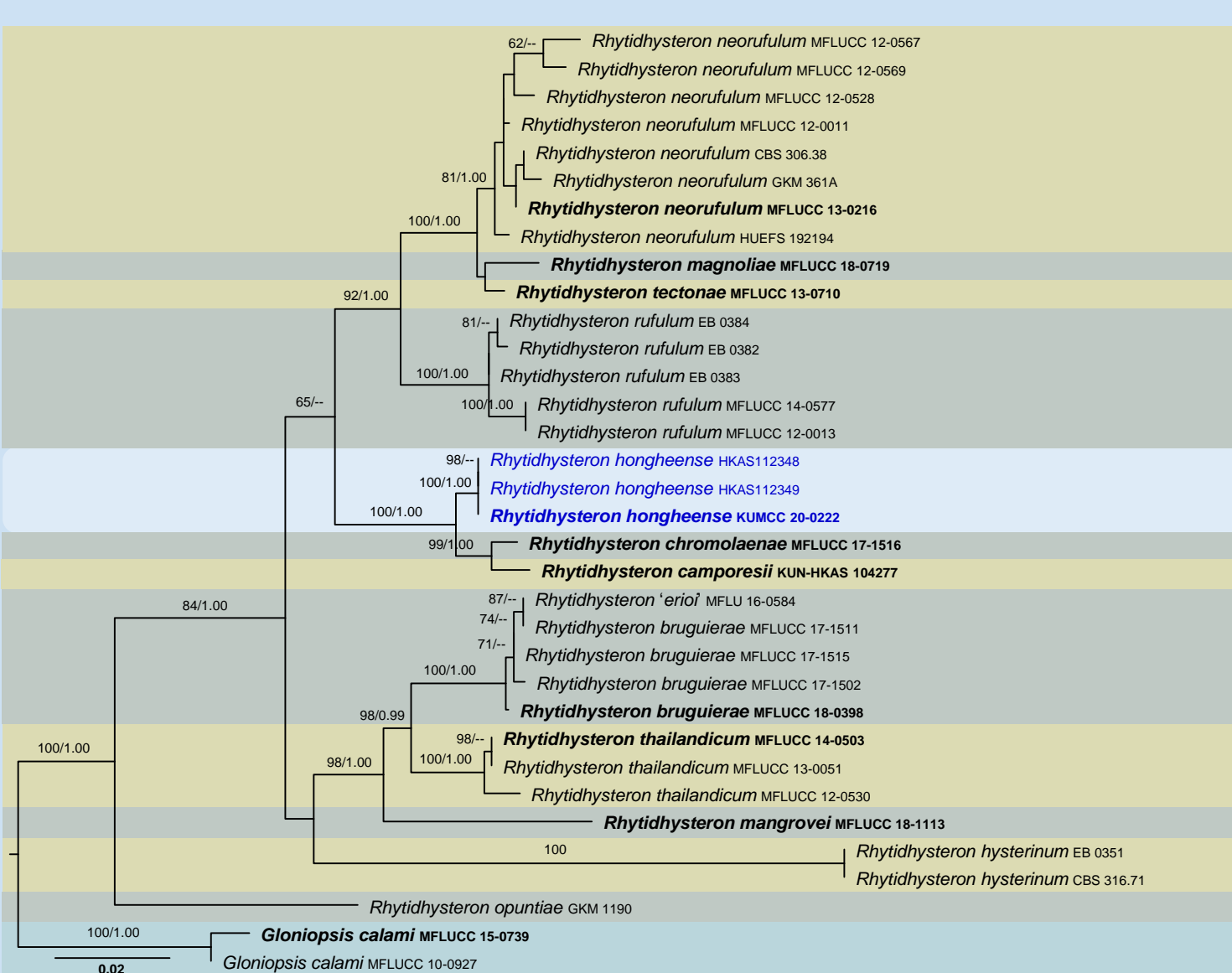


Figure 3. RAxML tree based on a combined dataset of partial SSU, LSU, ITS and tef1 DNA sequence analysis in Rhytidhysterium

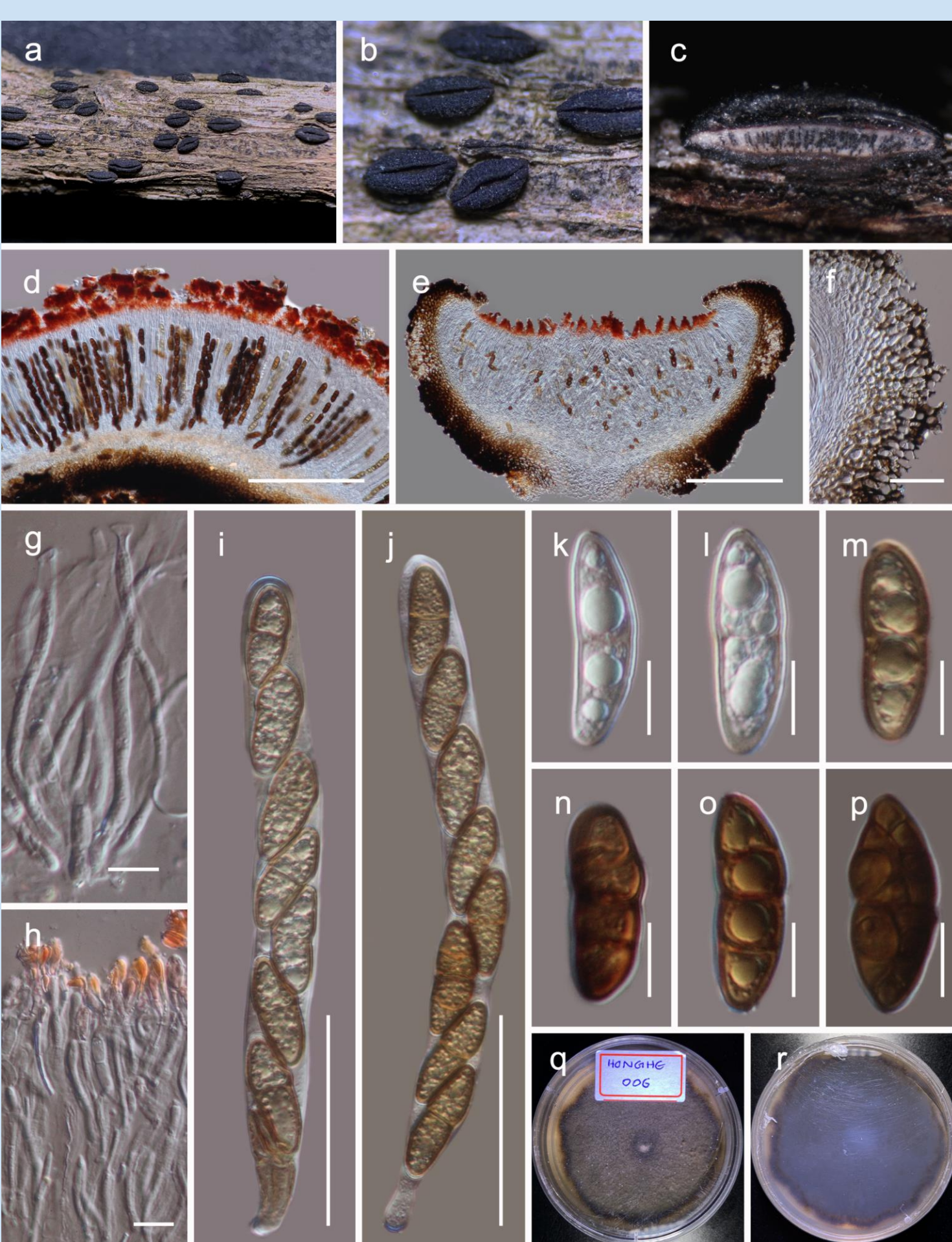


Figure 4. *Rhytidhysterium hongheensis*. (a, b) Appearance of hysterothecia on *Dodonaea viscosa*; (c, d) horizontal section of hysterothecium; (e) vertical section of hysterothecium; (f) cells of peridium; (g, h) pseudoparaphyses; (i, j) asci; (k-p) ascospores; (q, r) colony on PDA (r from the bottom). Scale bars: (d, e) 200 µm; (f, i, j) 50 µm; (g, h, k-p) 10 µm.

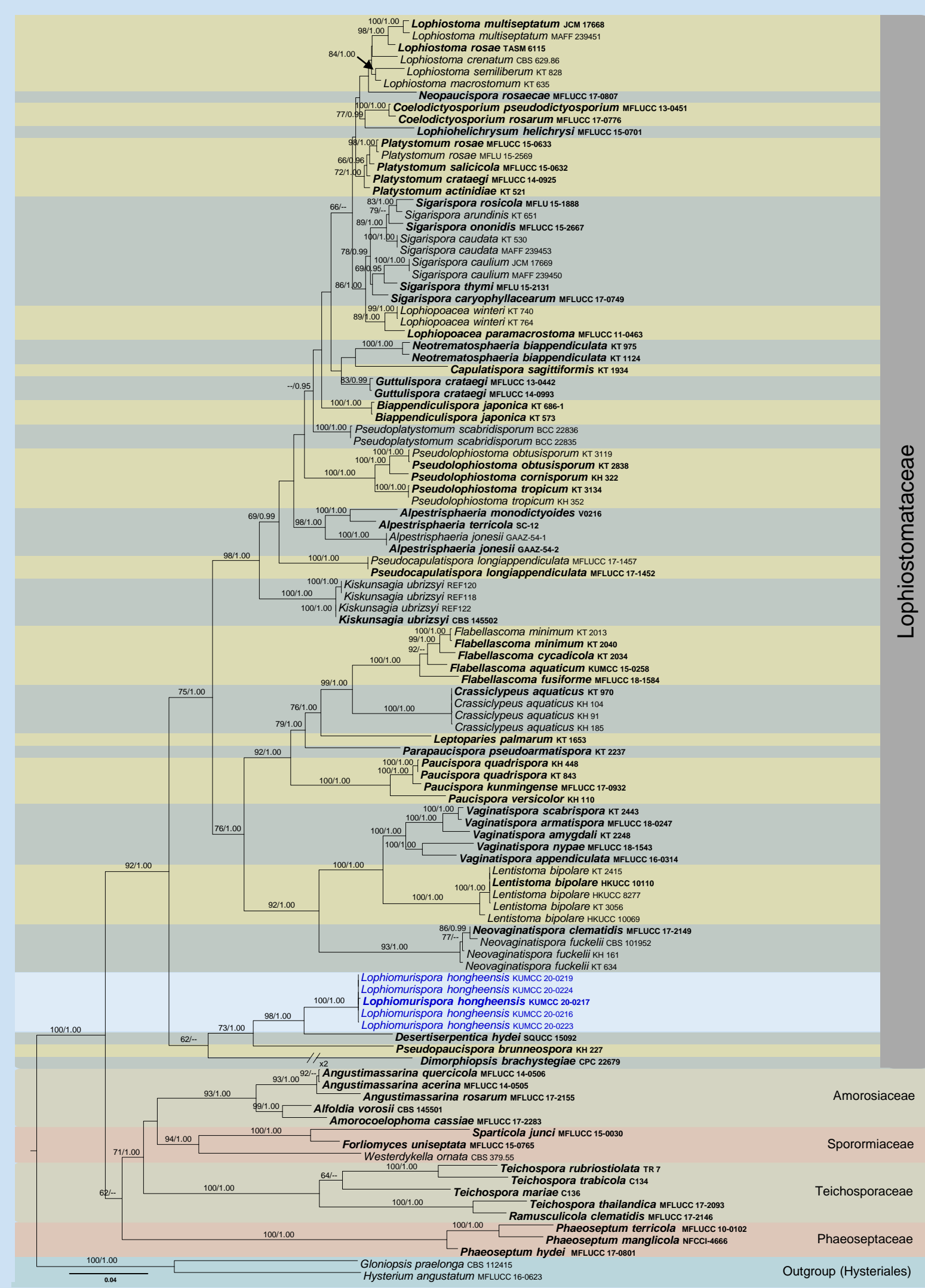


Figure 5. RAxML tree based on a combined dataset of partial SSU, LSU, ITS, tef1 and rpb2 DNA sequence analysis in Lophiostomataceae

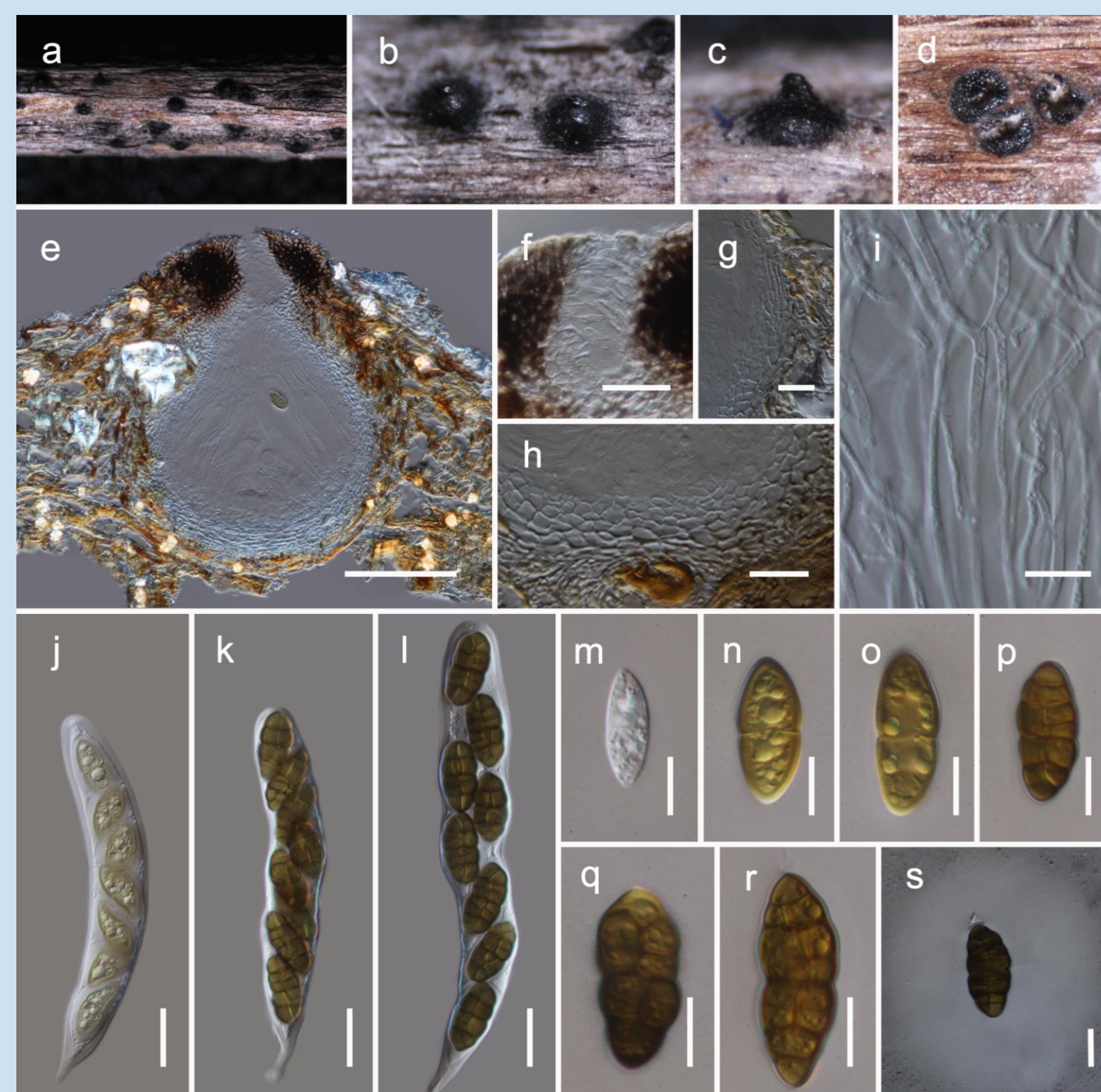


Figure 6. *Lophiomurispota hongheensis*. (a-c) Ascomata on *Dodonaea viscosa*; (d) cross section of ascomata; (e) vertical section of ascoma; (f) closeup of ostiole; (g, h) peridium; (i) pseudoparaphyses; (j-l) asci; (m-s) ascospores (s in Indian Ink); Scale bars: (e) 100 µm; (f-h, j-l) 20 µm; (i, m-s) 10 µm

Results

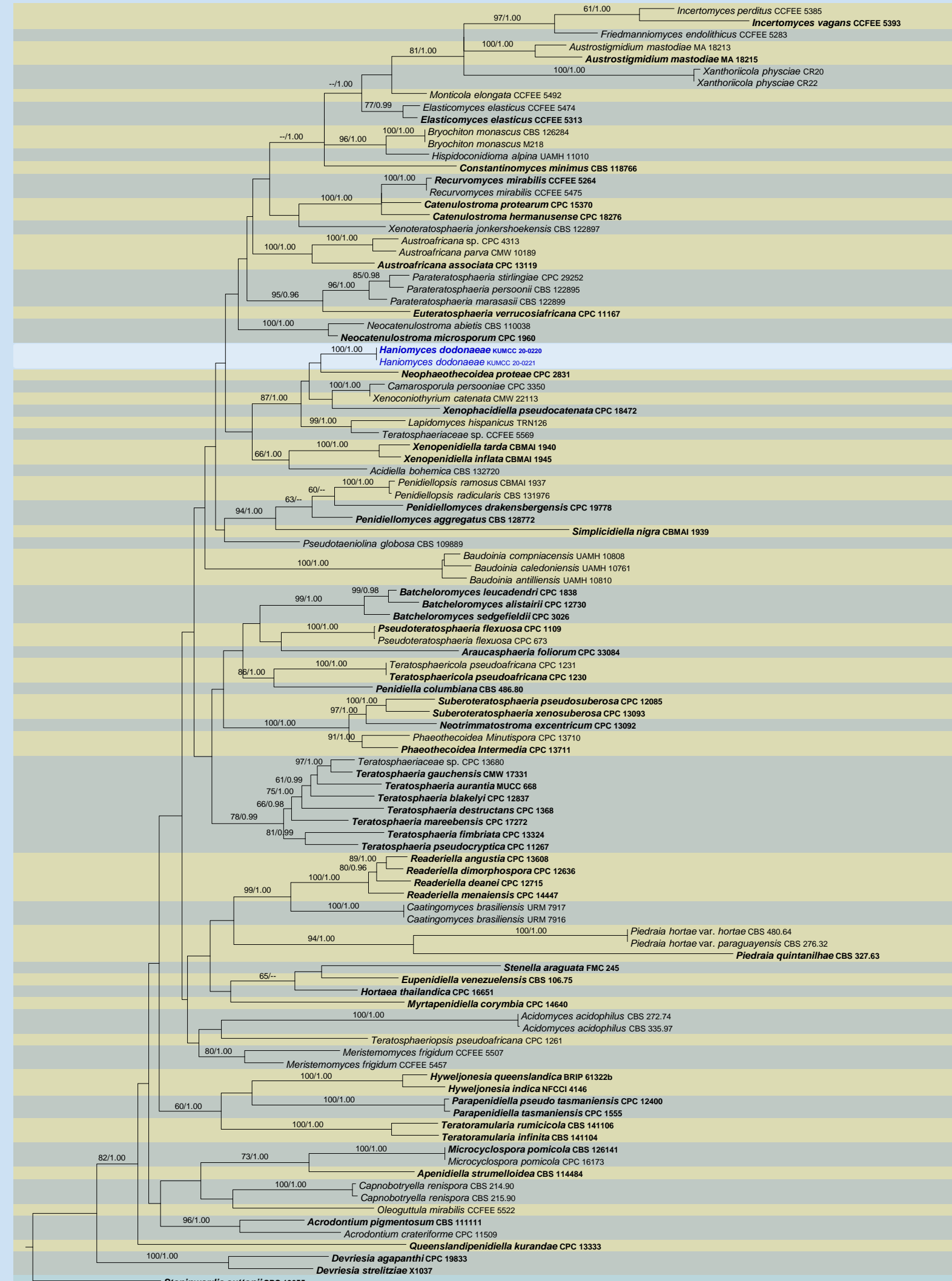


Figure 1. RAxML tree based on a combined dataset of partial LSU, ITS, rpb2, act, cal and tef1 DNA sequence analysis in Teratosphaeriaceae

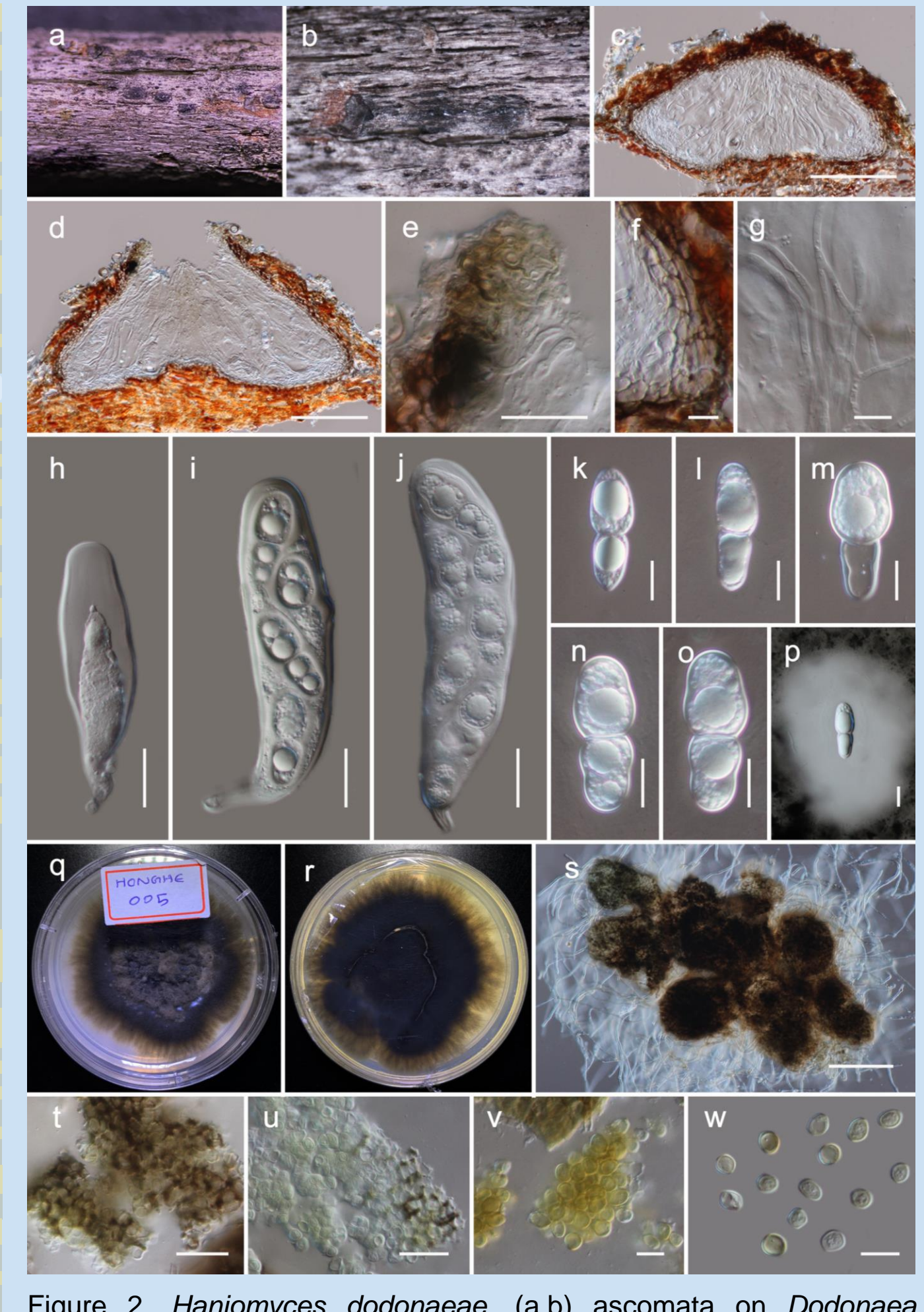


Figure 2. *Haniomyces dodonaeae*. (a, b) ascomata on *Dodonaea viscosa*; (c, d) vertical section of ascoma; (e) periphyses; (f) peridium; (g) pseudoparaphyses; (h-j) asci; (k-p) ascospores (p in Indian Ink); (q, r) colony on PDA (r from the bottom); (s) squashed pycnidia which were produced on PDA; (t) pycnidia wall; (u-w) conidia. Scale bars: (c, d) 100 µm; (e, h-j, t, u) 20 µm; (f, k-p, v, w) 10 µm; (s) 200 µm.

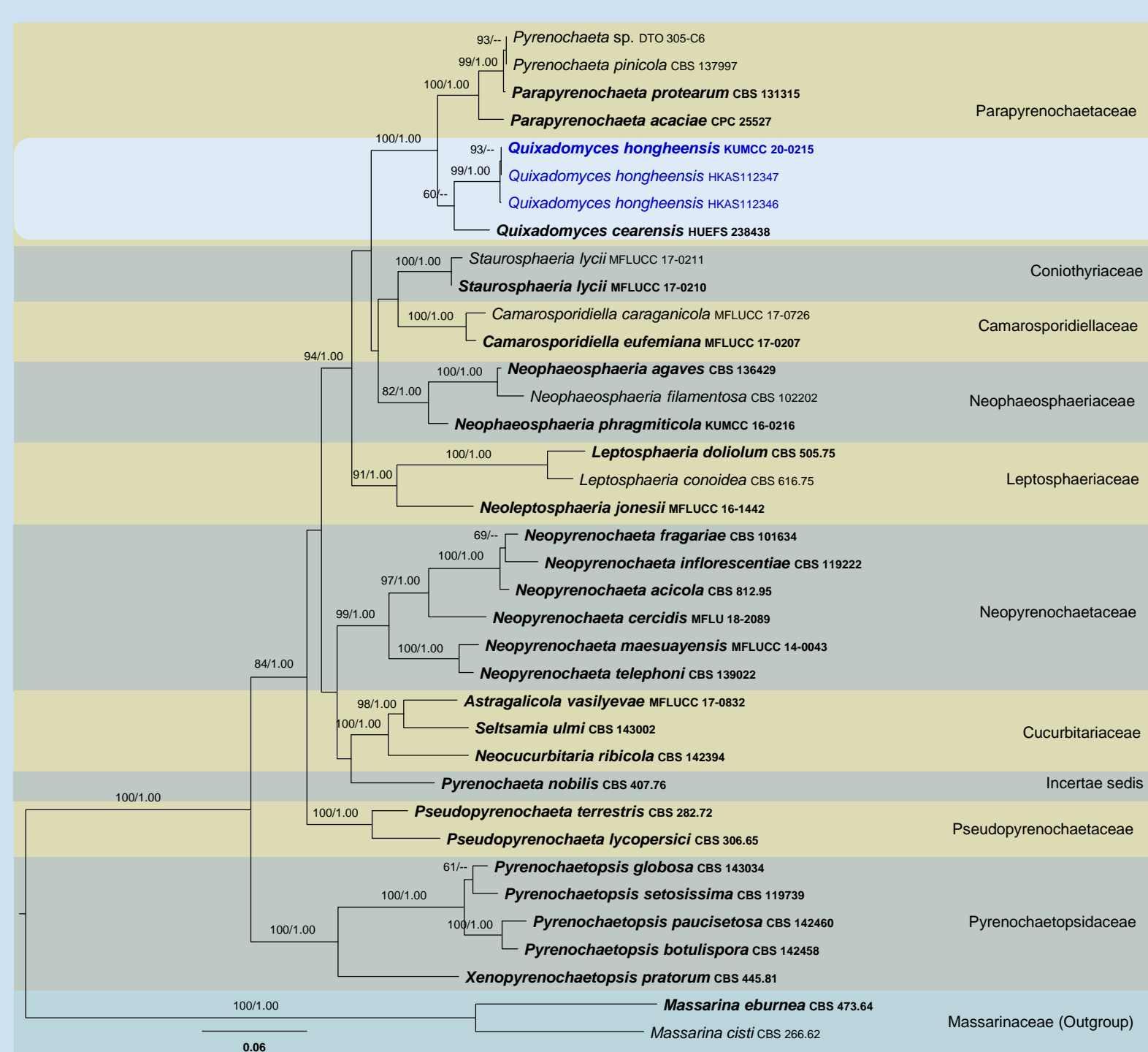


Figure 7. RAxML tree based on a combined dataset of partial LSU, SSU, ITS, rpb2, tef1 and tbtub DNA sequence analysis in Pleosporinae

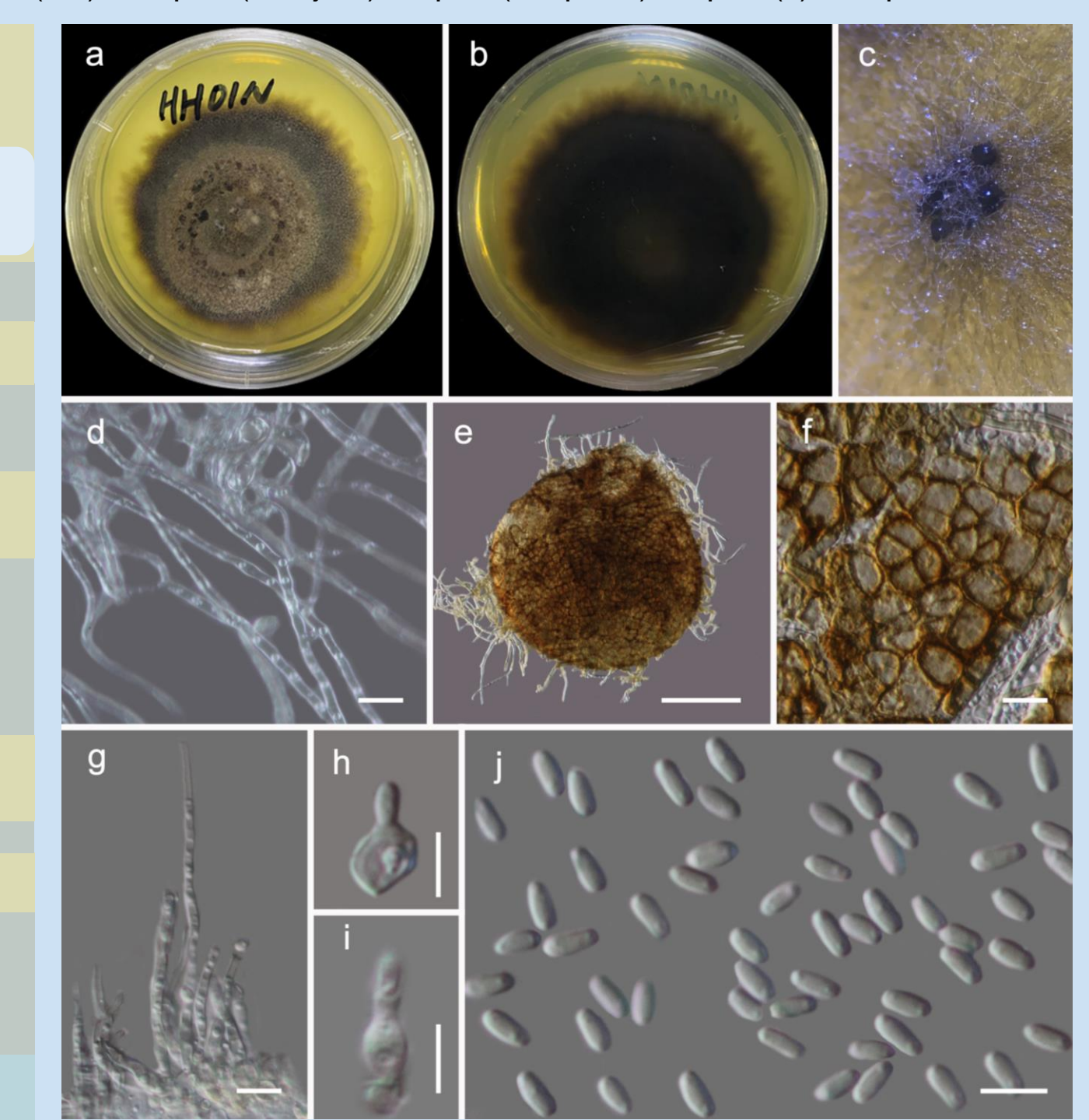


Figure 8. *Quixadomyces hongheensis*. (a, b) colony on PDA (b from the bottom); (c) pycnidia on PDA; (d) mycelia; (e) squashed pycnidia; (f) pycnidia wall; (g) paraphyses; (h, i) conidiophore; (j) conidia. Scale bars: (d, f, g) 10 µm; (e) 200 µm; (h-i) 5 µm

Discussion

Nearly a century's worth of taxonomic investigation into *Dodonaea viscosa* has yielded only 58 fungal records. These are mainly reported as saprobes or pathogens, but very few of these taxa are confirmed by both morphological and phylogenetic evidence. Many of these published records lack illustrations, descriptions or DNA sequence data, resulting in unclear taxonomic relationships. Even though *Dodonaea viscosa* is widely distributed across southwest and southern China, e.g., Fujian, Guangdong, Guangxi, Hainan, Sichuan and Yunnan, there is only one report for the fungus *Pseudocercospora mitteriana* on this host from China. Previous taxonomic studies have suggested that increased collections might lead to the discovery of many new fungal species, and we, too, believe that *Dodonaea* is likely teeming with fungal diversity. More *Dodonaea* collections across different geographic regions are urgently needed, along with accompanying work in culture isolation, morphological description, DNA sequence analyses, phylogenetic relationship investigation, and accurate identification and classification. This study provides a case study for *Dodonaea viscosa* as a worthwhile host for the further study of microfungi associations and hints that it may potentially host numerous unknown fungal species.

Reference

Wanasinghe DN, Mortimer PE, Xu J. Insight into the Systematics of Microfungi Colonizing Dead Woody Twigs of *Dodonaea viscosa* in Honghe (China). *Journal of Fungi*. 2021; 7(3):180.