CRISTIANE NASCIMENTO FIGUEIREDO

BIODIVERSITY AND CHITINASE PRODUCTION OF SOIL FUNGI IN THE CERRADO BIOME OF MINAS GERAIS

LAVRAS – MG
2025

CRISTIANE NASCIMENTO FIGUEIREDO

BIODIVERSITY AND CHITINASE PRODUCTION OF SOIL FUNGI IN THE CERRADO BIOME OF MINAS GERAIS

Tese apresentada à Universidade Federal de Lavras, como parte das exigências do Programa de Pós-Graduação em Microbiologia Agrícola, área de concentração em Microbiologia Agrícola para a obtenção do título de Doutora.

Prof. DSc. Victor Satler Pylro
Orientador

DSc. Jos Houbraken
Prof. DSc. Luís Roberto Batista
DSc.Pedro Crous
Coorientadores

LAVRAS – MG
2025

Ficha Catalográfica elaborada pelo Sistema de Geração de Ficha Catalográfica da Biblioteca Universitária da UFLA, com dados informados pelo(a) próprio(a) autor(a).

Figueiredo, Cristiane Nascimento .
Biodiversity and chitinase production of soil fungi in the Cerrado biome of Minas Gerais / Cristiane Nascimento Figueiredo. - 2025.
133 p. : il.

Orientador: Victor Satler Pylro
Coorientador: Jos Houbraken
Coorientador: Pedro Crous
Coorientador: Luís Roberto Batista

Tese (Doutorado) - Universidade Federal de Lavras, 2025.
Bibliografia.

1. enzymes. 2. filamentous fungi. 3. NGS. 4. new species. 5. taxonomy. I. Satler Pylro , Victor . II. Batista, Luís Roberto . III. Houbraken , Jos . IV. Crous, Pedro. V. Batista, Luís Roberto . VI. Universidade Federal de Lavras. VII. Título.

CRISTIANE NASCIMENTO FIGUEIREDO

BIODIVERSITY AND CHITINASE PRODUCTION OF SOIL FUNGI IN THE CERRADO BIOME OF MINAS GERAIS

BIODIVERSIDADE E PRODUÇÃO DE QUITINASE DE FUNGOS DO SOLO NO BIOMA CERRADO DE MINAS GERAIS

Tese apresentada à Universidade Federal de Lavras, como parte das exigências do Programa de Pós-Graduação em Microbiologia Agrícola, área de concentração em Microbiologia Agrícola para a obtenção do título de Doutora.

Aprovada em 26 de setembro de 2025.
Dr. Teotonio Soares De Carvalho UFLA
Dr. Jorge Teodoro de Souza UFLA
Dr. Leandro Nascimento Lemos CNPEM
Dr. Joziana Muniz de Paiva Barcante UFLA

Prof. DSc. Victor Satler Pylro
Orientador

LAVRAS – MG
2025

AGRADECIMENTOS

Após 4 anos de dedicação ao Curso de Doutorado em Microbiologia Agrícola, encerro um ciclo muito importante da minha trajetória profissional e pessoal. Começo agradecendo ao meu orientador, Professor Victor Pylro por todo suporte, confiança, ensinamentos e pela constante presença nessa longa jornada. Aos meus coorientadores Jos Houbraken e Pedro Crous, sou grata pela orientação, hospitalidade e ensinamentos durante o período de doutorado sanduíche no Instituto Westerdijk, Utrecht-Holanda. Meus agradecimentos também se estendem ao Professor Jorge Teodoro, ao meu coorientador, professor Luís Roberto e aos demais docentes e técnicos do programa do PPGMA, por cederem seus respectivos laboratórios, permitindo que esta pesquisa fosse desenvolvida. Agradeço aos professores Fátima Moreira e Teotonio Soares pela confiança e pelas contribuições relevantes nesse trabalho. Agradeço às equipes de Laboratórios, com menção especial a Bárbara, Mariana, Carlos, Caio, Camila, Wellington, Nicholas, Kelly, Giovanna, Harisson, Eduardo e Miriam pela colaboração e amizade.

Agradeço à Universidade Federal de Lavras (UFLA), ao Programa de Pós-Graduação em Microbiologia Agrícola, e à CAPES pela a concessão da bolsa de estudos no Brasil e no exterior por meio do Programa CAPES-PRINT. Este trabalho foi realizado com o apoio da Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Código de Financiamento 001. Além disso, este trabalho contou com o suporte financeiro do INCT Biodiversidade dos Solos (CNPq #406658/2022-6 e FAPEMIG #APQ-04011-24) e das Redes FAPEMIG (#RED-00181-23 e RED-00330-16). Agradeço aos profissionais de saúde mental, aos queridos psicólogos, Cintía e Pedro, por todo suporte, sensibilidade e cuidado.

Expresso minha profunda gratidão aos meus amados pais e as minhas queridas irmãs, pelo carinho e suporte incondicional, por sempre se fazerem presente em todos os momentos. Aos amigos que deixaram essa caminhada mais leve e inesquecível, tanto no Brasil quanto na Holanda Angélica, Sandy, Daniel, Cidinha, Daniela, Clara, Fabiana, Marcia, Harisson, Madalina, Lin, Yabin, Mariah, Mariana, Giovanna, Miriam, Hygor, Isabela, Welligton, Nicholas — agradeço o cuidado, troca e companheirismo. À minha família, pelo carinho. À família Lavrense com quem compartilhei a vida e o lar durante esse período — Mariana, Mariane, Maria Alice, Grazile, Hortênsia e Bianca. À minha doce Mara, pelo suporte incondicional, por acreditar em meu potencial, por seu amor e carinho. Ao meu amado e saudoso Diego, por todos os momentos de alegria compartilhados e por se fazer presente em todas as etapas desse trabalho, te levarei comigo até o fim. Por fim, agradeço profundamente a

todos os professores do programa e aos demais que contribuíram de alguma forma para a realização desse trabalho.

RESUMO

O Cerrado brasileiro é o segundo maior bioma do país e se estende principalmente na área central do Brasil, abrigando exuberante biodiversidade de plantas e animais. Entretanto, esse bioma já perdeu mais de 44% da sua vegetação nativa, em decorrência da mudança de uso do solo para produção de grãos, tais como soja, milho e café, além da produção de carne. O impacto da mudança no uso do solo sobre as comunidades fúngicas do Cerrado ainda é pouco compreendido, assim como a diversidade desses microrganismos e seu potencial enzimático. Desse modo, os principais objetivos deste estudo foram investigar e comparar a diversidade fúngica em solos de diferentes usos da terra, incluindo áreas agrícolas de cultivo de café regenerativo e convencional e áreas de vegetação nativa do Cerrado, no município de Patrocínio, em Minas Gerais. Além disso, objetivou-se testar o potencial quitinolítico e identificar, por taxonomia polifásica, isolados fúngicas oriundos do solo desses três diferentes sistemas de uso da terra. A técnica de diluição seriada foi utilizada para isolar um total de 1.481 cepas fúngicas de 60 amostras de solo. Alíquotas de 0,1 mL das diluições foram transferidas para meios de cultura padronizados, Dicloran Rosa Bengala Cloranfenicol (DRBC) e Dicloran Glicerol 18% (DG18), incubados a 25 °C por 7 dias. Por meio de Sequenciamento de Nova Geração (NGS), as amostras de solo oriundas do Cerrado nativo mostraram diferenças significativas entre as diversidades alfa e beta, as áreas do Cerrado nativo Ce1 e Ce2 apresentram a menor diversidade alfa entre as áreas analisadas (Chao1 consistentimente < 50). Entretanto, os grupos fúngicos foram similares entre as amostras do Cerrado, com maior abundância relativa do filo Basidiomycota, gêneros *Apiotrichum* e *Saitozyma*. Por outro lado, as áreas de cultivo de café, tanto convencional quanto regenerativo, apresentaram grupos fúngicos semelhantes, pertencentes ao filo Ascomycota. Em relação à produção de quitinase, o maior número de cepas fúngicas com atividade superior a 70 U min^{-1} foi isolado do solo de café convencional, sugerindo que o tipo de manejo pode influenciar a síntese dessa enzima. Sete novas espécies do gênero *Penicillium* foram descritas: *P. patrocinensis* (seção *Brevicompacta*), *P. guarae* (seção *Cinnamopurpurea*), *P. seriamae* (seção *Guizhouorum*), *P. beraoi* e *P. moreirae* (seção *Lanata-Divaricata*), *P. pequii* e *P. pluvialis* (seção *Ramosum*). Outros isolados foram identificados como registros de ocorrência de 29 espécies. Desse total, quatro foram identificadas utilizando sequências parciais dos genes *BenA, CaM e RPB2*, enquanto as 25 restantes foram identificadas apenas por meio da sequência parcial do gene *BenA*. Esses resultados reforçam que a mudança no uso do solo exerce forte impacto sobre as comunidades

fúngicas do Cerrado. Além disso, evidenciam que esse bioma constitui importante reservatório de novas espécies, cuja diversidade apresenta elevado potencial para aplicações biotecnológicas, especialmente na produção de enzimas hidrolíticas, como as quitinases.

Palavras-chave: enzimas; fungos filamentosos; NGS; novas espécies; taxonomia.

ABSTRACT

The Brazilian Cerrado is the second largest biome in the country and extends mainly across the central region of Brazil, harboring an exuberant biodiversity of plants and animals. However, this biome has already lost more than 44% of its native vegetation as a result of land-use change for the production of grains, such as soybeans, corn, and coffee, in addition to meat production. The impact of land-use change on the fungal communities of the Cerrado is still poorly understood, as is the diversity of these microorganisms and their enzymatic potential. Thus, the main objectives of this study were to investigate and compare fungal diversity in soils under different land uses, including agricultural areas of regenerative and conventional coffee cultivation, and areas of native Cerrado vegetation, in the municipality of Patrocínio, Minas Gerais. In addition, the study aimed to test the chitinolytic potential and to identify, through polyphasic taxonomy, fungal isolates obtained from the soil of these three different land-use systems. The serial dilution technique was used to isolate a total of 1.481 fungal strains from 60 soil samples. Aliquots of 0.1 mL of the dilutions were transferred to standardized culture media, Dichloran Rose Bengal Chloramphenicol (DRBC) and Dichloran Glycerol 18% (DG18), incubated at 25 °C for 7 days. Through Next-Generation Sequencing (NGS), soil samples from native Cerrado showed significant differences in both alpha and beta diversities; the native Cerrado areas Ce1 and Ce2 presented the lowest alpha diversity among the analyzed areas (Chao1 consistently < 50). However, fungal groups were similar among Cerrado samples, with a higher relative abundance of the phylum Basidiomycota, genera *Apiotrichum* and *Saitozyma*. On the other hand, coffee cultivation areas, both conventional and regenerative, presented similar fungal groups belonging to the phylum Ascomycota. Regarding chitinase production, the highest number of fungal strains with activity above 70 U min^{-1} was isolated from conventional coffee soil, suggesting that the type of management may influence the synthesis of this enzyme. Seven new species of the genus *Penicillium* were described: *P. patrocinensis* (section *Brevicompacta*), *P. guarae* (section *Cinnamopurpurea*), *P. seriamae* (section *Guizhouorum*), *P. beraoi* and *P. moreirae* (section *Lanata-Divaricata*), *P. pequii* and *P. pluvialis* (section *Ramosum*). Other isolates were identified as occurrence records of 29 species. Of this total, four were identified using partial sequences of the *BenA, CaM,* and *RPB2* genes, while the remaining 25 were identified only through the partial sequence of the *BenA* gene. These results reinforce that land-use change exerts a strong impact on the fungal communities of the Cerrado. Furthermore, they demonstrate that this biome constitutes an important reservoir of new species, whose diversity

presents high potential for biotechnological applications, especially in the production of hydrolytic enzymes such as chitinases.

Keywords: enzymes; filamentous fungi; NGS; new species; taxonomy.

INDICADORES DE IMPACTO

O desenvolvimento desse trabalho e os resultados obtidos contribuem no âmbito social, tecnológico, econômico e ambiental. Diante da constatação de que a mudança do uso do solo do Cerrado nativo impacta as comunidades fúngicas, demonstramos que, além da perda de uma rica biodiversidade de flora e fauna que correm risco de extinção, há potenciais riscos de perda de uma diversidade que ainda é parcialmente conhecida, os fungos do Cerrado. Do ponto de vista científico e tecnológico, identificamos sete novas espécies de *Penicillium* e diversos registros de ocorrência provenientes do Cerrado, o que amplia o conhecimento taxonômico dos fungos filamentosos dessa região, além de acessar um potencial enzimático pouco explorado com diversas aplicações biotecnológicas. Coletamos amostras de solo em fazendas, com a participação e o consentimento de produtores rurais, além de palestras enfatizando os resultados obtidos dos trabalhos desenvolvidos, favorecendo a aproximação entre a comunidade acadêmica e a comunidade local. Dessa forma, promovemos a compreensão da importância da biodiversidade microbiana do solo pela população local. Assim, fortalecemos a relação universidade-sociedade e geramos novos conhecimentos científicos, além de potenciais desenvolvimentos de biotecnologias baseadas em microrganismos do Cerrado. Esses impactos se inserem especialmente em áreas temáticas do Meio Ambiente, Tecnologia e Produção e Desenvolvimento Sustentável da Política Nacional de Extensão. Na área de Meio Ambiente, demonstramos o efeito da supressão da vegetação nativa nas comunidades fúngicas. No tema Tecnologia e Produção, destacamos a possibilidade de aplicar quitinases em práticas agrícolas sustentáveis. O trabalho também se enquadra na área de educação, formamos estudantes de graduação e pós-graduação envolvidos na pesquisa e promovemos atividades extensionistas por meio das coletas realizadas em fazendas, com interação direta com produtores rurais. Com relação aos Objetivos de Desenvolvimento Sustentável (ODS) da Organização das Nações Unidas (ONU), alinhamos o impacto desse trabalho com o ODS 2- Fome zero e agricultura sustentável: pela potencial contribuição ao desenvolvimento de práticas agrícolas mais sustentáveis por meio do uso de enzimas fúngicas quitinolíticas; ODS 12 - Consumo e produção responsáveis: incentivo ao desenvolvimento de alternativas sustentáveis na produção agrícola; ODS 13 - Ação contra a mudança global do clima: esse estudo demonstra o efeito da mudança do uso da terra nas

comunidades fúngicas do Cerrado; ODS 15 - Vida terrestre: descoberta de novas espécies de fungos filamentosos.

IMPACT INDICATORS

The development of this work and the results obtained contribute to the social, technological, economic, and environmental domains. In view of the finding that the change in land use from native Cerrado impacts fungal communities, we demonstrate that, in addition to the loss of a rich biodiversity of flora and fauna at risk of extinction, there are potential risks of losing a diversity that is still only partially known, the fungi of the Cerrado. From a scientific and technological perspective, we identified seven new species of *Penicillium* and several occurrence records from the Cerrado, which expand the taxonomic knowledge of filamentous fungi from this region, as well as access to an enzymatic potential that is still poorly explored, with diverse biotechnological applications. We collected soil samples on farms, with the participation and consent of rural producers, in addition to giving lectures emphasizing the results obtained from the developed works, promoting the connection between the academic community and the local community. In this way, we promoted the understanding of the importance of soil microbial biodiversity among the local population. Thus, we strengthened the university-society relationship and generated new scientific knowledge, in addition to potential developments of biotechnologies based on microorganisms from the Cerrado. These impacts are especially inserted in the thematic areas of Environment, Technology and Production, and Sustainable Development of the National Extension Policy. In the Environmental area, we demonstrated the effect of native vegetation suppression on fungal communities. In the Technology and Production theme, we highlighted the possibility of applying chitinases in sustainable agricultural practices. The work also fits into the field of Education, as we trained undergraduate and graduate students involved in the research and promoted extension activities through the collections carried out on farms, with direct interaction with rural producers. Regarding the United Nations (UN) Sustainable Development Goals (SDGs), we align the impact of this work with SDG 2 - Zero Hunger and Sustainable Agriculture: due to the potential contribution to the development of more sustainable agricultural practices through the use of fungal chitinolytic enzymes; SDG 12 - Responsible Consumption and Production: encouraging the development of sustainable alternatives in agricultural production; SDG 13 – Climate Action: this study demonstrates the effect of land-use change on fungal communities in the Cerrado; SDG 15 - Life on Land: discovery of new species of filamentous fungi.

SUMÁRIO

1. INTRODUÇÃO GERAL

Os fungos são altamente bem-sucedidos na colonização do solo em decorrência da sua notável plasticidade e habilidade de adaptação às condições adversas do ambiente. Assim, constituem um grupo diversificado, com centenas de espécies por grama de solo (FRAC et al., 2005; SCHLATTER et al., 2018). A maioria dos grupos fúngicos possui maior diversidade de espécies em ecossistemas tropicais, com exceção dos ectomicorrízicos e algumas classes desses microrganismos, cuja biodiversidade é mais elevada em ecossistemas temperados ou boreais (Tedersoo et al., 2014; Huang et al., 2023).

O Cerrado, um bioma tropical brasileiro, é considerado um *hotspot* da biodiversidade global, formado por um mosaico de diversas fitofisionomias que variam em composição e densidade de espécies e também é caracterizado por duas estações bem definidas ao longo do ano: um inverno seco e um verão chuvoso. O Cerrado cobre uma área aproximada de 2 milhões de km² e conecta-se à Caatinga, outro bioma brasileiro de vegetação aberta (MYERS et al., 2000; WERNECK, 2011). Estes dois biomas formam um corredor diagonal de florestas secas e, simultaneamente, constituem uma barreira biogeográfica entre as duas florestas tropicais úmidas, Amazônia e Atlântica (WERNECK, 2011).

O Cerrado é considerado a savana mais diversa do mundo e abriga rico patrimônio de recursos naturais, com milhares de espécies endêmicas adaptadas a solos ácidos e pobres em nutrientes (MYERS, 2000; FRANÇOSO et al., 2015; KLINK et al., 2020; MUNIZ et al., 2022). O Cerrado originalmente ocupava cerca de 24% do território brasileiro. No entanto, as mudanças no uso do solo, impulsionadas principalmente pela expansão da agricultura e da pecuária na região central do país, têm resultado na supressão de extensas áreas de vegetação nativa. Assim, a expansão da fronteira agrícola no Cerrado ameaça a existência desse bioma e de toda a sua biodiversidade, que permanece parcialmente desconhecida (RIBEIRO et al., 2009; IBGE, 2022). Apesar da grande importância ambiental e socioeconômica do bioma, a diversidade dos microrganismos do solo ainda é pouco documentada e explorada, especialmente no que diz respeito à biodiversidade fúngica (FALEIRO & LOYOLA, 2013; CALAÇA et al., 2020).

Os fungos, encontrados em todas as zonas climáticas do planeta, exercem um impacto amplo e profundo nos ecossistemas terrestres. Dentre os microrganismos presentes no solo, os fungos se destacam em decorrência de seu papel essencial nos processos biogeoquímicos, uma vez que atuam principalmente na decomposição da matéria orgânica e na ciclagem de nutrientes (BOER et al., 2005; CHOI & KIM, 2017).

Portanto, os fungos estão diretamente envolvidos na manutenção da fertilidade e na estruturação do solo, promovendo a agregação das partículas e, consequentemente, contribuindo para minimizar o processo de degradação desse substrato (REQUENA et al., 2001). A diversidade fúngica no solo é fortemente influenciada pelas condições climáticas e por elementos edáficos, tais como umidade, salinidade, serapilheira e pH (REQUENA et al., 2001; TARDY et al., 2015). As consequências para as comunidades fúngicas decorrentes da conversão de áreas de florestas tropicais nativas em terras agrícolas de monocultivo ainda não são bem compreendidas (BRINKMANN et al., 2019).

No estado de Minas Gerais, o Cerrado ocupava uma área de 54%, entretanto, as áreas remanescentes de vegetação nativa correspondem somente a 20,46% (IBGE, 2019; IEF, 2019). Entre as principais mudanças de uso do solo em áreas do Cerrado mineiro nativo, a produção de café se destaca. Essa cultura é cultivada em áreas extensas de monocultivo, utilizando principalmente, dois manejos de produção, regenerativo e convencional (BERNARDES, 2024). O cultivo regenerativo é um metódo que visa produzir alimentos de forma sustentável, preservando o meio ambiente, clima, recursos hídricos e saúde do solo (SHER et al., 2024). Por outro lado, o cultivo convencional é caracterizado por incluir práticas de revolvimento profundo do solo, aplicação de produtos químicos sintéticos, incluindo agrotóxicos e adubção, e organismos geneticamente modificados (GUILLEN-CRUZ et al., 2022).

Diante desse contexto, o objetivo deste trabalho foi investigar o impacto da mudança do uso do solo nas comunidades fúngicas do Cerrado e seu potencial na produção de enzimas quitinolíticas. Além disso, objetivou-se o isolamento e a identificação de fungos do solo, de áreas nativas do Cerrado e de áreas de cultivo de café regenerativo e convencional.

2. OBJETIVO

Este trabalho teve como objetivo investigar e comparar a diversidade fúngica em solos sujeitos a diferentes manejos e uso da terra, incluindo áreas agrícolas de cultivo de café regenerativo e convencional e áreas de vegetação nativa do Cerrado no município de Patrocínio, Minas Gerais.

2.1 Objetivos específicos

• Elucidar o impacto nas comunidades fúngicas do solo em função da conversão de áreas de Cerrado nativo em áreas de monocutivo de café, sob manejos convencional e regenerativo*;*

• Explorar o potencial biotecnológico quitinolítico dos isolados fúngicas oriundos do solo das áreas produtoras de café regenerativo e convencional e de Cerrado nativo;

• Identificar e descrever espécies por taxonomia polifásica de isolados pertencentes aos gêneros *Aspergillus, Talaromyces* e *Penicillium*;

3. REFERENCIAL TEÓRICO

3.1 Diversidade de fungos filamentosos e mudanças climáticas

Os fungos são os microrganismos pertencentes ao Reino Fungi e com base em estudos filogenéticos, estes organismos são agrupados em 18 filos, 23 subfilos, 74 classes, 215 ordens, 731 famílias e 5377 gêneros (TEDERSOO et al., 2018). Entretanto, outras análises sugerem que estes microrganismos possuem 12 filos e 224 ordens (JAMES et al., 2020). Aproximadamente 155.000 espécies fúngicas são conhecidas, aceitas e descritas, porém, a estimativa total do número de espécies fúngicas na Terra varia de 1.5 a 11 milhões de espécies (PHUKHAMSAKDA et al., 2022; KIRK, 2023; CAZABONNE et al., 2024) (WU et al., 2019; LUCKING et al., 2020).

Estes microrganismos evoluíram em três grandes papéis ecológicos, como saprófitos, parasitas e simbiontes mutualistas. Os saprófitos são os principais responsáveis pela decomposição da matéria orgânica, promovendo a ciclagem de nutrientes na biosfera (LIU et al., 2018). Os parasitas colonizam e infectam uma diversidade de espécies de plantas e animais (SEXTON & HOWLETT, 2006). Como mutualísticos, os fungos vivem em simbiose com diversos grupos de organismos como as algas e vegetais, formando os líquens e micorrizas, respectivamente (BLATRIX et al., 2009; TAYLOR et al., 2014). Esta associação mutualística com as plantas foi crucial para a colonização da Terra pelas gimnospermas e angiospermas na Terra (PIROZYNSKI & MALLOCH, 1975).

O Reino Fungi constitui o segundo maior grupo de eucariotos do planeta e são considerados organismos altamente diversificados e plásticos. Estes microrganismos são quimioheterotróficos e possuem larga variedade de ciclos de vida, morfogênese e metabolismos (CHOI & KIM, 2017). São amplamente distribuídos em todos os habitats e ecossistemas terrestres, e muitas espécies desse grupo são bem-adaptadas às condições ambientais extremas (CHAMBERGO & VALENCIA, 2016). Apesar da distribuição cosmopolita de múltiplas espécies desses microrganismos no globo terrestre, a maioria dos grupos taxonômicos fúngicos possuem maior diversidade e taxas de endemismo em ecossistemas tropicais (TEDERSOO et al., 2014). Nas áreas tropicais ocorrem as maiores atividades antropogênicas e consequentemente uma maior degradação do meio ambiente antropogênicas (MARTÍNEZ-RAMOS et al., 2016; ZHU et al., 2023).

As regiões tropicais estão situadas próximas à Linha do Equador, entre os trópicos de Câncer e Capricórnio e abrigam as florestas tropicais (SHANKAR & TRIPATHI, 2017).

Essas florestas são sumidouros de carbono e representam as comunidades mais ricas de espécies do planeta (PAN et al., 2011). Embora as populações florísticas e faunísticas estejam bem documentadas nesses ambientes, as comunidades microbianas, tais como as bactérias e os fungos são ainda poucas exploradas e parcialmente conhecidas (PEAY, BARALOTO & FINE, 2013). Além do conhecimento da diversidade fúngica ser limitado, padrões de vulnerabilidade em decorrência das mudanças ambientais, também permanecem desconhecidos, apesar desses microrganismos se mostrarem propensos a alterações da composição de suas comunidades no solo, diante de distúrbios ambientais climáticos e edáficos (TEDERSOO et al., 2022).

Assim como as plantas e animais, as comunidades fúngicas do solo podem ser sensíveis a fatores incluindo o estresse por altas temperaturas e secas prolongadas (Hannula & Veen, 2025). O número de espécies fúngicas incluídas na Lista Vermelha de Espécies Ameaçadas da IUCN™ ultrapassou 1.000, confirmando que a supressão contínua das florestas nativas resultado da mudança do uso do solo e o desenvolvimento urbano estão contribuindo para o declínio dessas espécies no planeta (disponível em: https://iucn.org/press-release/202503/first-1000-fungi-iucn-red-list-reveal-growing-threats-iucn-red-list).

Espécies fúngicas são especialmente importantes na composição da biodiversidade das florestas tropicais, pois são cruciais na decomposição da matéria orgânica, ciclagem de nutrientes, relações mutualísticas (micorrizas) e auxiliam na manutenção da diversidade de espécies vegetais nesses ecossistemas (SATISH, SULTANA & NANJUNDIAH, 2007; BAGCHI et al., 2014; AMMA et al., 2018). Portanto, a contínua degradação das florestas tropicais por diversos fatores, incluindo mudanças do uso do solo, extração de madeira, queimadas e secas severas, associadas às mudanças climáticas em curso, comprometem os serviços ecossistêmicos fundamentais para o equilíbrio da biosfera prestados por estes biomas e ameaçam a sua biodiversidade (BAYAS et al., 2022; LAPOLA et al., 2023). Assim, diversas espécies fúngicas endêmicas dessas regiões enfrentam potenciais riscos de extinção, incluindo espécies que ainda não foram descritas nesses biomas (BRINKMANN et al., 2019; LUGHADHA et al., 2020).

3.2 Importância e aplicação biotecnológica dos fungos filamentosos

Espécies de leveduras e de fungos filamentosos principalmente dos Filos Ascomycota e Basidiomycota, são essenciais nos mais diversos setores econômicos da sociedade e têm contribuído significativamente na saúde e bem-estar dos seres humanos, desde a antiguidade.

Embora os povos antigos não tivessem conhecimento dos processos fermentativos por microrganismos, a fabricação de bebidas fermentadas data desde 7.000 anos a.C na aldeia neolítica de Jiahu, China (MCGOVERN et al., 2004). Desde épocas passadas, leveduras do gênero *Saccharomyces* são utilizadas em larga escala na fabricação de pães, vinhos e cervejas (SICARD & LEGRAS, 2011). Outras espécies leveduriformes do gênero *Kluyveromyces*, com destaque para *Kluyveromyces lactis*, é considerada uma das leveduras mais importantes na indústria alimentícia, utilizada na produção de proteínas, metabólitos e enzimas que atuam na degradação da lactose (SPONHER et al., 2016).

Espécies de fungos filamentosos, tais como do gênero *Aspergillus* também se destacam na indústria de alimentos. *Aspergillus niger* é comercializado desde o ano de 1919 para produção de ácido cítrico, que é aplicado como aromatizante, conservante, acidulante e regulador de pH nas diversas indústrias alimentícias, bebidas e cosméticas (PAPAGIANNI, 2007; CAIRNS et al., 2018). As espécies *Aspergillus oryzae* e *Aspergillus sojae* são essenciais na produção de molhos de soja e koji, devido à capacidade de secretarem enzimas hidrolíticas da soja, conferindo sabores excepcionais ao produto final (BAMFORTH & COOK, 2019). Ainda na produção de alimentos, espécies de cogumelos e trufas são fontes de alimentos importantes na composição de uma dieta saudável e fungos filamentosos do gênero *Penicillium* são essenciais na produção de queijos. A espécie *Penicillium roqueforti* tem sido fundamental durante os últimos 4.000 anos na produção de queijos azuis, enquanto *Penicillium camemberti* é largamente utilizado na produção de queijos brancos (DEMAIN & MARTENS, 2017; ROPARS et al., 2020).

Os fungos também se destacam na produção de diversas enzimas e essa capacidade metabólica é explorada amplamente pelas indústrias. *Trichoderma reesei* é conhecido como o principal produtor de enzimas lignocelulolíticas, atuando na quebra da celulose (NOVY et al., 2019). Outras enzimas fúngicas, tais como proteases, pectinases, xilanases, lipases, amilases e fitases são comercialmente utilizadas em larga escala com ampla aplicação nas indústrias de papel, farmacêuticas, alimentícias, bio-branqueamento de polpa de madeira, bioprocessamento de têxteis, aditivos alimentares para aves e extração de óleos vegetais (KUHAD, GUPTA & SINGH, 2011; SINGH, 2019; BELLAOUCHI et al., 2021; JALIL et al., 2023). As enzimas utilizadas em diversas reações químicas como catalisadoras, contribuem na redução e gasto energético além de minimizar a poluição ambiental (MCKELVEY & MURPHY, 2017; LÓPEZ-LARGADA et al., 2025).

Além da produção de diversas enzimas importantes, os fungos também se destacam na produção de medicamentos. A penicilina, descoberta no ano de 1928, foi o primeiro

antibiótico utilizado no tratamento contra infecções acometidas por bactérias, principalmente durante a segunda guerra mundial (GAYNES, 2017). Esse antibiótico é produzido em larga escala industrialmente por espécies do gênero *Penicillium,* seção *Chrysogena* (HOUBRAKEN et al., 2012; SAWANT et al., 2024). A descoberta desse antibiótico impulsionou o desenvolvimento de mais pesquisas com fungos. Essas pesquisas levaram à descoberta de medicamentos, como ácido micofenólico, ciclosporina e griseofulvina (BROWN et al., 1976; FLOREY et al., 1946; MURTHY et al., 1999).

O ácido micofenólico é produzido por diversas espécies do gênero *Penicillium* incluindo *Penicillium brevicompactum, P. paxillin, P. rugulosum, P. canescens, P. roqueforti, P. viridicatum* and *P. glabrum* (WU et al., 2022). Esse medicamento possui ação antifúngica, antitumoral, antibacteriana e antipsoríase, além de atividade imunossupressora e antiviral contra o coronavírus 2 (KOBAYASHI et al., 2014; WANG et al., 2014; PATEL et al., 2020). Por outro lado, a ciclosporina, produzida principalmente por *Tolypocladium inflatum,* tem atividade imunossupressora e é amplamente utilizada no combate de doenças autoimunes (FALAH et al., 2024). As espécies filamentosas de *Penicillium griseofulvum* e *Aspergillus glaucus* são exploradas na produção do antifúngico griseofulvina e do composto anticancerígeno aspergiolida, respectivamente (TAO et al., 2009). Ainda na indústria farmacêutica, o microrganismo *Aspergillus japonicus* é utilizado na produção de probióticos (DEMAIN & MARTENS, 2017).

Na agricultura, os gêneros *Trichoderma, Beauveria, Metarhizium* e *Paecilomyces* são empregados como biopesticidas no controle biológico de fitopatógenos. Esses microrganismos atuam como alternativas sustentáveis à agricultura convencional, que depende intensamente do uso de agrotóxicos e adubação sintética. Assim, o uso desses grupos fúngicos contribuem para redução de impactos ambientais e promoção de sistemas agrícolas mais sustentáveis (POVEDA, ABRIL-URIAS e ESCOBAR, 2020; NASSARY, 2025). Ainda na agricultura, espécies dos gêneros *Aspergillus, Penicillium* e *Trichoderma* atuam na solubilização do fosfato, tornado esse macronutriente disponível para diversas culturas agrícolas, aumentando a peodução (LEI & ZHANG, 2015; YIN et al., 2015; LI et al., 2016).

Os fungos filamentosos também produzem uma diversidade de biocores que incluem várias classes químicas como, carotenóides, melaninas, flavinas, fenazinas, quinonas, monascinas, violaceína ou índigo (DUFOSSÉ et al., 2014). Esses corantes são uma alternativa sustentável aos corantes artificiais, uma vez que estes produzidos artificialmente são tóxicos, poluem o meio ambiente e têm efeitos carcinogênicos e imunossupressores (VENIL et al., 2020). Diferentemente do extrato de plantas e animais, o processo de produção dos pigmentos

fúngicos não depende de condições climáticas e sazonalidade, além de apresentarem baixo custos e possuem propriedade biodegradáveis, fácil processamento e resistência contra agentes bióticos e abióticos (EISENMAN & CASADEVALL, 2012; SEN et al., 2019; VENIL et al., 2020; DE MELO et al., 2024).

Espécies do gênero *Monascus* são utilizadas largamente em indústrias para a produção de três categorias de pigmentos vermelhos, amarelos e laranjas (CHAUDHARY et al., 2021). Esses pigmentos são utilizados como corantes em alimentos (LIN et al., 2008; MAPARI, THRANE & MEYER, 2010). A ampla diversidade metabólica funcional fúngica, destaca a importância e relevância não só ambiental desses microrganismos, mas também nos diversos setores da sociedade. Além da contribuição histórica no aumento da expectativa de vida e bem estar da sociedade, esses microrganismos possuem grande potencial para o desenvolvimento de soluções sustentáveis e inovadoras frente às demandas contemporâneas (DEMAIN & MARTENS, 2017).

3.3 Classificação e nomenclatura: breve história do gênero *Penicillium*

O gênero *Penicillium* pertence a ordem Eurotiales e possuem uma distribuição mundial com elevada plasticidade, conferindo adaptação nos mais diversos habitats ecológicos (TSANG et al., 2018). Muitas espécies desse gênero são de extrema importância com alto impacto social nas áreas da biotecnologia, médica e econômica (ADRIO & DEMAIN, 2003). Por exemplo, *Penicillium roqueforti* e *Penicillium camemberti* são explorados na produção de queijos azuis e brancos, respectivamente, enquanto *Penicillium rubens* é largamente utilizado na indústria farmacêutica na produção de antibióticos derivados de β-lactâmicos (POHL et al., 2020).

Penicillium foi introduzido por Heinrich Link em 1809 para descrever fungos assexuados que apresentavam estruturas reprodutivas semelhantes a um ‘penicillus’ (pincel) (NGUYEN & PHAM, 2021). Embora, este gênero tenha sido descrito originalmente como assexuado, algumas espécies de *Penicillium* produzem estruturas reprodutivas sexuadas (OJEDA-LÓPEZ et al., 2018; BÖHM et al., 2013). Em 1955, Benjamin introduziu o subgênero *Talaromyces* para incluir espécies de *Penicillium* que produzem conidióforos biverticilados simétricos e ascos contendo ascocarpos dispostos em cadeias curtas. Entretanto, no ano de 2011 Houbraken & Samson fizeram uso do princípio “um fungo - um nome” e baseados em uma filogenia de quatro genes (RPB1, RPB2, Tsr1 e Cct8) seguindo o princípio GCPSR (Genealogical Concordance Phylogenetic Species Recognition), mostraram que as

espécies pertencentes aos subgêneros *Penicillium biverticillium* e *Talaromyces* formam um clado monofilético distinto do clado monofilético de *Penicillium sensu stricto,* formado pelos subgêneros *Aspergilloides*, *Furcatum*, *Penicillium* e *Eupenicillium*. Além disso, o estudo também propôs a reorganização da antiga família *Trichocomaceae* em três famílias monofiléticas: *Aspergillaceae, Thermoascaceae* e *Trichocomaceae*.

Penicillium e *Talaromyces* foram reagrupados em famílias distintas, *Aspergillaceae* e *Trichocomaceae,* respectivamente (VISAGIE et al., 2024). Espécies pertencentes ao gênero *Talaromyces* apresentam estruturas reprodutivas sexuadas com paredes sem rigidez e compostas por múltiplas camadas de hifas entrelaçadas (HOUBRAKEN & SAMSON, 2011). Os ascomas amadurecem de forma mais rápida, dentro de algumas semanas. Em *Penicillium*, ocasionalmente as espécies produzem estruturas reprodutivas sexuadas e são semelhantes a esclerócios, formadas por células de paredes grossas que conferem rigidez e maior resistência. A maturidade pode durar meses e, frequentemente, os ascósporos não se formam (HOUBRAKEN & SAMSON, 2011). Com relação as fiálides, espécies de *Talaromyces* possuem essas estruturas mais estreitas, conhecidas como aculeadas ou lanceoladas, enquanto as espécies do gênero *Penicillium* apresentam essas estruturas mais largas, e são classificadas como ampuliformes (SAMSON et al., 2011). Dessa forma, as espécies de *Penicillium* e *Talaromyces* refletem uma divergência evolutiva dentro da ordem Eurotiales.

O gênero *Penicillium* era classificado tradicionalmente de acordo com os caracteres morfológicos. Entretanto, a identificação de espécies fúngicas sofreu alterações ao longo dos anos, devido a fatores que dificultam a distinção das espécies, tais como limitação do número de caracteres, pouca variabilidade intra e interespecífica, evolução convergente e espécies crípticas (RAJA et al., 2017). Assim, a diversidade taxonômica desse grupo de microrganismos pode ser subestimada. Desse modo, o sistema de classificação usado para a identificação de fungos mais preciso é o método de abordagem polifásica, que, além de aplicar a caracterização morfológica estritamente padronizada, envolve estudos filogenéticos multigênicos e dados de extrólitos (VISAGE et al., 2014). A identificação polifásica das espécies revolucionou a taxonomia fúngica e o esquema de classificação de um número expressivo de espécies já descritas do gênero *Penicillium* foi revisado (TSANG et al., 2018).

Penicillium possui 483 espécies descritas aceitas (VISAGIE et al., 2024). As espécies desse gênero são caracterizadas pelo padrão de ramificação apresentado por suas estruturas reprodutivas assexuadas. Desse modo, os conidióforos podem ser classificados em: conidióforos com fiálides solitárias, monoverticilado, divaricato, biverticilado, terverticilado e quaterverticilado (VISAGIE et al., 2024). O gene ITS é o “barcode” oficial para identificação

de espécies do gênero *Penicillium*, entretanto, em decorrência da limitação dessa região na distinção de espécies filogeneticamente relacionadas, o gene da β-tubulina (*BenA*) é considerado o melhor "barcode" secundário (VISAGIE et al., 2014).

Assim, o gênero *Penicillium* é considerado um dos grupos fúngicos mais importantes, principalmente devido às suas diversas aplicações biotecnológicas. Avanços na taxonomia desse gênero, impulsionados pela abordagem polifásica, permitem maior resolução na identificação e classificação de espécies, contribuindo para o conhecimento da sua diversidade, ecologia, evolução, além da possibilidade de descobrir novos metabólitos com o uso em diferentes áreas.

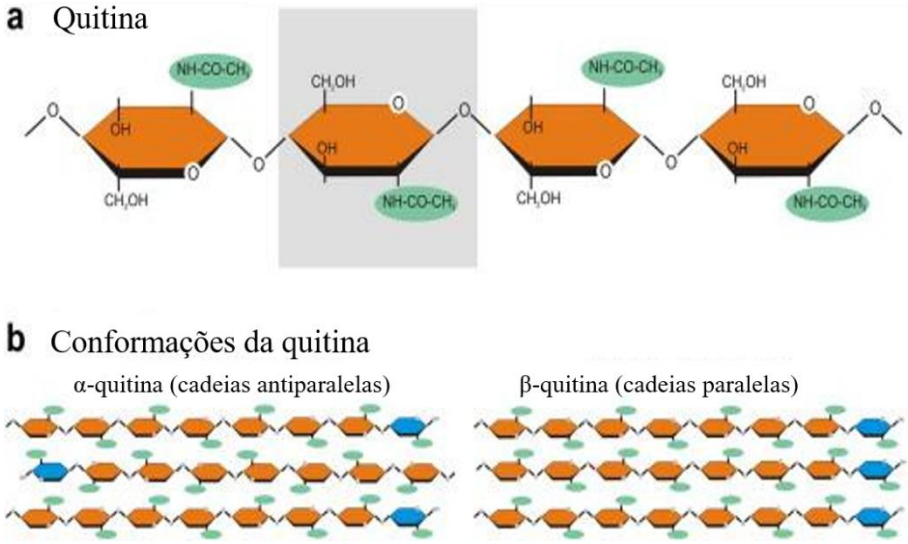
3.4 Quitina e quitinases e suas aplicabilidades

O termo quitina é oriundo de uma palavra grega que significa túnica, uma forma de vestimenta usada na Grécia Antiga. Esse biopolímero foi descoberto pela primeira vez em 1811 como uma substância encontrada em cogumelos, por Henri Braconnot (HARTL et al. 2011). A quitina é o segundo polissacarídeo mais abundante do planeta, considerada um biopolímero natural amplamente distribuído e pode ser encontrada nas principais fontes, nematóides, fungos, crustáceos, algas, insetos e moluscos (BHATTACHRYA et al., 2007; KARLSSON & STENLID 2008). Este biopolímero é composto por N-acetil D-glucosaminas (NAG) conectadas por ligação β-1,4-glicosídica (Figura 1) (KOTB, 2023). Na natureza, foram encontradas três formas cristalinas diferentes de quitina: α-, β- e γ (JANG et al., 2004).

As cadeias de α-quitina são a forma mais comum e ocorrem em estruturas rígidas, podendo ser encontradas em diferentes grupos de organismos, incluindo Arthropoda, Porifera, Bryozoa e nas paredes celulares de fungos (MINKE, BLACKWELL, 1978; WYSOKOWSKI et al., 2013). As formas de β- e γ-quitina são encontradas em estruturas flexíveis como as estruturas esqueléticas dos cefalópodes, parede celular das diatomáceas e no estômago da lula, respectivamente (KARLSSON & STENLID 2008, WANG et al. 2021).

A quitina e seu derivado desacetilado, a quitosana, são utilizados em diversas aplicações biomédicas devido às suas propriedades biodegradáveis, antimicrobianas, hidratantes e não tóxicas (JAYAKUMAR et al., 2010). Esses biopolímeros podem ser processados em diversos subprodutos, incluindo esferas (YUSOF, KHOR, 2000).

Figura 1 – Estrutura química da quitina.



Estrutura química da quitina. (a) A caixa cinza indica uma subunidade N-acetilglicosamina da cadeia de quitina. (b) Os dois principais tipos de quitina são caracterizados por um arranjo antiparalelo (α-quitina) e paralelo (β-quitina) das cadeias.

Fonte: adaptada de: Seidl, 2008.

A quitina é uma molécula insolúvel em água e em outras soluções; entretanto, este biopolímero pode ser decomposto por quitinases (E.C. 3.2.1.14). Essas enzimas são encontradas em diversos organismos, incluindo fungos, plantas e bactérias (KARTHIK et al., 2017). Em plantas e bactérias, as quitinases são cruciais principalmente nos mecanismos de defesa contra fungos e insetos (GOUGHENOUR et al., 2021; OMARI et al., 2024).

Em fungos, as quitinases desempenham papéis importantes na morfogênese, participando de atividades como o crescimento de hifas, a remodelação da parede celular durante os diferentes estágios da germinação de esporos e na competição ou defesa contra outros fungos (micoparasitismo), insetos (fungos entomopatogênicos) e nematoides (fungos que capturam nematoides) (RATHORE & GUPTA, 2015).

Em leveduras, a quitina representa apenas 1 a 2% do peso seco das células, embora em fungos filamentosos essa proporção possa chegar a 40% (BARTNICKI-GARCIA & LIPPMAN, 1969; LIU et al., 2021). A forma derivada da quitina, a quitosana, pode ser utilizada como fonte de carbono e nitrogênio por muitos grupos fúngicos (GÓMEZ-GAVIRIA & MORA-MONTES, 2024). Fungos entomopatogênicos têm a capacidade de infectar e matar insetos por meio de adesinas e enzimas líticas, incluindo quitinases, proteases e lipases (PAVEDA, 2021). O uso desses fungos como biopesticidas é uma alternativa ao processo de cultivo tradicional, que utiliza pesticidas e fungicidas em excesso (YAO et al., 2023). Alguns gêneros de fungos, como *Trichoderma*, *Metarhizium* e *Beauveria* são

conhecidos por controlar insetos-praga em lavouras (LITWIN, NOWAK, RÓŻALSKA, 2020).

Enquanto em genomas bacterianos existem de 2 a 4 genes codificadores de quitinases, os fungos filamentosos tipicamente possuem entre 10 e 25. As razões pelas quais os fungos apresentam essa quantidade ainda não foram bem elucidadas (SEIDL 2008). Essas quitinases fúngicas pertencem à família 18 das glicosídeos hidrolases (GH18) e possuem dominios e ordem distintas, resultando em uma grande diversidade estrutural e funcional (SEIDL 2008). O catabolismo da quitina ocorre em duas etapas, primeiramente envolve a clivagem do polímero por quitinases em oligossacarídeos, em seguida ocorre uma segunda clivagem em N-acetilglucosamina e monossacarídeos. As enzimas quitinolíticas são divididas em dois grupos principais: endoquitinases (E.C. 3.2.1.14) e exoquitinases. As endoquitinases dividem a quitina aleatoriamente em sítios internos, formando assim o dímero di-cetilquitobiose e multímeros solúveis de baixa massa molecular de GlcNAc. As exoquitinases foram divididas em Quitobiosidases (E.C. 3.2.1.29), que atuam na catalisação da liberação progressiva de di-acetilquitobiose começando na extremidade não redutora da microfibrila de quitina, e 1-4-β-glucosaminidases (E.C. 3.2.1.30), que quebram os produtos oligoméricos de endoquitinases e quitobiosidases, resultando em monômeros de GlcNAc (THAKU et al. 2023).

Anualmente, toneladas de quitina são produzidas na biosfera, resultado principalmente do alto consumo de camarão e caranguejo (DHILLON et al. 2013). A maioria desses resíduos quitinosos são incinerados ou aterrados, contribuindo significativamente na poluição ambiental (THOMAS et al. 2022). Em geral, a conversão química de resíduos quitinosos em quitina requer o uso de ácidos e bases fortes, como NaOH e HCl (HAHN et al. 2022). Esses metódos são prejudiciais ao meio ambiente e à saúde humana, além de impactar negativamente no produto final (KIM & PARK, 2015).

Em ecossistemas naturais, a degradação da quitina é realizada predominantemente por quitinases microbianas (THAKUR et al. 2023). Dessa forma, encontrar microrganismos produtores de potentes quitinases é fundamental para diversas aplicabilidades, incluindo solucionar o crescente problema do lixo marinho, além de possibilitar novos bioinsumos aplicados em uma produção agrícola sustentável.

3.5 Microbioma

O termo microbioma foi introduzido por Whipps e colaboradores em 1988 a partir de estudos ecológicos na rizosfera de plantas (BERG et al., 2020). O microbioma compreende

toda a comunidade de microrganismos vivos de um dado ambiente e moléculas produzidas por esses microrganismos, incluindo seus elementos estruturais (ácidos nucleicos, proteínas, lipídios, polissacarídeos), metabólitos (moléculas sinalizadoras, toxinas, moléculas orgânicas e inorgânicas) e moléculas produzidas por hospedeiros coexistentes (MARCHESI; RAVEL, 2015). Portanto, todos os elementos genéticos móveis, como fagos, vírus e DNA extracelular, devem ser incluídos no termo microbioma, tais como bactérias, archaea, fungos, algas e pequenos protistas que vivem em um dado ambiente (MARCHESI & RAVEL, 2015). Os microrganismos são extremamente abundantes nos solos e compõem o microbioma mais diversificado e complexo da Terra, podendo ser encontrado mais de 50.000 espécies por grama desse substrato (FIERER, 2017). Bactérias e fungos são geralmente os microrganismos dominantes no solo com mais biomassa do que protistas e archaea (BAR-ON, PHILLIPS & MILO, 2018). Além disso, um grama de solo pode conter 10^7 –10^9 partículas virais (FIERER, 2017).

A saúde do solo integra o conceito de saúde única, entretanto a derrubada das florestas nativas e a mudança do uso da terra alteram os atributos microbianos desse substrato (composição das comunidades microbianas e respiração basal do solo que controla a perda de carbono dos ecossistemas terrestres para a atmosfera) (BOND-LAMBERTY &THOMSON, 2010; CHEN, YANG & CHENG, 2022). O microbioma é um componente essencial do ecossistema do solo, influenciando processos como formação e fertilidade desse substrato, crescimento das plantas e tolerância ao estresse, renovação de nutrientes e armazenamento de carbono (DAN NAYLOR, RYAN MCCLURE & JANET JANSSON, 2022). Somado a esse fator, as mudanças climáticas antropogênicas ameaçam diretamente as contribuições microbianas aos serviços ecossistêmicos e na saúde do solo, plantas e animais (incluindo os humanos) - saúde única (BANERJEE & HEIJDEN, 2023). Nos últimos anos, o aumento do desmatamento das florestas e a mudança de uso da terra têm resultado substancialmente na degradação dos solos em decorrência da erosão, compactação e contaminação por agrotóxicos (TAMBURINI et al., 2020). O papel das comunidades microbianas deve ser compreendido e utilizado na proteção e melhoria da saúde do solo, práticas como diversificação de culturas, redução de agrotóxicos, fertilizantes sintéticos e cultivo intensivo contribuem para na manutenção da biodiversidade e a saúde do solo (FIERER, WOOD & MESQUITA, 2021).

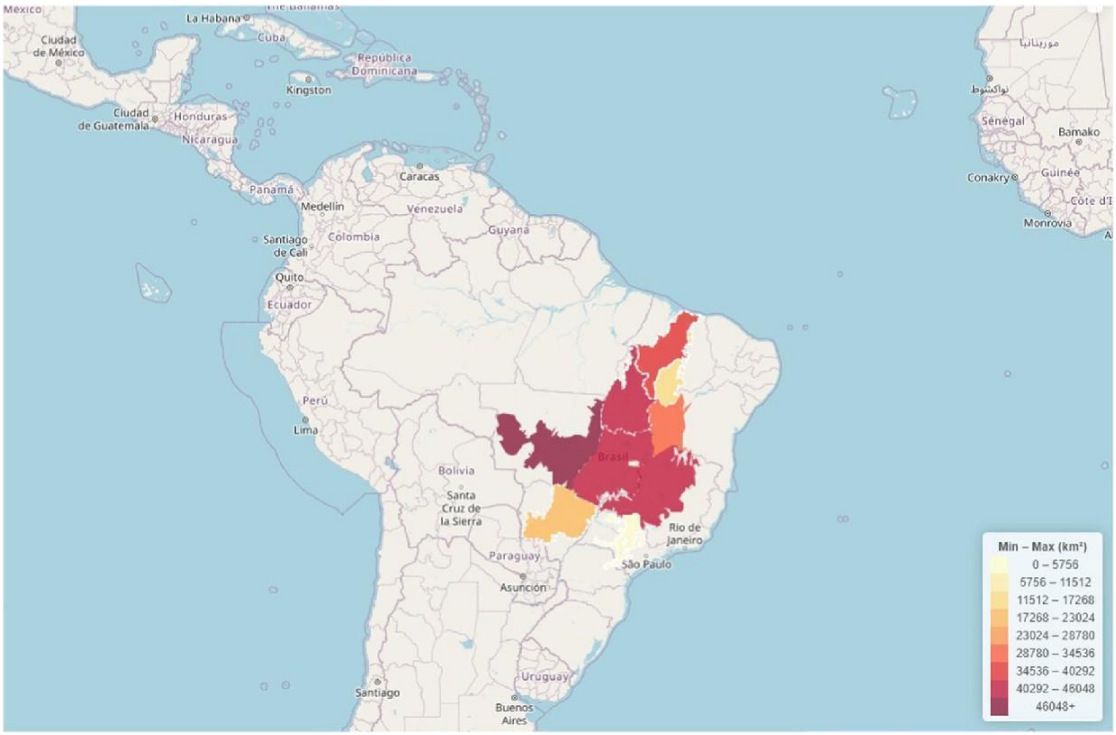
3.6 Bioma Cerrado

O Brasil é considerado um país tropical visto que a maior parte do seu território situa-

se entre a linha do equador e o trópico de capricórnio (MARTINELLI et al., 2021). Devido à posição no planeta, essa região é iluminada abundantemente o ano inteiro pelos raios solares e a precipitação é maior que 1.300 mm em grande parte do país. Esses fatores resultam em uma mega diversidade de flora e fauna, que se destaca mundialmente (MARTINELLI et al., 2021). O Brasil possui seis biomas bem definidos, e dentre esses, o Cerrado é o segundo maior bioma do país, formado por vários ecossistemas, desde campos abertos até florestas secas (KLINK et al., 2020). Essas mudanças na fitofisionomia não são determinadas pelo nível de pluviosidade, como observado nas savanas africanas, mas sim por fatores edáficos como fertilidade, grau de saturação e concentração de alumínio do solo e alterações causadas pelo fogo (LOPES & COX, 1977; AMORIM & BATALHA, 2006). Assim como em outras savanas tropicais, o clima do Cerrado é sazonal e altamente previsível, com duas estações bem definidas ao longo do ano: verão chuvoso de outubro a abril e inverno seco de maio a setembro (GARCIA et al., 2015).

Os solos são predominantemente do tipo latossolos, antigos, pobres em nutrientes, com altos níveis de ferro e alumínio e ácidos (pH 4.0-5.5) (VENDRAME et al., 2013). Entretanto, este bioma é a savana que exibe maior riqueza em diversidade botânica do mundo, abrigando 12.000 espécies de plantas nativas e várias espécies de insetos, peixes, anfíbios, aves e répteis endêmicos (KLINK & MACHADO, 2005; SANO, DE ALMEIDA, RIBEIRO, 2008). Localizado no planalto central do Brasil, o cerrado ocupa uma área de 2.036.448 Km^2 e abriga as cabeceiras e grandes porções de bacias hidrográficas importantes da América do Sul, os rios Paraguai-Paraná, Araguaia-Tocantins e São Francisco (FERREIRA et al., 2013). O cerrado incide nos estados de Piauí, Bahia, Maranhão, Goiás, Tocantins, Mato Grosso, Mato Grosso do Sul, Minas Gerais, Paraná, e São Paulo (Figura 2). Atrás somente da Mata Atlântica, o cerrado é o segundo bioma brasileiro que mais sofreu alterações com a ocupação humana. Com o aumento do consumo de alimentos, novas áreas são constantemente ocupadas e desmatadas para a produção de grãos e carnes, levando ao esgotamento progressivo dos recursos naturais da região (fig. 2) (DE CASTRO et al., 2008). A expansão agrícola tem convertido os ecossistemas em grandes áreas de monocultura de soja, algodão, café e eucalipto (LOPES, LIMA & REIS, 2021).

Figura 2 – Mapa do Cerrado no Brasil.



Mapa de distribuição do bioma Cerrado no Brasil e taxas de desmatamento de áreas nativas em quilômetros quadrados.

Fonte: INPE, 2020

As atividades antropogênicas praticadas recorrentes nessas áreas agrícolas resultam no desgaste do solo, por meio de diversos processos como compactação por maquinário, adubação química, salinização, contaminação por pesticidas, além de acidificação, diminuição de nutrientes no solo (AVELLANEDA-TORRES et al., 2022).

A composição e diversidade fúngica do solo pode ser impactada negativamente em áreas agrícolas, podendo resultar na perda de espécies. Esse fator aliado às mudanças climáticas que estão em curso podem atuar na completa extinção de linhagens fúngicas específicas (LUGHADHA et al., 2020). Em florestas tropicais esses fatores são mais críticos, já que estes ambientes são reservatórios de maior diversidade fúngica e taxa de endemicidade desses microrganismos. No Cerrado, considerado um dos biomas mais ameaçados do mundo e onde o conhecimento da diversidade fúngica é ainda limitado, taxóns fúngicos únicos correm risco de extinção.

3.7 Cultivo de café regenerativo e convencional

O café é a segunda bebida mais consumida do mundo (BAQUETA et al., 2024).

Pertencente à família Rubiaceae, e embora 124 espécies de plantas produtoras desse grão são reconhecidas, somente duas espécies são as principais cultivares utilizadas no mundo para a produção da bebida, *Coffea arabica* e *C. canephora* (YANG et al., 2011; DIAS et al., 2013, ASIALA et al., 2023). Essas espécies são arbustos tropicais perenes de origem africana com pequenas diferenças entre si, entretanto, o Arábica é considerado uma bebida mais aromática e saborosa quando comparada com a variedade Robusta, que por sua vez já apresenta mais resistência e, portanto, sua produção é considerada mais barata (SANTOS et al., 2021). A produção de cafezais ocorre principalmente de forma convencional, embora áreas de café regenerativas estão aumentando nos últimos anos (PONCET et al., 2024).

A produção de café convencional busca a alta produtividade e baixos custos e é caracterizada principalmente por extensas áreas de monocultura, com o uso intensivo de fertilização química do solo e agrotóxicos no controle de doenças (AZABARD, 2022; SUMBERG & GILLER, 2022). Esse tipo de produção impacta negativamente a saúde do solo, além de contribuir diretamente na emissão de gases do efeito estufa (PONCET et al., 2024). Por outro lado, a produção regenerativa visa sustentabilidade ambiental e alta produção baseada em interações biológicas, plantio direto, cobertura verde sem uso de adubação sintética e uso de pesticidas (GILLER et al., 2021). Algumas práticas realizadas em sistemas regenerativos são comuns em sistemas de produção orgânica e de agroflorestas (LE et al., 2021). Embora ainda haja confusão entre as práticas implementadas pelo os produtores e a teoria da agricultura regenerativa, esse sistema regenerativo na maioria das vezes se alinha com sustentabilidade, distinguindo de sistemas agrícolas convencionais (ALEXANDERSON et al., 2023).

O Brasil se destaca no cenário mundial como o maior produtor e exportador de café (VOLSI et al., 2019). Esta cultura foi introduzida no país em 1727, e no ano de de 1850 o Brasil já era responsável por 40% da produção mundial (MAPA, 2025). Ainda de acordo com o Ministério da da Agricultura e Agropecuária, a produção cafeeira é responsável pela produção de 8 milhões de empregos diretos e indiretos no País. E segundo dados da Associação Brasileira da Indústria de Café, no ano de 2024 o faturamento interno atingiu 36,82 bilhões (ABIC, 2024). Dentre os estados brasileiros, Minas Gerais se destaca como o maior produtor de café, responsável por 68 % da produção nacional, seguido por São Paulo com 16 % (DIAS, MARTINS, MARTINS, 2024).

As condições climáticas que favorecem o crescimento das plantas cafeeiras e alta produtividade, são temperaturas entre 14 e 26 °C, precipitação entre 1000 e 2700 mm com períodos secos de 1 a 3 meses, anualmente (DAVIS et al., 2006). Entretanto, devido às

mudanças climáticas em curso, as culturas cafeeiras têm experimentado condições climáticas adversas, tais como secas severas, longos períodos chuvosos e ondas de calor (VINECKY et al., 2017). A variabilidade climática é o fator principal que interfere nas flutuações de produtividade agrícola, por tanto mitigar os impactos (SANTOS et al., 2021). Diante da relevância econômica e social do café no Brasil e no mundo, é essencial estabelecer práticas de produção sustentável. Nesse contexto, sistemas agrícolas tais como orgânicos, agroflorestais e regenerativos surgem como alternativas a pratica convencional, e são capazes de promover a saúde do solo, redução de emissões de gases do efeito estufa e manutenção da produtividade em longo prazo.

REFERÊNCIAS

ADRIO, J., L.; DEMAIN, A., L. Fungal biotechnology. **International Microbiology**, v. 6, p.191–199, 2003.

ALEXANDERSON, MS.; LUKE, H.; LLOYD, DJ. Regenerative farming as climate action. **Journal of Environmental Management**, v. 347, 2023.

AMMA, S.; TOJU, H.; WACHRINRAT, C.; SATO, H.; TANABE, A., S.; ARTCHAWAKOM, T.; KANZAKI1, M. Composition and Diversity of Soil Fungi in Dipterocarpaceae-Dominated Seasonal Tropical Forests in Thailand. **Microbes and Enviroments**, v. 33, v. 2, p. 135-143, 2018.

AMORIM, P. K.; BATALHA, M. A. Soil characteristics of a hyperseasonal cerrado compared to a seasonal cerrado and a floodplain grassland: implications for plant community structure. **Brazilian Journal of Biology**, v. 66, p. 661-670, 2006.

ASIALA, H.; KARKKAINEN, E., JOKINEN, L.; LAAKSO, T., S.; RISCHER, H. Proof of Concept for Cell Culture-Based Coffee. **Journal Agricultural Food Chem**., v. 71, n. 47, 2023.

AZARBAD H. Conventional vs. Organic Agriculture-Which One Promotes Better Yields and Microbial Resilience in Rapidly Changing Climates? **Frontiers in Microbiology**, v. 9, n. 13, 2022. Doi: 10.3389/fmicb.2022.903500.

BAGCHI, R.; GALLERY, R., E.; GRIPENBERG, S.; GURR, S., J.; NARAYAN, L.; ADDIS, C., E.; FRECKLETON, R., P.; LEWIS, W., T. Pathogens and insect herbivores drive rainforest plant diversity and composition. **Nature**, v. 506, p. 85-88, 2014.

BAMFORTH, C., W.; COOK, D., **J. Food, Fermentation, and Micro-organisms**. 2ª Edição. Fevereiro, 2019.

BANERJEE, S.; HEIJDEN, M., G., A., V. Soil microbiomes and one health. **Nature Reviews Microbiology**, v. 21, p. 6–20, 2023.

BAQUETA MR., DINIZ PHGD., PEREIRA LL.; ALMEIDA, FLC.; VALDERRAMA, P.; PALLONE, JAL. An overview on the Brazilian *Coffea canephora* scenario and the current chemometrics-based spectroscopic research. **Food Research International**, v. 194, 2024.

BAR-ON, Y., M.; PHILLIPS, R.; MILO, R. The biomass distribution on Earth. **PNAS**, v. 115, n. 25, 2018.

BARTNICKI-GARCIA, S., AND E. LIPPMAN. Fungal morphogenesis: cell wall construction in Mucor rouxii. **Science**, v.165, p. 302-304, 1969.

BAYAS, J., C., L.; SEE, L.; GEORGIEVA, I.; SCHEPASCHENKO, D.; DANYLO, O.; DÜRAUER, M.; BARTL, H.; HOFHANSL, F.; ZADOROZHNIUK, R.; BURIANCHUK, M.; SIRBU, F.; MAGORI, B.; BLYSHCHYK, K.; BLYSHCHYK, V. Drivers of tropical forest loss between 2008 and 2019. **Scientific Data** n. 146, v. 9, 2022.

https://doi.org/10.1038/s41597-022-01227-3

BELLAOUCHI, R., ABOULOIFA, H., ROKNI, Y. et al. Characterization and optimization of extracellular enzymes production by Aspergillus niger strains isolated from date by-products. J **Genet Eng Biotechnol**, v. 19, n. 50, 2021. DOI: https://doi.org/10.1186/s43141-021-00145-y

BERG, G.; RYBAKOVA, D.; FISHER, D.; CERNAVA, T.; VERGÈS, M.-C., C.; CHARLES, T.; CHEN, X., Y.; COCOLIN, L.; EVERSOLE, K.; CORRAL, G., H. Microbiome definition re-visited: old concepts and new challenges. **Microbiome**, v. 8, n. 103, 2020.

BERNARDES, A. Cerrado Mineiro region has the largest regenerative coffee growing area in Brazil. **Revista Cultivar**, 24 maio 2024. Disponível em: https://revistacultivar.com/news/The-Cerrado-region-of-Minas-Gerais-has-the-largest-regenerative-coffee-growing-area-in-Brazil. Acesso em: 09 de Outurbo de 2025.

BHATTACHRYA D, NAGPURE A, GUPTA RK. Bacterial chitinase: Properties and potential. **Crit Rev Biotechnol**, p. 21–8, 2007. doi: 10.1080/07388550601168223.

BLATRIX, R.; BOUAMER, S.; MORAND, S.; SELOSSE, M., A. Ant-plant mutualisms should be viewed as symbiotic communities. **Plant Signal Behay**, v. 4, n. 6, p. 554-556, 2009.

BOND-LAMBERTY, B.; THOMSON, A. Temperature-associated increases in the global soil respiration record. **Nature**, v. 464, p. 579-582, 2010.

BRINKMANN, N.; SCHNEIDER, D.; SAHNER, J.; BALLAUFF, J.; EDY, N.; BARUS, H.; IRAWAN, B.; BUDI, S., W.; QAIM, M.; DANIEL, R.; POLLE, A. Intensive tropical land use massively shifts soil fungal communities. **Scientific Reports**, v. 9, n. 3403, 2019.

BROWN, A. G.; SMALE, T. C.; KING, T. J.; HASENKAMP, R.; THOMPSON, R. H. Crystal and molecular structure of compactin, a new antifungal metabolite from *Penicillium brevicompactum*. **Journal of the Chemical Society, Perkin Transactions** v. 1, n. 11, p. 1165-1170, 1976.

BOER, W.; FOLMAN, L., B.; SUMMERBELL, R., C.; BODDY, L. Living in a fungal world: impact of fungi on soil bacterial niche development. **FEMS Microbiology Reviews**, v. 29, n. 4, p. 795–811, 2005.

BÖHM, J.; HOFF, B.; O'GORMAN, C., M.; WOLFERS, S.; KLIX, V.; BINGER, D.; ZADRA, I.; KÜRNSTEINER, H.; PÖGGELER, S.; DYER, P., S.; KÜCK, U. Sexual reproduction and mating-type–mediated strain development in the penicillin-producing fungus *Penicillium chrysogenum*. **PNAS**, v. 110, n. 4, p. 1476-1481, 2013.

BOREL, J., F.; FEURER, C.; GUBLER, H., U.; STÄHELIN, H. Biological effects of cyclosporin A: A new antilymphocytic agent. **Agents and actions**, v. 6, p. 468-475,1976.
BUTLER, R., A. The Amazon Rainforest: The World's Largest Rainforest. Mongaby, 2020.

CAIRNS, C., T.; NAI, C.; MEYER, V. How a fungus shapes biotechnology: 100 years of

Aspergillus niger research. **Fungal Biology and Biotechnology**, v. 5, n. 13, 2018.

CALAÇA, F., J., S.; SOUSA, D., G.; BELÉM-JUNIOR, J., S.; FAQUIM, R., C., P.; XAVIER-SANTOS, S.; SILVA-NETO, C., M.; SOUZA, M., M., O. Perception of fungi by farmers in the Cerrado. **Brazilian Journal of Biology**, v. 82, 2022.

CAPORASO, J., G.; BITTINGER, K.; BUSHMAN, F., D; DESANTIS, T., Z.; ANDERSEN, G., L.; KNIGHT, R. PyNAST: a flexible tool for aligning sequences to a template alignment. **Bioinformatics**, v.26, p. 266–267, 2010.

CASTELLANI, A., A. Maitenance na cultivation of the common pathogenic fungi of man. In sterile distilled water. Further researches. **Journal of Tropical Medicine**, v. 70, p. 181-184, 1967.

CHAMBERGO, S., F.; VALENCIA, E., Y. Fungal biodiversity to biotechnology. **Applied Microbiology and Biotechnology**, v. 100, n. 6, 2016.

CHAUDHARY, V.; KATYAL, P.; POONIA, A.K.; KAUR, J.; PUNIYA, AK.; PANWAR, H. Natural pigment from *Monascus*: The production and therapeutic significance. **Journal of Applied Microbiology**, v. 133, n. 1, 2021. DOI: https://doi.org/10.1111/jam.15308.

CHEN A., J.; HUBKA, V.; FRISVAD, J., C.; VISAGIE, C., M.; HOUBRAKEN, J.; MEIJER, M.; VARGA, J.; DEMIREL, R.; JURJEVIĆ, Ž.; KUBÁTOVÁ, A.; SKLENÁŘ, F.; ZHOU1, Y., G.; SAMSON, R., A. Polyphasic taxonomy of *Aspergillus* section *Aspergillus* (formerly Eurotium), and its occurrence in indoor environments and food. **Studies in Mycology**, v. 88, p. 37-135, 2017.

CHEN, Q.; YANG, F.; CHENG, X. Effects of land use change type on soil microbial attributes and their controls: Data synthesis. **Ecological Indicators**, v. 138, 2022.

CHEN, Y.; XI, J.; XIAO, M.; WANG, S.; CHEN, W.; LIU, F.; SHAO, Y.; YUAN, Z. Soil fungal communities show more specificity than bacteria for plant species composition in a temperate forest in China. **BMC Microbiology**, v. 22, n. 208, 2022.

CHOI, J.; KIM, S.,-H. A genome Tree of Life for the Fungi kingdom. **PNAS,** v. 114, n. 35, p. 9391-9396, 2017.

COLE, M., B.; AUGUSTIN, M., A.; ROBERTSON, M., J.; MANNERS, J., M. The science of food security. **NPJ Science of Food**, v. 2, n. 14, 2018.

COLLI, G., R.; VIEIRA, C., R.; DIANESE, J., C. Biodiversity and conservation of the Cerrado: recent advances and old challenges. **Biodiversity and Conservation**, v. 29, p. 1465–1475, 2020.

DAVIS A. P., GOVAERTS R., BRIDSON D. M., STOFFELEN P. An annotated taxonomic conspectus of the genus Coffea (Rubiaceae). **Botanical J. Linnean** Soc, v. 152, p. 465–512, 2006.

DEMAIN, A.; MARTENS, E. Production of valuable compounds by molds and yeasts. **The Journal of Antibiotics**, v. 70, p. 347–360, 2017.

DE MELO PEREIRA DÍ, GURGEL RS, DE SOUZA ATF, MATIAS RR, DE SOUZA FALCÃO L, CHAVES FCM, DA SILVA GF, MARTÍNEZ JG, DE LIMA PROCÓPIO RE, FANTIN C, ALBUQUERQUE PM. Isolation and Identification of Pigment-Producing Endophytic Fungi from the Amazonian Species Fridericia chica. **Journal of Fungi**, v. 10, n. 1. Doi: 10.3390/jof10010077.

DHILLON GS, KAUR S, BRAR SK, VERMA M. Green synthesis approach: extraction of chitosan from fungus mycelia. **Crit Rev Biotechnol**, v. 33, p. 379–403, 2013. DOI: https://doi.org/10.3109/07388551.2012.717217

DIAS CM, OLIVEIRA FA, MADUREIRA FD, SILVA G, SOUZA WR, CARDEAL ZL, et al. Multi-residue method for the analysis of pesticides in Arabica coffee using liquid chromatography/tandem mass spectrometry. **Food Addit Contam, Part A**, v. 30, n. 1308, 2013. DOI: 10.1080/19440049.2013.801088

DIAS, C., G.; MARTINS, F., B.; MARTINS, M., A. Climate risks and vulnerabilities of the Arabica coffee in Brazil under current and future climates considering new CMIP6 models. **Science of The Total Environment**, v. 907, 2024.

DUFOSSÉ, L.; FOUILLAUD, M.; CARO, Y.; MAPARI, S., A., S.; SUTTHIWONG, N. Filamentous fungi are large-scale producers of pigments and colorants for the food industry. **Current Opinion in Biotechnology**, v. 26, p. 56-61, 2014.

EDGAR, R., C. UPARSE: sequências OTU altamente precisas de leituras de amplicons microbianos. **Nature Methods**, v. 10, p. 996–998, 2013.

EISENMAN, H., E.; CASADEVALL, A. Synthesis and assembly of fungal 31melanin. **Applied Microbiology and Biotechnology**, v. 93, n. 3, p. 931-940,2012.

EVANSA, T., L.; COSTA, M.; TOMAS, W., M.; CAMILO, A., R. Large-scale habitat mapping of the Brazilian Pantanal wetland: A synthetic aperture radar approach. **Remote Sensing of Environment,** v. 155, p. 89-108, 2014.

EVERITT, B., S.; **Cluster Analysis**. Edward Arnold, 3 edition, 1993.

FALAH, F., SAMIE, A., MORTAZAVI, S.A. et al. Bio-synthesis, purification and structural analysis of Cyclosporine-A produced by *Tolypocladium inflatum* with valorization of agro-industrial wastes. **Scientific Report**, v. 14, n. 12540, 2024. DOI: https://doi.org/10.1038/s41598-024-63110-y

FALEIRO, F., V.; LOYOLA, R., D. Socioeconomic and political trade-offs in biodiversity conservation: a case study of the Cerrado Biodiversity Hotspot, Brazil. **Diversity and Distribuitions**, 2013.

FIERER, N. Embracing the unknown: disentangling the complexities of the soil microbiome. **Nature Reviews Microbiology**, v. 15, p. 579–590, 2017.

FIERER, N.; WOOD, S., A.; MESQUITA, C., P., B. How microbes can, and cannot, be used to assess soil health. **Soil Biology and Biochemistry**, v. 153, 2021.

FLOREY, H.W.; ADELAIDE, M. B.; JENNINGS, M. A.; OXFD, B.M.; GILLIVER, K.; CAMB, B. A.; SANDERS, A. G.; LOND, M. B. Mycophenolic acid an antibiotic from *Penicillium brevicompactum* dierckx. **Lancet**, p. 46-49, 1946.

FRAC, M.; HANNULA, S., E.; BEŁKA, M.; JĘDRYCZKA, M. Fungal Biodiversity and Their Role in Soil Health. **Frontiers in Microbiology**, 2018.

FRANÇOSO, R. D.; BRANDÃO, R.; NOGUEIRA, C. C.; SALMONA, Y. B.; MACHADO, R. B.; COLLI, G. R. Habitat loss and the effectiveness of protected areas in the cerrado biodiversity hotspot. **Natureza & Conservação**, v. 13, n. 1, p. 35-40, 2015.

GARCIA, W. M.; ZAGO, B. W.; NUNES, J. R. S.; CARVALHO, I. F. Diversity of filamentous fungi in Cerrado soil under native vegetation. **Revista Brasileira de Biociências**, v. 13, n. 4, p. 245-249, 2015.

GAYNES, R. The Discovery of Penicillin—New Insights After More Than 75 Years of Clinical Use. **Emerging Infectious Diseases, v. 23, n. 5, p. 849, 2017.**

GILLER KE, HIJBEEK R, ANDERSSON JA, SUMBERG J. Regenerative Agriculture: An agronomic perspective. **Outlook Agricultural**, v. 50, n. 1, p. 13-25, 2021. Doi: 10.1177/0030727021998063.

GODFRAY, H., C., J.; CROZ, D., M., D.; ROBINSON, S. Food system consequences of a fungal disease epidemic in a major crop. Philosophical transactions of the Royal Society of London. Series B, **Biological sciences**, v. 371, n. 1709, 2016.

GÓMEZ-GAVIRIA, M.; MORA-MONTES, H., M. Exploring the potential of chitin and chitosan in nanobiocomposites for fungal immunological detection and antifungal action. **Carbohydrate Research**, v. 543, 2024.

GOUGHENOUR, K., D.; WHALIN, J.; SLOT, J., C.; RAPPLEYE, C., A. Diversification of Fungal Chitinases and Their Functional Differentiation in Histoplasma capsulatum. Molecular **Biology and Evolution**, v. 38, n. 4, p. 1339–1355, 2021. DOI: https://doi.org/10.1093/molbev/msaa293.

GUEVARA-SUAREZ, M.; SUTTON, A., D.; CANO-LIRA, J., F.; GARCÍA, D.; MARTIN-VICENTE, A.; WIEDERHOLD, N.; GUARRO, J.; GENÉA, J. Identification and Antifungal Susceptibility of *Penicillium*-Like Fungi from Clinical Samples in the United States. **Journal of Clinical Microbiology**, v. 54, n. 8, p. 2155-2161, 2016.

GUILLEN-CRUZ G, TORRES-ARREOLA R, SANCHEZ-MEJIA Z, FLORES-RENTERIA D. The effect of conventional and sustainable agricultural management practices on carbon and water fluxes in a Mexican semi-arid region. **Peer Journal**, v. 10, 2022. DOI: 10.7717/peerj.14542.

HAHN T, TAFI E, VON SEGGERN N, FALABELLA P, SALVIA R, THOMA J, FEBEL E, FIJALKOWSKA M, SCHMITT E, STEGBAUER L, ZIBEK S. Purification of chitin from pupal exuviae of the black soldier fly. **Waste Biomass Valor**, v. 13, p. 1993–2008, 2022. https://doi.org/10.1007/s12649-021-01645-1

HAMPL, V.; PAVLICEK, A.; FLEGR, J. Construction and bootstrap analysis of DNA fingerprinting-based phylogenetic trees with the freeware program FreeTree: application to trichomonad parasites. **International Journal Systematic and Evolutionary Microbiology**, v.51, p.731-735, 2001.

HANNULA, SE., VEEN, GF. Drought has short-term effects on soil fungal communities leading to long-term effects on soil functions. **Soil Biology and Biochemistry**, v. 209, 2025. DOI: https://doi.org/10.1016/j.soilbio.2025.109893.

HARTL L, ZACH S, SEIDL-SEIBOTH V. Fungal chitinases: diversity, mechanistic properties and biotechnological potential. **Appl Microbiol Biotechnol**, v. 93, n. 2, p. 533-43, 2012. Doi: 10.1007/s00253-011-3723-3.

HOUBRAKEN, J.; SAMSON, R., A. Phylogeny of Penicillium and the segregation of Trichocomaceae into three families. **Studies in Mycology**, v. 70, n. 1, p. 1-51, 2011.

HOUBRAKEN J, FRISVAD JC, SEIFERT KA, OVERY DP, TUTHILL DM, VALDEZ JG, SAMSON RA. New penicillin-producing *Penicillium* species and an overview of section *Chrysogena*. **Persoonia**. 2012 Dec;29:78-100. doi: 10.3767/003158512X660571.

INSTITUTO BRASILEIRO DE GEOGRAFIA ESTATÍSTICA (**IBGE**). Disponível em: < https://educa.ibge.gov.br/jovens/conheca-o-brasil/territorio/18307-biomas-brasileiros.html Acessado em: 15 de Agosto de 2022.

INSTITUTO ESTADUAL DE FLORESTAS (**IEF**). Cobertura vegetal de Minas Gerais. Disponível em: http://www.ief.mg.gov.br/florestas. Acesso em: 09 de outubro de 2025.

HUANG, Z.; SU, Y.; LIN, S.; WU, G.; CHENG, H.; HUANG, G. Elevational patterns of microbial species richness and evenness across climatic zones and taxonomic scales. **Ecology and Evolution**, v. 13, n. 10, 2023.

JAMES, T., Y.; STAJICH, J., E.; HITTINGER, C., T.; ROKAS, A. Toward a Fully Resolved Fungal Tree of Life. **Annual Review of Microbiology**, v. 74, p. 291-313, 2020. DOI: https://doi.org/10.1146/annurev-micro-022020-0518352020

JANG, M. K., KONG, B. G., JEONG II, Y., LEE, C. H. & NAH, J. W. Physicochemical characterization of α-chitin, β-chitin, and γ-chitin separated from natural resources. **Journal of Polymer Science Part A Polymer Chemistry,** v. 42, p. 3423–3432, 2004.

JAYAKUMAR, R.; PRABAHARAN, M.; NAIR, S., V.; TAMURA, H. Novel chitin and chitosan nanofibers in biomedical applications. **Biotechnology Advances**, v. 28, n. 1, p. 142-150.

JALIL, M.; ZAKARIA, MT.; SALIKIN, NA.; N.H. et al. Assessment of cultivation parameters influencing pectinase production by Aspergillus niger LFP-1 in submerged fermentation. J Genet **Eng Biotechnol**, v. 21, 45, 2023. https://doi.org/10.1186/s43141-023-00510-z

KARLSSON M, STENLID J. Comparative evolutionary histories of the fungal chitinase

gene family reveal non-random size expansions and contractions due to adaptive natural selection. **Evol Bioinform Online**, v. 4, p. 47–60, 2008. doi: 10.4137/ebo.s604

KARTHIK N, BINOD P, PANDEY A. Chitinases. In: Ashok P, Sanfeeta N, Carlose R Soccol (eds) Current Developments in Biotechnology and Bioengineering: production isolation and purification of industrial products. **Elseivie**r, pp 335–368, 2017.

KIM Y, PARK RD. Progress in bioextraction processes of chitin from crustacean biowastes. **J Korean Soc Appl Biol Chem**, v. 58, p. 545–554, 2015. https://doi.org/10.1007/s13765-015-0080-4

KIRK, P. Species fungorum plus. Royal Botanic Gardens, Kew. **Checklist dataset**, 2023. https://doi.org/10.15468/ts7wsb

KLINK, C. A.; MACHADO, R. B. Conservation of the Brazilian cerrado. **Conservation biology**, v. 19, n. 3, p. 707-713, 2005.

KLINK, C., A.; SATO, M., N.; CORDEIRO, G., G.; RAMOS, M., I., M. The Role of Vegetation on the Dynamics of Water and Fire in the Cerrado Ecosystems: Implications for Management and Conservation. **Plants Basel**, v. 9, n. 12, p. 1803, 2020.

KOBAYASHI A, YAMAMOTO I, NAKADA Y, KIDOGUCHI S, MATSUO N, TANNO Y, OHKIDO I, TSUBOI N, YAMAMOTO H, YOKOYAMA K, YOKOO T. Successful treatment of BK virus nephropathy using therapeutic drug monitoring of mycophenolic acid. **Nephrology**, v. 19, p. 37–41, 2014. doi: 10.1111/nep.12249.

KOTB, E.; ALABDALALL, A., H.; ALGHAMDI, A., I.; ABABUTAIN, I., M.; ALDAKEEL, S., A.; AL-ZUWAID, S., K.; ALGARUDI, B., M.; ALGARUDI, S, M.; AHMED, A., A.; ALBARRAG, A., M. Screening for chitin degrading bacteria in the environment of Saudi Arabia and characterization of the most potent chitinase from Streptomyces variabilis Am1. **Scientific Reports,** v. 13, n. 11723, 2023.

KUHAD, R., C.; GUPTA, R.; SINGH, A. Microbial Cellulases and Their Industrial Applications. **Enzyme Research**, v. 2011, 2011.

LANGE, L. The importance of fungi and mycology for addressing major global challenges. **IMA Fungus**, v. 5, n. 2, p. 463–471, 2014.

LAPOLA, D., M.; PINHO, P.; BARLOW, J.; ARAGÃO, L., O., A., C.; BERENGUER, E.; CARMENTA, R.; LIDDY, H., M.; SEIXAS, H.; SILVA, C., V., J.; WALKER, W., S et al. The drivers and impacts of Amazon forest degradation. **Science**, v. 379, n. 6630, 2023. DOI: 10.1126/science.abp8622.

LE QV., COWA, S.; JOVANOVIC, G.; LE, LDT. A Study of Regenerative Farming Practices and Sustainable Coffee of Ethnic Minorities Farmers in the Central Highlands of Vietnam. Frontiers Sustain. **Food Syst**., v. 5, 2021. DOI: https://doi.org/10.3389/fsufs.2021.712733.

LEI Z., ZHANG Y. Q. Effects of phosphate solubilization and phytohormone production of Trichoderma asperellum Q1 on promoting cucumber growth under salt stress**. J. Integr.**

Agric. 14, 1588–1597, 2015. Doi: 10.1016/S2095-3119(14)60966-7.

LI Z., BAI T., DAI L., WANG F., TAO J., MENG S., et al. A study of organic acid production in contrasts between two phosphate solubilizing fungi: *Penicillium oxalicum* and *Aspergillus Niger*. **Scientific Reports**, v. 6, n. 25313, 2016. doi: 10.1038/srep25313

LIN Y. L.; WANG T. H.; LEE M. H.; SU N. W. Biologically active components and nutraceuticals in the Monascus-fermented rice: a review. **Appl. Microbiol. Biotechnol**, v. 77, n. 5, p. 965–973, 2008. 10.1007/s00253-007-1256-6

LITWIN, A.; NOWAK, M.; RÓŻALSKA, S. Entomopathogenic fungi: unconventional applications. **Reviews in Environmental Science and Bio/Technology**, v. 19, p. 23-42, 2020. 10.1007/s11157-020-09525-1

LIU, Y., WU, Q., WU, X., ALGHARIB, S. A., GONG, F., HU, J., LUO, W.,ZHOU, M., PAN, Y., YAN, Y., & WANG, Y. Structure, prepara-tion, modification, and bioactivities of β-glucan and mannan fromyeast cell wall: A review. **International Journal of Biological Macro-molecules**, 173, 445–456, 2021. https://doi.org/10.1016/j.ijbiomac.2021.01.125

LIU, P.; LIN, A.; ZHANG, G.; ZHANG, J.; CHEN, Y.; SHEN, T.; ZHAO, J.; WEI, D.; WANG, W. Enhancement of cellulase production in Trichoderma reesei RUT-C30 by comparative genomic screening. **Microbial Cell Factories**, v. 18, n. 81, 2019.

LOPES, A. S.; COX, F. R. Cerrado Vegetation in Brazil: An Edaphic Gradient 1. **Agronomy Journal**, v. 69, n. 5, p. 828-831, 1977.

LÓPEZ-LEGARDA, X.; TRIANA-PEÑALOZA, A.V.; FLÓREZ-RESTREPO, M.A.; CASTAÑEDA-CASTAÑO, A.; SEGURA-SÁNCHEZ, F. **CHAPTER 15 - Fungal enzymes production through the utilization of by-product**, Editor(s): Jameel R. Al-Obaidi, Noor Baity Saidi, Ali Z. Al-Saffar, In Developments in Applied Microbiology and Biotechnology, Fungal Macromolecule Applications in Life Sciences, Academic Press, 2025, p. 353-378. DOI: https://doi.org/10.1016/B978-0-443-30102-5.00015-6.

LOURENÇO-DE-MORAES, R.; LISBOA, B., S.; DRUMMOND, L., O.; MOURA, C., C., M.; MOURA, G., J., D.; LYRA, M.; GUARNIERI, M.; MOTT, T.; HOOGMOED, M., S.; SANTANA, D., J. A New Species of the Genus Adelophryne (Anura: Eleutherodactylidae: Phyzelaphryninae) from the Atlantic Forest of Northeastern Brazil**. Herpetologica**, v. 77, n. 2, 164-175, 2021.

LÜCKING, R.; AIME, M., C.; ROBBERTSE, B.; MILLER, A., N.; ARIYAWANSA, H., A.; AOKI, T.; CARDINALI, G.; CROUS, P., W et al. Unambiguous identification of fungi: where do we stand and how accurate and precise is fungal DNA barcoding? **Ima Fungus**, v. 11, n. 14, 2020.

LUGHADHA, E., N.; BACHMAN, S., P.; LEÃO, T., C., C.; FOREST, F.; HALLEY, J., M.; MOAT, J.; ACEDO, C.; BACON, K., L.; BREWER, R., F., Q.; GÂTEBLÉ, G.; GONÇALVES, S., C.; GOVAERTS, R. Et al. Extinction risk and threats to plants and fungi. **Plants People Planet**, v. 2, n. 5, p. 389-408, 2020.

MAPARI, S., A., S.; THRANE, U.; MEYER, A., S. Fungal polyketidenazaphilone pigments as future natural food colorants? **Trends Biotechnology**, v. 28, n. 6, p. 7-300, 2010.

MARTINELLI, L., A.; NARDOTO, G., B.; SOLTANGHEISI, A.; REIS, C., R., G. et al. Determining ecosystem functioning in Brazilian biomes through foliar carbon and nitrogen concentrations and stable isotope ratios. **Biogeochemistry**, v. 154, p. 405-423, 2021.

MARTÍNEZ-RAMOS, M.; ORTIZ-RODRÍGUEZ, I., A.; PIÑERO, D.; DIRZO, R.; SARUKHÁN, J. ANTHROPOGENIC disturbances jeopardize biodiversity conservation within tropical rainforest reserves. **PNAS**, v. 113, n. 19, p. 5323-5328, 2016. DOI: https://doi.org/10.1073/pnas.1602893113

MCGOVERN, P., E.; ZHANG, J.; TANG, J.; ZHANG, Z.; HALL, G., R.; MOREAU, R., A.; NUÑEZ, A.; BUTRYM, E., D.; RICHARDS, M., P.; WANG, C.-S.; CHENG, G.; ZHAO, Z.; WANG, C. Fermented beverages of pre- and proto-historic China. **PNAS**, v. 101 n. 51, p. 17593–17598, 2004.

MCKELVEY, S., M.; MURPHY, R., A. Biotechnological Use of Fungal Enzymes. **Fungi: Biology and Applications**, 2017.

MINKE, R. & BLACKWELL, J. The structure of α-chitin. **Journal of Molecular Biology**, v. 120, p. 167–181, 1978.

MILLER, M. A., PFEIFFER, W. & SCHWARTZ, T. Creating the CIPRES science gateway for inference of large phylogenetic trees. In Proceedings of the Gateway Computing Environments **Workshop (GCE)** (eds Miller, M. A. et al.) 1–8 (IEEE, 2010).

MELLO, S., S.; PASTORE, J., B. Ornamental flora of the Cerrado in landscape architecture: a portrait of its practical application. **Ornamental Horticulture**, v. 27, n. 1, 2021.

MYERS, N.; MITTERMEIER, R., A.; MITTERMEIER, CC., G.; FONSECA, G., A., B.; KENT, J. Biodiversity hotspots for conservation priorities. **Nature**, v. 403, p. 853-858, 2000.

MUNIZ, A., C.; PIMENTA, R., J., G.; CRUZ, M., V.; RODRIGUES, J., G.; BUZATTI, R., S., O.; HEUERTZ, M.; LEMOS-FILHO, J., P.; LOVATO, M., B. Hybrid zone of a tree in a Cerrado/Atlantic Forest ecotone as a hotspot of genetic diversity and conservation. **Ecology and Evolution**, v. 12, n. 1, 2022.

NAYLOR, D.; MCCLURE, R.; JANSSON, R. Trends in Microbial Community Composition and Function by Soil Depth. **Microorganisms**, v. 10, n. 3, 2022.

NASSARY, EK. Fungal biocontrol agents in the management of soil-borne pathogens, insect pests, and nematodes: Mechanisms and implications for sustainable agriculture. **The microbe**, v. 7, 2025. DOI: https://doi.org/10.1016/j.microb.2025.100391.

NOVY V, NIELSEN F, SEIBOTH B, NIDETZKY B. The influence of feedstock characteristics on enzyme production in *Trichoderma reesei*: a review on productivity, gene regulation and secretion profiles. **Biotechnol Biofuels,** v. 12, n. 238, 2018. doi: 10.1186/s13068-019-1571-z.

OJEDA-LÓPEZ, M.; CHEN, W.; EAGLE, C., E.; GUTIÉRREZ, G.; JIA, W., L.; SWILAIMAN, S., S.; HUANG, Z.; PARK, H.-S.; YU, J.-H.; CÁNOVAS, D.; DYER, P., S. Evolution of asexual and sexual reproduction in the aspergilli. **Studies in Mycology**, v. 91, p. 37-59, 2018.

OLIVEIRA, G., C.; FRANCELINO, M., R.; ARRUDA, D., M.; FERNANDES-FILHO, E., I.; SCHAEFER, C., E., G., R. Climate and soils at the Brazilian semiarid and the forest-Caatinga problem: new insights and implications for conservation. **Environmental Research Letters**, v.14, n. 10, 2019.

OMARI, F., A.; ALZANDI, A., A.; HIJAB, L., Y., A., A.; AL-HAZMI, N., E.; ELAGIB, S., M.; ALMIMAN, B., F.;ALHARTHI, F.; NAGUIB, D., M. Production of chitinase in elicited tomato cell suspensions and its application as a biopesticide and fungicide against soil-borne pests and fungi. **Physiological and Molecular Plant Pathology**, v. 134, 2024.

PAN, Y.; BIRDSEY, R., A.; FANG, J.; HOUGHTON, R.; KAUPPI, P., E.; KURZ, W., A.; PHILLIPS, O., L.; SHVIDENKO, A.; LEWIS, S., L.; CANADELL, J., G.; CIAIS, R.; JACKSON, R., B.; PACALAA, S., W.; MCGUIRE, A., D.; PIAO, S.; RAUTIAINEN, A.; SITCH, S.; HAYES, D. A Large and Persistent Carbon Sink in the World's Forests. **Science,** v. 333, n. 6045, p. 988-993, 2011.

PAPAGIANNI, M. Advances in citric acid fermentation by Aspergillus niger: Biochemical aspects, membrane transport and modeling. **Biotechnology Advances**, v. 25, n. 3, p. 244-263, 2007.

PATEL G, THAKUR NS, KUSHWAH V, PATIL MD, NILE SH, JAIN S, KAI G, BANERJEE UC. Mycophenolate co-administration with quercetin via lipid-polymer hybrid nanoparticles for enhanced breast cancer management. **Nanomedicine**, v. 24, 2020. Doi: 10.1016/j.nano.2019.102147.

PEAY, K., G.; BARALOTO, C.; FINE, P., V., A. Strong coupling of plant and fungal community structure across western Amazonian rainforests. **The ISME Journal**, v. 7, p. 1852-1861, 2013.

PHUKHAMSAKDA, C., NILSSON, R.H., BHUNJUN, C.S. et al. The numbers of fungi: contributions from traditional taxonomic studies and challenges of metabarcoding. **Fungal Diversity**, v. 114, p. 327–386, 2022. https://doi.org/10.1007/s13225-022-00502-3

PIROZYNSKI, K. A. & MALLOCH, D. W. The origin of land plants: a matter of mycotropism. **Biosystems**, v. 6, p. 153–164, 1975.

POHL, C.; POLLI, F.; SCHÜTZE, T.; VIGGIANO, A.; MÓZSIK, L.; JUNG, S.; VRIES, M.; BOVENBERG, R., O., A., L.; MEYER, V.; DRIESSEN, A., J., M. A *Penicillium rubens* platform strain for secondary metabolite production. **Scientific Reports**, v. 10, n. 7630, 2020.

PONCET, V.; ASTEN PV, MILLET CP, VAAST P, ALLINNE C. Which diversification trajectories make coffee farming more sustainable? **Current Opinion in Environmental Sustainability**, v. 68, 2024.

POVEDA, J.; ABRIL-URIAS, P.; ESCOBAR, C. Biological Control of Plant-Parasitic Nematodes by Filamentous Fungi Inducers of Resistance: *Trichoderma*, Mycorrhizal and Endophytic Fungi. **Frontiers in microbiology**, 2020.

POVEDA, J. *Trichoderma* as biocontrol agent against pests: New uses for a mycoparasite. **Biological Control**, v. 159, 2021.

RATHORE, A., S.; GUPTA, R., D. Chitinases from Bacteria to Human: Properties, Applications, and Future Perspectives. **Enzyme Research**, v. 2015, n. 1, 2015.dx.doi.org/10.1155/2015/791907.

REQUENA, N.; PEREZ-SOLIS, E.; AZCÓN-AGUILAR, C.; JEFFRIES, P.; BAREA, J., M. Management of Indigenous Plant-Microbe Symbioses Aids Restoration of Desertified Ecosystems. **Applied and Environmental Microbiology**, v. 67, n. 2, p. 495-498, 2001.

RIBEIRO, M., C.; METZGER, J., P.; MARTENSEN, A., C.; PONZONI, F., J.; HIROTA, M., M. The Brazilian Atlantic Forest: How much is left, and how is the remaining forest distributed? Implications for conservation. **Biological Conservation**, v. 142, p. 1141-1153, 2009.

ROPARS, J.; DIDIOT, E.; VEGA, RCR.; BENNETOT, B.; COTON, M.; POIRIER, E.; COTON, E.; SNIRC, A.; PRIEUR, S., L.; GIRAUD, T. Domestication of the Emblematic White Cheese-Making Fungus Penicillium camemberti and Its Diversification into Two Varieties. **Current Biology**, v. 30, n. 22, p. 4441-4453, 2020. https://doi.org/10.1016/j.cub.2020.08.082.

SAMSON, R., A.; YILMAZ, N.; HOUBRAKEN, J.; SPIERENBURG, H.; SEIFERT, K., A.; PETERSON, S., W.; VARGA, J.; FRISVAD, J., C. Phylogeny and nomenclature of the genus *Talaromyces* and taxa accommodated in *Penicillium* subgenus *Biverticillium*. **Studies in mycology**, v. 70, n. 1, p. 159-183, 2011.

SANTOS DG, COELHO CCS, FERREIRA ABR, FREITAS-SILVA O. Brazilian Coffee Production and the Future Microbiome and Mycotoxin Profile Considering the Climate Change Scenario**. Microorganisms**, v. 9, n. 4, p. 858, 2021. Doi: 10.3390/microorganisms9040858

SATISH, N.; SULTANA, S.; NANJUNDIAH, V. Diversity of soil fungi in a tropical deciduous forest in Mudumalai, southern India. **Current Science**, v. 93, n. 5, p. 669-677, 2007.

SAWANT, A.M., NAVALE, V.D. & VAMKUDOTH, K.R. Genome sequencing and analysis of penicillin V producing *Penicillium rubens* strain BIONCL P45 isolated from India. **Int Microbiol** 27, 1473–1484, 2024. https://doi.org/10.1007/s10123-024-00491-0

SCHLATTER, D., C.; KAHL, K.; CARLSON, B.; HUGGINS, D., R.; PAULITZ, T. Fungal community composition and diversity vary with soil depth and landscape position in a no-till wheat-based cropping system. **FEMS Microbiology Ecology**, v. 94, n. 7, 2018.

SEIDL V. Chitinases of filamentous fungi: a large group of diverse proteins with multiple

physiological functions. **Fungal Biol Rev**, v. 22, p. 36–42, 2008. doi: 10.1016/j.fbr.2008.03.002

SEN T., BARROW C.J., DESHMUKH S.K. Microbial pigments in the food industry—Challenges and the way forward. **Frontiers**, n. 6, v. 7, 2019 Doi: 10.3389/fnut.2019.00007.

SEXTON, A., C.; HOWLETT, B., J. Parallels in fungal pathogenesis on plant and animal hosts. **Eukaryotic Cell**, v. 5, n. 12, p. 1941-1949, 2006.

SHANKAR, U.; TRIPATHI, A., K. Rainforests north of the Tropic of Cancer: Physiognomy, floristics and diversity in 'lowland rainforests' of Meghalaya, India. **Plant diversity**, v. 39, n. 1, p. 20-36, 2017.

SHER, A., LI, H., ULLAH, A. et al. Importance of regenerative agriculture: climate, soil health, biodiversity and its socioecological impact. **Discover Sustainability**, v. 5, n. 462, 2024. DOI: https://doi.org/10.1007/s43621-024-00662-z.

SHI, L.; DOSSA, G.; O.; PAUDEL, E.; ZANG, H.; XU, J.; HARRISON, R., D. Changes in Fungal Communities across a Forest Disturbance Gradient. **Applied and Environmental Microbiology**, v. 85, n. 12, 2019.

SICARD, D.; LEGRAS, J., L. Bread, beer and wine: Yeast domestication in the Saccharomyces sensu stricto complexPain, bière et vin: domestication des levures du complexe d'espèces Saccharomyces sensu stricto. **Comptes Rendus Biologies**, v. 334, n. 3, p. 229-236, 2011.

SOTERRONI, A., C.; RAMOS, F., M.; MOSNIER, A.; FARGIONE, J.; ANDRADE, P., R.; BAUMGARTEN, L.; PIRKER, J.; OBERSTEINER, M.; KRAXNER, F.; POLASKY, S. Expanding the Soy Moratorium to Brazil's Cerrado. **Science Advances**, v. 5, n. 7, 2019.

SPOHNER, S., C.; SCHAUM, V.; QUITMANN, H.; CZERMAK, P. *Kluyveromyces* lactis: An emerging tool in biotechnology. **Journal of Biotechnology**, v. 222, p. 104-116, 2016. DOI: https://doi.org/10.1016/j.jbiotec.2016.02.023.

SUMBERG J, GILLER KE. What is 'conventional' agriculture? **Global Food Security**, v. 32, 2022.

TARDY, V.; CHABBI, A.; CHARRIER, X.; BERRANGER, C.; REIGNIER, T.; DEQUIEDT, S.; FAIVRE-PRIMOT, C.; TERRAT, S.; RANJARD, L.; MARON, P., A. Land Use History Shifts In Situ Fungal and Bacterial Successions following Wheat Straw Input into the Soil. **Plos one**, v. 10, n. 6, 2015.

TAO, K.; DU, L.; SUN, X.; CAI, M.; ZHU, T.; ZHOU, X.; GU, Q.; ZHANG, Y. Biosynthesis of aspergiolide A, a novel antitumor compound by a marine-derived fungus Aspergillus glaucus via the polyketide pathway. **Tetrahedron Letters**, v. 50, n. 9, p. 1082-1085, 2009.

TAMBURINI, G.; BOMMARCO, R.; WANGER, T., C.; KREMEN, C.; HEIJDEN M., G., A., V., D.; LIEBMAN, M.; HALLIN, S. Agricultural diversification promotes multiple ecosystem services without compromising yield**. Science advances**, v. 6, n. 45, 2020.

TAYLOR, D. L.; HOLLINGSWORTH, T. N.; MCFARLAND, J. W.; LENNON, N. J.; NUSBAUM, C.; RUESS, R. W. A first comprehensive census of fungi in soil reveals both hyperdiversity and fine–scale niche partitioning. **Ecological Monographs**, v. 84, n.1, p. 3-20, 2014.

TEDERSOO, L.; MIKRYUKOV, V.; ZIZKA, A.; BAHRAM, M.; HAGH-DOUST, N.; ANSLAN, S.; PRYLUTSKYI, O.; DELGADO-BAQUERIZO, M.; MAESTRE, F., T.; PÄRN, J.; ÖPIK, M.; MOORA, M.; ZOBEL, M.; ESPENBERG, M.; MANDER, U.; KHALID, A., N.; CORRALES, A.; AGAN, A.; VASCO-PALACIOS, A.-M.; SAITTA, A.; RINALDI, A., C et al. Global patterns in endemicity and vulnerability of soil fungi. **Global Change Biology**, 2022.

TEDERSOO, L.; SÁNCHEZ-RAMÍREZ, S.; KÕLJALG, U.; BAHRAM, M.; DÖRING, M.; SCHIGEL, D.; MAY, T.; RYBERG, M.; ABARENKOV, K. High–level classification of the fungi and a tool for evolutionary ecological analyses. **Fungal Diversity**, v. 90, n. 1, p. 135–159, 2018.

THAKUR, D., BAIRWA, A., DIPTA, B. et al. An overview of fungal chitinases and their potential applications. **Protoplasma**, v. 260, p. 1031–1046, 2023. DOI: https://doi.org/10.1007/s00709-023-01839-5

TSANG, C., C.; TANG, J., Y., M.; LAU, S., K., P.; WOO, P., C., Y. Taxonomy and evolution of *Aspergillus, Penicillium* and *Talaromyces* in the omics era – Past, present and future. **Computational and Structural Biotechnology Journal**, v. 16, p. 197-210, 2018.

THOMAS, R., FUKAMIZO, T. & SUGINTA, W. Bioeconomic production of high-quality chitobiose from chitin food wastes using an in-house chitinase from *Vibrio campbellii*. **Bioresour. Bioprocess**, v. 9, n. 86, 2022. https://doi.org/10.1186/s40643-022-00574-8

VENIL, C., K.; VELMURUGAN, P.; DUFOSSÉ, L.; DEVI, P., R.; RAVI, A., V.; Fungal Pigments: Potential Coloring Compounds for Wide Ranging Applications in Textile Dyeing. **Journal of fungi**, v. 6, n. 68, 2020.

VINECKY F., DAVRIEUX F., MERA A. C., ALVES G. S. C., LAVAGNINI G., LEROY T., et al. Controlled irrigation and nitrogen, phosphorous and potassium fertilization affect the biochemical composition and quality of Arabica coffee beans. **J. Agric. Sci**., v. 155, p. 902-918, 2017.

VISAGIE, C., M.; HOUBRAKEN, J.; FRISVAD, J., C.; HONG, S., B.; KLAASSEN, C., H.; PERRONE, G.; SEIFERT, K., A.; VARGA, J.; YAGUCHI, T.; SAMSON, R., A. Identification and nomenclature of the genus *Penicillium*. **Studies in Mycology**, v. 78, p. 343-371, 2014. DOI: 10.1016/j.simyco.2014.09.001.

VISAGIE, C., M.; YILMAZ, N.; KOCSUBÉ, S.; FRISVAD, J., C.; HUBKA, V.; SAMSON, R., A.; HOUBRAKEN, J. A review of recently introduced *Aspergillus, Penicillium, Talaromyces* and other *Eurotiales* species. **Studies in Mycology**, v. 107, p. 1-66, 2024. DOI: 10.3114/sim.2024.107.01

VOLSI B, TELLES TS, CALDARELLI CE, CAMARA MRGD. The dynamics of coffee

production in Brazil**. PLoS One**, v. 14, n. 7, 2019. Doi: 10.1371/journal.pone.0219742.

VU, D.; GROENEWALD, M.; VRIES, M.; GEHRMANN, T.; STIELOW, B.; EBERHARDT, U.; AL-HATMI, A.; GROENEWALD, J., Z.; CARDINALI, G.; HOUBRAKEN, J.; BOEKHOUT, T.; CROUS, P., W.; ROBERT, V.; VERKLEY, G., J., M. Large-scale generation and analysis of filamentous fungal DNA barcodes boosts coverage for kingdom fungi and reveals thresholds for fungal species and higher taxon delimitation. **Studies in Mycology**, v. 92, p.135-154, 2019.

WANG Y, ZHOU X, DEBING Y, CHEN K, VAN DER LAAN LJ, NEYTS J, JANSSEN HL, METSELAAR HJ, PEPPELENBOSCH MP, PAN Q. Calcineurin inhibitors stimulate and mycophenolic acid inhibits replication of hepatitis E virus. **Gastroenterol**, p. 1775–1783, 2014. doi: 10.1053/j.gastro.2014.02.036.

WANG C, ZENG ZQ, ZHUANG WY. Comparative molecular evolution of chitinases in ascomycota with emphasis on mycoparasitism lifestyle. **Microb Genom**, v. 7, n. 9, 2021. doi: 10.1099/mgen.0.000646.

WERNECK, F., P. The diversification of eastern South American open vegetation biomes: Historical biogeography and perspectives. **Quaternary Science Reviews**, v. 30, n. 13-14, p. 1630-1648, 2011.

WILDE, A., H.; DOBBE, J., C., G.; MEER, Y., V.; THIEL, V.; NARAYANAN, K.; MAKINO, S.; SNIJDER, E., J.; HEMERT, M., J., V. Cyclosporin A inhibits the replication of diverse coronaviruses. **Journal of general virology**, v. 92, p. 2542-2548, 2011.

WYSOKOWSKI, M., BAZHENOV,V., V.; TSURKAN, M., V.; GALLI, R.; STELLING, A., L.; STÖCKER, H.; KAISER,S.; NIEDERSCHLAG, E.; GÄRTNER, G.; BEHM, T.; ILAN, M.; PETRENKO, A.; JESIONOWSKI, T.; EHRLICH, H. Isolation and identification of chitin in three-dimensional skeleton of Aplysina fistularis marine sponge. **International Journal Biology Macromolecules**. 62, 94–100 (2013).

WU Q, LI M, BILAL M, YANG Y, ZHANG J, LI X. Enhanced Production of Mycophenolic Acid from *Penicillium brevicompactum* via Optimized Fermentation Strategy. **Appl Biochem Biotechnol**, v. 194, p. 3001-3015, 2022. doi: 10.1007/s12010-022-03886-9.

WU, B.; HUSSAIN, M.; ZHANG, W.; STADLER, M.; LIU, X.; XIANG, M. Current insights into fungal species diversity and perspective on naming the environmental DNA sequences of fungi. **Mycology: An International Journal on Fungal Biology**, v. 10, n. 3, p. 127-140, 2019.

YANG XWJ, XU DC, QUI JW, MA Y, CUI J. Simultaneous determination of 69 pesticide residues in coffee by gas chromatography-mass spectrometry**. Food Anal Methods**, v. 4, p. 186–95, 2011. 10.1007/s12161-010-9155-3.

YAO, X.; GUO, H.; ZHANG, K.; ZHAO, M.; RUAN, J.; CHEN, J. *Trichoderma* and its role in biological control of plant fungal and nematode disease. **Frontiers in Microbiology**, v. 14. doi.org/10.3389/fmicb.2023.1160551

YIN Z., SHI F., JIANG H., ROBERTS D. P., CHEN S., FAN B. Phosphate solubilization and promotion of maize growth by *Penicillium oxalicum* P4 and *Aspergillus Niger* P85 in a calcareous soil. **Can. J. Microbiol**. 61, 913–923, 2015. doi: 10.1139/cjm-2015-0358

YUSOF, N., L., B., M.; LIM, L., Y.; KHOR, E. Preparation and characterization of chitin beads as a wound dressing precursor. **Journal of Biomedical Materials Research**, v. 54, n.1, p. 1-148, 2000.

ZHU, L.; LI, W.; CIAIS, P.; HE, J.; CESCATTI, A.; SANTORO, M.; TANAKA, K.; CARTUS, O.; ZHAO, Z.; XU, Y.; SUN, M.; WANG, J. Comparable biophysical and biogeochemical feedbacks on warming from tropical moist forest degradation. **Nature Geoscience**, v. 16, p. 244–249, 2023.

SEGUNDA PARTE

Artigo 1 - Redigido conforme norma do periódico científico - Versão preliminar	
Título do artigo:	Chitinase production and effects of land use change on fungal communities in the soil of the Brazilian Cerrado
Autores:	Cristiane Nacimento Figueiredo Mariana Prósperi de Oliveira Paula Clara Resende de Souza Castro Fabiana Franca Passamani Teotonio Soares de Carvalho Harisson de Souza Guimaraes Luís Roberto Batista Daniele Cabral Michel Daniel Kumazawa Morais Fatima Maria de Souza Moreira Victor Satler Pylro
Periódico:	Applied Soil Ecology
ISSN	
DOI	

Artigo 1

Chitinase production and effects of land use change on fungal communities in the soil of the Brazilian Cerrado

Cristiane Nacimento Figueiredo[1], Mariana Prósperi de Oliveira Paula[1], Clara Resende de Souza Castro[1], Fabiana Passamani[1], Teotonio Soares de Carvalho[1], Harisson de Souza Guimaraes[1], Luís Roberto Batista[1], Daniele Cabral Michel[1], Daniel Kumazawa Morais[2], Fatima Maria de Souza Moreira[1], Victor Satler Pylro[1,*]

1, Federal University of Lavras, MG, BR. Emails: cristiane.figueiredo1@estudante.ufla.br, clararsc1@gmail.com, marianaprosperi.tp@gmail.com, fabireinis@gmail.com, teotonio.carvalho@ufla.br, harisson.souza1@estudante.ufla.br, luisrb@ufla.br, danielecamichel@gmail.com, fmoreira@ufla.br, victor.pylro@ufla.br. *Correspondence: Victor Satler Pylro, victor.pylro@ufla.br ORCID: 0000-0003-2154-9150

2, Norwegian School of Fishery Sciences, UiT The Arctic University of Norway, Tromsø, Norway. Email: daniel.morais@uit.no

Abstract

The Brazilian Cerrado is the most biodiverse savanna in the world. However, a large portion of the native vegetation of this biome has been converted into cultivated areas, including coffee plantations. The impact of land-use change on soil fungal communities in the Cerrado is still not well understood. This study evaluated, using next-generation sequencing, the diversity and ecological aspects of soil fungal communities in areas of native Cerrado vegetation and in regenerative and conventional coffee plantations. Furthermore, we assessed the chitinolytic enzymatic potential of fungal strains isolated from soils under these three different land-use types. The Cerrado soil samples showed significant differences in both alpha and beta diversity. Areas Ce1 and Ce2 presented the lowest alpha diversity, while samples Ce3 and Ce4 presented similar alpha diversity to some regenerative and conventional coffee areas. However, fungal groups were similar across the Cerrado areas, with the Basidiomycota phylum being the most abundant relatively, with *Apiotrichum* and *Saitozyma* standing out. In contrast, coffee cultivation areas, both conventional and regenerative, harbored similar fungal groups belonging to the phylum Ascomycota. Regarding chitinase production, the strains originating from native Cerrado soil and regenerative coffee plantations showed the highest chitinase production. These results indicate that land-use

change impacts soil fungal communities in the Cerrado and that this biome harbors a fungal diversity with high biotechnological potential.

Keywords: Brazilian savanna, Desforestation, Enzyme, Fungal biodiversity.

1. INTRODUCTION

The Cerrado is a global biodiversity hotspot and is also considered the largest and most threatened tropical savanna in the world (Silva & Bates, 2002; Strassburg et al., 2017). In the last three decades, agricultural expansion has driven extensive land-use changes in the biome, converting more than half of the native areas into large monoculture fields of maize, wheat, soybean, cotton, sugarcane, coffee, and livestock production (IBGE, 2018; Grande, Aguiar, Machado, 2020; MapBiomas, 2025). Characterized by marked seasonality, with dry winters and rainy summers, the Cerrado plays crucial ecological roles, in addition to harboring a rich diversity with several endemic species of plants and animals (Coman et al., 2024). Despite its flora and fauna being well documented, the fungal diversity of the Cerrado is little known, although these microorganisms show higher diversity and endemism in tropical ecosystems (Tedersoo et al., 2014; Noriler et al., 2018; Mikryukov et al., 2023).

The impact of land-use change in tropical biomes on soil fungal communities is still not well understood, however, the deforestation of native areas alters the quantity and quality of litter, the physicochemical properties of the soil, and the local microclimate (Hartmann et al., 2012; Mollier et al., 2024). These factors are the main determinants of fungal diversity, therefore, land-use change may threaten yet-undiscovered fungal species with extinction (Hartmann et al., 2012; IUCN, 2025). The loss of fungal species compromises ecological functions and interactions that are still poorly understood, besides reducing the availability of molecules and enzymes with high biotechnological potential, such as chitinases. Fungal communities from Cerrado soils present adaptations to adverse environmental conditions, including low humidity, acidic pH, and recurrent fires, which are associated with the production of thermostable and acid-tolerant enzymes with high catalytic efficiency, characteristics of great industrial interest (Gomes et al., 2001; Castro et al., 2008; Araujo et al., 2017; Zhang et al., 2021).

Chitinases (E.C. 3.2.1.14) degrade chitin, a polysaccharide responsible for the rigidity of the fungal cell wall. Its deacetylated derivative, chitosan, acts as a carbon and nitrogen source for fungal species and has broad biomedical applicability due to its biodegradable, antimicrobial, moisturizing, and non-toxic properties (Yusof & Khor, 2000; Jayakumar et al.,

2010; Gómez-Gaviria & Mora-Montes, 2024). In the Kingdom Fungi, chitinases are fundamental for competition among microorganisms, mycoparasitism, nutrition, and cell wall remodeling (Goughenour et al., 2021; Omari et al., 2024). In entomopathogenic fungi, chitinases degrade the chitin of the insect exoskeleton, promoting systemic infection and consequently host death (Paveda, 2021). In this context, biopesticide fungi emerge as a sustainable alternative to conventional agriculture, with some fungal genera, including *Trichoderma*, *Metarhizium* and *Beauveria*, already showing efficiency in pest control (Litwin, Nowak, Różalska, 2020).

Thus, investigating the impact of land-use change on fungal communities and exploring the chitinolytic potential in biodiverse and little-explored ecosystems such as the Cerrado allows the evaluation of species loss, the identification of new fungal lineages, and access to a reservoir of biomolecules with potential for sustainable agricultural production. Therefore, this study aimed to evaluate the impact of deforestation on soil fungal communities and to investigate the chitinase production capacity of isolates from the Cerrado biome.

2. MATERIAL AND METHODS

2.1 Study sites and sampling

Soil samples were collected during the rainy season in two areas located in the municipality of Patrocínio, Minas Gerais, Brazil (Figure 1) (the geographic coordinates of the sampling sites are provided in Supplementary Material 1). Each area comprised three land-use systems: native Cerrado, conventional coffee plantations, and regenerative coffee plantations. Soil sampling consisted of two paired coffee plots, each pair including one conventional and one regenerative plantation. Coffee plots within each pair shared the same cultivar, planting age, and irrigation system (when present), differing only in management practices. For each pair of coffee plots, one transect was established in the adjacent native Cerrado, resulting in six transects per area and twelve transects across the experiment. Within each transect, soil samples were obtained from five distinct points spaced 50 meters apart. At each sampling point, subsamples were collected and homogenized to form a single composite sample, which was stored in sterile plastic bags.

Soil sampling was carried out using an auger at a depth of 0–15 cm. A total of 60 samples (20 from each land-use system) were collected and stored at –20 °C for subsequent molecular analyses.

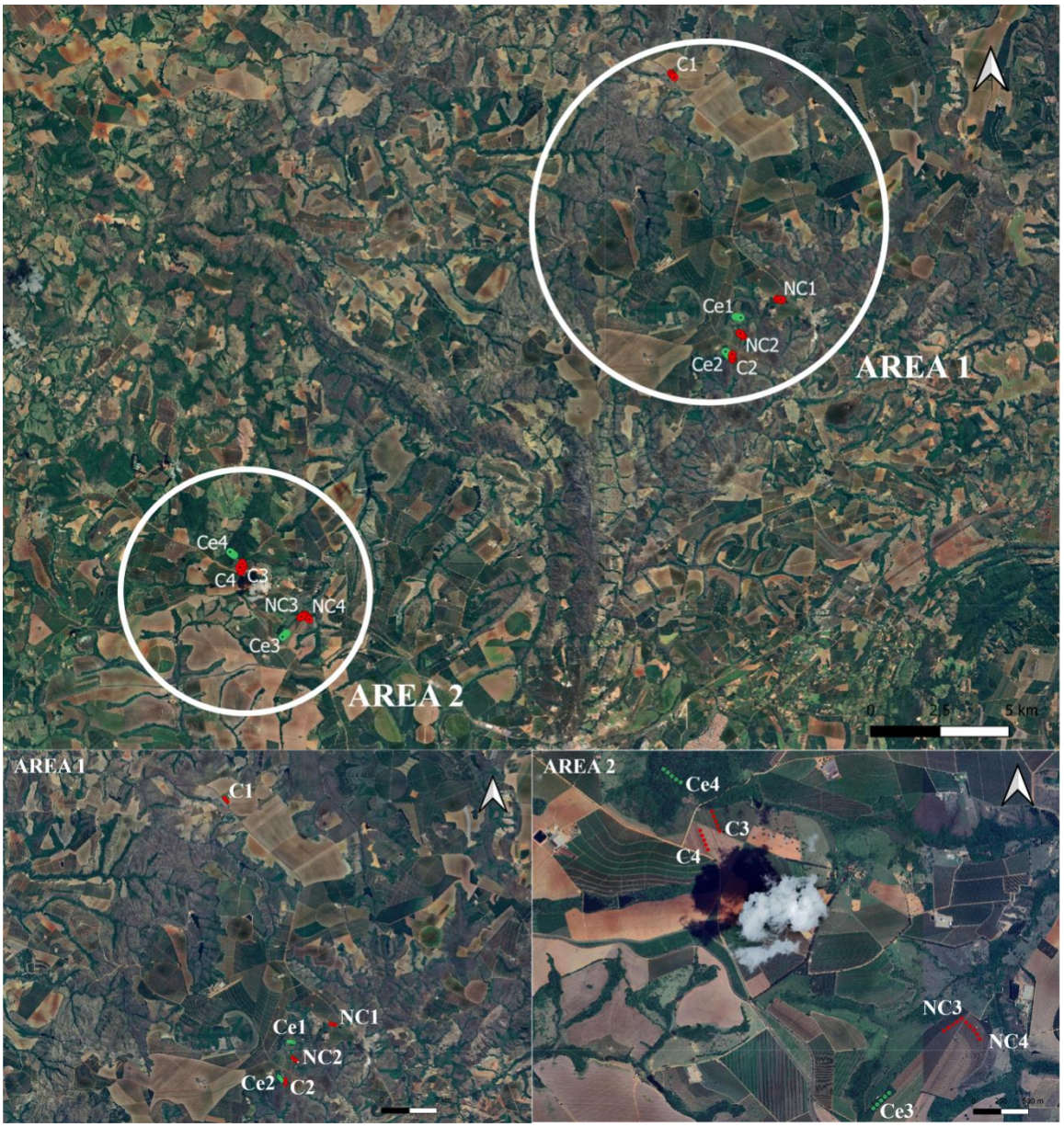


Figure 1. Sampling areas. Area 1: Ce1 - Cerrado; C1 - conventional coffee, cultivar Catuaí 144, irrigated; NC1 - regenerative coffee, cultivar Catuaí 144, irrigated; Ce2 - Cerrado; C2 - conventional coffee, cultivar Topázio, irrigated; NC2 - regenerative coffee, cultivar Topázio, irrigated. Area 2: Ce3 - Cerrado; C3 - conventional coffee, cultivar Catuaí 62, non-irrigated; NC3 - regenerative coffee, cultivar Catuaí 66, non-irrigated; Ce4 - Cerrado; C4 - conventional coffee, cultivar IBC 12/IAC 125, irrigated; NC4 - regenetative coffee, cultivar IBC 12/IAC 125, irrigated.

2. Fungal community analysis

2.2.1 Extraction of DNA total

The DNA from each sample soil was extracted and purified using the DNeasy PowerSoil Pro Kit (Qiagen, Germany), following the manufacturer's instructions. The purity

of the extracted DNA was observed by spectrophotometry using the Nanodrop One equipment (Nanodrop Technologies, Wilmington, DE, USA) (260/280 nm), and quantification was performed with the Qubit® 4.0 fluorimeter, using Qubit® dsDNA HS Assay Kit (Invitrogen TM).

2.2.2 Metataxonomics, analysis of data and statistics

The structure of the microbial communities in soil samples was analyzed using metataxonomics. The ITS region was amplified using the primers ITS1F/ITS2 (Kemler et al., 2013), according to the Ion Amplicon library preparation manual, PCR amplification was performed with barcodes attached to an A sequence from the Ion adapter and a trP1 sequence from the Ion adapter. The ITS region was amplified using the following protocol: 3 min at 96°C, followed by 30 cycles of 30 sec at 96°C, 45 sec at 45°C, and 1 min at 72°C, with a final extension of 7 min at 72°C. Amplicons containing adapters and barcode sequences were purified using AMPureBeads (1.2×, Beckman Coulter). Emulsion PCR was performed using the Ion OneTouch 2™ system and the Ion Template PGM™ OT2 400 kit (Life Technologies). Sequencing of the amplicon libraries was conducted on an Ion 318™ Chip Kit v2 with the PGM Ion Torrent sequencer (Life Technologies). Post-sequencing, the PGM software filtered the Ion Torrent fastq files to remove polyclonal and low-quality sequences.

The operational taxonomic unit (OTU) table was generated using the UPARSE pipeline, following the Brazilian Microbiome Project recommendations (Pylro et al., 2014). Reads were truncated to 200 base pairs, and quality filtering allowed a maximum error rate of 1.5. Filtered reads were dereplicated and clustered with a 97% similarity threshold, and representative sequences were obtained for each OTU (Edgar, 2013). Taxonomic classification was performed using QIIME with the UCLUST method, referencing the UNITE database (version 10.0) (Kõljalg et al. 2005). Alpha diversity was assessed using the Chao1 richness index, while beta diversity was calculated with the Bray-Curtis index (Bray and Curtis, 1957; Chao, 1984). A Principal Coordinate Analysis (PCoA) was performed to visualize microbial community dissimilarity between samples, and a PERMANOVA test was applied to assess the statistical significance of the observed differences. Finally, the web-based tool Microbiome Analyst was used to generate abundance and diversity plots, providing a comprehensive visual representation of microbial community composition and diversity dynamics (Dhariwal et al., 2017).

2.3 Evaluation of chitinase production

2.3.1 Fungal isolation

The serial dilution technique was used to isolate a total of 1.481 fungal strains from 60 soil samples (20 samples from the native Cerrado, 20 from the conventional coffee production area and 20 from the regenerative coffee production area). 25 g of each soil sample was diluted in 225 ml of 1% peptone water. Then, 0.1 mL aliquots of the dilutions were transferred to standardized culture media, Dicloran Rose Bengal Chloramphenicol (DRBC) and Dicloran Glycerol 18% (DG18). After 7 days of incubation at 25 °C, the colonies were characterized and transferred to MEA (Malt Extract Agar) and PDA (Potato Dextrose Agar) culture media at 25 °C for 7 days. After obtaining pure colonies, the isolates were preserved using the Castellani method (1967) and frozen in 20% glycerol.

2.3.2 Preparation of colloidal chitin and culture media

Colloidal chitin was prepared from shrimp shells and commercial chitin (Sigma-Aldrich, Germany) using the method described by Roberts and Selitrennikoff (ROBERTS & SELITRENNIKOFF, 1988). A total of 300 mL of concentrated HCl (38%) was added to 50 g of chitin and incubated at 4 °C overnight. Subsequently, ice-cold 95% ethanol was added to neutralize the acid and kept overnight at 4 °C. Distilled water was then added, and the mixture was centrifuged at 8.000 g for 7 min at 4 °C. The pellet was washed with sterile distilled water until the alcohol odor was completely removed. The resulting colloidal chitin was pasty, with 90–95% moisture content, neutral pH, and was stored at 4 °C.

For the production of chitinase in liquid medium, fungal isolates were cultured in test tubes (5 mL) containing 1.0% colloidal chitin, 0.03% $MgSO_4 \cdot 7H_2O$, 0.3% $(NH_4)_2SO_4$, 0.2% KH_2PO_4, 0.1% citric acid monohydrate, 0.02% Tween-80, and 0.015% bromocresol purple. The pH was adjusted to 4.7 and the medium was autoclaved at 121 °C for 15 min (AGRAWAL & KOTASTHANE, 2012). The fungal isolates were reactivated in MEA culture medium, and after 7–10 days of growth, fresh culture plugs (8 mm) were inoculated into the liquid chitin medium and incubated at 28 °C for 10 days in a shaker incubator (160 rpm).

2.3.3 Assay enzimatic and spectrophotometric determination of chitinase activity

Chitinolytic activity was assessed by measuring the release of reducing sugars from colloidal chitin, used as a substrate. The reaction mixture, consisting of 0.1 ml of 1% coilodal chitin (neutral pH) and 0.1 ml of the crude enzyme extract, was incubated for 1 h at 40°C in a water bath. After the incubation period, the enzymatic reaction was stopped with the addition of 0.6 ml of 3,5-dinitrosalicylic acid, followed by heating at 100°C for 5 min. A standard curve was generated from serial dilutions of N-acetyl-D-glucosamine (from 0 to 5 mg mL^{-1}) and was used to determine the concentration of reducing sugars. One unit (U) of chitinase activity was defined as the amount of enzyme required to release 1 mg of N-acetyl-D-glucosamine from chitin/min. Absorbance was measured at 530 nm using a UV spectrophotometer, along with serial dilutions of N-acetyl-D-glucosamine and blanks.

3. RESULTS

3.1 Diversity analysis of soil samples

The alpha diversity of fungal communities was evaluated based on ITS gene sequences, using the Chao1 (Figure 2), Shannon (Fig. S1), and Observed (Fig. S2) indices. The analysis of variance show significant differences in fungal richness among the types of land use (ANOVA, F = 18.914, p = 7.3175e-14; F = 14.527, p = 8.6883e-12; F = 34.826, p = 3.992e-19, respectively). Post-hoc comparisons indicated that the native Cerrado soil samples (Ce1 and Ce2) showed the lowest diversity values, indicating fungal communities with lower taxon richness. In contrast, the highest alpha diversity was recorded in the regenerative coffee areas (NC3 and NC4). The remaining Cerrado samples (Ce3 and Ce4) showed diversity levels comparable to those found in regenerative and conventional coffee systems. This pattern was observed across the different Chao1, Shannon, and Observed indices analyzed (Fig. 2, S1 and S2).

Based on the results of the permutational multivariate analysis of variance (PERMANOVA) and using different beta diversity indices, Bray-Curtis, Jaccard, and Jensen-Shannon, significant effects were observed in the composition of the fungal community. The ordination plot showed the formation of three distinct clusters (Figure 2). The native Cerrado samples (Ce1 and Ce2) showed clear separation from the others along Axis 1, standing out for their uniqueness in relation to the other areas. This indicates significant differences in the fungal composition between the native Cerrado and the soils of the coffee-producing area. A

second group comprised of soil samples, including regenerative and conventional coffee systems (C1, NC1, C2, and NC2), independently of the coffee variety.

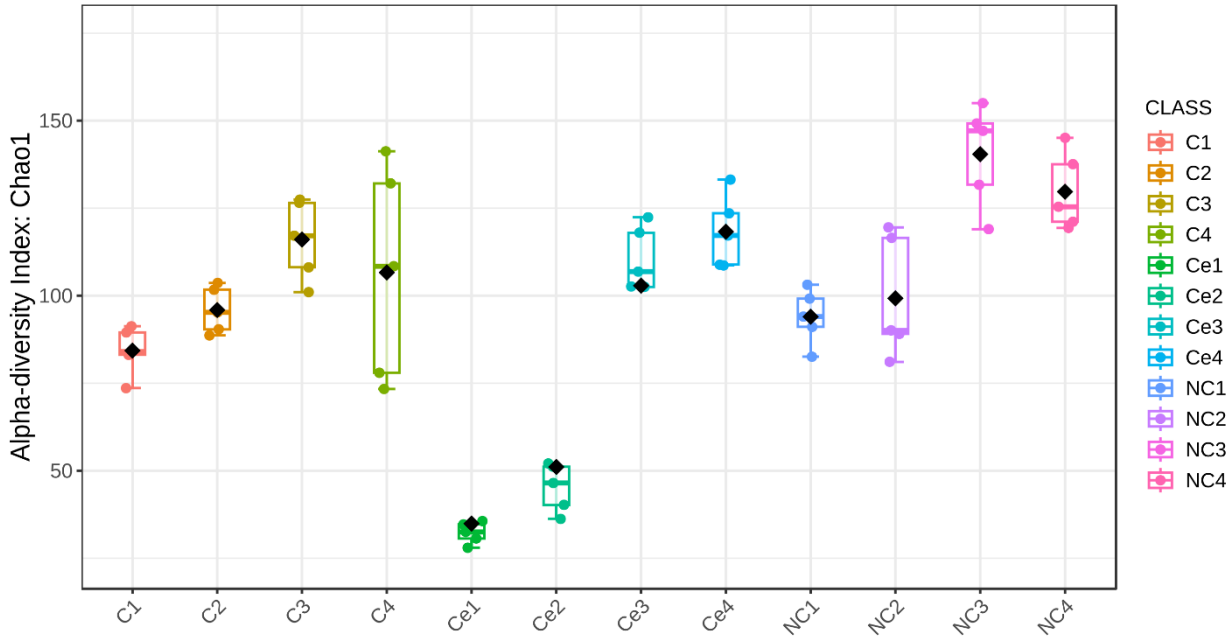


Figure 2. Comparative analysis of Alpha Diversity (Chao1 index) based on ITS gene sequencing in different soil treatments. Boxplots show Chao1 diversity for each land use type (n = 4 eplicates for each land use type). Y-axis: Alpha Diversity Index: Chao1 (higher values indicate greater diversity). X-axis: sample land use types (C1–C4: conventional coffee; Ce1–Ce4: native Cerrado; NC1–NC4: regenerative coffee). Colors identify the sample land use types, as shown in the legend. P-value: 8.6883e-12.

These areas are located closer together, indicating that the fungal community may be influenced by local soil characteristics and that the beta diversity of these microorganisms is similar across the different coffee management systems, whether conventional or regenerative (Fig. 2). A third group consisted of samples that included two subareas of native Cerrado (Ce3 and Ce4), two regenerative coffee production areas (NC3 and NC4), and two conventional coffee production areas (C3 and C4). All of these areas are also located closer together (Fig. 2). Also, in the other indixes, Jaccard and Jensen-Shannon, the cluster patterns are consistent, indicating that agricultural management and local edaphic conditions may influence the composition and structure of fungal communities (Fig. S3 and S4).

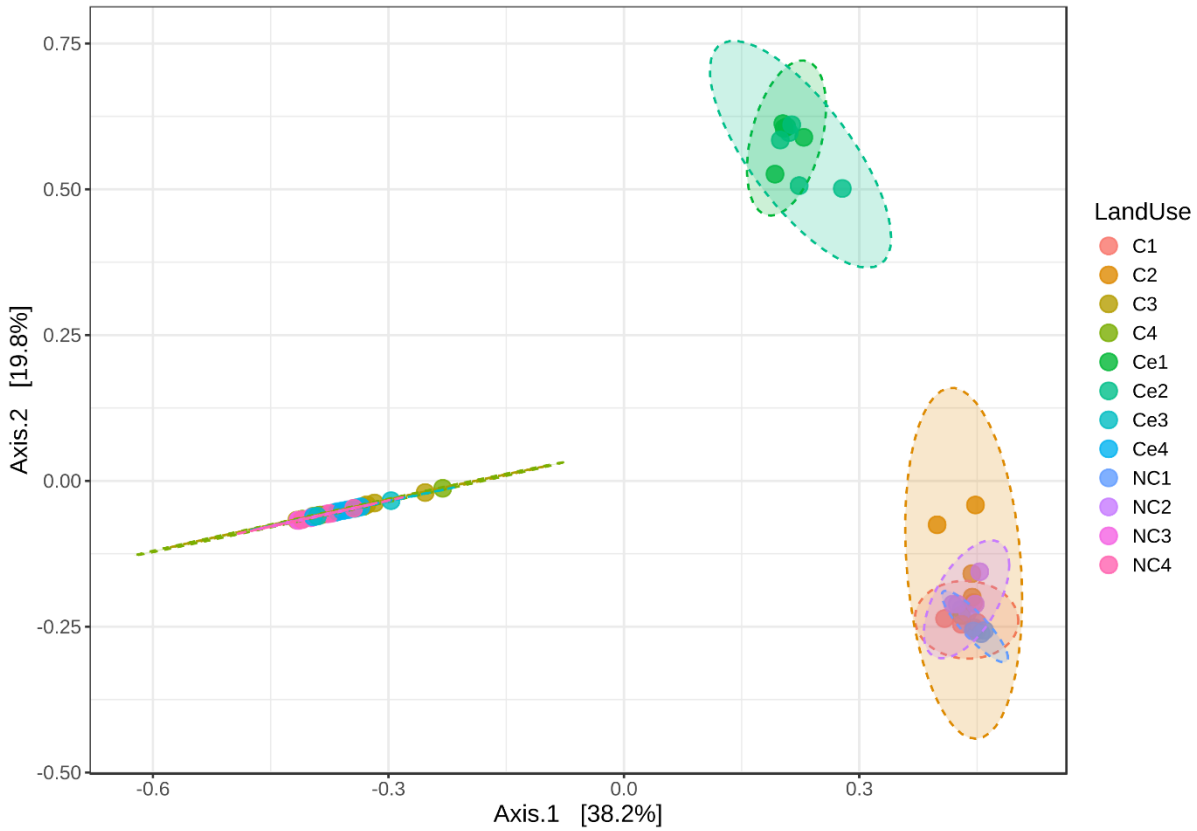
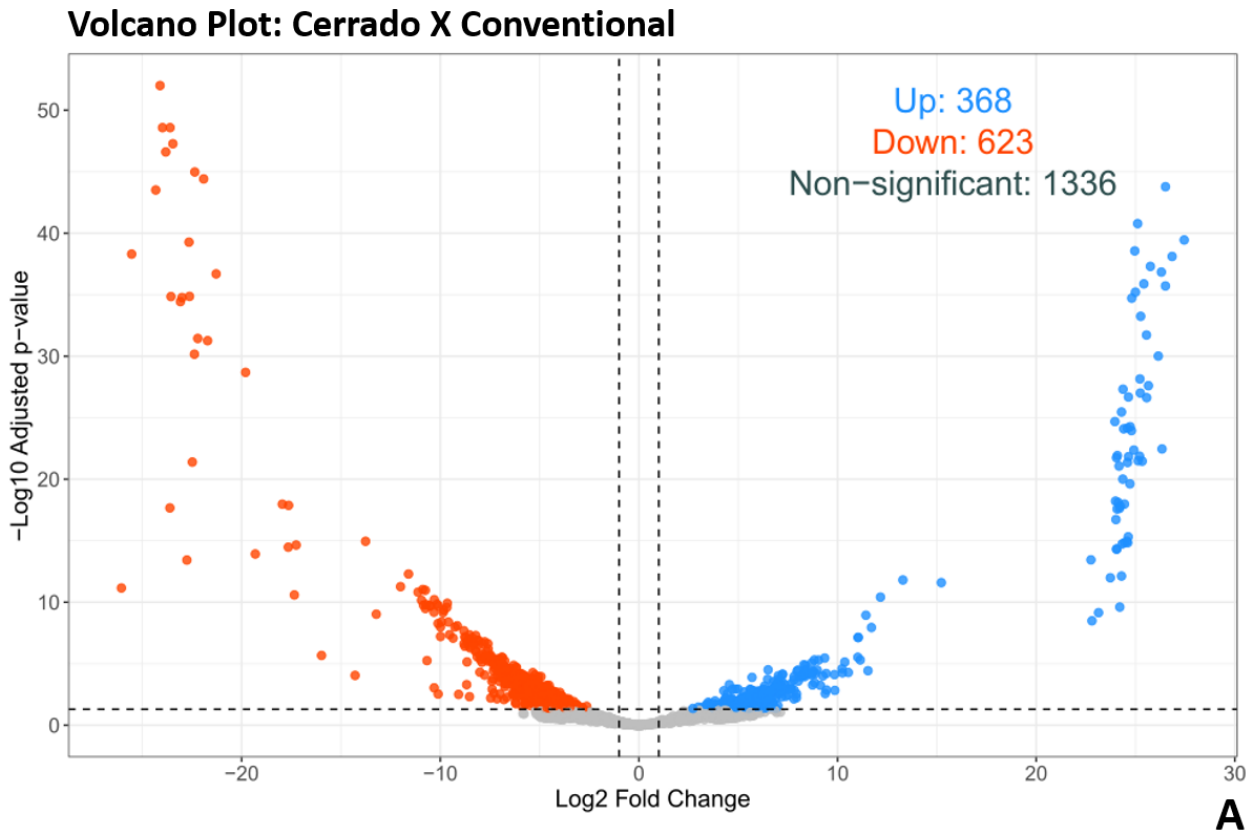
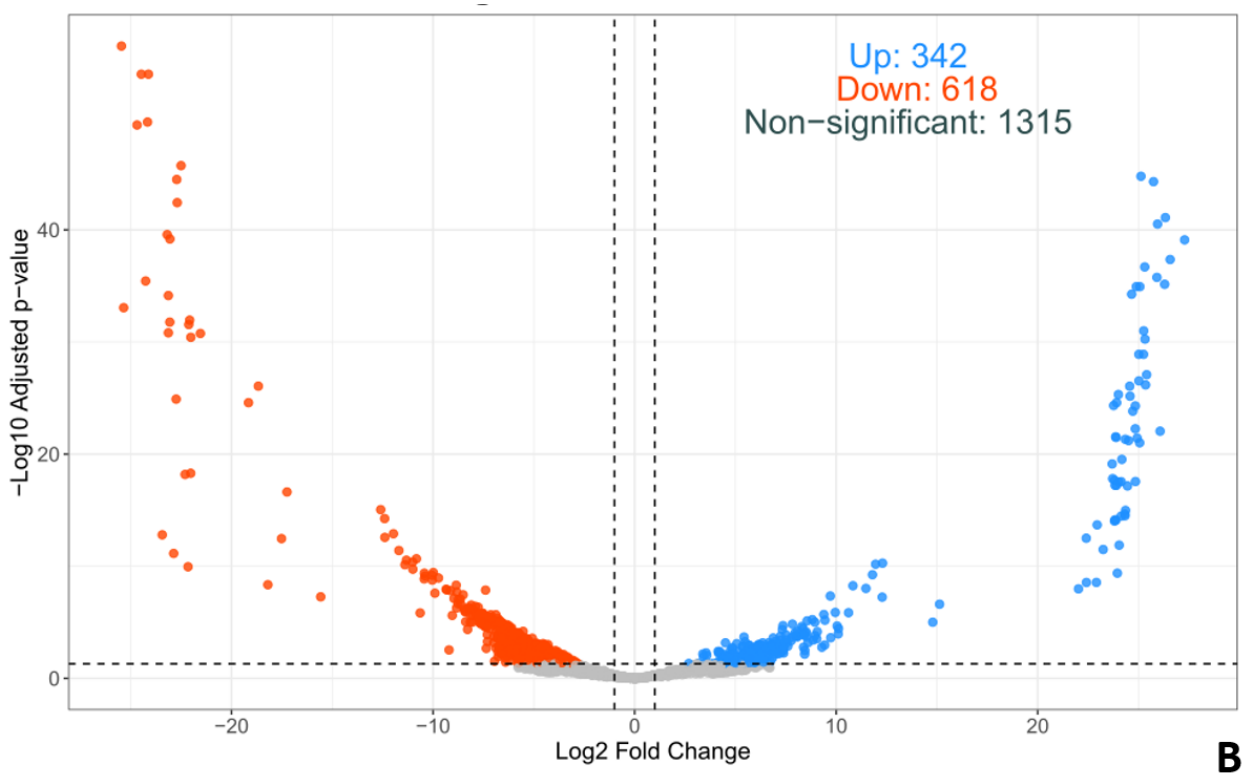


Figure 3. Beta-diversity analysis of fungal community in soil samples using the Bray-Curtis index and PCoA based on ITS gene sequencing. Closer clusters indicate greater similarity in fungal community structure between samples. C1–C4: conventional coffee; Ce1–Ce4: native Cerrado; NC1–NC4: regenerative coffee. Colors identify the sample land use types, as shown in the legend, and multivariate analysis of variance (PERMANOVA) indicated significant differences between land use types ($p = 0.001$).

A detailed examination of the fungal community composition in the analyzed areas of native Cerrado and conventional and regenerative coffee plantations, based on modeling OTU abundance using DESeq2, corroborates the alpha diversity data. A higher number of taxa are more abundant under conventional coffee management (623) compared to native Cerrado (368) (Figure 4a). The comparison between native Cerrado and regenerative coffee plantations (Figure 4b) shows that 618 taxa are more abundant in regenerative coffee, whereas 342 taxa are more abundant in native Cerrado. The comparison between soil samples from regenerative and conventional coffee areas (Figure 4C) shows that most taxa (1451) occur in both management systems, with no significant differences. However, 62 fungal taxa exhibited significantly higher abundance in regenerative coffee areas, whereas 83 taxa were significantly more abundant in conventional coffee areas (Fig. 4C).


Volcano Plot: Cerrado X Conventional
Up: 368
Down: 623
Non−significant: 1336
−Log10 Adjusted p−value
Log2 Fold Change
50
40
30
20
10
0
−20
−10
0
10
20
30

A


Volcano Plot: Cerrado X Regenerative
Up: 342
Down: 618
Non−significant: 1315
−Log10 Adjusted p−value
Log2 Fold Change
40
20
0
−20
−10
0
10
20

B

.

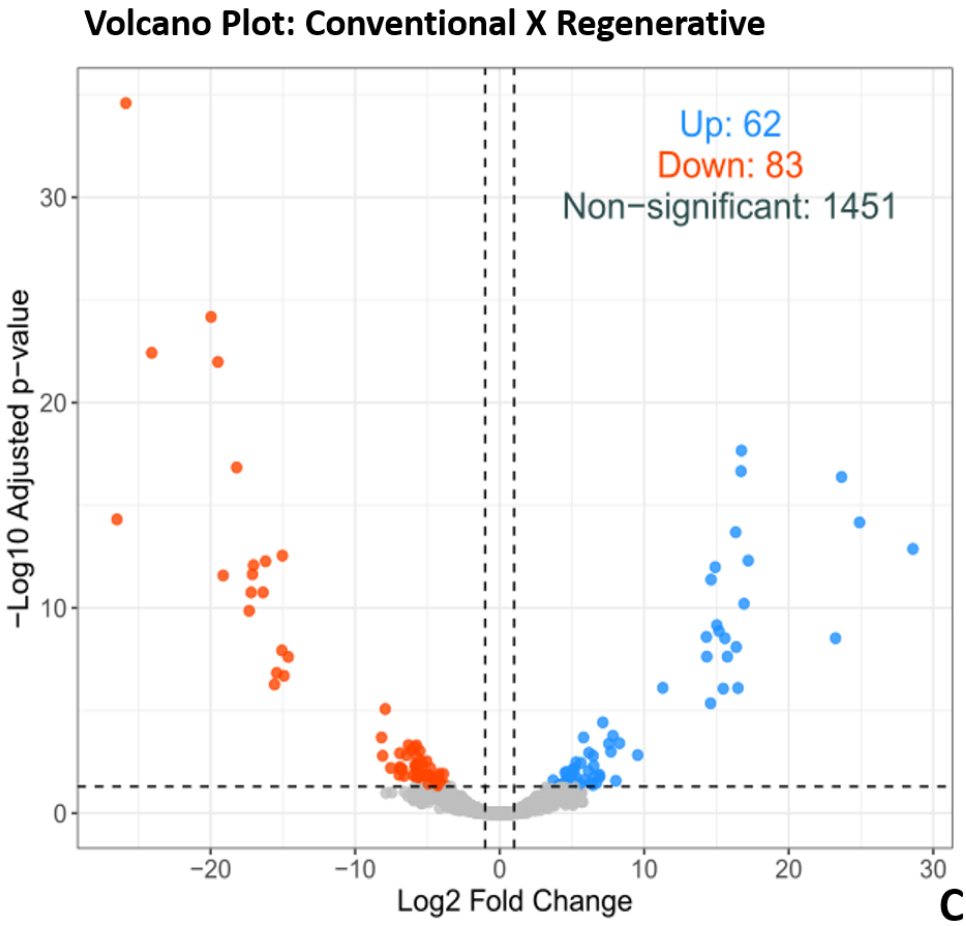


Figure 4. Volcano plots from DESeq2 analysis comparing the abundance of OTUs among different land uses, native Cerrado vs. conventional coffee (Fig. 4A); native Cerrado vs. regenerative coffee (Fig. 4B); and conventional coffee vs. regenerative coffee (Fig. 4C). The points represent individual OTUs. Colored points (above the dotted line) indicate statistically significant taxa (p-value < 0.05). The logarithmic variation (x-axis) shows the magnitude of enrichment; positive values indicate taxa more abundant in coffee systems, while negative values indicate enrichment in native Cerrado. A greater number of taxa were significantly enriched in both coffee systems compared to the native Cerrado (Fig. 4A and 4B). In the comparison between coffee areas, 1.451 taxa show no significant difference (p > 0.05). However, 62 taxa are more abundant in the regenerative system, while 83 taxa are more abundant in the conventional system (Fig. 4C).

3.2 Taxonomic classification of soil treatments

Four to six fungal phyla were detected in soil samples from native Cerrado, conventional coffee, and regenerative coffee (Figure 5). Although the native Cerrado samples (Ce1, Ce2, Ce3 and Ce4) varied in alpha and beta diversity, the phylum Basidiomycota showed the highest relative abundance in samples from these areas, particularly in samples Ce1 and Ce2. In contrast, the phylum Ascomycota exhibited the highest relative abundance in soil samples from conventional and regenerative coffee cultivation, peaking in the samples C1, NC1 and NC2 (Figure 5). These results indicate that, regardless of management

conventional or regenerative, similar fungal groups are favored in areas with these land-use systems.

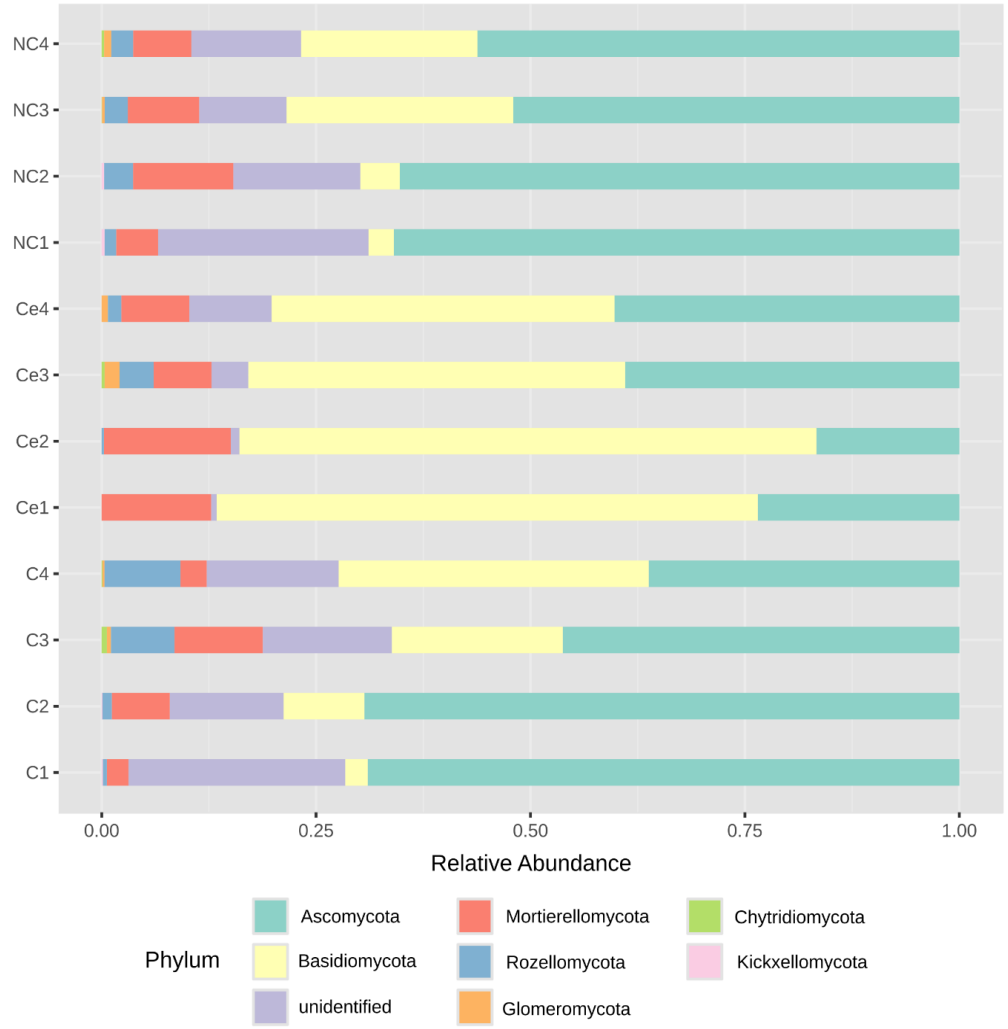


Figure 5. Taxonomic composition of the soil fungal community at the phylum level in areas of native Cerrado vegetation (Ce1–Ce4), conventional coffee cultivation (C1–C4), and regenerative coffee cultivation (NC1–NC4). Bars represent the relative abundance of the major fungal phyla identified.

Genus-level analyses show clear patterns in fungal community composition across native Cerrado and coffee management systems (Figure 6). In soil samples from native Cerrado and one area of conventional coffee (C4), *Saitozyma* was the most abundant taxon, followed by *Apiotrichum* (Figures 6 and 7). The genera *Metarhizium* and *Trichoderma* exhibited high to intermediate relative abundances in Cerrado areas, however, within the cultivated systems, *Metarhizium* was more abundant in regenerative coffee soils, whereas *Trichoderma* prevailed in conventional coffee areas. Other genera, including *Fusarium*, *Pyrenochaetopsis*, *Acremonium*, *Cladopholophora*, *Sedosporium*, *Wardomyces*, *Exophiala*,

Cladosporium, *Clavaria*, and *Aspergillus*, showed intermediate to high abundance in conventional and regenerative coffee systems. Except for *Clavaria*, all these genera belong to the phylum Ascomycota. These relative abundance patterns were further supported by LEfSe analysis (Figure 6), which identified the same taxa as potential biomarkers differentiating native Cerrado, conventional coffee, and regenerative coffee systems.

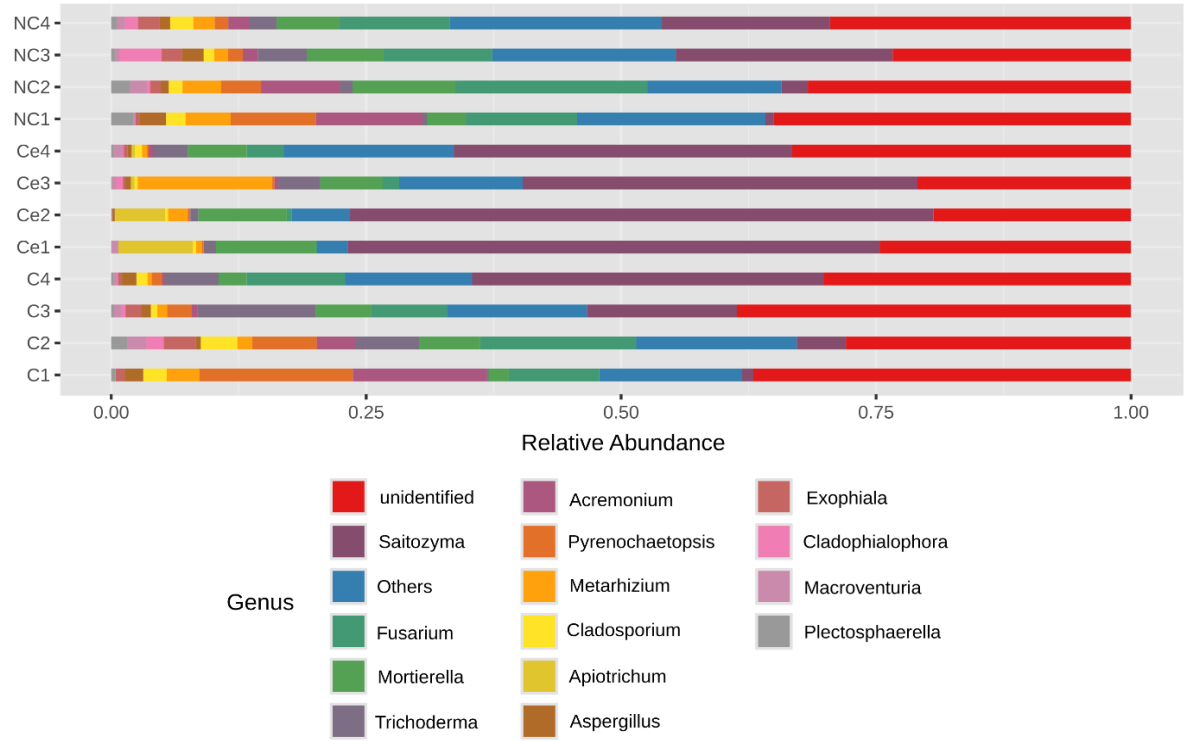


Figure 6. Taxonomic composition of the soil fungal community at the genera level in areas of native Cerrado vegetation (Ce1–Ce4), conventional coffee cultivation (C1–C4), and regenerative coffee cultivation (NC1–NC4). Bars represent the relative abundance of the major fungal genera identified.

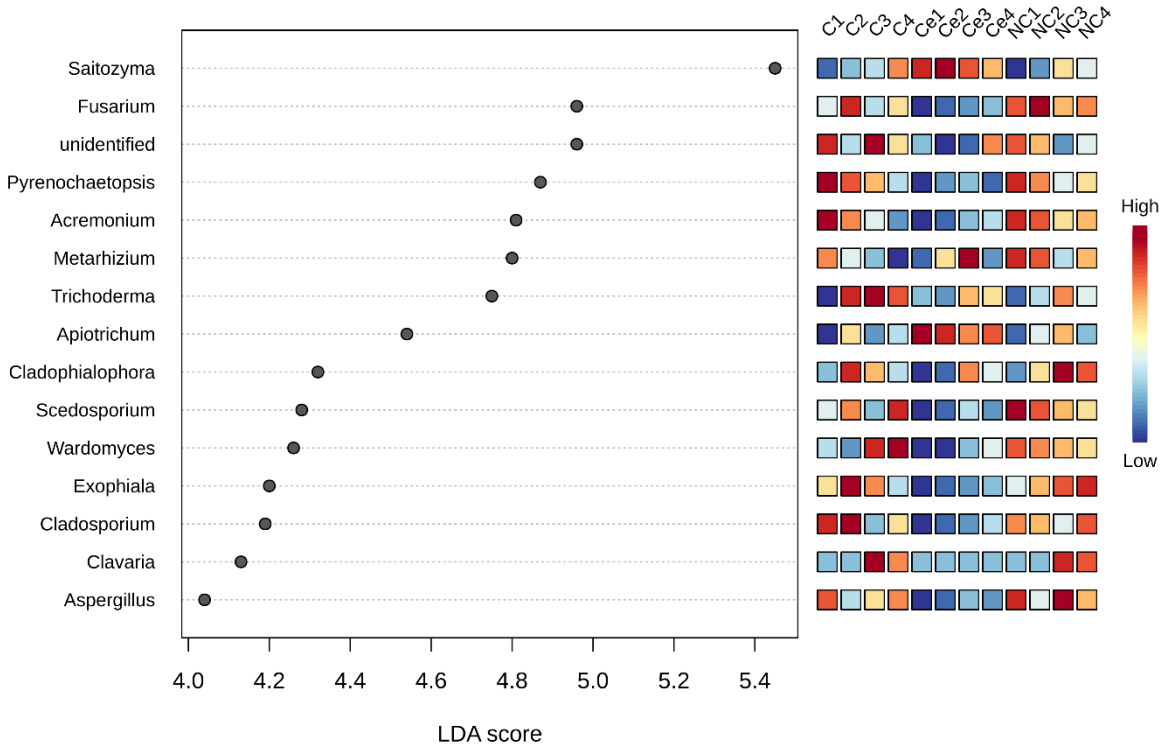


Figure 7. Biomarker discovery based on LEfSe analysis of fungal communities at the genus level across native Cerrado, conventional, and regenerative coffee systems. Differentially abundant taxa were identified using the Kruskal–Wallis test ($p < 0.05$) followed by Linear Discriminant Analysis (LDA), with genera showing the highest effect sizes highlighted. The plot displays genera with significantly different relative abundances among land-use types, serving as potential fungal biomarkers of each management system.

3.3 Chitinolytic activity of fungal isolates

A total of 1.481 fungal isolates were evaluated for chitinase production. Of these, 646 strains were obtained from soil samples of native Cerrado, 386 from conventional coffee areas, and 449 from regenerative coffee management areas. Fungal isolates from different land uses showed variation in chitinase production with significant differences based on the Scott-Knott clustering test ($p < 0.05$). The fungal isolates from conventional coffee cultivation areas formed two groups (Fig. 8). The group of fungal isolates (blue) produced a lower amount of the enzyme, while the second group (orange) showed higher chitinase production, with isolate 147 standing out, which produced 127 U min^{-1} (Fig. 8).

Fungal isolates from native Cerrado areas formed seven distinct groups, indicating significant differences in enzyme production among these isolates. The fungal isolates of the first group (dark blue) showed lower enzyme production under the tested conditions, while the isolates represented by the yellow color (group 7) stood out with higher chitinase production, ML280b and RM55, which produced 178 and 192 U min^{-1}, respectively (Fig. 9).

The fungal isolates from regenerative coffee cultivation showed the greatest variation in chitinase production. The Scott-Knott clustering test ($p < 0.05$) formed fourteen distinct groups. The smallest group of fungal isolates that produced chitinase is represented by the dark blue color, while the highest chitinase producer (225 U min^{-1}) under the tested conditions, isolate IR280, is represented by the orange color (Fig. 10).

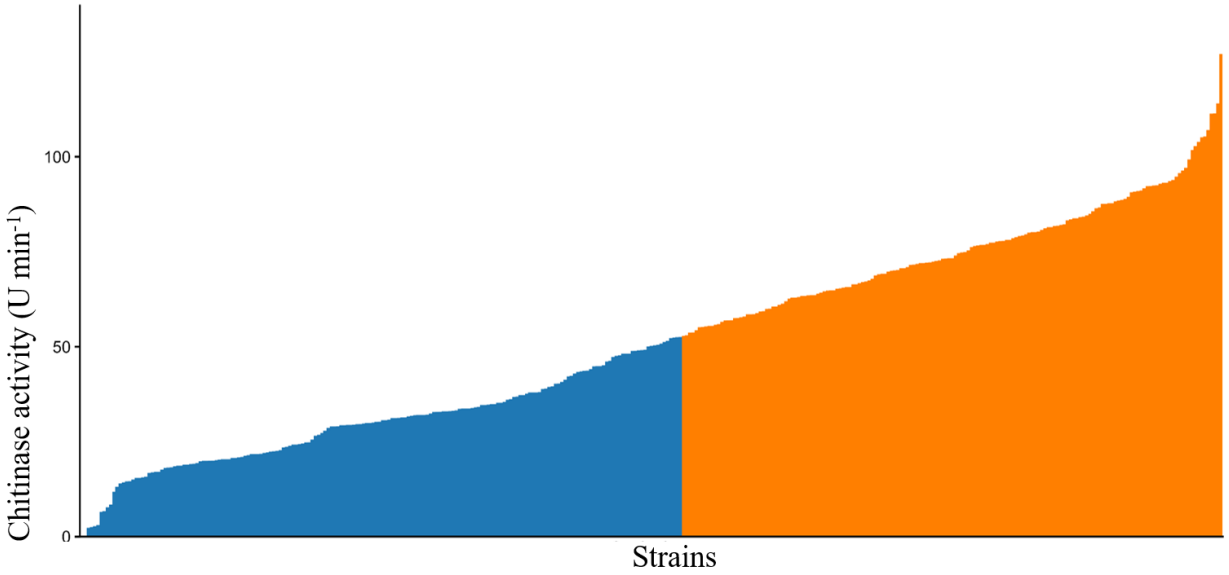


Figure 8. Chitinase activity U min^{-1} of fungal isolates obtained from conventional coffee soil. The colors differ from each other along the X-axis due to significant differences in enzymatic production among the isolates ($p < 0.05$) according to the Scott-Knott means test. The enzymatic activity ranged from the lowest (blue) to the highest (orange) level of production, with emphasis on strain 147, which produced 127 U min^{-1}.

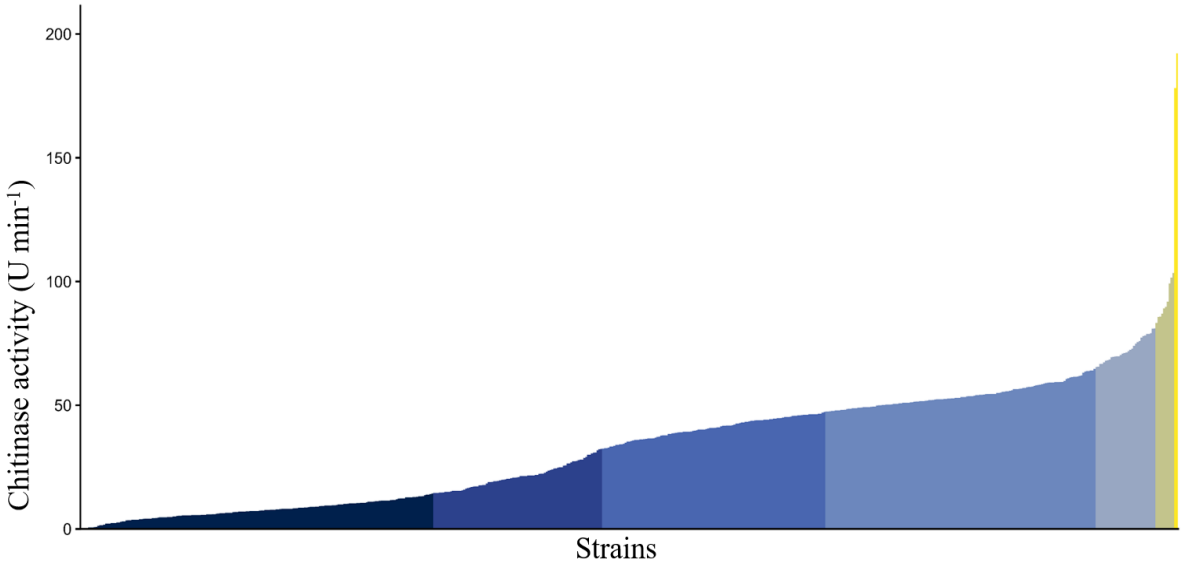


Figure 9. Chitinase activity U min^{-1} of fungal isolates obtained from native Cerrado soil. The colors differ from each other along the X-axis due to significant differences in enzymatic production among the isolates ($p < 0.05$) according to the Scott-Knott means test. The enzymatic activity ranged from the lowest (dark blue) to the highest (yellow) level of production, with emphasis on strains ML280b and RM55, which produced 178 and 192 U min^{-1}, respectivelly.

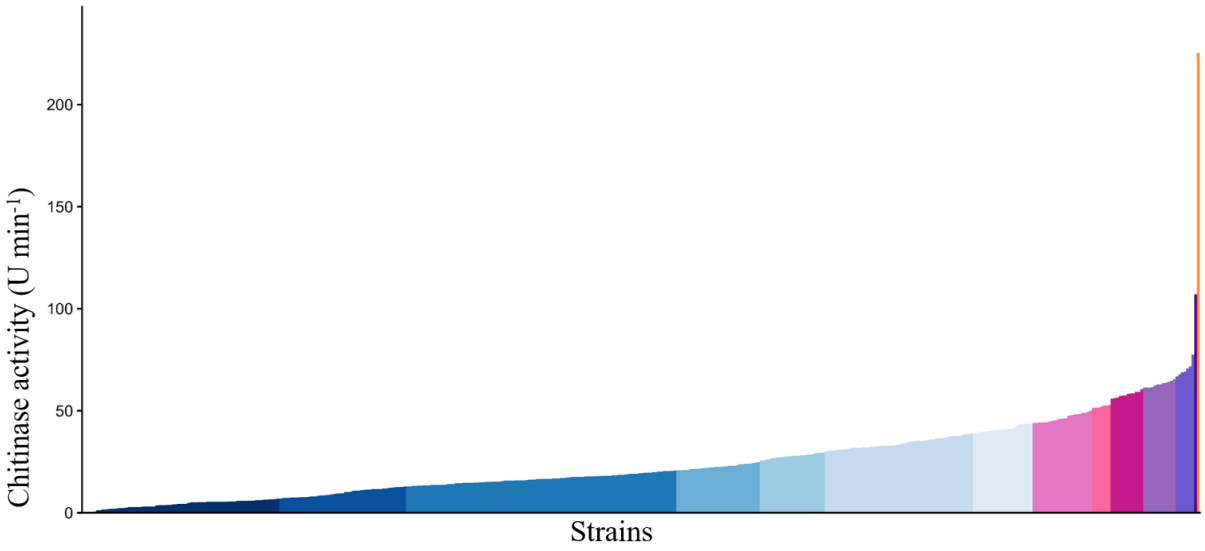


Figure 10. Chitinase activity U min^{-1} of fungal isolates obtained from regenerative coffee soil. The colors differ from each other along the X-axis due to significant differences in enzymatic production among the isolates ($p < 0.05$) according to the Scott-Knott means test. The enzymatic activity ranged from the lowest (dark blue) to the highest (orange) level of production, with emphasis on strains IR280, which produced 225 U min^{-1}.

4. DISCUSSION

Knowledge about how land-use changes affect soil fungal communities in the Brazilian Cerrado, one of the world's biodiversity *hotspots*, is still limited (Brinkmann et al. 2019). In this context, this work provides new insights into how different coffee production systems and native Cerrado areas harbor distinct fungal communities, both in terms of richness and taxonomic composition.

According to the results, alpha diversity is lower in areas of the native Cerrado (Ce1 and Ce2) and higher in areas of conventional and regenerative coffee plantations. This difference can be explained by the Intermediate Disturbance Hypothesis (IDH), which proposes that maximum species diversity is achieved when ecological disturbances occur at intermediate frequency or intensity (Kang et al., 2024). In coffee plantations, both conventional and regenerative, the application of chemical fertilizers, manure, or compost, combined with the accumulation of organic matter in the soil, since coffee is a perennial crop, creates favorable conditions for increased fungal diversity. Similar results were found by Zhang and colleagues (2018), indicating that conventional agricultural soils may have greater microbial diversity than in areas of native vegetation, where resource availability is limited.

It is worth noting that, although the ITS region is the official barcode for fungi, analyses using this gene may present limitations in assessing alpha diversity, given the number of copies of the nuclear ribosomal operon varies among different fungal taxa, ranging from 14 to over 1.000, and the high intragenomic variability related to this factor (Schoch et al., 2012; Lofgren et al., 2019; Cedeño-Sanchez et al., 2024). Furthermore, some fungal groups have low amplification success when general fungal ITS primers are used, and DNA extraction efficiency can vary between fungal lineages (Amend et al. 2010; Tedersoo et al., 2015, 2018).

Our beta diversity results demonstrate a distinct pattern in fungal composition between native Cerrado soil samples. The well-defined cluster for samples from this biome Ce1 and Ce2, indicates that the fungal community composition of this environment is distinct from the agricultural areas. In contrast, other Cerrado samples Ce3 and Ce4, formed a cluster with soil samples from conventional and regenerative coffee cultivation. These areas are relatively close and therefore share similar edaphic characteristics, moreover, these Cerrado fragments may be affected by edge effects, such as the influx of dust and fungal propagules originating from coffee cultivation areas, since these areas exhibit a high abundance of the phylum Ascomycota, a group characterized by rapid growth and the production of thousands of easily

dispersed spores. For example, these samples from native Cerrado (Ce3 and Ce4) showed medium abundance of the genus *Trichoderma*, whereas in Cerrado subareas from area 1, the abundance of this genus is low (Figure 7).

Despite these differences, the analyzed native Cerrado areas exhibit a high relative abundance of the phylum Basidiomycota, with emphasis on *Apiotrichum* and *Saitozyma*, particularly in subareas Ce1 and Ce2. These genera are yeast-like fungi, and the taxon *Apiotrichum* generally consists of species associated with soil and water, however, some species of this genus have been found in various substrates, such as caves and plant leaves, while others have been isolated from grape wine and other fermented alcoholic beverages (Li et al., 2020; Takashima et al., 2020; Chen et al., 2022; Wei et al., 2022; Crous et al., 2024).

The genus *Saitozyma* is highly abundant in native Cerrado samples and shows medium abundance in one subarea of conventional and regenerative coffee cultivation (C4 and NC3). These data are consistent with previous reports describing the occurrence of this genus in both native vegetation and anthropized soils, suggesting that members of *Saitozyma* are widely distributed (Moreira & Vale, 2017). In another study, Figueiredo and collaborators (2023) demonstrated that the species *Saitozyma podzolica* is highly abundant in preserved areas of the Cerrado and proposed its potential as a bioindicator of soil quality. The predominance of the genus *Saitozyma* in our dataset reinforces the importance of this yeast in the soil fungal communities of this biome, and this taxonomic lineage may play an important role in ecosystem functioning.

In coffee-producing areas, under both management systems, the phylum Ascomycota is highly abundant. This may be due to rapid colonization, production of large numbers of spores, and the easy dispersal of some genera within this phylum, such as *Aspergillus*, *Cladosporium* and *Fusarium* (Egidi et al. 2019). These results are similar to those of a 2010 study and demonstrate that Ascomycetes remained the most abundant fungi in agricultural soils (Deacon et al. 2006, Manici et al. 2024).

Although the relative abundance of the genera is generally similar among the coffee plantations, the genus *Trichoderma* is more abundant in most subareas of conventional coffee, whereas in most subareas of regenerative coffee, the genus *Metarhizium* appears with medium or high abundance. The genus *Trichoderma* has a high competitive capacity in disturbed environments, and some studies show that this group of microorganisms is resistant and adapts well to sites with synthetic fertilization and is resistant to fungicides (Küpper et al. 2022, Mulatu et al. 2022, Cun et al. 2024). These practices are common in conventionally managed areas. In contrast, some studies have shown that the greater abundance of

entomopathogenic fungi, such as species of the genus *Metarhizium*, occurs in soils of organically managed fields compared to conventionally managed fields (Miętkiewski et al. 1997, Klingen et al. 2002, Clifton et al. 2015). Other studies suggest that the application of organic fertilizers may be the main determining factor for the increase in the abundance of *Metarhizium* spp. (Klingen et al. 2002, Fernández-Bravo et al. 2021).

The lower abundance of basidiomycetes in agricultural areas may be due to the absence of thick woody debris in the litter biomass, resulting in low lignin residues (Bail et al. 2022). Meanwhile, native Cerrado areas showed a high abundance of Basidiomycetes. This group of microorganisms plays fundamental roles in forests, as they are deeply involved in the decomposition of wood, leaf litter, and soil organic matter. These ecological processes are essential for terrestrial carbon cycling (Casieri et al. 2010, Anastasi et al. 2019).

These data demonstrate that native Cerrado soils have distinct composition and fungal structures than those found after land use change for coffee cultivation, whether under regenerative or conventional management. Despite the fungal community is similar between both systems analyzed in these studies, regenerative management can provide environmental, economic, and human health benefits, while in conventional cultivation, the continuous use of pesticides and synthetic fertilizers can contaminate soil and water bodies (Kopittke et al. 2019, Sher et al. 2024, Nuruzzaman et al. 2025). Furthermore, the use of fungicides in coffee cultivation can influence the production of mycotoxins by filamentous fungi, since the population numbers of fungal species known to produce these metabolites are high (Brzonkalik et al., 2011; Greeff-Laubscher et al., 2019). One of the main mycotoxin-producing species in coffee are species of the genus *Aspergillus*, section *Circumdati* (Gil-Serna et al. 2020). During the isolation of fungi from soil in this study, a large population number of section *Circumdati* strains was observed in conventional coffee soil samples, while in regenerative coffee soil samples, in some plates of culture media, the population of this group was very low and sometimes absent (Fig. S5).

Some genera that are highly abundant in coffee growing areas under conventional and regenerative management are already known for their high chitinase production, including species from the genera *Trichoderma, Fusarium, Acremonium, Cladosporium* and *Aspergillus* (Sherief et al. 1991, Nuero 1995, Karlsson et al. 2008, Brzezinska & Jankiewicz 2012, Chung et al. 2019, Mohiddin et al. 2021). Chitinases are essential enzymes for fungi with several functions. Among the fungal isolates tested for chitinase production, the areas of conventional coffee cultivation stand out, since they have the largest number of fungal strains producing higher amounts of this enzyme under the tested conditions. Although fungal diversity is

similar between conventional and regenerative coffee cultivation areas, most of the isolates from regenerative systems showed lower chitinase production. This result may be correlated with the use of pesticides in conventional crops. Some studies suggest that these agrochemicals cause stress in the soil microbial community, inducing competition (Baudy et al. 2021, Walder et al. 2022, Cruz et al. 2024). Chitinase is an enzyme used by species of mycoparasitic fungi that compete for space and natural resources (Wang et al., 2021).

On the other hand, the highest chitinase-producing strain comes from regenerative coffee soil, followed by two strains isolated from native Cerrado areas. Although the largest number of isolates were obtained from areas of native Cerrado vegetation, a small group of fungal isolates produced higher amounts of chitinase under the tested conditions. In natural forest soils, fungal enzymes, including cellulases, hemicellulases, pectinases, peroxidases, acid phosphatases, laccase, and chitinase, are involved in the degradation and decomposition of organic matter (Perez-Moreno & Read, 2000; Pritsch & Garbaye, 2011). Forests are complex environments, and therefore, specific fungal groups are specialized in the production of certain enzymes. For example, white rot fungi are considered the most efficient at degrading lignin, while ascomycota species focus on degrading cellulose and chitin (Cerro et al. 2021, Ferreira & Soares 2023, Leifheit et al. 2024). In forest soils, chitinolytic fungi act by breaking down chitin, present in fungal cell walls and arthropod exoskeletons, into organic nitrogen sources (Pritsch & Garbaye, 2011).

These results demonstrate that land-use changes in the Brazilian Cerrado significantly alter the diversity and composition of soil fungal communities. Furthermore, although the highest chitinase-producing strain originated from regenerative coffee cultivation soil, fungi from native Cerrado also showed good chitinase producers, with two strains standing out. These findings highlight the importance of preserving a biome as threatened as the Cerrado, and how this rich ecosystem harbors an unexplored enzymatic potential.

ACKNOWLEDGMENTS

This work was carried out with the support of the Coordination for the Improvement of Higher Education Personnel – Brazil (CAPES) – Funding Code 001. In addition, this work received financial support from the INCT Soil Biodiversity (CNPq #406658/2022-6 and FAPEMIG #APQ-04011-24) and the FAPEMIG Networks (#RED-00181-23 and RED-00330-16).

.

REFERENCES

Agrawal, T.; Kotasthane, A., S. Chitinolytic assay of indigenous *Trichoderma* isolates collected from different geographical locations of Chhattisgarh in Central India. Springerplus, v. 1, n. 73, 2012.

Amend AS, Seifert KA, Bruns TD. 2010. Quantifying microbial communities with 454 pyrosequencing: does read abundance count? Mol Ecol 19:5555–5565. doi: 10.1111/j.1365-294X.2010.04898.x.

Anastasi A, Vizzini A, Prigione V, Varese GC. Wood degrading fungi: morphology, metabolism and environmental applications. In: Chauhan AK, Varma A (eds) A textbook of molecular biotechnology. I.K. International, New Delhi, pp 957–993, 2009. ISBN: 9789380026374

Bail J, Gomez JAM, de Oliveira Vaz GC, de Castro WAC, Bonugli-Santos RC. Structural and functional changes in the fungal community of plant detritus in an invaded Atlantic Forest. BMC Microbiol. 2022 Jan 5;22(1):10. doi: 10.1186/s12866-021-02431-8.

Baldrian, P.; Bell-Dereske, L.; Lepinay, C.; Větrovský, T.; Kohout, P. Fungal communities in soils under global change. Studies in Mycology, v. 103, p. 1–24, 2022.

Baudy, P., Zubrod, J.P, Konschak, M., Röder, N., Nguyen, TH., Schreiner, VC., Baschien, C., Schulz, R., Bundschuh, M. Environmentally relevant fungicide levels modify fungal community composition and interactions but not functioning. Environmental Pollution, v. 285, 2021. DOI: https://doi.org/10.1016/j.envpol.2021.117234.

Berrin, J-G.; Navarro, D.; Couturier, M.; Olivé, C.; Grisel, S.; Haon, M.; Taussac, S.; Lechat, C.; Courtecuisse, R.; Favel, A.; Coutinho, P., M.; Lesage-Meessen, L. Exploring the Natural Fungal Biodiversity of Tropical and Temperate Forests toward Improvement of Biomass Conversion. Applied and Environmental Microbiology, v. 78, n. 18, 2012.

Bever, J., D.; Mangan, S., A.; Alexander, H., M. Maintenance of Plant Species Diversity by Pathogens. Annual review of ecology, evolution, and systematics, v.46, 2015.

Bray, J. R.; Curtis, J. T., 1957. An Ordination of the Upland Forest Communities of Southern Wisconsin. Ecological Monographs. 27(4), pp. 325–349. https://doi.org/10.2307/1942268

Brinkmann, N.; Schneider, D.; Sahner, J.; Ballauff, J.; Edy, N.; Barus, H.; Irawan, B.; Wilarso, B.; Qaim, M.; Daniel, R.; Polle, A. Intensive tropical land use massively shifts soil fungal communitie. Scientific Reports, v. 9, n. 3403, 2018.

Brzezinska MS, Jankiewicz U. Production of antifungal chitinase by Aspergillus niger LOCK 62 and its potential role in the biological control. Curr Microbiol. 2012 Dec;65(6):666-72. doi: 10.1007/s00284-012-0208-2

Brzonkalik K, Herrling T, Syldatk C, Neumann A. 2011. The influence of different nitrogen and carbon sources on mycotoxin production in Alternaria alternata. Int J Food Microbiol [Internet]. 147:120–126

Bueno C., G.; Moora, M; Gerz, M.; Davison, J.; Öpik, M.; Pärtel, M.; Helman, A.; Ronk, A.; Kuhn, I.; Zobel, M. Plant mycorrhizal status, but not type, shifts with latitude and elevation in Europe. Global Ecology Biogeography, v. 26, p. 690–762, 2017.

Casieri, L., Anastasi, A., Prigione, V. et al. Survey of ectomycorrhizal, litter-degrading, and wood-degrading Basidiomycetes for dye decolorization and ligninolytic enzyme activity. Antonie van Leeuwenhoek 98, 483–504 (2010). DOI: https://doi.org/10.1007/s10482-010-9466-9

Cedeño-Sanchez, M., Cheng T., Lambert C., et al. 2024. "Unraveling Intragenomic Polymorphisms in the High-Quality Genome of Hypoxylaceae: A Comprehensive Study of the rDNA Cistron." Mycological Progress 23, no. 1: 5. 10.1007/s11557-023-01940-2.

Cerro, C. Dong T., Salvachúa, D. Intracellular pathways for lignin catabolism in white-rot fungi. PNS, v. 118, n. 9, 2021. DOI: https://doi.org/10.1073/pnas.2017381118

Chao, A., 1984. Nonparametric Estimation of the Number of Classes in a Popu-lation. Scandinavian Journal of Statistics. 11(4), pp. 265–270, 1984.

Chen GM, Li WL, Tong SG, Qiu YT, Han JZ, Lv XC, Ai LZ, Sun JY, Sun BG, Ni L. Effects of the microbial community on the formation of volatile compounds and biogenic amines during the traditional brewing of Hongqu rice wine. Current research Food Science, v. 6, n. 5, p. 1433-1444, 2022. doi: 10.1016/j.crfs.2022.08.020.

Chung, D., Baek, K., Bae, S.S. et al. Identification and characterization of a marine-derived chitinolytic fungus, Acremonium sp. YS2-2. J Microbiol. 57, 372–380 (2019). https://doi.org/10.1007/s12275-019-8469-0

Colman, C., B.; Guerra, A.; Almagro, A.; Roque, F., O.; Rosa, I., M., D.; Fernandes, G., W.; Oliveira, P., T., S. Modeling the Brazilian Cerrado land use change highlights the need to account for private property sizes for biodiversity conservation. Scientific Reports, n. 14, 2024.

Crous PW, Jurjević Ž, Balashov S, De la Peña-Lastra S, Mateos A, Pinruan U, Rigueiro-Rodríguez A, Osieck ER, Altés A, Czachura P, Esteve-Raventós F, Gunaseelan S, Kaliyaperumal M, Larsson E, Luangsa-Ard JJ, Moreno G, Pancorbo F, Piątek M, Sommai S, Somrithipol S, Asif M, Delgado G, Flakus A, Illescas T, Kezo K, Khamsuntorn P, Kubátová A, Labuda R, Lavoise C, Lebel T, Lueangjaroenkit P, Maciá-Vicente JG, Paz A, Saba M, Shivas RG, Tan YP, Wingfield MJ, Aas T, Abramczyk B, Ainsworth AM, Akulov A, Alvarado P, Armada F, Assyov B, Avchar R, Avesani M, Bezerra JL, Bhat JD, Bilański P, Bily DS, Boccardo F, Bozok F, Campos JC, Chaimongkol S, Chellappan N, Costa MM, Dalecká M, Darmostuk V et al. Fungal Planet description sheets: 1614-1696. Fungal Syst Evol. 2024 Jun;13:183-440. doi: 10.3114/fuse.2024.13.11

Cruz, J.A., Camenzind, T., Xu, B. *et al.* Limited role of fungal diversity in maintaining soil processes in grassland soil under concurrent fungicide stress. *Environ Sci Eur* 36, 156 (2024). https://doi.org/10.1186/s12302-024-00983-w

Deacon, L. J., Pryce-Miller, E. J., Frankland, J. C., Bainbridge, B. W., Moore, P. D., and Robinson, C. H. (2006). Diversity and function of decomposer fungi from a grassland soil. Soil Biol. Biochem. 38, 7–20. doi: 10.1016/j.soilbio.2005.04.013

de Castro, A.P., Quirino, B.F., Pappas, G. et al. Diversity of soil fungal communities of Cerrado and its closely surrounding agriculture fields. Archives of Microbiology, v. 190, p. 129–139 (2008). https://doi.org/10.1007/s00203-008-0374-6

Dhariwal, A., et al. MicrobiomeAnalyst: a web-based tool for comprehensive statistical, visual and meta-analysis of microbiome data. Nucleic Acids Research, Volume 45, Issue W1, 3 July 2017, Pages W180–W188. DOI: https://doi.org/10.1093/nar/gkx295

Edgar, R. C., 2013. UPARSE: highly accurate OTU sequences from 2893 mi-crobial amplicon reads. Nature Methods. 10, 996–998. https://doi.org/10.1038/nmeth.2604.

Egidi E, Delgado-Baquerizo M, Plett JM, Wang J, Eldridge DJ, Bardgett RD, Maestre FT, Singh BK. A few Ascomycota taxa dominate soil fungal communities worldwide. Nat Commun. 2019 May 30;10(1):2369. doi: 10.1038/s41467-019-10373-z.

Ferreira de Araujo, A.S., Bezerra, W.M., dos Santos, V.M. et al. Fungal diversity in soils across a gradient of preserved Brazilian Cerrado. Journal of Microbiology, v. 55, p. 273–279 (2017). https://doi.org/10.1007/s12275-017-6350-6

Ferreira, J.M., Soares, FEF. Entomopathogenic fungi hydrolytic enzymes: A new approach to biocontrol? Journal of Natural Pesticide Research, v. 3, 2023. DOI: https://doi.org/10.1016/j.napere.2023.100020.

Gomes, E., Costa, M., de Paula, R., Azevedo, R., Silva, F., Noronha, E., Ulhoa, C., Monteiro, V., Cardoza, R., Gutiérrez, S., Silva, R. The Cerato-Platanin protein Epl-1 from *Trichoderma harzianum* is involved in mycoparasitism, plant resistance induction and self cell wall protection. Scientific Reports, v. 5, n. 17998, 2016. https://doi.org/10.1038/srep17998

Gomes, R. C., Sêmedo, L.T.S, Soares, R.M.A, L.F. Linhares, C.J. Ulhoa, C.S. Alviano, R.R.R. Coelho, Purification of a thermostable endochitinase from Streptomyces RC1071 isolated from a cerrado soil and its antagonism against phytopathogenic fungi, Journal of Applied Microbiology, Volume 90, Issue 4, 1 April 2001, Pages 653–661, https://doi.org/10.1046/j.1365-2672.2001.01294.x

Figueiredo, M. A., da Silva, T. H., Pinto, O. H. B., Leite, M. G. P., de Oliveira, F. S., Messias, M. C. T. B., Rosa, L. H., Câmara, P. E. A. S., Lopes, F. A. C., & Kozovits, A. R. (2023). Metabarcoding of soil fungal communities in rupestrian grassland areas preserved and degraded by mining: implications for restoration. *Microbial Ecology*, 85, 1045–1055. https://doi.org/10.1007/s00248-023-02177-y

Gil-Serna J, Vázquez C, Patiño B. The Genomic Regions That Contain Ochratoxin A Biosynthetic Genes Widely Differ in *Aspergillus* Section *Circumdati* Species. Toxins (Basel), v. 12, n. 12, 2020. doi: 10.3390/toxins12120754

Grande, T., O.; Aguiar, L., M., S.; Machado, R., B. Heating a biodiversity hotspot: connectivity is more important than remaining habitat. Landscape Ecology, 35, p. 639-657, 2020.

Greeff-Laubscher, M., R.; Beukes, I,; Marais, G., J.; Jacobs, K. Mycotoxin production by three different toxigenic fungi genera on formulated abalone feed and the effect of an aquatic environment on fumonisins. Mycology: An International Journal On Fungal Biology, v. 11, n. 2, p. 105-117, 2019.

Hartl L, Zach S, Seidl-Seiboth V. Fungal chitinases: diversity, mechanistic properties and biotechnological potential. Applied Microbiology and Biotechnology, v. 93, n. 2, p. 533-543, 2011. doi: 10.1007/s00253-011-3723-3

Hartmann M, Howes CG, VanInsberghe D, Yu H, Bachar D, Christen R, Henrik Nilsson R, Hallam SJ, Mohn WW. Significant and persistent impact of timber harvesting on soil microbial communities in Northern coniferous forests. ISME J, v. 6, n. 12, p. 2199-218, 2012. doi: 10.1038/ismej.2012.84.

IUCN. First 1,000 fungi on the IUCN Red List reveal growing threats. 2025. Disponível em: https://iucn.org/press-release/202503/first-1000-fungi-iucn-red-list-reveal-growing-threats-iucn-red-list. Accessed in: 8 out. 2025.

Kang, H.; Kim, S.; Song, K.; Kwon, M.-J.; Lee, J. Intermediate Disturbances Enhance Microbial Enzyme Activities in Soil Ecosystems. Microorganisms 2024, 12, 1401. https://doi.org/10.3390/microorganisms12071401

Khangura R, Ferris D, Wagg C, Bowyer J. Regenerative agriculture—a literature review on the practices and mechanisms used to improve soil health. Sustainability, v. 15, n. 2338, 2023.

Kemler, M.; et al., 2013. Ion Torrent PGM as Tool for Fungal Community Analysis: A Case Study of Endophytes in Eucalyptus grandis Reveals High Taxonomic Diversity. PLOS ONE. 8(12), e81718. https://doi.org/10.1371/journal.pone.0081718

Kõljalg U, Larsson K-H, Abarenkov K, Nilsson RH, Alexander IJ, Eberhardt U, Erland S, Høiland K, Kjøller R, Larsson E et al. UNITE: a database providing web-based methods for the molecular identification of ectomycorrhizal fungi. New Phytologist, v. 166, p. 1063–1068, 2005.

Kopittke, PM., Menzies, NW., Wang, P., McKenna, BA., Lombi, E. Soil and the intensification of agriculture for global food security. Environment International, v. 132, 2019. DOI: https://doi.org/10.1016/j.envint.2019.105078.

Lehmann, A.; Zheng, W., Rillig, M., C. Soil biota contributions to soil aggregation. Nature Ecology & Evolution, v. 1, p.1828–1835, 2017.

Leifheit, EF, Tessa Camenzind, Anika Lehmann, Diana R Andrade-Linares, Max Fussan, Sophia Westhusen, Till M Wineberger, Matthias C Rillig, Fungal traits help to understand the decomposition of simple and complex plant litter, FEMS Microbiology Ecology, Volume 100, Issue 5, May 2024, fiae033, https://doi.org/10.1093/femsec/fiae033

Li AH, Yuan FX, Groenewald M, Bensch K, Yurkov AM, Li K, Han PJ, Guo LD, Aime MC, Sampaio JP, Jindamorakot S, Turchetti B, Inacio J, Fungsin B, Wang QM, Bai FY. Diversity and phylogeny of basidiomycetous yeasts from plant leaves and soil: Proposal of two new orders, three new families, eight new genera and one hundred and seven new species. Stud Mycol. 2020 Jan 28;96:17-140. doi: 10.1016/j.simyco.2020.01.002

Lindahl B D, Taylor A F S. Occurrence of N-acetylhexosaminidase-encoding genes in ectomycorrhizal basidiomycetes. New Phytologist, v. 164, n. 1, p. 193-199, 2004.

Lofgren, L. A. , Uehling J. K., Branco S., Bruns T. D., Martin F., and Kennedy P. G.. 2019. “Genome-Based Estimates of Fungal rDNA Copy Number Variation Across Phylogenetic Scales and Ecological Lifestyles.” Molecular Ecology 28, no. 4: 721–730. 10.1111/mec.14995

Lorito M, Woo SL, Garcia I, Colucci G, Harman GE, Pintor-Toro JA, Filippone E, Muccifora S, Lawrence CB, Zoina A, Tuzun S, Scala F. Genes from mycoparasitic fungi as a source for improving plant resistance to fungal pathogens. PNAS, v. 95, n. 14, p. 7860-7865, 1998. doi: 10.1073/pnas.95.14.7860.

Manici, LM., Caputo, F., Fornasier, F., Paletto, A., Ceotto, E., De Meo, I. Ascomycota and Basidiomycota fungal phyla as indicators of land use efficiency for soil organic carbon accrual with woody plantations. Ecological Indicators, v. 160, 2024. DOI: https://doi.org/10.1016/j.ecolind.2024.111796.

MapBiomas Brasil | Notícias [WWW Document]. URL (2025) (accessed 27.6.25) https://mapbiomas.org/noticias

Martin, M., F. Heijden, m., G., A., V., D. The mycorrhizal symbiosis: research frontiers ingenomics, ecology, and agricultural application. New Phytologist, v. 242. 2024.

McLaughlin, A.; Mineau, P. The impact of agricultural practices on biodiversity. Agriculture, Ecosystems & Environment, v. 55, n. 3, p. 201-212, 1995.

Mikryukov, V.; Dulya, O.; Zizka, A.; Bahram, M.; Hagh-Doust, N.; Anslan, S.; Prylutskyi, O.; Delgado-Baquerizo, M.; Maestre, F., T.; Tedersoo, l. Connecting the multiple dimensions of global soil fungal diversity. Science Advances, v. 9, n. 48, 2023. DOI: 10.1126/sciadv.adj8016

Mohiddin FA, Padder SA, Bhat AH, Ahanger MA, Shikari AB, Wani SH, Bhat FA, Nabi SU, Hamid A, Bhat NA, Sofi NR, Waza SA, Hamid B, Parveen S, Hussain A, Bhat AN, Ali OM, Dar MS, Abdel Latef AAH. Phylogeny and Optimization of Trichoderma harzianum for Chitinase Production: Evaluation of Their Antifungal Behaviour against the Prominent Soil Borne Phyto-Pathogens of Temperate India. Microorganisms. 2021 Sep 15;9(9):1962. doi: 10.3390/microorganisms9091962.

Mollier, S.; Kunstler, G.; Dupouey, J., L.; Mulero, S.; Bergès, L. Forest management and former land use have no effect on soil fungal diversity in uneven-aged mountain high forests. Annals of Forest Science v. 81, n. 2, 2024. https://doi.org/10.1186/s13595-023-01218-3

Montgomery, R., Biklé, A., Archuleta, R., Brown, P., Jordan, J. Soil health and nutrient density: preliminary comparison of regenerative and conventional farming. Peer J, v. 10, n. 12848, 2022. DOI: 10.7717/peerj.12848

Moreira, GAM., Vale, HMM. Occurrence of Yeast Species in Soils under Native and Modified Vegetation in an Iron Mining Area. Revista Brasileira Ciencias do Solo, 2017. DOI: https://doi.org/10.1590/18069657rbcs20170375

Nampally, m., Rajulu, m., Gillet, d., Suryanarayanan, t., Moerschbache, b. A High Diversity in Chitinolytic and Chitosanolytic Species and Enzymes and Their Oligomeric Products Exist in Soil with a History of Chitin and Chitosan Exposure. Biomed Research International, 2015.

Ngaba, M., Mgelwa, A., Gurmesa, G., A.; Uwiragiye, Y., Zhu, F., Qiu, Q., Fang, Y., Hu, B., Rennenberg, H. Meta-analysis unveils differential effects of agroforestry on soil properties in different zonobiomes. Plant Soil, v. 496, p. 589–607, 2024. DOI: https://doi.org/10.1007/s11104-023-06385-w

Noriler, S., A.; Savi, D., C.; Aluizio, R.; Palácio-Cortes, A., M.; Possiede, Y., M.; Glienke, C. Bioprospecting and Structure of Fungal Endophyte Communities Found in the Brazilian Biomes, Pantanal, and Cerrado. Frontiers in Microbiology, v. 9, n. 1526, 2018. doi: 10.3389/fmicb.2018.01526

Nuero, O.M. Production of chitinase by *Fusarium* species. Current Microbiology 30, 287–289 (1995). https://doi.org/10.1007/BF00295503

Nuruzzaman, MD., Bahar, MDM., Naidu, R. Diffuse soil pollution from agriculture: Impacts and remediation. Science of The Total Environment, v. 962, 2025. DOI: https://doi.org/10.1016/j.scitotenv.2025.178398.

Perez-Moreno J, Read DJ. Mobilization and transfer of nutrients from litter to tree seedlings via the vegetative mycelium of ectomycorrhizal plants. New Phytol, v. 145, p. 301–309, 2000.

Pritsch, K.; Garbaye, J. Enzyme secretion by ECM fungi and exploitation of mineral nutrients from soil organic matter. Annals of Forest Science, v. 68, p. 25–32, 2011.

Produção agrícola e pecuária municipal: censo agropecuário, produção e extração vegetal da silvicultura. Rio de Janeiro Disponível em: https://sidra.ibge.gov.br/home/pms/brasil (2018).

PYLRO, V.S.; et al. Brazilian Microbiome Project: revealing the unexplored microbial diversity challenges and prospects. Microbial Ecology, v. 67, n. 2, p. 237-241, 2014.

Roberts, W., K.; Selitrennikoff, C., P. Plant and Bacterial Chitinases Differ in Antifungal Activity. Microbiology, v. 134, n. 1, 1988. DOI: https://doi.org/10.1099/00221287-134-1-169

Satish N., Sultana S., and Nanjundiah V., Diversity of soil fungi in a tropical deciduous forest in Mudumalai, southern India, Current Science, v. 93, n. 5, p. 669–677, 2007. DOI: 2-s2.0-34748887898

Schoch CL, Seifert KA, Huhndorf S, Robert V, Spouge JL, Levesque CA, Chen W; Fungal Barcoding Consortium; Fungal Barcoding Consortium Author List. Nuclear ribosomal internal transcribed spacer (ITS) region as a universal DNA barcode marker for Fungi. Proc Natl Acad Sci U S A., v. 109, n. 16, p. 6241-6246, 2012. doi: 10.1073/pnas.1117018109

Sher, A., Li, H., ullah, A., Hamid, y., Nasir, b., Zhang, j. Importance of regenerative agriculture: climate, soil health, biodiversity and its socioecological impact. Discover Sustainability, v. 5, n. 462, 2024. DOI: https://doi.org/10.1007/s43621-024-00662-z

Sherief, A.A., El-Sawah, M.M.A. & Abd El-Naby, M.A. Some properties of chitinase produced by a potent Aspergillus carneus strain. Appl Microbiol Biotechnol 35, 228–230 (1991). https://doi.org/10.1007/BF00184691

Silva, J., M., C.; Bates, J., M. Biogeographic patterns and conservation in the South American Cerrado: A Tropical Savanna Hotspot: The Cerrado, which includes both forest and savanna habitats, is the second largest South American biome, and among the most threatened on the continent. Bioscience, n. 52, p. 225–234, 2002. doi: 10.1641/0006-3568(2002).
Steidinger BS, Crowther TW, Liang J, Nuland MEV, Werner GDA, Reich PB, et al. Climatic controls of decomposition drive the global biogeography of forest-tree symbioses. Nature, v. 569, p. 404–408, 2019.

Sternhagen, E., Black, K., Hartmann, E., Shivega, W., Johnson, P., McGlynn, R., Schmaltz, L., Keller, R., Vink, S., Aldrich-Wolfe, L. Contrasting Patterns of Functional Diversity in Coffee Root Fungal Communities Associated with Organic and Conventionally Managed Fields. Applied and Environmental Microbiology, v. 86, n. 11, 2020.

Strassburg, B., B., N.; T. Brooks, R. Feltran-barbieri, A. Iribarrem, R. Crouzeilles, R. Loyola, A.E. Latawiec, F.J.B.O. Filho, C.A.M. Scaramuzza, F.R. Scarano, F. Soares-Filho, A. Balmford. Moment of truth for the Cerrado hotspot. Native Ecology Evolution, n. 0099, 2017. 10.1038/s41559-017-0099

Takashima, M., Kurakado, S., Cho, O., Kikuchi, K., Sugiyama, J., & Sugita, T. (). Description of four Apiotrichum and two Cutaneotrichosporon species isolated from guano samples from bat-inhabited caves in Japan. International Journal of Systematic and Evolutionary Microbiology, v. 70, n. 8, 2020. DOI: https://doi.org/10.1099/ijsem.0.004277

Tedersoo, L., Bahram M., Põlme S., et al. 2015. “Response to Comment on “Global Diversity and Geography of Soil Fungi”.” Science 349, no. 6251: 936. 10.1126/science.aaa5594.
Tedersoo, L. Tooming-Klunderud A., and Anslan S. 2018. “PacBio Metabarcoding of Fungi and Other Eukaryotes: Errors, Biases and Perspectives.” New Phytologist 217, no. 3: 1370–1385. 10.1111/nph.14776.

Tedersoo, L.; Bahram, M.; Põlme, S.; Kõljalg, U.; Yorou, N., S.; Wijesundera, R.; Ruiz, L., V.; Vasco-Palacios, A., M.; Thu, P., Q.; Suija, A.; Smith, M., E.; Sharp, C.; Saluveer, E.;

Saitta, A.; Rosas, M.; Riit, T.; Ratkowsky, D.; Pritsch, K.; Põldmaa, K.; Piepenbring, M.; Phosri, C.; Peterson, M et al. Global diversity and geography of soil fungi. Science, v. 346, n. 6213, 2014. DOI: 10.1126/science.1256688

Treseder, K., K.; Lennon, J., T. Fungal traits that drive ecosystem dynamics on land. Microbiology and Molecular Biology Reviews, v. 79, n. 2, p. 243-262, 2015.

Walder, F. Schmid, MW., Riedo, J., Valzano-Held, AY., Banerjee, S., Büchi, L., Bucheli, TD., van der Heijden, MGA.Soil microbiome signatures are associated with pesticide residues in arable landscapes. Soil Biology and Biochemistry, v. 174, 2022. DOI: https://doi.org/10.1016/j.soilbio.2022.108830

Wu L, Chen J, Wu H, Wang J, Wu Y, Lin S, Khan MU, Zhang Z, Lin W. Effects of consecutive monoculture of Pseudostellaria heterophylla on soil fungal community as determined by pyrosequencing. Scientific Report, v. 6, n. 26601, 2016. doi: 10.1038/srep26601.

Wei R-T, Chen N, Ding Y-T, et al. (2022a). Correlations between microbiota with physicochemical properties and volatile compounds during the spontaneous fermentation of Cabernet Sauvignon (*Vitis vinifera* L.) wine. LWT - Food Science and Technology 163: 113529.

YU, X.; DIJKSTRA, F., A. Carbon and nitrogen dynamics affected by litter and nitrogen addition in a grassland soil: Role of fungi. European Journal of Soil Biology, v. 100, 2020.

Zhang, W., Ma, J., Yan, Q., Jiang, Z. Yang, S. Biochemical characterization of a novel acidic chitinase with antifungal activity from *Paenibacillus xylanexedens* Z2–4. International Journal of Biological Macromolecules, v. 182, p. 1528-1536, 2021.

Zhang X., Johnston ER., Barberán A., Ren Y., Wang Z., Han X. Effect of intermediate disturbance on soil microbial functional diversity depends on the amount of effective resources. Environmental Microbiology, v. 20, n. 10, p.3862-3875, 2018. doi: 10.1111/1462-2920.14407

Supplemental Information for:

Chitinase production and effects of land use change on fungal communities in the soil of the Brazilian Cerrado

Cristiane Nacimento Figueiredo[1], Mariana Prósperi de Oliveira Paula[1], Clara Resende de Souza Castro[1], Fabiana Passamani[1], Teotonio de Carvalho[1], Harisson Guimaraes[1], Daniele Cabral Michel[1], Fatima Moreira[1], Victor Satler Pylro[1,*]

Geographic coordinates

Area/Identification of the Transect	Variety	Planting Date	Management Practice	Latitude	Longitude
AREA 1					
NC 1	IBC 12/IAC 125	2015	Organic with irrigation	18°46’13.56”	46°56’02.76”
C1	IBC 12/IAC 125	2017	Conventional with irrigation	18°41’36.03”	46°58’14.67”
NC2	Catuaí 62	2010	Organic without irrigation	18°46’56.89”	46°56’49.78”
C1	Catuaí 62	2014	Conventional without irrigation	18°47’24.95”	46°57’01.12”
Ce1	Cerrado nativo			18°46’35.21”	46°56’53.92”
Ce2	Cerrado nativo			18°47’20.55”	46°57’07.30”
AREA 2					
NC3	Catuaí 144	2008	Regenerative with irrigation	18°52’45.33”	47° 05’51.91”
C3	Catuaí 144	2007	Conventional with irrigation	18°51’41.91”	47°07’05.51”
NC4	Topázio	2008	Regenerative with irrigation	18°52’47.00”	47°05’45.53”
C4	Topázio	2007	Conventional with irrigation	18°51’48.11”	47°07’08.96”
Ce3	Cerrado nativo			18°53’08.77”	47°06’14.29”
Ce4	Cerrado nativo			18°51’28.32”	47°07’18.95”

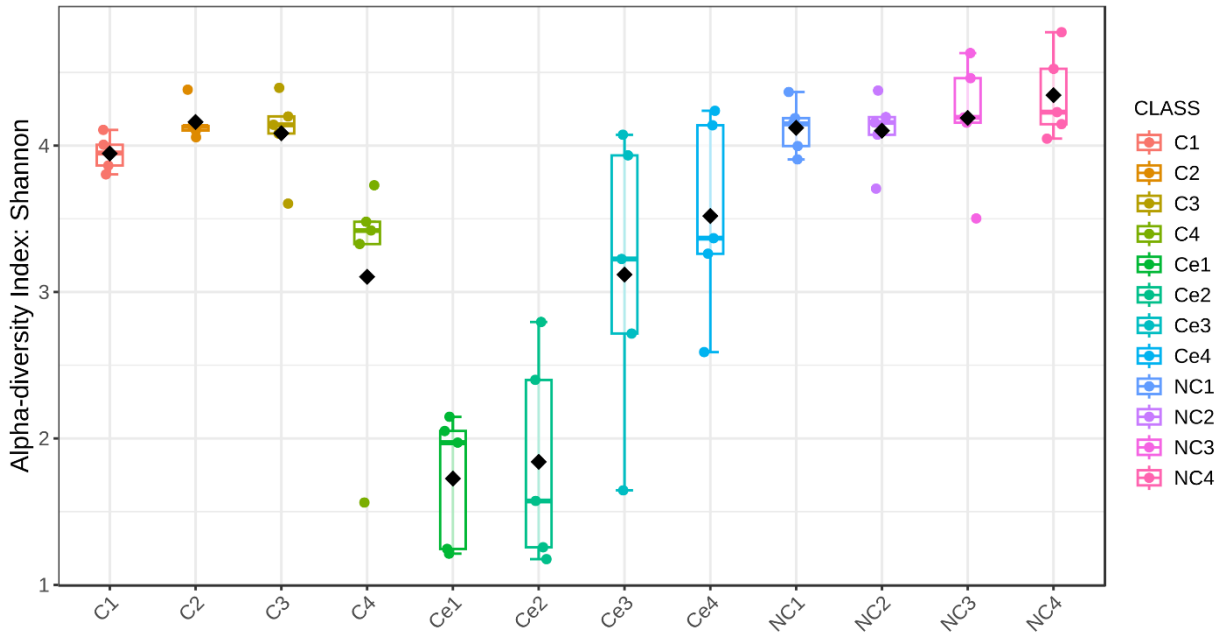


Figure S1. Comparative analysis of Alpha Diversity (Shannon index) based on ITS gene sequencing in different soil treatments. Boxplots show Shannon diversity for each land use type (n = 4 replicates for each land use type). Y-axis: Alpha Diversity Index: Shannon (higher values indicate greater diversity). X-axis: sample land use types (C1–C4: conventional coffee; Ce1–Ce4: native Cerrado; NC1–NC4: regenerative coffee). Colors identify the sample land use types, as shown in the legend. P-value: 8.6883e-12.

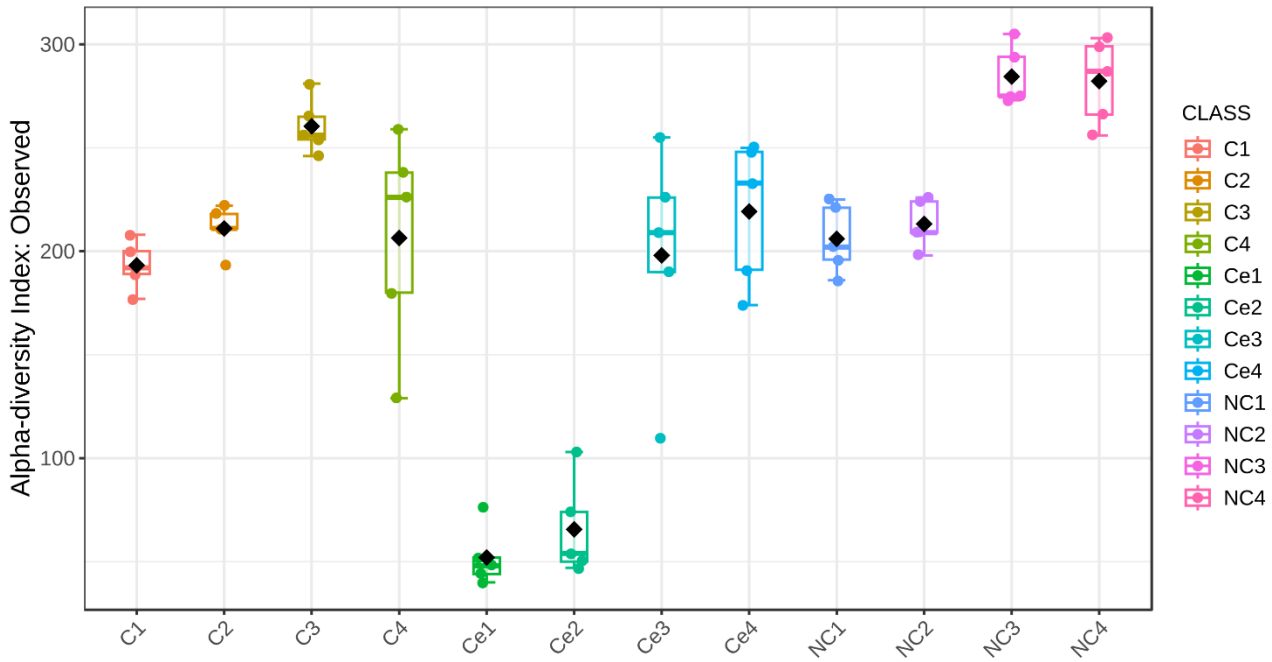


Figure S2. Comparative analysis of Alpha Diversity (Observed index) based on ITS gene sequencing in different soil treatments. Boxplots show Observed diversity for each land use type (n = 4 replicates for each land use type). Y-axis: Alpha Diversity Index: Observed (higher values indicate greater diversity). X-axis: sample land use types (C1–C4: conventional coffee; Ce1–Ce4: native Cerrado; NC1–NC4: regenerative coffee). Colors identify the sample land use types, as shown in the legend. P-value: 8.6883e-12.

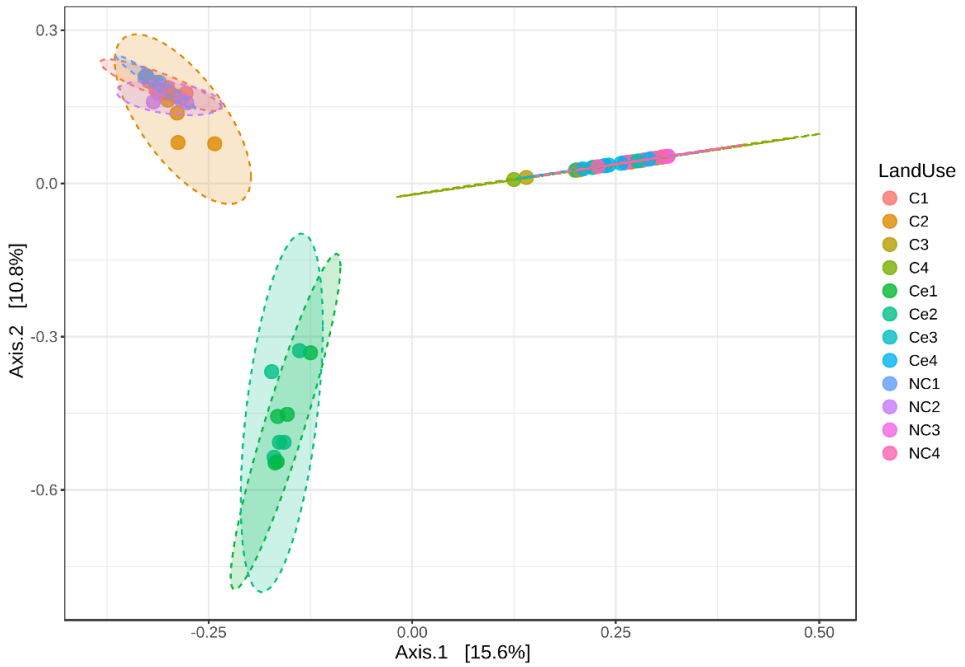


Figure S3. Beta-diversity analysis of fungal community in soil samples using the Jaccard index and PCoA based on ITS gene sequencing. Closer clusters indicate greater similarity in fungal community structure between samples. C1–C4: conventional coffee; Ce1–Ce4: native Cerrado; NC1–NC4: regenerative coffee. Colors identify the sample land use types, as shown in the legend, and multivariate analysis of variance (PERMANOVA) indicated significant differences between land use types (p = 0.001).

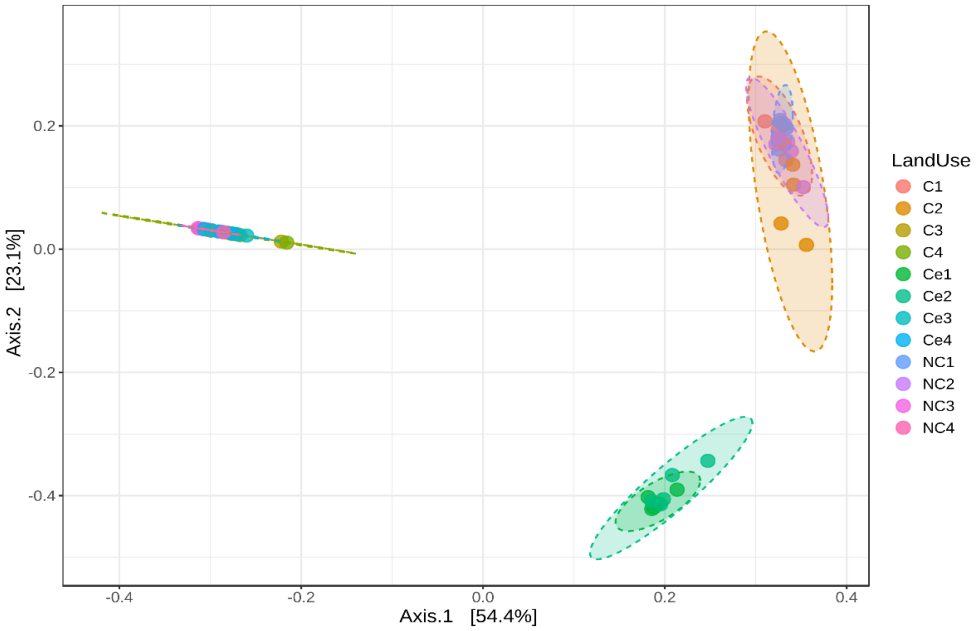


Figure S4. Beta-diversity analysis of fungal community in soil samples using the Jensen-Shannon index and PCoA based on ITS gene sequencing. Closer clusters indicate greater similarity in fungal community structure between samples. C1–C4: conventional coffee; Ce1–Ce4: native Cerrado; NC1–NC4: regenerative coffee. Colors identify the sample land use types, as shown in the legend, and multivariate analysis of variance (PERMANOVA) indicated significant differences between land use types (p = 0.001).

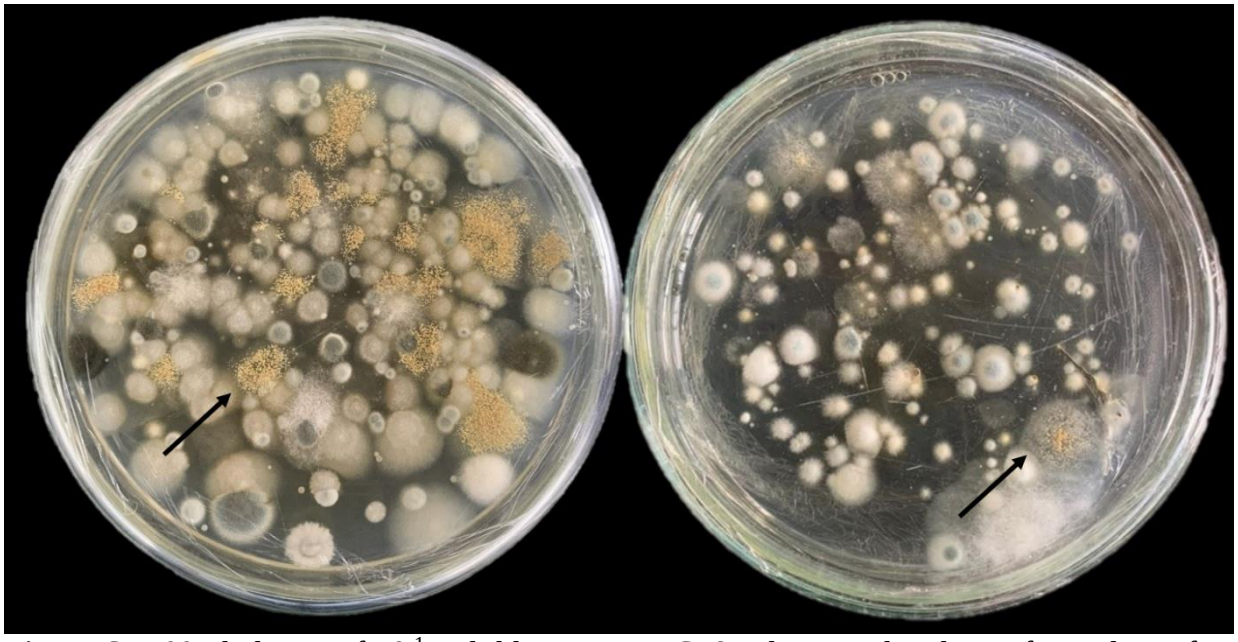


Figure S5.100 µl aliquot of 10^{-1} soil dilutions on DG18 culture media plates after 7 days of incubation at 25 °C. Left, soil dilution from a conventional coffee cultivation area (15 *Aspergillus* strains, section *Circumdati*); right, diluted soil from regenerative coffee (3 *Aspergillus* isolates, section *Circumdati*). Black arrows indicate isolates of the genus *Aspergillu*s from section *Circumdati*.

Artigo 2 - Redigido conforme norma do periódico científico - Versão preliminar	
Título do artigo:	Description of novel species and occurrence records of the genus *Penicillium* in the Brazilian Cerrado
Autores:	Cristiane Nacimento Figueiredo Fabiana Reinis Franca Passamani Harisson de Souza Guimaraes Ya Bin Zhou Bart Krak Teotonio Soares de Carvalho Jorge Teodoro de Souza Luís Roberto Batista Pedro Crous Jos Houbraken Victor Satler Pylro
Periódico:	Persoonia
ISSN	
DOI	

Artigo 2

Description of novel species and occurrence records of the genus *Penicillium* in the Brazilian Cerrado

C.N. Figueiredo[1], F.R. Passamani[1], H.S. Guimarães[1], Y.B. Zhou[2], B. Krak[2], T.S. Carvalho[1], J.T. de Souza[1], L.R. Batista[1], P.W. Crous[2], J. Houbraken[2], V.S. Pylro[1,*]

[1]Federal University of Lavras, Lavras, MG, BR. Emails: cristiane.figueiredo1@estudante.ufla.br, harisson.souza1@estudante.ufla.br; teotonio.carvalho@ufla.br; jorge.souza@ufla.br; luisrb@ufla.br; victor.pylro@ufla.br;

[2]Westerdijk Fungal Biodiversity Institute, Utrecht, The Netherlands. Emails: y.zhou@wi.knaw.nl; b.kraak@wi.knaw.nl; p.crous@wi.knaw.nl; j.houbraken@wi.knaw.nl;

[*]Corresponding authors: V.S. Pylro, victor.pylro@ufla.br; J. Houbraken, j.houbraken@wi.knaw.nl;

Abstract: *Penicillium* species are cosmopolitan and are isolated from diverse substrates, including soil, water, air, plant debris, and food products. The diversity of the Cerrado fungal community remains unexplored, including the *Penicillium* genus. To explore fungal diversity in Cerrado soils, 1,481 fungal strains were isolated using serial dilution techniques in DRBC and DG18. Out of this total, 166 isolates belong to the genera *Aspergillus*, *Penicillium* and *Talaromyces* were identified. The new species were identified through polyphasic taxonomy, combined morphological characteristics, and multigene phylogenies based on ITS, BenA, CaM, and RPB2 data. Seven new species are described, *Penicillium patrocinensis* in section *Brevicompacta*, *Penicillium guarae* in *Cinnamopurpurea*, *Penicillium seriamae* in *Ghizhourum*, *Penicillium beraoi* and *Penicillium moreirae* in *Lanata-Divaricata*, *Penicillium pequii* and *Penicillium pluviallis* in sect. *Ramosum*. The remaining isolates were identified as occurrence records of 29 species. Of this number, 4 species were identified using partial sequences of the genes *BenA*, *CaM*, and *RPB2*, and the remaining 25 species were identified only by the partial sequence of the *BenA* gene. The identification of several species in the Cerrado biome, including seven new species, demonstrates the potential for a large diversity of filamentous fungi that remains unexplored in this biome.

Keywords: Brazilian savanna, Biodiversity, Filamentous fungi, Taxonomy.

Citation: Figueiredo CN, Passamani FR, Guimarães HS, Zhou YB, Krak B, Carvalho TS, Souza JT, Batista LR, Crous PW, Houbraken J, Pylro VS (2025). Description of new species and ocurrence record genus *Penicillium* in the Brazilian Cerrado. *Persoonia* **XX**: xx–xx. doi: 10.3114/persoonia.2025.XX.xx

1. INTRODUCTION

The fungi are successful in soil in consequence of their high plasticity and ability to adapt to adversity, their population densities vary colony-forming units per gram of soil (CFU g^{-1} soil) (Bhattarai et al. 2015). These microorganisms play as the main decomposers of organic matter, therefore contribute directly to soil fertility and nutrient cycling, and they are essential for biosphere balance (Alves et al. 2021). Although tropical forests are the most diverse biomes on the planet, the biodiversity of filamentous fungi remains underresearched (Berrin et al. 2012, Sullivan et al. 2017). The Cerrado is one of Brazilian tropical biome considerate as a hotspot of global biodiversity, since is home a rich property nature resource with thousands endemic species at risk of extinction (Colli et al. 2020, Rodrigues et al. 2022).

The Cerrado typically has two seasons well defined, dry winter and rainy summer. This biome was distributed over 24% Brazilian national territory. However, agriculture and livestock farming have suppressed large areas of native vegetation cover, being converted into monoculture areas manly of crops such as coffee, soybeans, cotton and eucalyptus (Oliveira et al. 2019, Rodrigues et al. 2022). The expansion of the agricultural frontier combined with ongoing climate change, threatens the existence of Cerrado biome and all its biodiversity, which has not yet been fully explored, such as filamentous fungi (Strassburg et al. 2017). Little information is available about the diversity filamentous fungi in Cerrado soil, some studies shows that Ascomycota and Basidiomycota species are predominant in this biome, representing approximately 70% of the total fungal community (Castro et al. 2008, Jr & Sánchez-García 2019).

Penicillium is a widespread *Ascomycota* genus, and it was established by Link in 1809, belongs Aspergilaceae family, this genus is divided in two subgenera, 32 sections, 89 series and has 535 accept described species (Houbraken et al. 2020, Visagie et al. 2024). *Penicillium* has a worldwide distribution, isolated from several substrates, such indoor environments, food, soil, seawater, macroalgae and decomposition material (Visagie et al. 2014, Park et al. 2016, Gonçalves et al. 2019, Park et al. 2020).

Penicillium is well recognized for its economic importance. The production of antibiotics, such as penicillin produced by *P. rubens* was the first antibiotic used against diseases caused by gram-positive bacteria (Houbraken et al. 2011). In the food industry *P.camemberti* is used for white cheese (Camembert and Brie) and *P. roqueforti* for blue cheese (roquefort, gorgonzola, stilton), *P. nalgiovense* and *P. salamii* are used as starter

cultures for sausage fermentation (Magistà et al. 2016, Ropars et al. 2020, Cleere et al. 2024). Previous studies show that *Penicillium* stands out as one of the genera with the highest incidence in the Cerrado soil, however this diversity has not yet been fully elucidated. Thus, in this study, our aim was to identify and describe strains of genus *Penicillium*, isolated from Cerrado soil samples, collected in Patrocinio, Minas Gerais, Brazil.

2. MATERIAL AND METHODS

2.1 Sampling and isolation

A total of 166 fungal isolates were identified by polyphasic taxonomy out of 1,481 fungal strains, isolated from soil samples of native Cerrado, conventional coffee, and regenerative coffee. These samples were collected during the rainy season of 2023, between January and March, in the municipality of Patrocínio, state of Minas Gerais, Brazil (the geographic coordinates of the sampling sites are provided in Supplementary Material 1). The serial dilution plating method was used to isolate the fungal strains, 225 mL peptone water 1% was added 25 g of soil. Two successive dilutions were prepared, and 0.1 mL of each diluted suspension was spread in triplicate onto Dichloran 18 % glycerol agar (DG18) and Rose Bengal Agar + Chloramphenicol + Dichloran (DRBC). Plates were incubated at 25 °C for 7 days. All strains were deposited in the Microbiological Resources Unit-UMICRO, located at Federal University of Lavras- UFLA, Minas Gerais, Brazil. Type living cultures were deposited in the working culture collection of the Applied and Industrial Mycology department (DTO) of the Westerdijk Institute Fungal Biodiversity, Utrecht, The Netherlands.

2.2 Morphology observations

Morphological characterization of each species was made according to the incubation patterns standardized by Visagie et al. 2014. Culture media were dispensed in 90 mm Petri dishes with a volume of 20 mL. Strains were inoculated onto culture media from spore suspensions in a three-point pattern using a micropipette. Each species was characterized from observed morphological characters on Czapek yeast autolysate agar (CYA), malt extract agar (MEA), yeast extract sucrose agar (YES), dichloran 18 % glycerol agar (DG18), Oatmeal agar (OA), CYA supplemented with 5 % NaCl (CYAS) and creatine sucrose agar (CREA) grown

at 25 °C, with additional CYA plates incubated at 30 and 37 °C. After 7 days of incubation, strains were described according to size, texture, presence or no acid, soluble pigment and exudates, presence sulcate, color of mycelia, obverse and reverse colony colors. Colour names and codes refer to the Methuen Handbook of Colour (Kornerup & Wanscher 1967).

For microscopy, the slides were performed from cultures grown 7 days on MEA, with 60% lactic acid used as mounting fluid. The structures such as conidiophore, phialide, stipes, metuale (when present) and conidia were examined using light-microscope (Olympus BX50 and Zeiss Axioskop 2 Plus). Pictures of these structures were taken using Nikon NIS-elements D v. 4.0 and measured with ImagePro v. 6.0. Photoplates were made using Adobe® Photoshop® Creative 2024.

2.3 DNA extraction, sequencing and phylogenetic reconstruction

For DNA isolation, the strains were grown on MEA by 3 days at 25 °C. Then part of mycelia of each strain was harvested for total genomic DNA isolation using Kit DNeasy® UltraClean® Microbial Kit (Qiagen, Hilden, Germany). The pairs of primers ITS1/ITS4 (White et al. 1990) were in used to amplify the fragments internal transcribed spacer region (ITS), beta-tubulin (*BenA*) was amplified with BT2a/BT2b (Glass & Donaldson, 1995), calmodulin (*CaM*) with CMD5/CMD6 or CF1/CMD6 (Hong et al. 2006), and the second largest subunit of RNA polymerase II (RPB2) was amplified using the primers RPB2-F1/RPB2-7CR_1 (Houbraken et al. 2020). PCR reactions had a final volume of 13 µL, consisting of 1 µL of template DNA was mixed with 0.63 µL dimethylsulfoxide (DMSO, 5% w/w), 1.25 µL Taq polymerase buffer, 0.98 µL dNTPs (1 mM), 0.25 µL of each primer (10 µM), 0.38 µL MgCl2 (50 mM) and 0.05 µL of BioTaq DNA polymerase (5 U µL).The thermocycling conditions for amplifications had an initial denaturation step at 95 °C for 5 min, followed by 35 cycles of denaturation at 95 °C for 30 s, annealing at 55 °C (ITS, *BenA*, *CaM*) or 48 °C (RPB2) for 30 s, extension at 72 °C for 75 s, and final extension at 72 °C for 5 min, followed by cooling to 10 °C. PCR products were visualized in 1% agarose gel electrophoresis. Then, the strands (forward and reverse) were sequenced using the BrilliantDye Terminator v. 3.1 Cycle Sequencing Kit (NimaGen, Nijmegen, The Netherlands). The sequencing was performed using an Applied Biosystems™ 3730xl DNA Analyzer (ThermoFisher Scientifc, CA, USA). The SeqMan Pro v. 15 program was used to assemble the consensus sequences for each locus.

The phylogenetic relationships of the isolates within the sections were inferred from a

combined dataset of the *BenA, CaM* and *RPB2* gene sequences. Single gene datasets were also analyzed to assess concordance among the three phylograms. Partial sequences of the *BenA, CaM* and *RPB2* genes obtained from fungal isolates and from previously described species within each section were selected. Multiple sequence alignments for each gene were generated with MAFFT v. 7.427, and subsequently edited in MEGA X (Kumar et al., 2018), where the evolutionary model selection and phylogenetic inference by Maximum Likelihood (ML) were performed with 1000 bootstrap replicates. For multilocus analyses, the aligned sequences of the *BenA, CaM* and *RPB2* genes were concatenated using SequenceMatrix. Bayesian inference was conducted with MrBayes v. 3.2.6 using a Markov Chain Monte Carlo (MCMC) algorithm, and the optimal nucleotide substitution model for each locus was determined with MrModelTest v. 2.2. Four simultaneous Markov chains were run for 10 million generations, with trees sampled every 1000th generation, or until the average standard deviation of split frequencies dropped below 0.01. The first 25% of the sampled trees were discarded as burn-in, and the remaining trees were used to calculate posterior probabilities (PP). Resulting trees were visualized and edited with FigTree v. 1.4.2 and Adobe Illustrator CS5. *P. chrysogenum* CBS 306.48, *P. sclerotiorum* NRRL2074, *P. griseolum* NRRL2671, *P. glabrum* CBS 125543, *P. stolkieae* CBS 315.67 served as an outgroup.

3. RESULTS

3.1 Phylogeny

The phylogenetic results show that this study contains seven new species and four occurrence records belonging to the genus *Penicillium*. One of these new species belongs to section *Brevicompactum*, one to section *Cinnamopurpurea*, one to section *Ghizhourum*, two to section *Lanata-Divaricata* and two to section *Ramosum*. The isolates that are occurrence records belong to sections *Guizhourum* (3), *Exilicaulis* (1), *Fasciculata* (1) and *Sclerotiorum* (1).

Section *Brevicompacta* and *Ramosum*

The analyses of the concatenated dataset (*BenA, CaM*, and *RPB2*) comprised 15 species from Section *Brevicompactum* and 22 species from Section *Ramosum*, each with different gene lengths, totaling 1.967 aligned characters (*BenA*: 457 bp, *CaM*: 464 bp, *RPB2*:

1,046 bp) (Figure 1). Section *Brevicompacta* is formed by four distinct series, with the new species *P. patrocinensis* phylogenetically related to *P. tularense*, series *Tularensia* (Fig. 1). In the phylogenetic analyses of individual genes, the new species, together with *P. tularense*, consistently formed a well-supported clade, with mostly high support values (>99% bs, 1.00 pp), except for ITS. The section *Ramosum* is composed of five series. The two new species in this section, *P. pequii* and *P. pluviallis* are phylogenetically related to *P. scabrosum* and *P. xyleborini*, respectively, and are part of different series (Fig. 1). In individual gene phylogenetic analyses, the new species consistently formed a well-supported clade with *P. scabrosum* and *P. xyleborini*, with high support values (Fig. 1)

Section *Cinnamopurpurea*

The analyses of the concatenated dataset (*BenA*, *CaM* and *RPB2*) comprised 32 species of Section *Cinnamopurpurea*, each with different gene lengths, totaling 2.139 aligned characters (*BenA*: 478 bp, *CaM*: 543 bp, *RPB2*: 1.118 bp) (Figure 2). Section *Cinnamopurpurea* is formed by 4 distinct series, with the new species *P. guarae* phylogenetically related to the species *P. pusillum*, *P. jiangxiense* and *P. tealli* series *Jiangxiensia* (Fig. 2). In phylogenetic analyses of individual genes, the new species, together with phylogenetically related species, consistently formed a well-supported clade, with mostly high support values (100% bs, 1.00 pp), except for ITS.

Section *Guizhouorum*

The analyses of the concatenated dataset (*BenA*, *CaM*, and *RPB2*) included 14 species from section *Charlesia*, one species from section *Guizhouorum* and one species from section *Eremophila* (Figure 3). Each gene had different lengths, totaling 2,028 aligned characters (*BenA*: 462 bp, *CaM:* 517 bp, *RPB2*: 1.049 bp) (Fig. 3). In the section *Guizhouorum*, there was only one described species, *P. guizhouense*. In the phylogenetic analyses of individual genes, the new species, *P. seriamae*, is phylogenetically related to *P. guizhouense*, consistently forming a well-supported clade, with predominantly high support values (> 100% bp, 1.00 pp), except for ITS. The three strains presented identical *RPB2*, *BenA* and *Cam* sequences, with sequence similarity of 100%, 99,5% and 100% with *P. guizhouense*, respectivelly. All strains formed a monophyletic group with *P. guizhouense* (Fig. 3).

Section *Lanata-Divaricata*

The analyses of the combined dataset (*BenA, CaM*, and *RPB2*) included 127 species of section *Lanata-Divaricata* (Figure 4). Each gene presented different lengths, totaling 2,028 aligned characters (*BenA*: 463 bp, *CaM:* 542 bp, *RPB2:* 1,027 bp). Section *Lanata-Divaricata* is formed by 5 distinct series, with the new species *P. moreirae* and *P.beraoi* phylogenetically related to the species *P. reverso-vinaceum*, belonging to the series *Janthinella* (Fig. 4). In phylogenetic analyses of individual genes, the novel species, together with the phylogenetically related species *P. reverso-vinaceum*, consistently formed a well-supported clade, with mostly high support values (>97% bs, 1.00 pp), except for ITS.

Sections *Exilicaulis, Fasciculata* and *Sclerotiorum*

Phylogenetic trees based on the combined sequence data from the three loci, *RPB2, BenA* and *CaM*, indicate that fungal strains DTO 510-F2, DTO 515-A2 and DTO 514-E8 formed a monophyletic clade to the species *P. menonorum, P. dabashanicum* and *P. daejeonium*, respectively (Figure 5, 6 and 7). The strain DTO 510-F2 has 98% sequence similarity to *P. menonorum* at the *BenA*, 96% at the *CaM* locus and 98% at the *RPB2*.

The multigene phylogeny (Fig. 4) indicated that the isolate DTO 515-A2 forms a clade with the species *Penicillium dabashanicum*. The *BenA* region of these two isolates did not differ from each other, while the sequences of *CaM* and *RPB2* differed in six and two nucleotide positions, respectively. The combined dataset of both the *BenA, CaM* and *RPB2* loci, section *Sclerotiorum* (Fig. 7) shows the strain DTO 514-E8, formed a monophyletic group with *Penicillium daejeonium*. Based on a megablast search of NCBIs GenBank nucleotide database, partial sequences of the *CaM* and *BenA loci,* have 99% and 98% similarity to *P. daejeonium*, respectively.

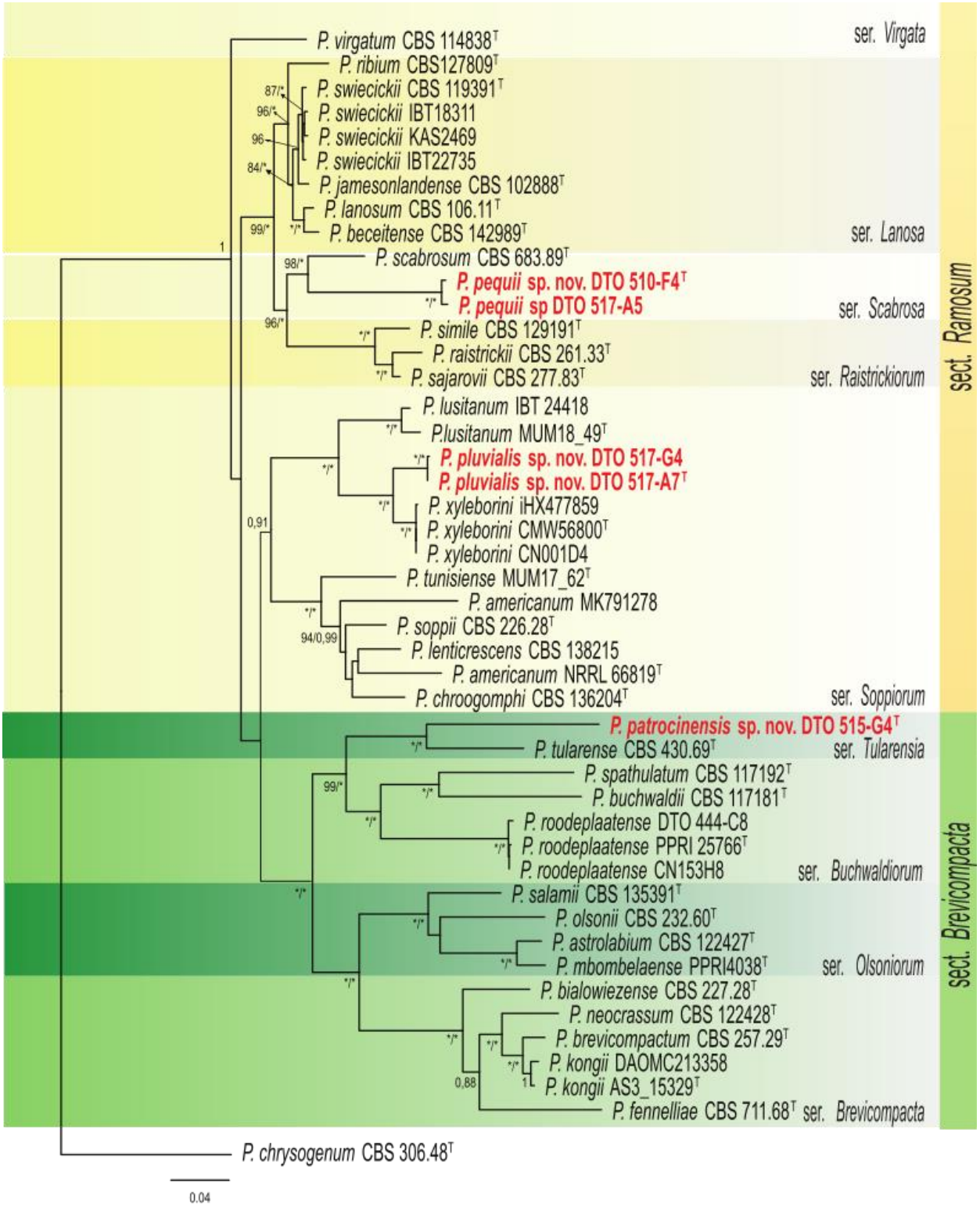


Figure 1. Maximum likelihood (IQ-TREE) phylogeny of the species belonging to the *Brevicompacta* and *Ramosum* sections inferred from combined, partial *CaM, rpb2* and *BenA* loci. The tree is rooted to *Penicillium chrysogenum* CBS 306.48. Numbers at the nodes are IQ-TREE ultra-fast bootstrap (BS) (UFBoot) values ≥ 70 %, followed by RAxML BS ≥ 70 %, and MrBayes posterior probability (PP) ≥ 95. Coloured names indicate strains that belong to the new species. The scale bar represents the number of substitutions per site.

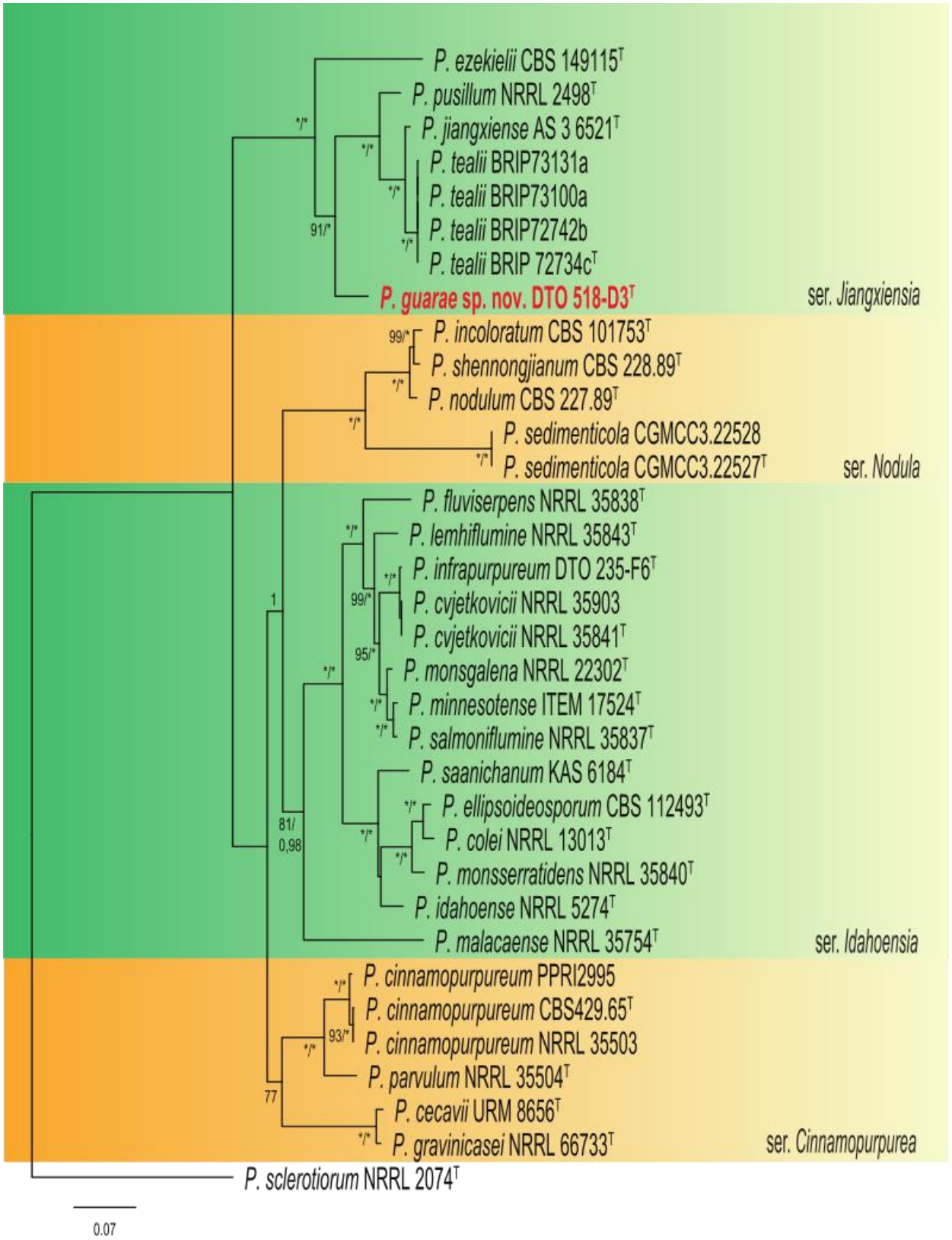


Figure 2. Maximum likelihood (IQ-TREE) phylogeny of the species belonging to the *Cinnamopurpurea* section inferred from combined, partial *CaM*, *rpb2* and *BenA loci*. The tree is rooted to *Penicillium sclerotiorum* NRRL2074. Numbers at the nodes are IQ-TREE ultra-fast bootstrap (BS) (UFBoot) values ≥ 70 %, followed by RAxML BS ≥ 70 %, and MrBayes

posterior probability (PP) ≥ 95. Coloured names indicate strains that belong to the new species. The scale bar represents the number of substitutions per site.

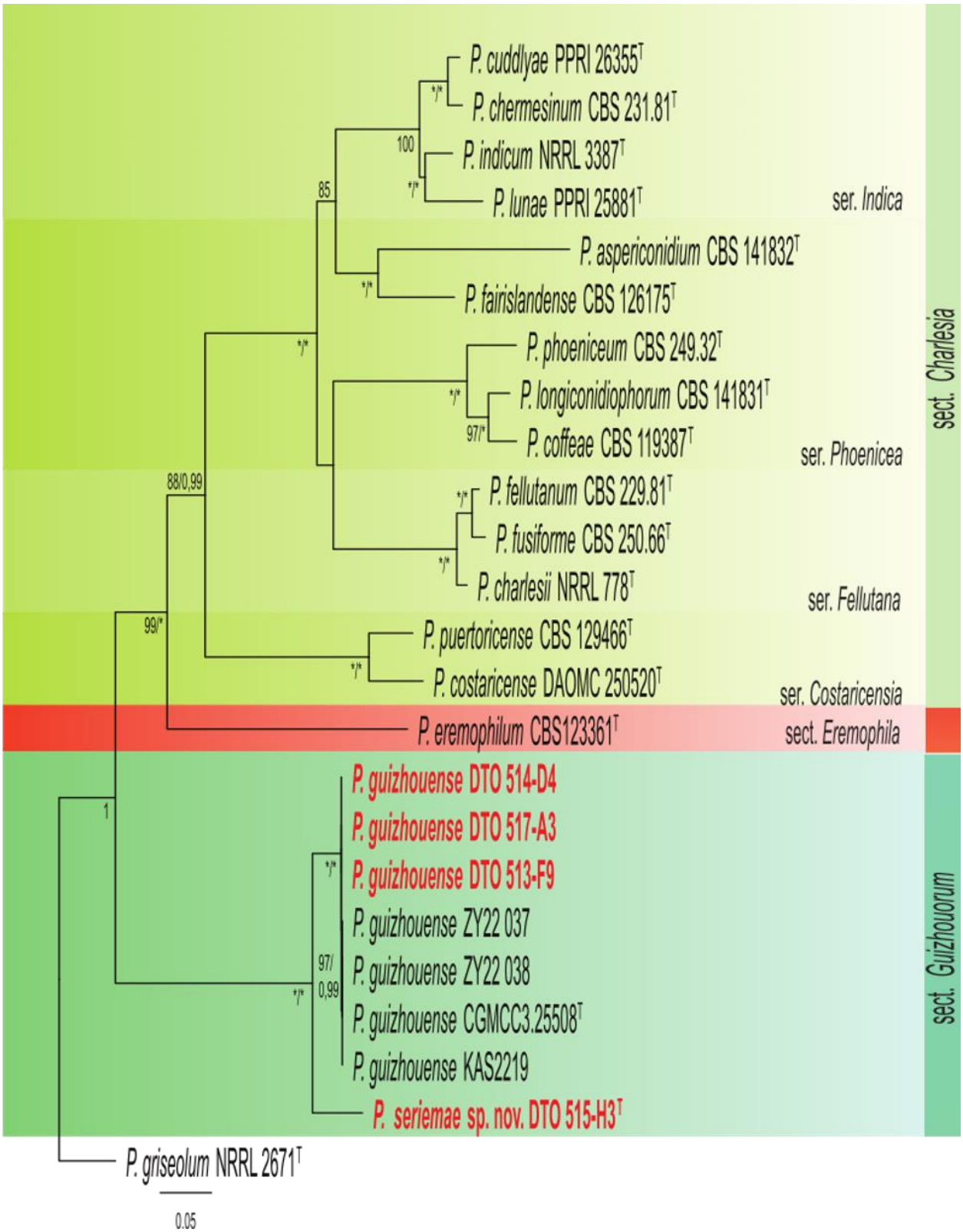


Figure 3. Maximum likelihood (IQ-TREE) phylogeny of the species belonging to the *Charlesia, Eremophila* and *Guizhouorum* sections inferred from combined, partial *CaM*, *rpb2* and *BenA loci*. The tree is rooted to *Penicillium griseolum* NRRL2671. Numbers at the nodes are IQ-TREE ultra-fast bootstrap (BS) (UFBoot) values ≥ 70 %, followed by RAxML BS ≥ 70 %, and MrBayes posterior probability (PP) ≥ 95. Coloured names indicate strains that belong to the new species. The scale bar represents the number of substitutions per site.


P. coffeatum CS10-15T
P. vasconiae CBS 339.79T
P. annulatum CBS 135126T
P. soliforme CGMCC.3.18806T
P. amazonense COAD3476T
P. rolfsii CBS 368.48T
P. terrarumae CBS 131811T
P. piscarium CBS 362.48T
P. ochrochloron CBS 357.48T
P. rotoruae CBS 145838T
P. svalbardense EXF 1307T
P. flaviroseum CGMCC.3.18805T
P. subrubescens DTO 188-D6T
P. camponoti DAOMC 250557T
P. subrutilans CS20-14T
P. pulvillorum CBS 280.39T
P. excelsum ITAL 7572T
P. bissettii DAOMC 167011T
P. fructuariae-cellae ITEM 18276T
ser. Rolfsiorum
P. hainanense CGMCC.3.18798T
P. melanosporum FMR 17424T
P. ehrlichii CBS 324.48T
P. meloforme CBS 445.74T
P. siccitolerans FMR 17381T
P. levitum CBS 345.48T
P. coeruleum CBS 141.45T
P. elleniae CBS 118135T
P. caperatum CBS 443.75T
P. malacosphaerulum CBS 135120T
P. reticulisporum CBS 122.68T
P. nordestinense URM 8423T
P. javanicum CBS 341.48T
P. silvertonense PPRI 25784T
P. setosum WSR 62T
P. uruguayense FMR 14490T
P. lineolatum CBS 188.77T
P. potchefstroomense PPRI 8668T
P. curticaule CBS 135127T
P. brefeldianum CBS 235.81T
P. limosum CBS 339.97T
P. michoacanense FMR 17612T
P. ortum CBS 135669T
P. ashbyae MST FP22310T
P. cremeogriseum CBS 223.66T
P. ludwigii CBS 417.68T
P. janthinellum CBS 340.48T
P. zhanjiangense CGMCC.3.25206T
P. soli KUMCC 18_0202T
P. glaucoroseum DTO 225-E8T
P. yunnanense CGMCC.3.18794T
P. raperi CBS 281.58T
P. koreense KACC 47221T
P. tengii CS27-03T
P. beraoi sp. nov. DTO 511-A1
P. beraoi sp. nov. DTO 517-A4
P. beraoi sp. nov. DTO 510-F3
P. beraoi sp. nov. DTO 513-C2T
P. reverso-vinaceum SJ70
P. reverso-vinaceum URM 8967T
P. reverso-vinaceum URM 9078
P. moreirae sp. nov. DTO 515-G9
P. moreirae sp. nov. DTO 513-B9
P. moreirae sp. nov. DTO 514-F4
P. moreirae sp. nov. DTO 513-C3T
P. moreirae sp. nov. DTO 513-B2
P. donggangicum AS3 15900T
P. limoniforme CGMCC.3.22512T
ser. Janthinella

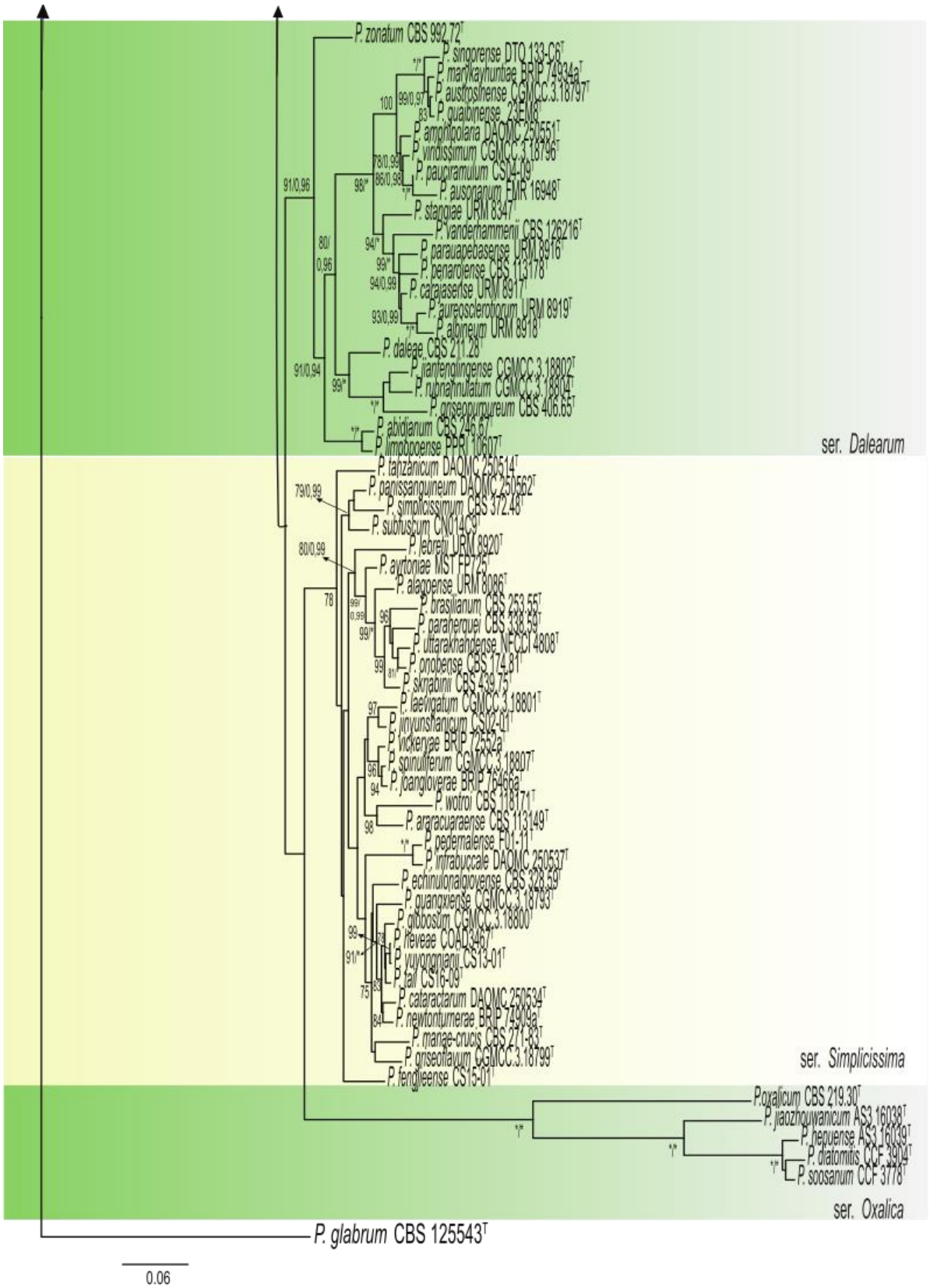


Figure 4. Maximum likelihood (IQ-TREE) phylogeny of the species belonging to the *Lanata-Divaricat*a section inferred from combined, partial *CaM, rpb2* and *BenA* loci. The tree is rooted to *Penicillium glabrum* CBS125543. Numbers at the nodes are IQ-TREE ultra-fast bootstrap (BS) (UFBoot) values ≥ 70 %, followed by RAxML BS ≥ 70 %, and MrBayes posterior probability (PP) ≥ 95. Coloured names indicate strains that belong to the new species. The scale bar represents the number of substitutions per site.

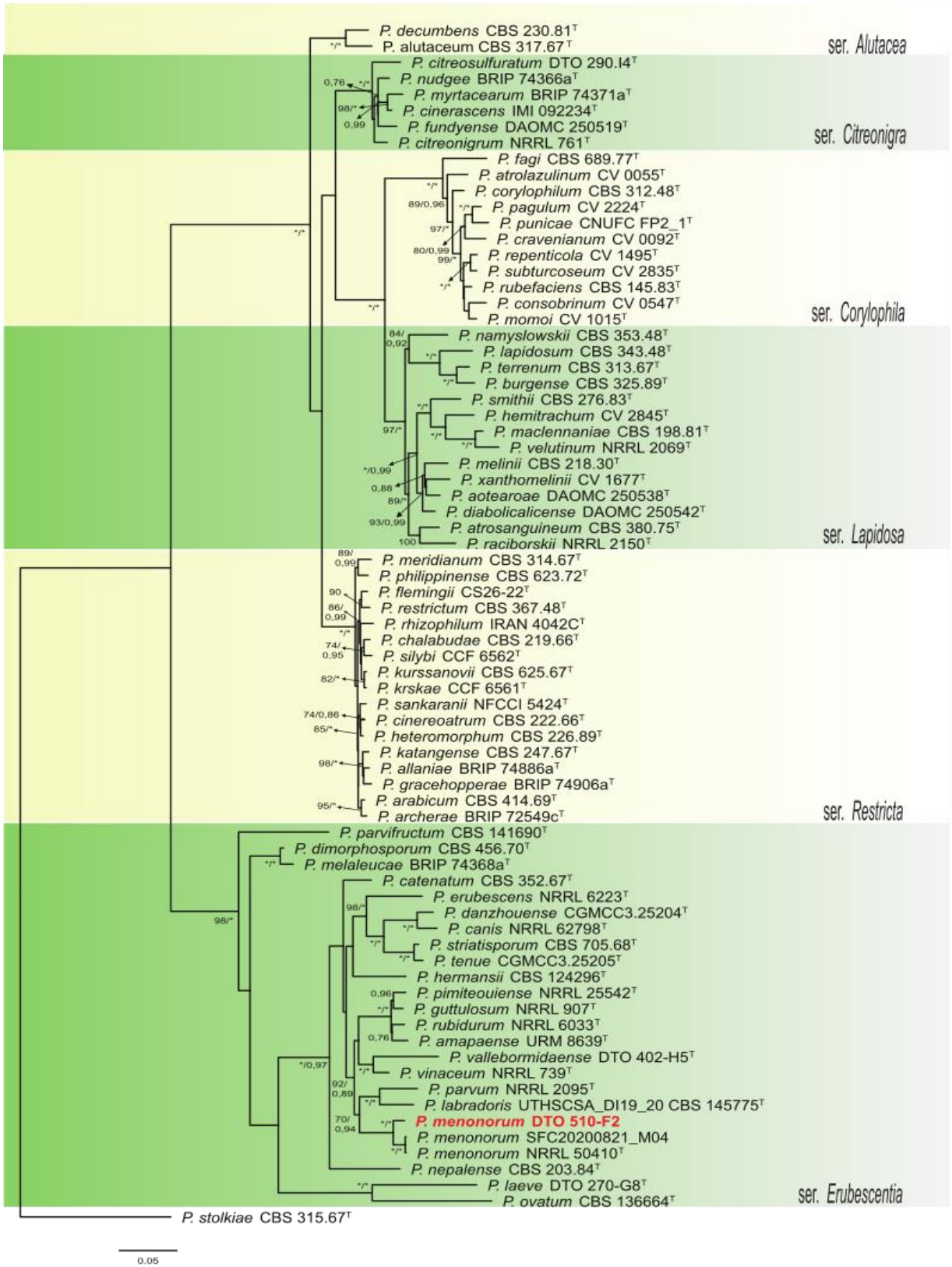


Figure 5. Maximum likelihood (IQ-TREE) phylogeny of the species belonging to the *Exilicaulis* section inferred from combined, partial *CaM, rpb2* and *BenA loci*. The tree is rooted to *Penicillium stolkieae* CBS 315.67. Numbers at the nodes are IQ-TREE ultra-fast bootstrap (BS) (UFBoot) values ≥ 70 %, followed by RAxML BS ≥ 70 %, and MrBayes posterior probability (PP) ≥ 95. Coloured name indicate strain from Cerrado that is ocurrence record species. The scale bar represents the number of substitutions per site.

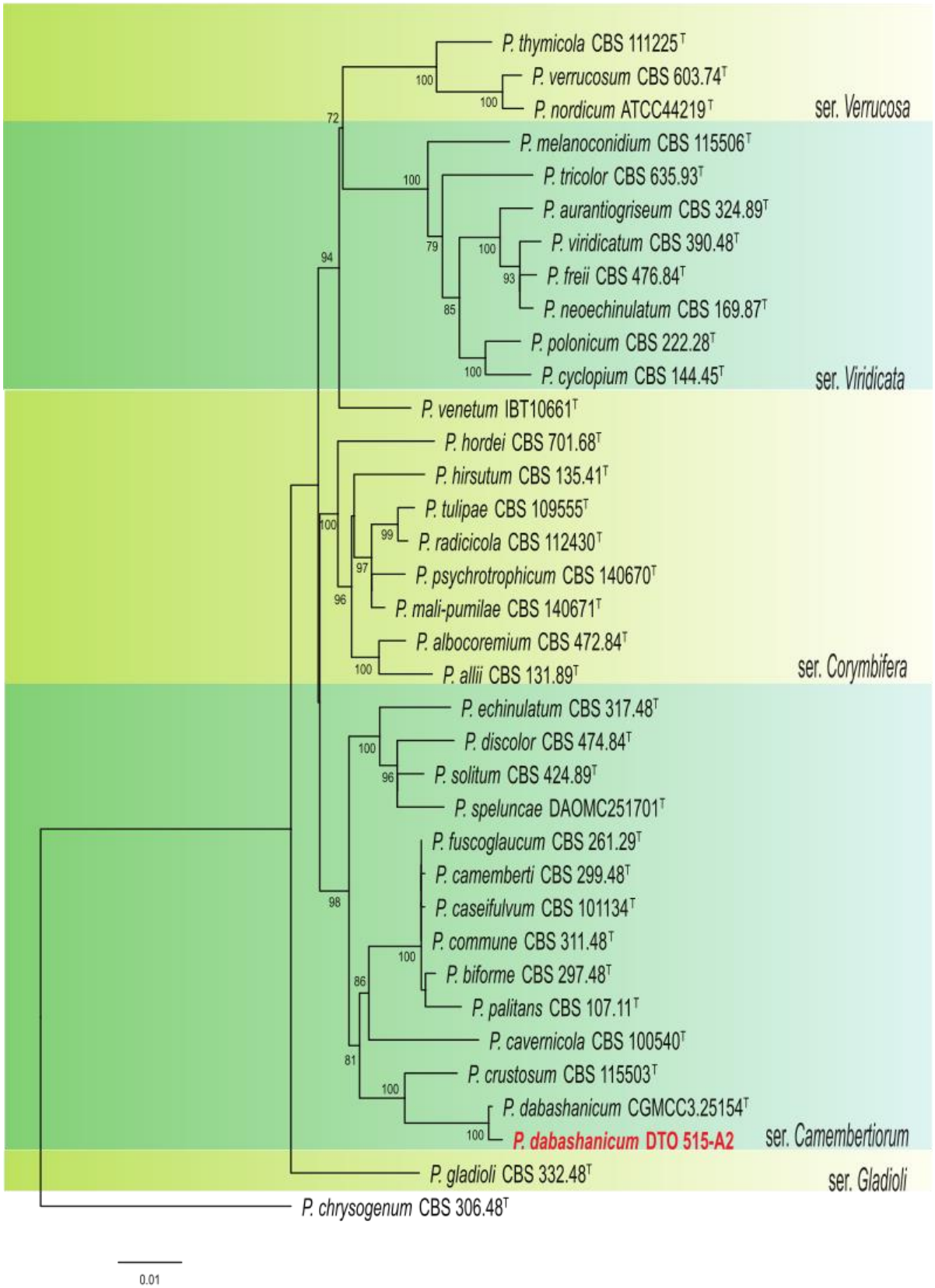


Figure 6. Maximum likelihood (IQ-TREE) phylogeny of the species belonging to the *Fasciculata* section inferred from combined, partial *CaM*, *rpb2* and *BenA loci*. The tree is rooted to *Penicillium chrysogenum* CBS 306.48. Coloured name indicate strain from Cerrado that is ocurrence record species. The scale bar represents the number of substitutions per site.

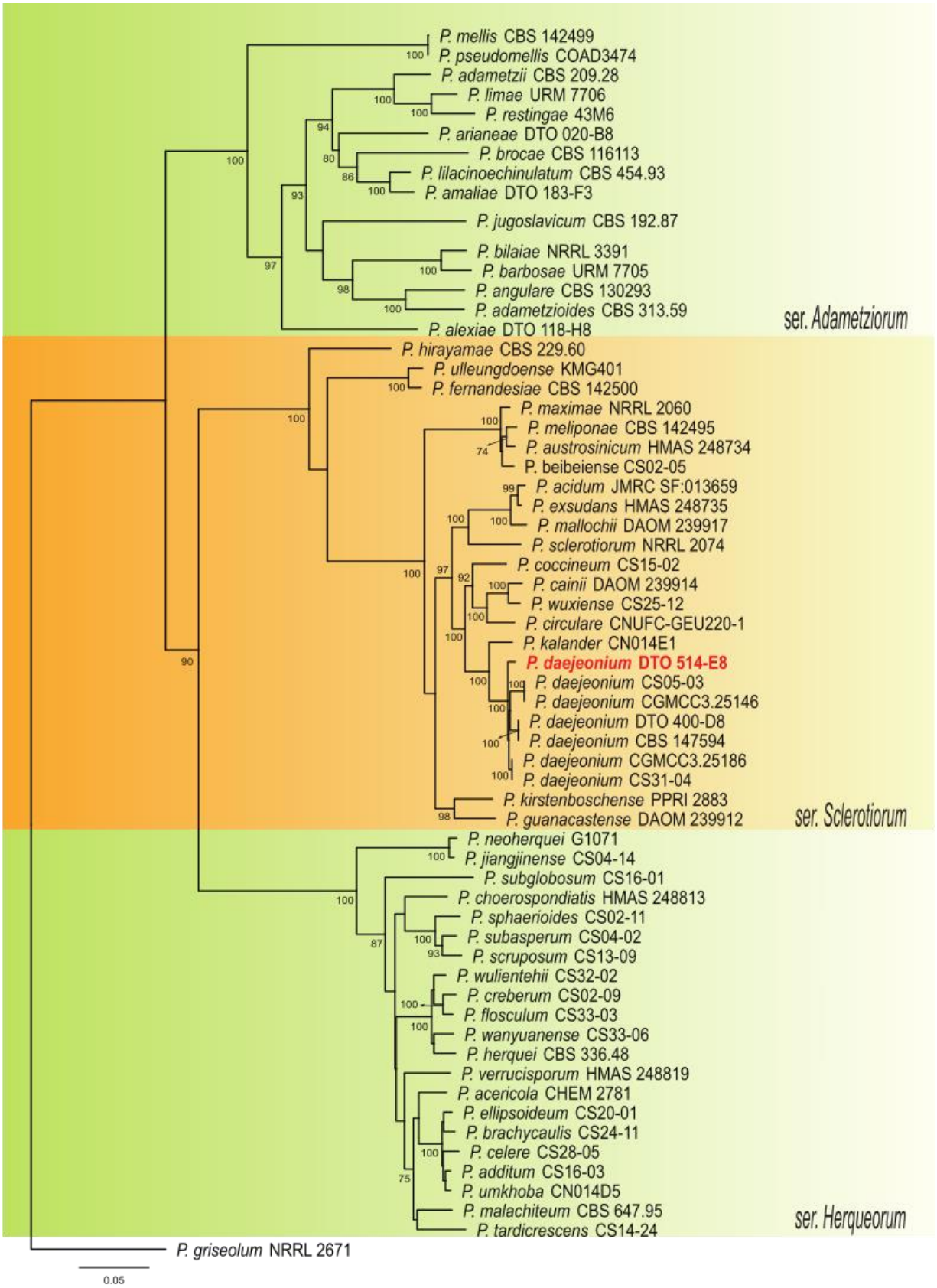


Figure 7. Maximum likelihood (IQ-TREE) phylogeny of the species belonging to the *Sclerotiorum* section inferred from combined, partial *CaM*, *rpb2* and *BenA loci*. The tree is rooted to *Penicillium griseolum* NRRL 2671. Coloured name indicate strain from Cerrado that is ocurrence record species. The scale bar represents the number of substitutions per site.

3.2 Taxonomy

Penicillium patrocinensis Figueiredo, Zhou, Batista, Houbraken, *sp. nov*. Fig. 8.

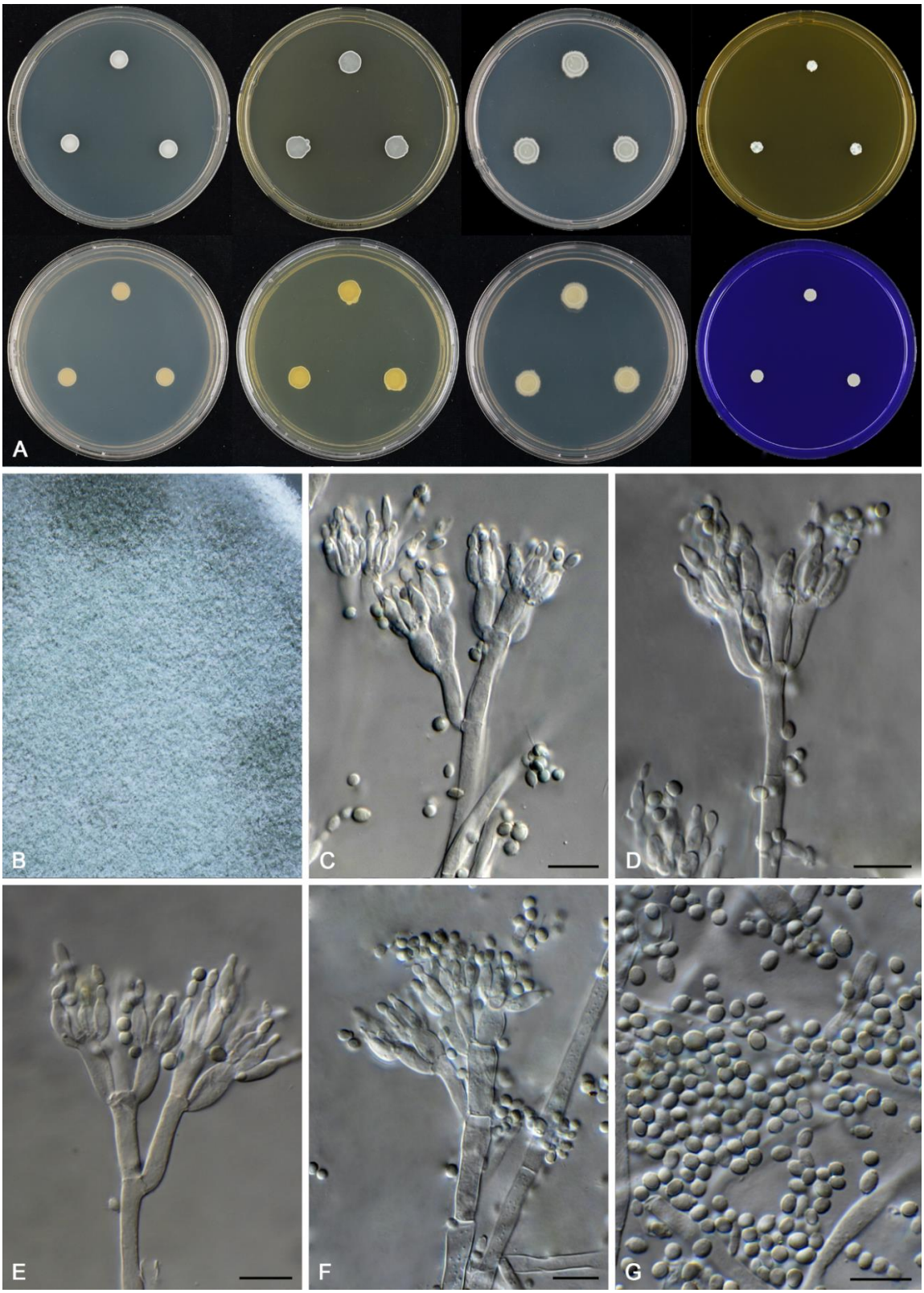


Figure 8. *Penicillium patrocinensis* (DTO 515-G4). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 µm.

Etymology. Latin, *Penicillium patrocinensis*: named, in reference Patrocínio, the municipality in Minas Gerais State, Brazil, where the type strain was isolated from soil in area of regenerative coffee cultivation.

Diagnosis: Colonies restricted on all culture media. No acid production in CREA and no growth at 30 and 37 °C in CYA. Conidiophores mmostly biverticillate and terverticillate; stipes slightly rough walled, occasionally smooth; phialides measuring 5.8–11.5 × 2.4–3.6 µm; conidia finely roughened, subglobose to ellipsoidal, occasionally apiculate, measuring 3–3.5 × 2.5–3 µm. Sclerotia are not produced.

Colony morphology (7 d, in mm): CREA 5–6; CYA 8; CYA 30°C no growth; CYA 37°C no growth; CYAS 9–12; DG18 10–12; MEA 5–6; OA 6–7; YES 10–12.

Culture characteristics: *CYA 25 °C, 7 d*: Colonies raised at center, margins low, narrow, entire; mycelia white; texture velutinous; sporulation poor to moderate; conidia *en masse* pale blue to pastel blue (23A3-23A4); soluble pigments absent; exudates absent; reverse dull yellow to champagne (3B3-4B4); *CYAS 25 °C, 7 d*: Colonies raised, margins low, narrow, irregular; mycelia white; texture velutinous; sporulation moderate, conidia *en masse* sky grey to water blue (23B-24C5); soluble pigments absent; exudates absent; reverse olive grey to yellowish white (2E2-3A2); *MEA 25 °C, 7 d*: colonies slightly raised; margins low, narrow, entire; mycelium white; colony texture velotunious; sporulation poor; conidia *en masse* greyish blue to dull blue (23B4-23D4); exudates absent; soluble pigments absent; reverse pompeian yellow to golden brown (5C6-5D7); *YES 25 °C, 7 d*: Colonies deep, crateriform, sometimes randomly, margins low, narrow, irregular; mycelium white; colony texture velutinous; sporulation moderate; conidia *en masse* greyish green to dull green (25C3-25D4); exudates absent; soluble pigments absent; reverse olive (3D4-3E5); *DG18 25 °C, 7 d*: colonies raised at center; margins low, narrow, irregular; mycelium white; colony texture velutinous; sporulation moderate; conidia *en masse* sky grey to greyish green (23B2-25D4); exudates absent; soluble pigments absent; reverse greyish yellow to olive grey (2C3-2E2) at center and greenish grey (27C2) in the margin; *OA 25 °C, 7 d*: Colonies plane, margin entire; mycelium white; colony texture floccose; sporulation moderate; conidia *en masse* greyish yellow (25B3-25C5); exudates absent; soluble pigments greyish yellow; *CREA 25 °C, 7 d*: poor growth, acid production absent, base formation absent.

Micromorphology: Conidiophores mostly biverticillate and terverticillate also present; stipes slightly rough walled, occasionally smooth 138–408 × 2.9–4.8 µm; branches/rami 2–3 when present, 11.6–23.5 × 3–5 (–7.5) µm; metulae divergent, 2–7 per stipe/branch 7–15.7 × 3.2–7.2 µm; phialides ampulliform, 5.8–11.5 × 2.4–3.6 µm (8.0 ± 1.1 × 2.9 ± 0.2); conidia finely

roughened, subglobose to ellipsoidal, occasionally apiculate, 2.4–4.2 × 2–3 (3.0 ± 0.3 × 2.4 ± 0.2), n = 50.

Typus: **Brazil**, Minas Gerais, Patrocinio, 18°52'45"S 47°05'53"W, 965 m a.s.l., from soil Cerrado, in February. 2023, *C.N. Figueiredo* (**holotype** DTO515-G4 ex-type culture URMICRO 11842 = MC186).

Notes: Penicillium patrocinensis is characterized by colonies restricted to all culture media. No acid production in CREA and no growth at 30 and 37 °C in CYA. *Penicillium tularense* is also no acid producer in CREA and shows no growth at 37°C in CYA, but is easily distinguished from *P. patrocinensis* due to its growth rate in culture media and in CYA at 30°C. Furthermore, the new species has larger metulae and conidia.

Based on a megablast search of NCBIs GenBank nucleotide database, the closest hits using the ITS sequence are *Penicillium malodoratum* (strain CBS 490.65, GenBank OL711860; Identities 847/873 (97%), six gap), *Penicillium sajarovii* (strain CBS 144777, GenBank MK226542; Identities 846/873 (97%), five gaps) and *Penicillium spathulatum* (strain CBS 117192, GenBank MH863015; Identities 840/865 (97%), seven gaps). The closet hits using the rpb2 sequence are *Penicillium* sp. (strain strain SYPF 8431, GenBank MF588914); Identities 1030/1078 (96%), two gaps) and *Penicillium tularense* (strain CBS 430.69, GenBank JN121516); Identities 866/953 (91%), three gaps). The closest hits using the *CaM* sequence are *Penicillium sp.* (strain strain SYPF 8431, GenBank MF588906; Identities 424/464 (91%), seven gaps) and *Penicillium tularense* (strain AS3.14006, GenBank KC427155; Identities 393/469 (84%), gaps 20/469(4%)). The closest hits using the *BenA* sequence are *Penicillium catenatum* (strain MUT<ITA>:3930, GenBank MK050517; Identities 430/459 (94%), six gaps) and *Penicillium tularense* (strain CBS 43169, GenBank AY674433; Identities 340/405 (84%), 12 gaps).

Penicillium pequii Figueiredo, Zhou, Batista, Houbraken, *sp. nov*. Fig. 9.

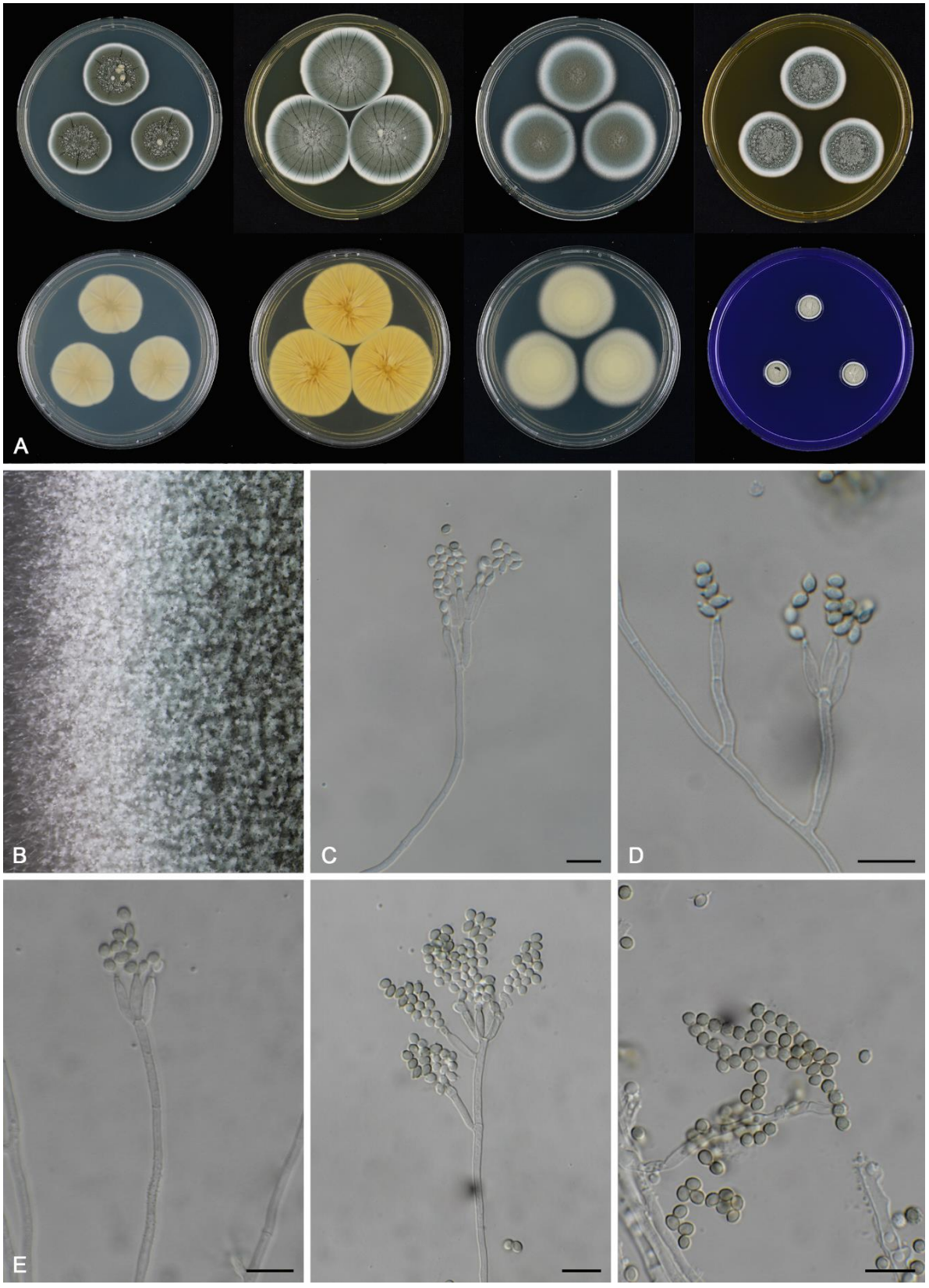


Figure 9. *Penicillium pequii* (DTO 510-F4^{T}). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 μm.

Etymology. Latin, *pequii*: named in reference to the tree (*Caryocar brasiliense*) that is a symbol and a typical species of the Brazilian Cerrado, wich produces the frui known as "pequi', of great cultural, ecological, and economic importance in the region.

Diagnosis: Colonies on CYA 28–31 mm, CYA 30 °C 9–45 and CYA 37 °C no growth. Acid not produced on CREA. Conidiophores monoverticillate, and biverticillate, occasionally terverticillate; 5 µm; phialides ampullifrom, 2–14 per metulae, 7–12 × 2–3.2 µm; conidia finely roughened, ellipsoidal to fusiform, 2.6–4 × 2–3 µm; Sclerotia are not produced.

Colony morphology (7 d, in mm): CREA 12–15; CYA 28–31; CYA 30°C 9–14; CYA 37°C no growth; CYAS 29–32; DG18 28–37; MEA 25–30; OA 23–26; YES 32–42.

Culture characteristics: CYA 25 °C, 7 d: Colonies moderately deep, radially sulcate, raised at center; margins low, narrow, entire; mycelia white; texture velutinous; sporulation dense, conidia *en masse* dull green (25E3-29E3); soluble pigments sometimes inconspicuous yellow; exudates clear; reverse yellowish grey to dull yellow (2B2-3B3); *CYAS 25 °C, 7 d*: Colonies moderately deep, radially and concentrically sulcate, margin entire; mycelium white; colony texture velutinous; dense sporulation; conidia *en masse* greenish grey to dull green (26D2-27E3); exudates absent; soluble pigments absent; reverse yellowish grey to dull yellow (3B2-3B3); *MEA 25 °C, 7 d*: Colonies moderately deep to colonies plane, slightly raised at the center, sometimes radially sulcate, margin entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* dull green to greyish green (25D4-25E5); exudates clear; soluble pigments absent; reverse honey yellow to brownish yellow (5C6-5C7) at center and light orange (5A5) in the margin; *YES 25 °C, 7 d*: Colonies moderately deep, raised at center, radially sulcate, margin entire; mycelium white; colony texture velutinous; sporulation dense; conidia en masse dull green (25E3-28E4); exudates absent; soluble pigments absent; reverse greyish yellow (3B4); *DG18 25 °C, 7 d*: Colonies plane, raised at the center, margin entire; mycelium white; colony texture velutinous; sporulation dense; conidia en masse olive to greyish green (2E3-25C5), sometimes dull green (25E4); exudates absent; soluble pigments absent; reverse pale yellow to greenish green (1A3-26C2); *OA 25 °C, 7 d*: Colonies plane, margin entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish green (25C5-25D5); exudates clear; soluble pigments absent; *CREA 25 °C, 7 d*: poor to moderate growth, acid production absent, base formation absent.

Micromorphology: conidiophore monoverticillate and biverticillate, occasionally terverticillate; stipes smooth, occasionally rough-walled, 10–172 × 2–2.7 µm; metulae cylindrical, 7–21 × 2–3.5 µm; phialides ampullifrom, 2–14 per metulae, 7–12 × 2–3.2 µm (8.9 ± 0.9 × 2.7 ± 0.2); conidia finely roughened, ellipsoidal to fusiform, 2.6–4 × 2–3 µm, (3.1 ±

0.3 × 2.5 ± 0.2), n = 36.

Typus: **Brazil**, Minas Gerais, Patrocinio, 18°52'45"S 47°05'53"W, 965 m a.s.l., from soil native Cerrado, in February. 2023, *C.N. Figueiredo* (**holotype** DTO 510-F4 ex-type culture URMICRO 11748 = MM13).

Notes: Penicillium pequii is characterized by the production of a clear exudate in CYA, MEA, and OA and growth at 30 °C in CYA. *P. pequii* is easily distinguished from *Penicillium scabrosum* due to its production of monoverticillate and terverticillate conidiophores, in addition to having ellipsoidal to fusiform conidia. *P. scabrosum* has a lower growth rate on culture media and only biverticillate conidiophores. Based on a megablast search of NCBIs GenBank nucleotide database, the closest hits using the ITS sequence are *Penicillium scabrosum* (strain NNIBRFG1470, GenBank MT995062; Identities 864/877 (99%), three gaps) and *Penicillium sajarovii* (strain CBS 144777, GenBank MK226542; Identities 861/875 (98%), one gap). The closet hits using the rpb2 sequence are *Penicillium lanosum* (strain CBS 106.11, GenBank KU904356); Identities 991/1089 (91%), no gaps), *Penicillium jamesonlandense* (strain CN093B7, GenBank MZ984124); Identities 958/1051 (91%), two gaps) and *Penicillium swiecickii* (strain CN093F5, GenBank MZ984133); Identities 957/1051 (91%), two gaps). The closest hits using the *CaM* sequence are *Penicillium scabrosum* (strain CBS 683.89, GenBank FJ530987; Identities 382/438 (87%), 24/438 (5%) gaps) and *Penicillium biourgeianum* (strain NRRL 865, GenBank AY484828; Identities 335/381 (88%), 15/381 (3%) gaps). The closest hits using the *BenA* sequence are *Penicillium swiecickii* (strain DAOM 214775, GenBank DQ285608; Identities 403/446 (90%), 10 gaps) and *Penicillium lanosum* (strain CBS 260.55, GenBank DQ285623; Identities 404/448 (90%), 12 gaps).

Penicillium pluvialis Figueiredo, Zhou, Batista, Houbraken, *sp. nov*. Fig. 10.

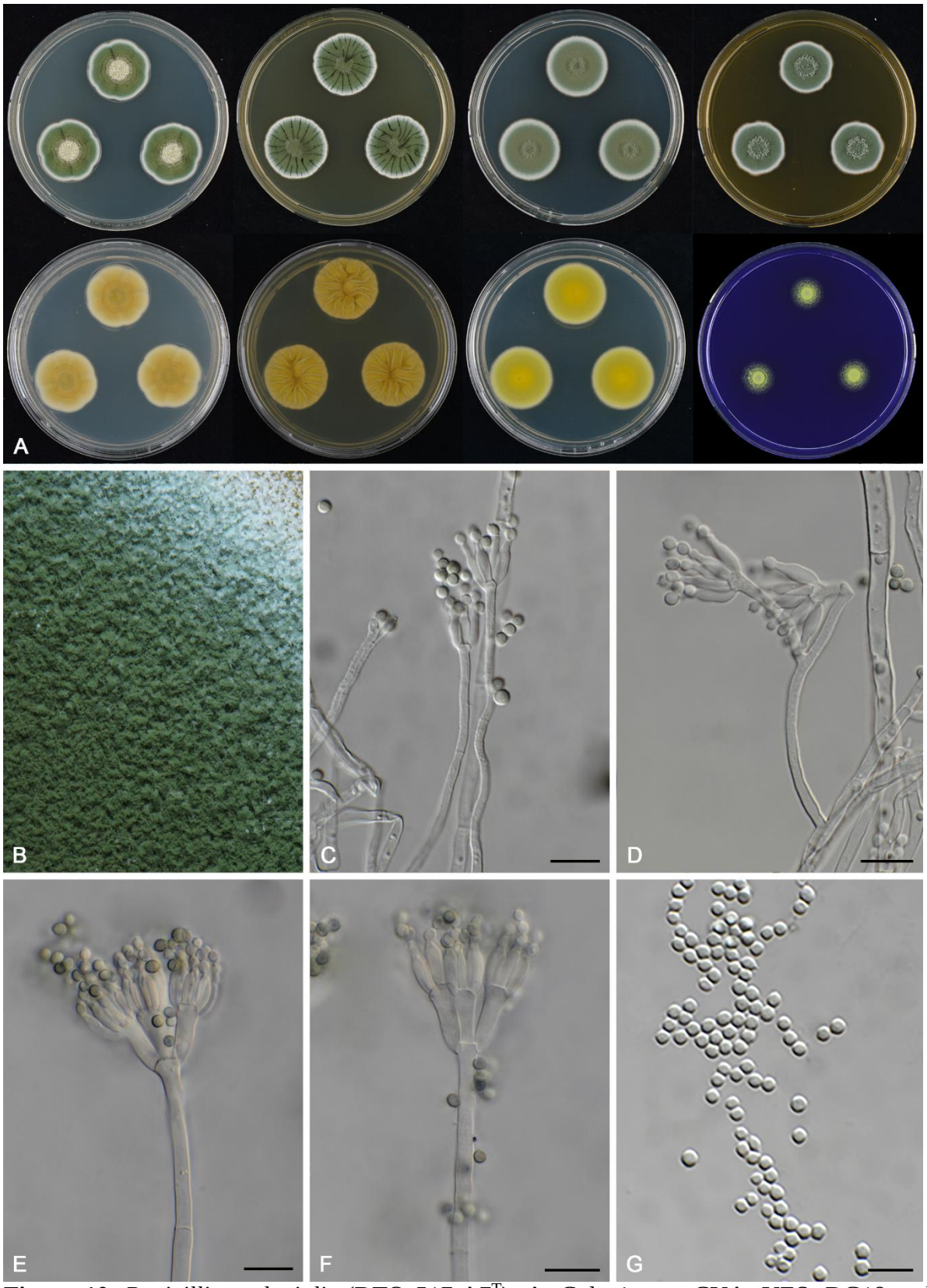


Figure 10. *Penicillium pluvialis* (DTO 517-A7^{T}). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 µm.

Etymology. Latin, pluvialis: named, in reference to the rainy season in which the soil samples

were collected (January-March) and to the marked seasonality of precipitation that characterizes the Cerrado biome.

Diagnosis: Penicillium pluvialis is characterized by its abundant sporulation on culture media and growth at 30 °C in CYA. This specie presents conidiophore biverticillate, occasionally monoverticillate and terverticillate, stipes smooth-walled, measuring 20–302 µm, conidia finely roughened to rough, globose to subglobose, measuring 2.7–3.8 × 2.2–3 µm.

Colony morphology (7 d, in mm): CREA 15–17; CYA 25–27; CYA 30°C 5–8; CYA 37°C no growth; CYAS 28–29; DG18 23–27; MEA 19–24; OA 18–20; YES 27–28.

Culture characteristics: CYA 25 °C, 7 d: colonies moderately deep, sometimes raised at center, concentrically and radially sulcate; margins low, narrow, entire; mycelia white; texture velutinous; sporulation dense, conidia *en masse* greyish green (28E6); exudates clear to inconspicuously yellow; soluble pigments absent; reverse yellowish grey to greyish yellow (3C2-3C5); *CYAS 25 °C, 7 d*: Colonies raised at center, radially sulcate, margin entire; mycelium white; colony texture velutinous; dense moderate; conidia *en masse* greyish green (25D5-25E6); exudates and soluble pigments absent; reverse greenish grey (26B2-27C2); *MEA 25 °C, 7 d*: Colonies slightly raised at center, radially sulcate margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish green (26D5); exudates and soluble pigments absent; reverse olive brown (4D7-4D8); *YES 25 °C, 7 d*: Colonies raised at center, concentrically and radially sulcate margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish green (28E5); exudates absent and soluble pigments absent; reverse light yellow (2A5); *DG18 25 °C, 7 d*: Colonies low, plane, slightly raised at center, margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish grey (28E5); exudates and soluble pigments absent; greyish yellow (2B6) at center and olive yellow (2C6) in margin; *OA 25 °C, 7 d*: Colonies low, plane, margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish grey (26D6); exudates sometimes clear soluble pigments light brown; *CREA 25 °C, 7 d*: moderate growth, acid production absent, base formation absent.

Micromorphology: Conidiophore biverticillate, occasionally monoverticillate and terverticillate; stipes smooth walled, 20–302 × 2–3.8 µm; metulae cylindrical, 2–6 per stipe, 8.7–20 × 2.7–4.7 µm; phialides ampulliform, 6.2–10.3 × 2.7–4 µm (8.4 ± 0.9 × 3.1 ± 0.2); conidia finely roughened to rough, globose to subglobose, 2.7–3.8 × 2.2–3 (3.1 ± 0.2 × 2.6 ± 0.2), n = 40.

Typus: **Brazil**, Minas Gerais, Patrocinio, 18°52'45"S 47°05'53"W, 965 m a.s.l., from soil

native Cerrado, in February. 2023, *C.N. Figueiredo* (**holotype** DTO 517-A7 ex-type culture URMICRO 11872 = MM214).

Notes: A multigene phylogeny resolves *Penicillium pluvialis* as a close relative of *Penicillium xyleborini* in sect *Ramosum* series *Soppiorum*. The new species growth in CYA at 30 °C and produces condiophore biverticillate, occasionally monoverticillate and terverticillate, compared no growth in CYA at 30°C and conidiophore biverticillate to terverticillate of *P. xyleborini*. Based on a megablast search of NCBIs GenBank nucleotide database, the closest hits using the ITS sequence are *Penicillium xyleborini* (strain CN001D4, GenBank MW504356; Identities 848/855 (99%), one gap), *Penicillium scabrosum* (strain NNIBRFG1470, GenBank MT995062; Identities 856/871 (98%), three gaps) and *Penicillium sajarovii* (strain CBS 144777, GenBank MK226542; Identities 847/864 (98%), two gaps). The closet hits using the rpb2 sequence are *Penicillium xyleborini* (strain CN001D4, GenBank MW480824); Identities 1018/1064 (96%), no gaps), *Penicillium lusitanum* (strain CN093C6, GenBank MZ984142); Identities 977/1051 (93%), three gaps) and *enicillium lanosum* (strain CBS 106.11, GenBank KU904356); Identities 989/1098 (90%), no gaps). The closest hits using the *CaM* sequence are *Penicillium xyleborini* (strain CN001D4, GenBank MW480823; Identities 449/461 (97%), no gaps) and *Penicillium virgatum* (strain IBT 24423, GenBank KY989078; Identities 415/481 (86%), gaps 19/481 (3%)). The closest hits using the *BenA* sequence are *Penicillium xyleborini* (strain CN001D4, GenBank MW480817; Identities 410/428 (96%), one gap) and *Penicillium virgatum* (strain IBT 19272, GenBank KY989033; Identities 387/445 (87%), gaps 20/445 (4%)).

Penicillium guarae Figueiredo, Zhou, Batista, Houbraken, *sp. nov*. Fig. 11.

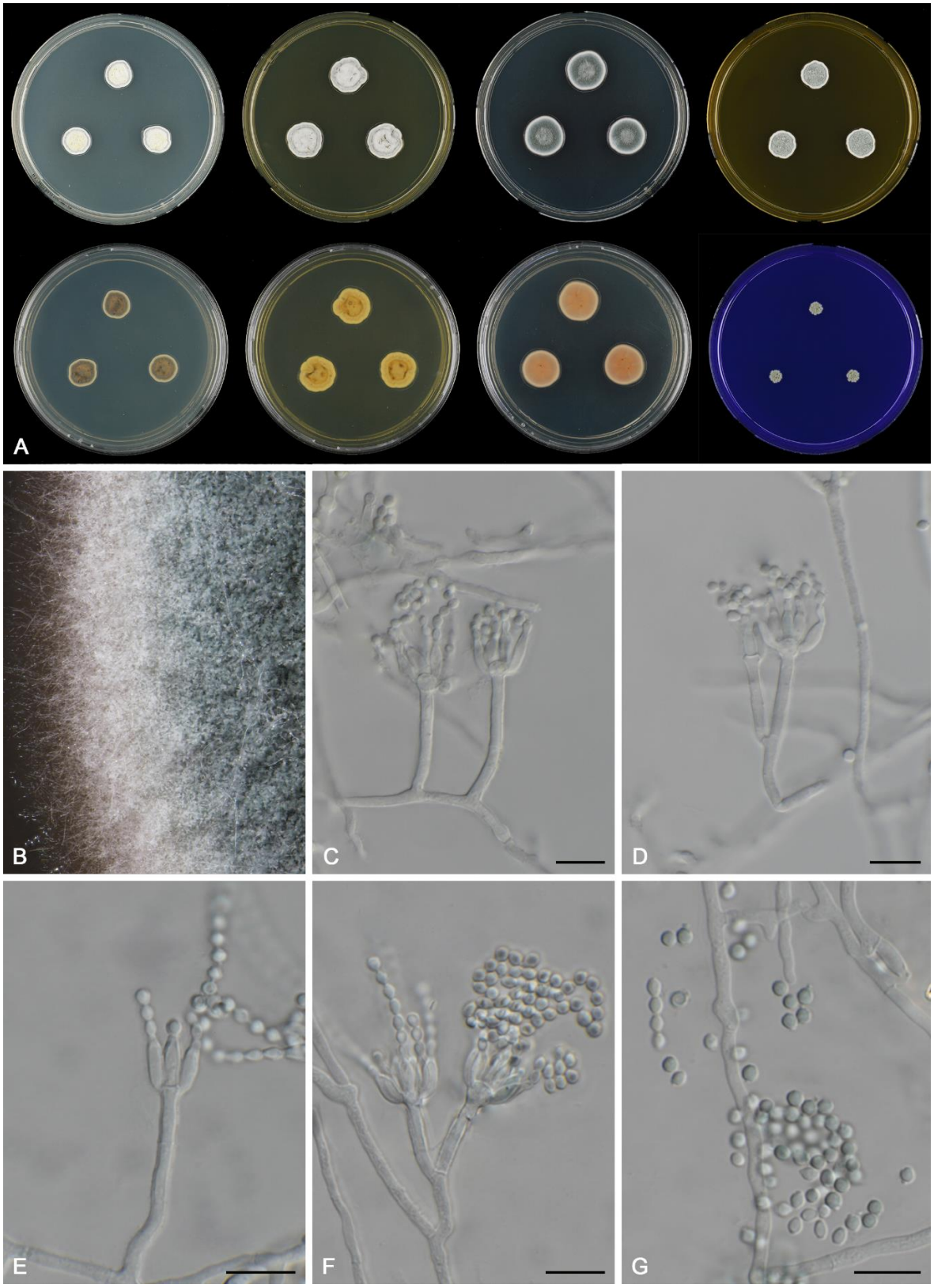


Figure 11. *Penicillium guarae* (DTO 518-D3). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 μm.

Etymology. Latin, *guarae*: named in reference to the maned wolf (*Chrysocyon brachyurus*),

an emblematic mammal and symbol of the Brazilian Cerrado, currently threatened with exctinction due to the devastation and fragmentation of the remaining areas of this biome.

Diagnosis: Colonies restricted on all culture media, no growth above 37 °C. Conidiophores monoverticillate, occasionally biverticillate; stipes smooth walled with slightly vesiculate ending; phialides 5.5–10 × 2.1–2.9 µm; conidia smooth walled, ellipsoidal to subglobose, grayish green to dark green, 2.3–3 × 1.6–2.2 µm; Sclerotia are not produced.

Colony morphology (7 d, in mm): CREA 6-7; CYA 12–13; CYA 30°C 9–10; CYA 37°C no growth; CYAS 15-16; DG18 12–13; MEA 12; OA 15; YES 16.

Culture characteristics: CYA 25 °C, 7 d: Colonies moderately deep, irregularly sulcate, slightly crateriform, margin entire; mycelium white; colony texture velvety; poor sporulation; conidia *en masse* greyish green (27C3); exudates absent; soluble pigments absent; reverse olive yellow (3D8) in the center and greenish grey (27E2) in the periphery; *CYAS 25 °C, 7 d*: Colonies moderately deep, radially sulcate, slightly crateriform, margin entire; mycelium white; colony texture velvety; moderate sporulation; conidia *en masse* olive (3E3); exudates absent; soluble pigments slightly; reverse grey (3F1); *MEA 25 °C, 7 d*: Colonies plane, margin entire; mycelium white; colony texture velvety; sporulation moderate; conidia *en masse* olive (2E3); exudates golden yellow; soluble pigments absent; reverse light brown (6D7) in the center and brown (6E5) in the periphery; *YES 25 °C, 7 d*: Colonies moderately deep, irregulary sulcate, crateriform, margin entire; mycelium white; colony texture velvety; sporulation poor; conidia *en masse* grey (3C1); exudates absent; soluble pigments absent; reverse yellowish brown (5D6); *DG18 25 °C, 7 d*: Colonies moderately deep, radially sulcate, margin entire; mycelium white; colony texture velvety; sporulation dense; conidia *en masse* full green (27E4); exudates absent; soluble pigments present; reverse light brown (5D7); *OA 25 °C, 7 d*: Colonies plane, margin entire; mycelium white; colony texture velvety; sporulation poor; conidia *en masse* dark green (28F8); exudates clear; soluble pigments present; *CREA 25 °C, 7 d*: poor growth, acid production absent, base formation absent.

Micromorphology: Conidiophores monoverticillate, occasionally biverticillate; stipes smooth walled, 12.5–59 × 2–3 µm; metulae cylindrical, 7.4–17 × 2.2–3.1 µm; vesicle 3.2–5.4 × 4–6.5 µm; phialides ampulliform, 3–8 per metulae, 5.5–10 × 2.1–2.9 µm; conidia smooth walled, ellipsoidal to subglobose, grayish green to dark green, 2.3–3 × 1.6–2.2 in diam.

Typus: **Brazil**, Minas Gerais, Patrocinio, 18°52'45"S 47°05'53"W, 965 m a.s.l., from soil native cerrado, in January, 2023, *C.N. Figueiredo* (**holotype** DTO 518-D3 ex-type culture URMICRO URMICRO 11858 = ML261).

Notes: Penicillium guarae phylogenetically belongs to section *Cinnamopurpurea* and, like

other members of this section, grows restrictedly on CYA and MEA and predominantly produces monoverticillate conidiophores. This new species is sister to a clade containing *P. Jiangxiense*, *P. Pusillum* and *P. tealii*. *Penicillium jiangxiense* can be differentiated from *P. guarae* by its ability to grow at 37 °C and the production of small, smooth-walled conidia (2–2.5 × 1.5–2 μm) (Kong & Liang 2003). *Penicillium pusillum* and *Penicillium tealii* produces sclerotia brownish and yellowish to orange, respectivelly (Tan et al., 2022). These characters are not shared with *P. guarae.* Based on a megablast search of NCBIs GenBank nucleotide database, the closest hits using the ITS sequence are *P. ezekielii* (strain DTO_463-A7, GenBank ON723773; Identities 839/867 (97%), 13 gaps), *Penicillium adametzii* (strain CN014G1, GenBank OR164452; Identities 834/868 (96%) eight gaps) and *Penicillium roseopurpureum* (strain CBS 148.83, GenBank MH861558; Identities 836/872(96%), 10 gaps). The closet hits using the rpb2 sequence are *P. pusillum* (strain NRRL 2498, GenBank KF932995); Identities 920/1012 (91%), five gaps), *P. jiangxiense* (strain DTO 309-A7, GenBank MN969122); Identities 908/1010 (90%), seven gaps), *P. ezekielii* (strain DTO 065-D2, GenBank ON920784; Identities 834/942 (89%), no gaps). The closest hits using the *BenA* sequence are *P. tealii* (strain BRIP 72735b, GenBank OP039553; Identities 436/484 (90%), 12 gaps), *P. jiangxiense* (strain AS:3.6521, GenBank KJ890409; Identities 413/458 (90%), fourteen gaps) and *P. ezekielii* (strain DTO 437-A7, GenBank ON920779; Identities 428/486 (88%), 22 gaps).

Penicillium seriamae Figueiredo, Zhou, Batista, Houbraken, *sp. nov*. Fig. 12.

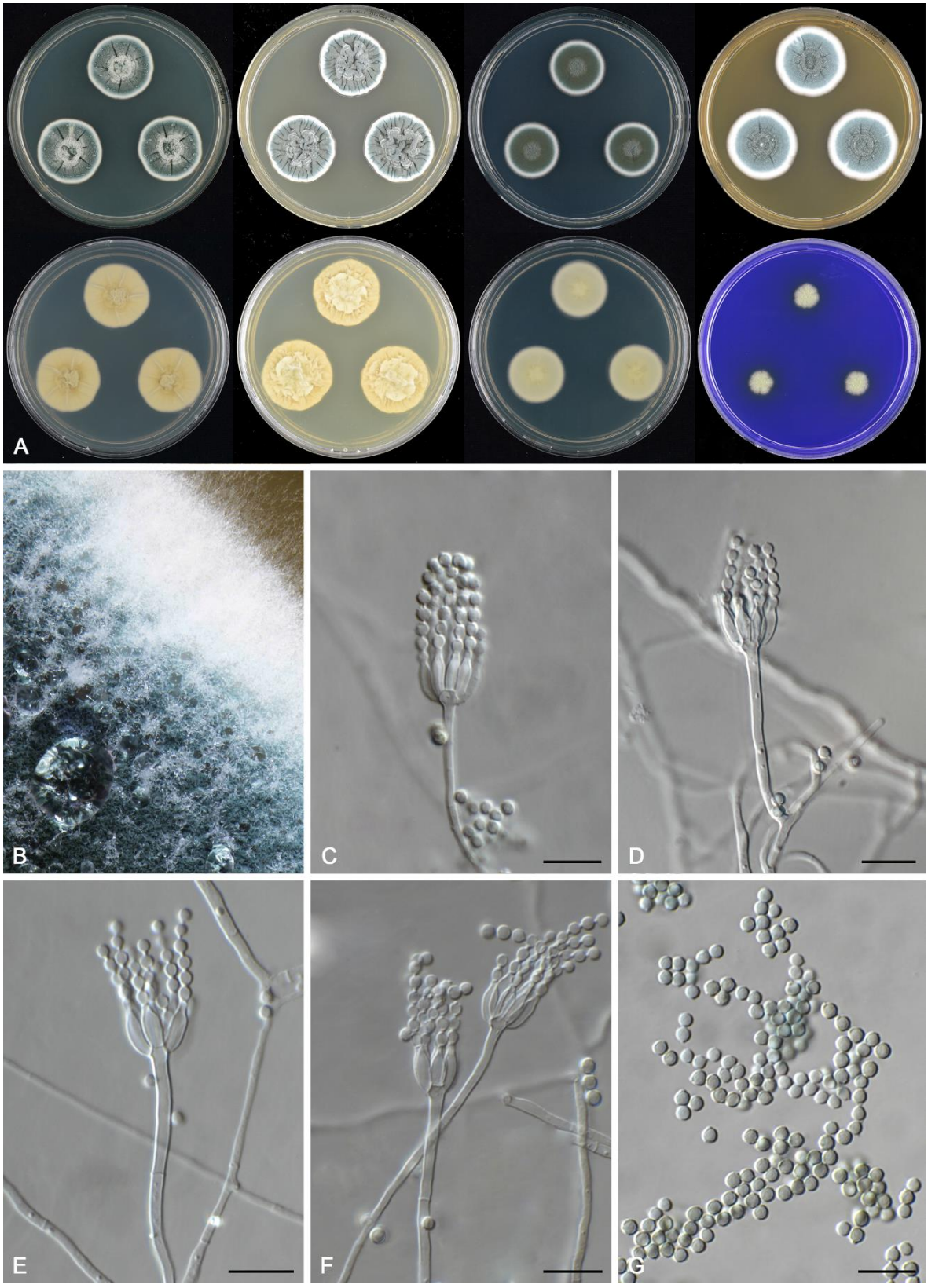


Figure 12. *Penicillium seriemae* (DTO 515-H3). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 µm.

Etymology. Latin, seriemae: named, in reference to the seriema (*Cariama cristata*), a bird

species typical of the Brazilian Cerrado biome, symbolising the native fauna.

Diagnosis: Exduate clear appearance on most media. Conidiophores monoverticillate; stipes occasionally rough-walled, vesiculate; phialides 7–18 per stipe; conidia finely roughened to rough walled, globose to subglobose, 2.2–3 × 2–2.8 μm.

Colony morphology (7 d, in mm): CREA 12–14; CYA (32) 26–28; CYA 30°C (10) 17–18; CYA 37°C no growth; CYAS (17) 22–24; DG18 26–29; MEA 31–32; OA 24–25; YES 34–35.

Culture characteristics: *CYA 25 °C, 7 d*: Colonies moderately deep, slightly crateriform at center, radially sulcate, margins low, entire; mycelium white; colony texture velvety; sporulation dense; conidia *en masse* greyish yellow to greyish turquoise (24D5-24E5); exudate clear; soluble pigments inconspicuous orange; reverse greyish yellow to brownish orange (4B3-5C3); *CYA 30 °C, 7 d*: Colonies moderately deep, slightly crateriform at center, randomly sulcate, margins low, entire; mycelium white; colony texture velvety and floccose at center; sporulation poor to moderate; conidia *en masse* light turquoise to greyish turquoise (23A4-24E5); exudate clear; soluble pigments inconspicuous orange; reverse orange grey to brownish orange (5B2-5C4); *CYAS 25 °C, 7 d*: Colonies moderately deep, slightly raised at center, randomly sulcate, margins low, entire; mycelium white; colony texture velvety; sporulation dense; conidia *en masse* greyish turquoise (24D4); exudate absent; soluble pigments inconspicuous orange; reverse greyish yellow (4B3); *MEA 25 °C, 7 d*: Colonies moderately deep, radially and concentrically sulcate, crateriform; margins low, wide, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish turquoise (24E4-24E6); exudates clear; soluble pigments absent; reverse yellowish orange to brownish orange (4B7-5C6); *YES 25 °C, 7 d*: Colonies moderately deep, raised at the center, randomly sulcate, margins low, narrow, entire; mycelium white; colony texture velvety and floccose at center; sporulation dense; conidia *en masse* dull green to greyish yellow (25E4-25E5); exudates absent; soluble pigments absent; deep yellow to orange red (4B5); *DG18 25 °C, 7 d*: Colonies plane, slightly raised at the center, slightly radially sulcate at center, margin low, entire; mycelium white; colony texture velvety; sporulation dense; conidia *en masse* greyish green (25E5); exudates absent; soluble pigments absent; reverse greenish grey (27B2); *OA 25 °C, 7 d*: Colonies plane, margin low, entire; mycelium white; colony texture velvety; sporulation dense; conidia *en masse* greyish green (25E5); exudates clear; soluble pigments inconspicuous brown; *CREA 25 °C, 7 d*: moderate growth, acid production present, base formation absent.

Micromorphology: Conidiophores strictly monoverticillate; stipes occasionally rough-walled,

20–133 × 1.5–2.2 µm, vesicle (3–4± 0.3); phialides occasionally rough walled, ampulliform, 7–18 per stipe, 5.7–16 × 2.2–3 µm (8.0 ± 0.9 × 2.6 ± 0.2); conidia finely roughened to rough walled, globose to subglobose, 2.2–3 × 2–2.8 (3.0 ± 0.2 × 2.4 ± 0.2), n = 35.

Typus: **Brazil**, Minas Gerais, Patrocinio, 18°47'17"S 46°57'09"W, 965 m a.s.l., from soil native Cerrado, in February. 2023, *C.N. Figueiredo* (**holotype** DTO 515-H3 ex-type culture URMICRO 11873 = MPe257).

Notes: Penicillium seriemae phylogenetically belongs to section *Guizhouorum* (Zhang et al., 2024) and like another member of this section, strictly produces monoverticillate conidiophores and conidias in moderately long chains. *P. seriemae* is phylogenetically closely related to *Penicillium guizhouense*, but can be distinguished by the latter's ability to produce larger vesicles (10 µm in diameter) and conidia measuring 4–6 × 3–5.5 µm. Based on a megablast search of NCBIs GenBank nucleotide database, the closest hits using the ITS sequence are *Penicillium hirayamae* (strain CBS 238.65, GenBank MH858553; Identities 841/871 (97%), 9 gaps), *Penicillium adametzii* (strain CN014G1, GenBank OR164452; Identities 844/876 (96%), nine gaps) and *Penicillium senticosum* (strain CBS 329.71, GenBank MH860152; Identities 839/869 (97%), 7 gaps). The closet hits using the CaM sequence are *Penicillium guizhouense* (strain CGMCC 3.25508, GenBank OR828450); Identities 452/505 (90%), six gaps), *Penicillium* sp. (strain XM1-7-2, GenBank ON193185); Identities 284/329 (86%), 16 gaps), *Penicillium yezoense* (strain KMM 5004, GenBank PP446172; Identities 284/329 (86%), 15 gaps). The closest hits using the rpb2 sequence are *Penicillium guizhouense* (strain ZY 22.035, GenBank OR842931; Identities 982/1030 (95%), no gaps), *Penicillium* sp. (strain LS-2020a, GenBank MN627763; Identities 982/1031 (95%), no gaps) and *Penicillium glabrum* (strain SZ-22-12/3, GenBank OR664141; Identities 892/1049 (85%), 6 gaps). The closest hits using the BenA sequence are *Penicillium* sp. (strain UFMGCB 12385, GenBank MN045350; Identities 371/403 (92%), 3 gaps), *Penicillium* sp. (strain UFMGCB 12429, GenBank MN045352; Identities 371/403 (92%), 3 gaps), and *P. guizhouense* (strain ZY 22.035, GenBank OR843219; Identities 383/422 (91%), 3 gaps).

Penicillium moreirae Figueiredo, Zhou, Batista, Houbraken, *sp. nov*. Fig. 13.

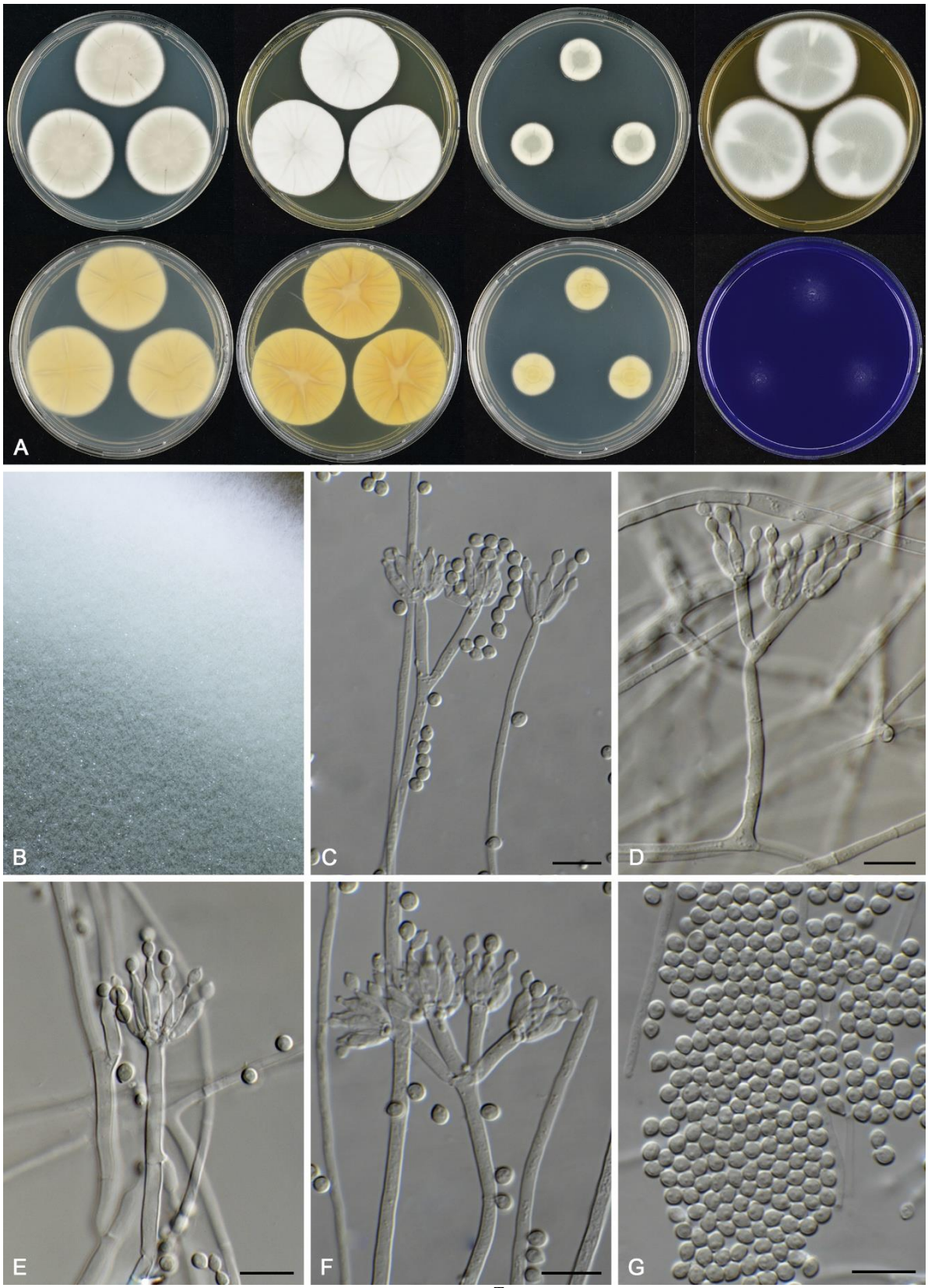


Figure 13. *Penicillium moreirae* (DTO 513-C3^{T}). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 μm.

Etymology. Latin, moreirae: in reference to Professor Fátima Maria de Souza Moreira (Federal University of Lavras, Minas Gerais, Brazil), for her remarkable contributions to soil microbiology, microbial ecology, and biodiversity, and for her distinguished scientific career dedicated to advancing soil science nationally and internationally.

Diagnosis: Textura veolutinous dominate colony appearance on most media. Conidiophores monoverticillate and biverticillate; stipes smooth to slightly rough walled, measuring 23–360 (–537) × 1.5–3.0 µm; phialides ampulliform 6.7–14 × 2.0–3.0 µm; conidia smooth walled, subglobose to apiculate, occasionally fusiform, 2.7–3.6 × 2.1–2.8 µm.

Colony morphology (7 d, in mm): CREA 22–34; CYA (31) 39–43; CYA 30°C (19) 32–38; CYA 37°C no growth; CYAS 17–24; DG18 19–20; MEA 38–49; OA 40–50; YES 40–47.

Culture characteristics: *CYA 25 °C, 7 d*: Colonies moderately deep, radially and concentrically sulcate; margins low, narrow, entire; mycelia white; texture velutinous; sporulation poor to moderate, conidia *en masse* turquoise grey (24C2) at center and greenish grey (25B2) in the margin; soluble pigments absent; exudates clear, sometimes absent; reverse greyish yellow to brownish orange (4B3-5C3); *CYAS 25 °C, 7 d*: Colonies raised at center, radially and randomly sulcate; margins low, narrow, entire; mycelium white; colony texture velutinous; poor sporulation; conidia *en masse* greyish white to greyish green (25B1-25C3); exudates clear, sometimes present; soluble pigments absent; reverse pale yellow to light yellow (4A3-4A4), some strains light brown (5D5); *MEA 25 °C, 7 d*: Colonies moderately deep, slightly radially sulcate, margins low, wide, entire; mycelium white; colony texture velutinous; sporulation poor to moderate; conidia *en masse* greenish grey (26C2), baby blue (23B3) at center and dull yellow (3B3) in the margin, some strains greyish turquoise (24C3) at center and greyish green (25B3) in the margin; sometimes exudates clear; soluble pigments absent; reverse yellow ochre (5C7) at center and amber yellow (4A6) in the margin; *YES 25 °C, 7 d*: Colonies moderately deep, radially and randomly sulcate, margins low, entire; mycelium white; colony texture velutinous; sporulation poor, sometimes absent; conidia *en masse* baby blue to greyish blue (23B3-23C4), some strains pastel blue (23A4); exudates sometimes clear; soluble pigments absent; reverse greyish yellow butter yellow to greyish yellow (4A5-4B6); *DG18 25 °C, 7 d*: Colonies moderately deep, radially and concentrically sulcate, margins low, entire; mycelium white; colony texture velutinous; sporulation moderate; conidia *en masse* aquamarine greyish green (24B3-25C4); exudates absent; soluble pigments absent; reverse greyish yellow (4C3) at center and yellowish grey (4B2) in the margin, some strains champagne (4A4); *OA 25 °C, 7 d*: Colonies plane, margins low, fimbriate; mycelium white; colony texture velutinous; sporulation poor to moderate,

sometimes absent; conidia *en masse* greyish yellow to greenish grey (4C3-28C2); exudates absent; soluble pigments sometimes present orange; *CREA 25 °C, 7 d*: good growth, acid production absent, base formation absent.

Micromorphology: Conidiophores monoverticillate and biverticillate; stipes smooth to slightly rough walled, 23–360 (–537) × 1.5–3.0 µm; metulae cylindrical, 2–4 per stipe, 7–28 × 1.8–3.0 µm; phialides ampulliform, 2–12 per metulae, 6.7–14 × 2.0–3.0 µm (9.1 ± 1.7 × 2.5 ± 0.2); conidia smooth walled, subglobose to apiculate, occasionally fusiform, 2.7–3.6 × 2.1–2.8 (3.0 ± 0.2 × 2.5 ± 0.2), n = 38.

Typus: **Brazil**, Minas Gerais, Patrocinio, 18°46'58"S 46°56'47"W, 965 m a.s.l., from soil Cerrado, in March, 2023, *C.N. Figueiredo* (**holotype** DTO 513-C3 ex-type URMICRO 11781 = CR100).

Notes: According to multigene phylogenies, *Penicillium moreirae* is phylogenetically related to *Penicillium reverso-vinaceum* and *Penicillium beiraoi*. These species also differ morphologically. *P. moreirae* has subglobose to apiculate conidia, occasionally fusiform and smaller, whereas *Penicillium beiraoi* has larger and strictly fusiform conidia. On the other hand, *P. moreirae* no producer of sclerotia and no growth on CYA at 37 °C, compared to the subglobose, ellipsoidal, or irregular sclerotia and growth on CYA at 37 °C of *P. reverso-vinaceum.* Based on a megablast search of NCBIs GenBank nucleotide database, the closest hits using the ITS sequence are *Penicillium soli* (strain CMV004H8, GenBank MK450683; Identities 862/866 (99%), 3 gaps), *Penicillium ortum* (strain CMV007H9, GenBank MK450708; Identities 858/861 (99%), two gaps) and *Penicillium koreense* (strain CMV006G6, GenBank MK450699; 859/863 (99%), two gaps). The closet hits using the CaM sequence are *Penicillium reverso-vinaceum* (strain URM 8967, GenBank PP806635); Identities 542/560 (97%), no gaps), *Penicillium* sp. (strain CS27-03, GenBank OR051297); Identities 542/560 (97%), two gaps), *Penicillium raperi* (strain CMV006I1, GenBank MK451642; Identities 543/562 (97%), four gaps). The closest hits using the rpb2 sequence are *Penicillium koreense* (strain DTO 347-C1, GenBank MN969159; Identities 975/998 (98%), no gaps), *P. koreense* (strain CMV006G6, GenBank MK450842; Identities 973/997 (98%), no gaps) and *Penicillium raperi* (strain CMV006H9, GenBank MK450850; Identities 971/999 (97%), no gaps). The closest hits using the BenA sequence are *Penicillium reverso-vinaceum* (strain URM 9078, GenBank PP977222; Identities 473/491(96%), no gaps), *Penicillium sp*. ES-2024a (strain VTT D-231709, GenBank PQ252672; Identities 473/491(96%), 1 gap), and *Penicillium donggangicum* (strain AS3.15900, GenBank MZ004914; Identities 459/473 (97%), 1 gap).

Penicillium beraoi Figueiredo, Zhou, Batista, Houbraken, *sp. nov*. Fig. 14.

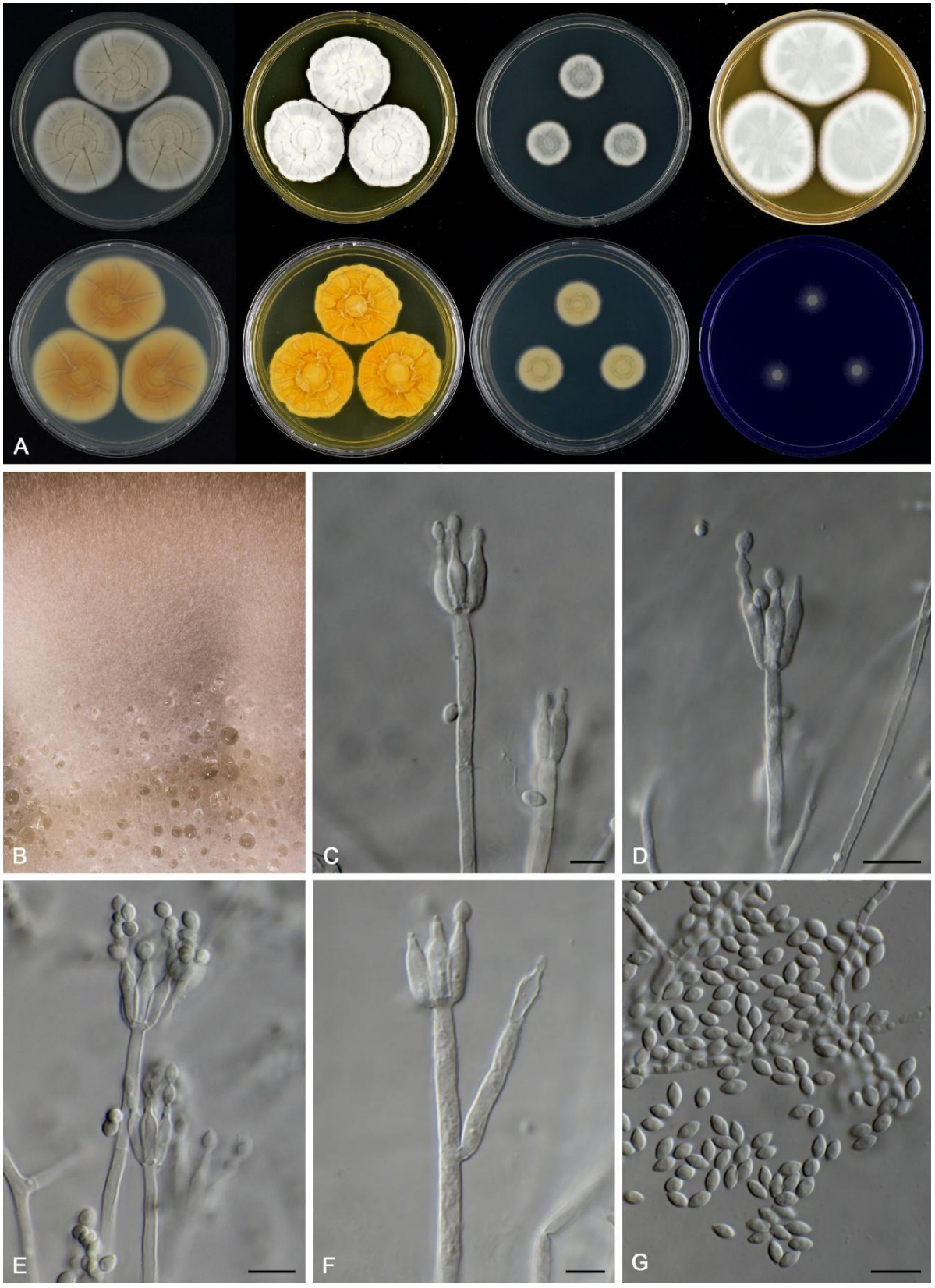


Figure 14. *Penicillium beraoi* (DTO 513-C2^{T}). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 μm.

Etymology. Latin, beraoi: named, in reference to the Professor and researcher Paulo Sérgio Lacerda Beirão, doctor and science administrator, in recognition of his contributions to science and research in Brazil, especially during his presidency of the Research Support Foundation of the State of Minas Gerais (FAPEMIG) between 2020–2023 and his support for projects aimed at combating the COVID-19 pandemic.

Diagnosis: *P. breaoi* is characterized by conidia coloration varies from gray to greenish gray and turquoise. Conidiophores monoverticillate, occasionally biverticillate; stipes smooth walled, occasionally slightly rough-walled, 34–316 × 2–3.5 µm; phialides ampulliform, 7.8–15 × 2.5–3.5 µm; conidia smooth walled, fusiform, 3–4.6 × 2–3 µm.

Colony morphology (7 d, in mm): CREA 21-35; CYA 34–42; CYA 30°C 29–30; CYA 37°C no growth; CYAS 20-28; DG18 18-22; MEA 30-48; OA 46-56; YES 27-49.

Culture characteristics: CYA 25 °C, 7 d: Colonies moderately deep, sometimes slightly raised at center radially and concentrically sulcate; margins low, narrow, entire; mycelia white; texture velutinous to floccose; sporulation poor to moderate, conidia en masse greyish turquoise to greyish green (24C4-25C2) at center and greenish grey (25B2) in the margin; soluble pigments absent; exudates sometimes clear; reverse greyish yellow to brownish orange (4B3-5B5), sometimes topaz to grayish orange (5C5-6B6); CYAS 25 °C, 7 d: Colonies moderately deep, radially and concentrically sulcate, sometimes randomly and slightly raised at center; margins low, narrow, fimbriate; mycelium white; colony texture velutinous; poor sporulation; conidia en masse grey (2B1); exudates absent; soluble pigments absent; reverse greyish yellow (4B3-4B4) to greyish orange (5B4); MEA 25 °C, 7 d: Colonies moderately deep, radially and concentrically sulcate, margins low, lobade; mycelium white; colony texture velutinous to floccose; sporulation poor to dense; conidia en masse grey to greyish green (2B1-25C3); sometimes exudates clear; soluble pigments absent; reverse yellowish brown (5D8) at center and light yellow (4A5) in the margin; YES 25 °C, 7 d: Colonies moderately deep, randomly sulcate, sometimes crateriform; margins low, entire, sometimes irregular; mycelium white; colony texture velutinous; sporulation poor; conidia en masse grey to turquoise white (2B1-24A2) and sometimes greenish grey (25B2); exudates absent; soluble pigments absent; reverse greyish yellow to topaz (4B6-5C6); DG18 25 °C, 7 d: Colonies moderately deep, radially and concentrically, margins low, entire; mycelium white; colony texture velutinous to floccose; sporulation poor to moderate; conidia en masse light yellow to greenish grey (2A5-26C2); exudates absent; soluble pigments absent; reverse dull yellow (3B3-3B4) and sometimes light yellow (3A5) at center and pale yellow (3A3) in

the periphery; OA 25 °C, 7 d: Colonies plane, margins low, fimbriate; mycelium white; colony texture velutinous to floccose; sporulation poor to moderate; conidia en masse greenish grey (26B2-27B2); exudates absent; soluble pigments absent; CREA 25 °C, 7 d: moderate growth, acid production absent, base formation absent.

Micromorphology: Conidiophores monoverticillate, occasionally biverticillate; stipes smooth walled, occasionally slightly rough-walled, 34–316 × 2–3.5 µm; phialides ampulliform, 2–9 per stipe, 7.8–15 × 2.5–3.5 µm, (10.5 ± 1.2 × 2.9 ± 0.2); conidia smooth walled, fusiform, 3–4.6 × 2–3 µm, (3.8 ± 0.4 × 2.3 ± 0.3), n = 40.

Typus: **Brazil**, Minas Gerais, Patrocinio, 18°51'28"S 47°07'18"W, 965 m a.s.l., from soil Cerrado, in February, 2023, *C.N. Figueiredo* (**holotype** DTO 513-C2 ex-type culture URMICRO 11778 = 220).

Notes: Penicillium beraoi is characterized by fusiform conidia and clear exudates at 25°C CYA. Clear exudates also occur in *P. moreirae*. *P. beraoi* was distinguished from *P. moreirae* based on more abundant conidiogenesis in YES, shorter stipes, and colony size in CYA at 30°C. Compared to *Penicillium reverso-vinaceum*, *P. beraoi* does not produce sclerotia and does not exhibit the dark vinaceous, characteristics typical of the species *P. reverso-vinaceum*. Based on a megablast search of NCBIs GenBank nucleotide database, the closest hits using the ITS sequence are *Penicillium* sp. (strain 28 BRO-2013, GenBank KF367540; Identities 879/881 (99%), one gap), *Penicillium janthinellum* (strain F-13, GenBank AB293968; Identities 879/884 (99%), three gaps) and *Penicillium soli* (strain CMV004H8, GenBank MK450683; Identities 862/863 (99%), no gaps). The closet hits using the rpb2 sequence are *Penicillium koreense* (strain CMV006G6, GenBank MK450842); Identities 1029/1049 (98%), no gaps), *Penicillium raperi* (strain CMV006H9, GenBank MK450850); Identities 1027/1049(98%), no gaps), *P. raperi* (strain CT105, GenBank PV791183; Identities 1023/1049 (98%), no gaps). The closest hits using the *CaM* sequence are *P. reverso-vinaceum* (strain URM 8967, GenBank PP806635; Identities 536/562 (95%), no gaps), *Penicillium raperi* (strain CMV006H9, GenBank MK451641; Identities 535/562 (95%), two gaps) and *Penicillium tengii* (strain CS27-03, GenBank OR051297; Identities 534/561 (95%), two gaps). The closest hits using the *BenA* sequence are *Penicillium* sp (strain 54E, GenBank MT732931; Identities 488/491 (99%), no gaps), *Penicillium* sp. (strain C2281B, GenBank PP915710; Identities 481/489 (98%), gaps) and *P. reverso-vinaceum* (strain URM 9078, GenBank PP977222; Identities 479/491 (98%), no gaps).

The species descriptions for the occurrence records were compared with the original description of each species. The isolates from Cerrado DTO 513-F9, DTO 514-D4 and DTO 517-A3 (Fig. 15) and the type isolate of the species *P. guizhouense* have most of the similar characteristics, except for the number of phialides per stip, which the isolates presented slightly higher numbers.

Strain DTO 510-F2 formed a monophyletic clade with *P. menonorum* (Fig. 5). The morphological characteristics of strain DTO 510-F2 (Figure 16) are similar to those of the type isolate of the species *P. menonorum*, except for growth in CYA at 37°C, where the type strain of the species exhibits high growth rates, and isolate DTO 510-F2 does not grow at this temperature.

DTO 515-A2 forms a monophyletic clade with the species *P. dabashanicum* (Fig. 6). The results of morphological analyses comparing strain DTO 515-A2 (figure 17) with the original description for *P. dabashanicum* show that most morphological characteristics are similar; however, the type isolate of the species exhibits slightly larger stipes and strictly terverticillate and quaterverticillate conidiophores, in addition to a lower growth rate in culture medium. The strain DTO 514-E8 (figure 18) of the *P. daejeonium* species also has morphological characteristics similar to the type isolate of the species, such as acid production in CREA and microscopic characteristics, with the exception of growth in CYA at 30°C of isolate DTO 514-E8, whereas the type isolate of the species does not grow.

Penicillium guizhouense Zhi Y. Zhang & Y. F. Han, Mycosphere, 2024. (Figure 15)

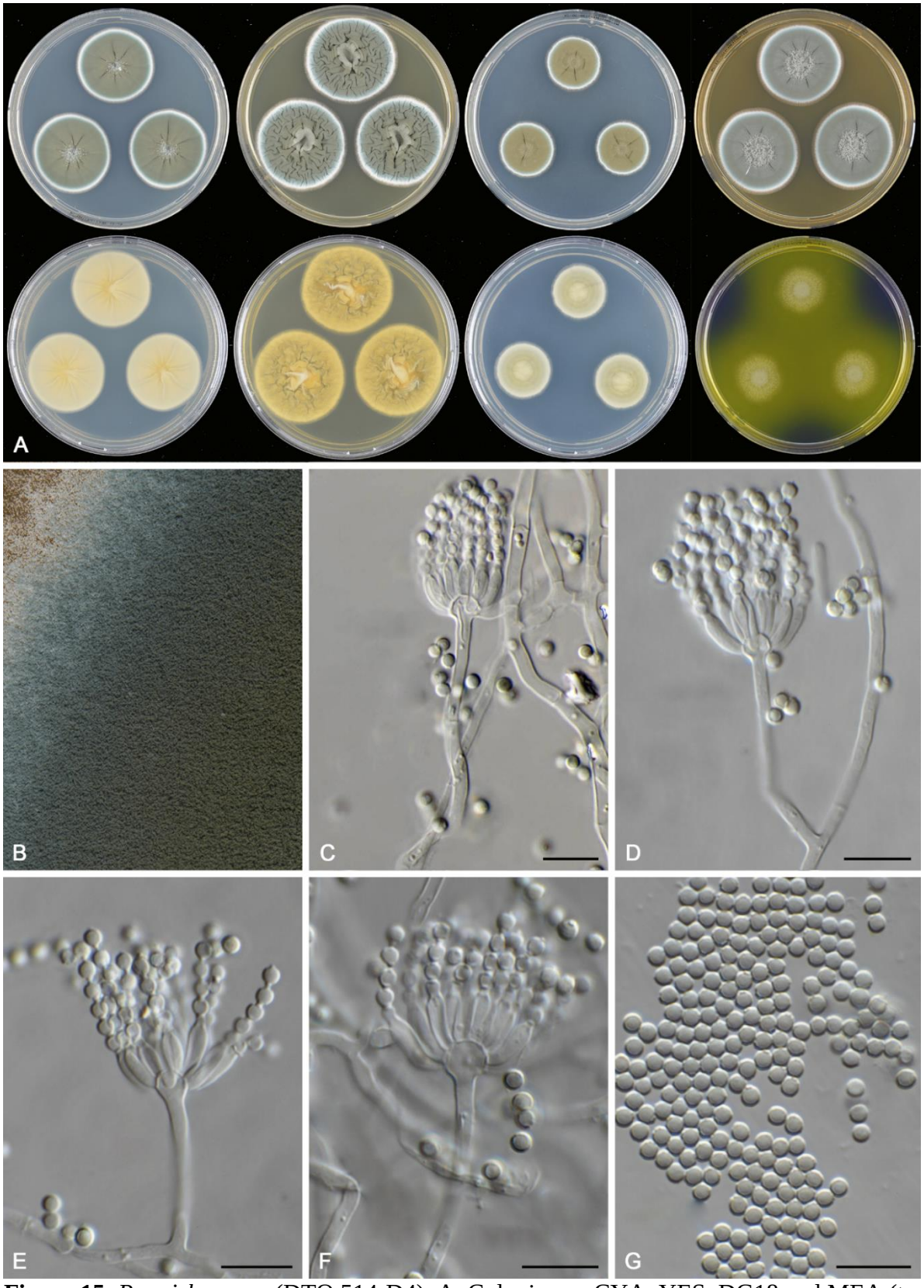


Figure 15. *P. guizhouense* (DTO 514-D4). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 μm.

Culture characteristics: CYA 25 °C, 7 d: colonies moderately deep, radially sulcate; margins low, narrow, entire; mycelia white; texture velutinous; sporulation dense, conidia *en masse* greyish green (28E6); exudates and soluble pigments absent; reverse yellowish grey to greyish yellow (3C2-3C5); *MEA 25 °C, 7 d*: colonies moderately deep, radially sulcate margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish green (26D5); exudates and soluble pigments absent; reverse olive brown (4D7-4D8); *YES 25 °C, 7 d*: Colonies crateriform, irregularly sulcate. margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish green (28E5); exudates absent and soluble pigments absent; reverse light yellow (2A5); *DG18 25 °C, 7 d*: colonies moderately deep, radially and concentrically sulcate, margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish grey (28E5); exudates and soluble pigments absent; reverse greyish yellow (2B6); *CREA 25 °C, 7 d*: moderate growth, acid production present, base formation absent.

Penicillium menonorum S.W. Peterson, IMA Fungus, 2011. (Figure 16)

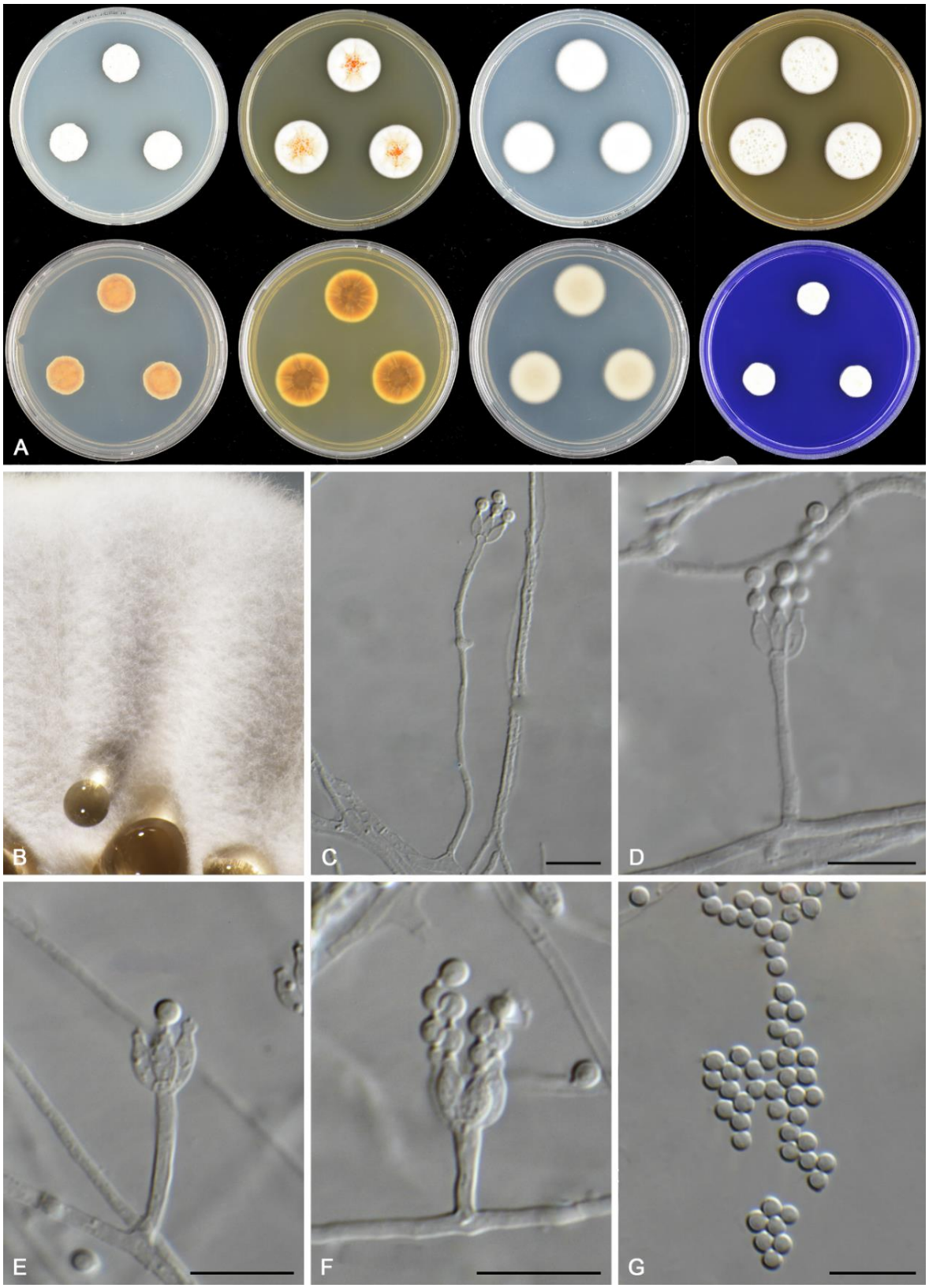


Figure 16. *Penicillium menonorum* (DTO 510-F2). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 µm.

Colony morphology: CREA 14; CYA 17–18; CYA 30°C 9–11; CYA 37°C no growth; CYAS 12-21; DG18 23-24; MEA 30-31; OA 27; YES 25-26.

Colony characters: CYA 25 °C, 7 d: Colonies raised at center, radially sulcate, margins low, narrow, entire; mycelia white; texture velutinous; sporulation absent; soluble pigments absent; exudates absent; reverse greyish orange (5B4);; *MEA 25 °C, 7 d*: colonies slightly raised at the center, radially sulcate, margin entire; mycelium white; colony texture floccose; sporulation poor; conidia en masse greenish white (25A2); exudates yellow; soluble pigments absent; reverse light brown (6D6) at center and greyish orange (6B3) in the periphery; *YES 25 °C, 7 d*: Colonies moderately deep, irregulary and radially sulcate, margin entire; mycelium white; colony texture floccose; sporulation absent; exudates sometimes present light orange; soluble pigments sometimes present light orange; reverse greyish yellow to orange (4B5-6A6); *DG18 25 °C, 7 d:* colonies slightly raised at the center and slightly radially sulcate, margin entire; mycelium white; colony texture floccose; sporulation moderate; conidia en masse olive to pale blue (23A3); exudates absent; soluble pigments absent; reverse greyish yellow (4A3-4B4; *CREA 25 °C, 7 d:* moderate growth, acid production absent, base formation absent.

Micromorphology: conidiophores strictly monoverticillate; stipes smooth to slightly rough walled, 6–51 × 1.3–2 µm; phialides ampulliform, 2–4 per stipe, 4–7 × 1.8–2.7 µm (5 ± 0.6 × 2.3 ± 0.2); conidia smooth to slightly rough valled, globose to subglobose, 2–2.6 × 1.8–2.6 µm (2.2 ± 0.1 × 2.1 ± 0.1), n = 40.

Penicillium dabashanicum X.C. Wang & W.Y. Zhuang, Journal of fungi, 2023. (Figure 17)

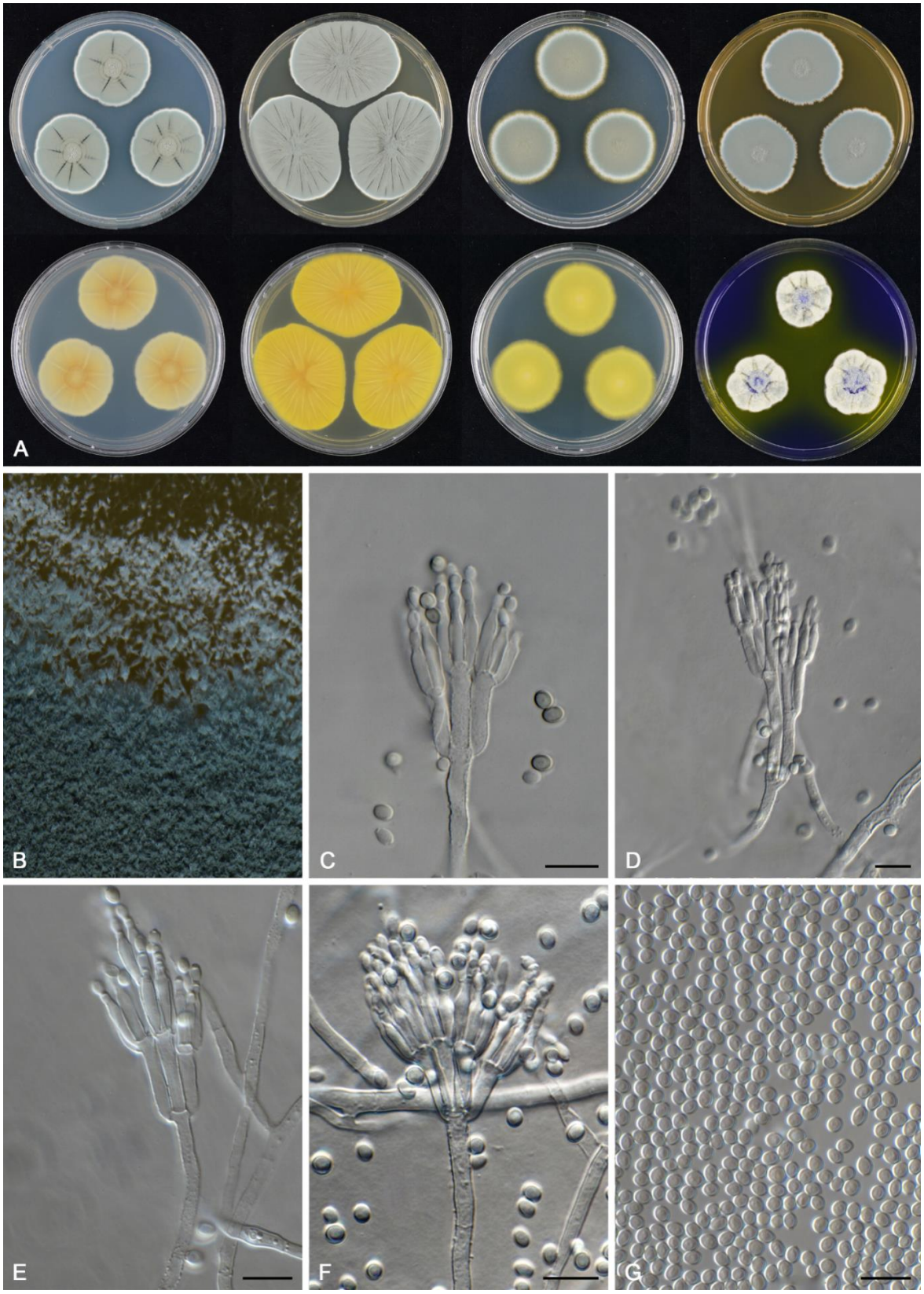


Figure 17. *Penicillium dabashanicum* (DTO 515-A2). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 µm.

Colony morphology (7 d, in mm): CREA 26–28; CYA 34–35; CYA 30°C 21–25; CYA 37°C no growth; CYAS 33–34; DG18 31–36; MEA 35–38; OA 32–37; YES 49–51.

Colony characters: *CYA 25 °C, 7 d*: Colonies moderately deep, radially and concentrically sulcate; margins low, narrow, entire; mycelia white; texture velutinous; sporulation dense, conidia *en masse* dull green (25D4); exudates clear sometimes; soluble pigments absent; reverse dull yellow to mustard yellow (3B3-3B6), sometimes caramel at center (6C6) and wax white (2B3) in the periphery; *MEA 25 °C, 7 d*: Colonies low, plane, sometimes slightly raised at center; margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* dull green (25D4); exudates absent; soluble pigments absent; reverse yellowish orange to yolk yellow (4B7-4B8); *YES 25 °C, 7 d*: Colonies moderately deep, slightly raised at center, radially sulcate; margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation dense; conidia *en masse* greyish green to dull green (25C3-25D4); exudates absent; soluble pigments absent; reverse naples yellow to lemon (3B7-3B8); *DG18 25 °C, 7 d*: Colonies plane, slightly raised at the center; margins low, narrow, entire; mycelium white, sometimes yellow; colony texture velutinous; sporulation dense; conidia *en masse* dull yellow to greyish green (25D4-25D5); exudates absent; soluble pigments absent; reverse olive yellow (2C6); *CREA 25 °C, 7 d*: good growth, acid production present, base formation present at center of the colony.

Micromorphology: conidiophores biverticillate, occasionally monoverticillate and terverticillate; stipes occasionally rough-walled, 34–150 (–246) × 3.4–4.8 µm; metulae cylindrical, 2–5 per stipe, 11.7–22 × 3–5 µm; phialides ampulliform, 8–15 × 2.8–4 µm (10.7 ± 1.5 × 3.3 ± 0.3); conidia smooth walled, globose to ellipsoidal, 3.7–5 × 2.5–4 µm (4.1 ± 0.3 × 3.2 ± 0.2).

Penicillium daejeonium S.H. Yu & H.-K. Sang, Journal of Microbiology, 2013. (Fig. 18)

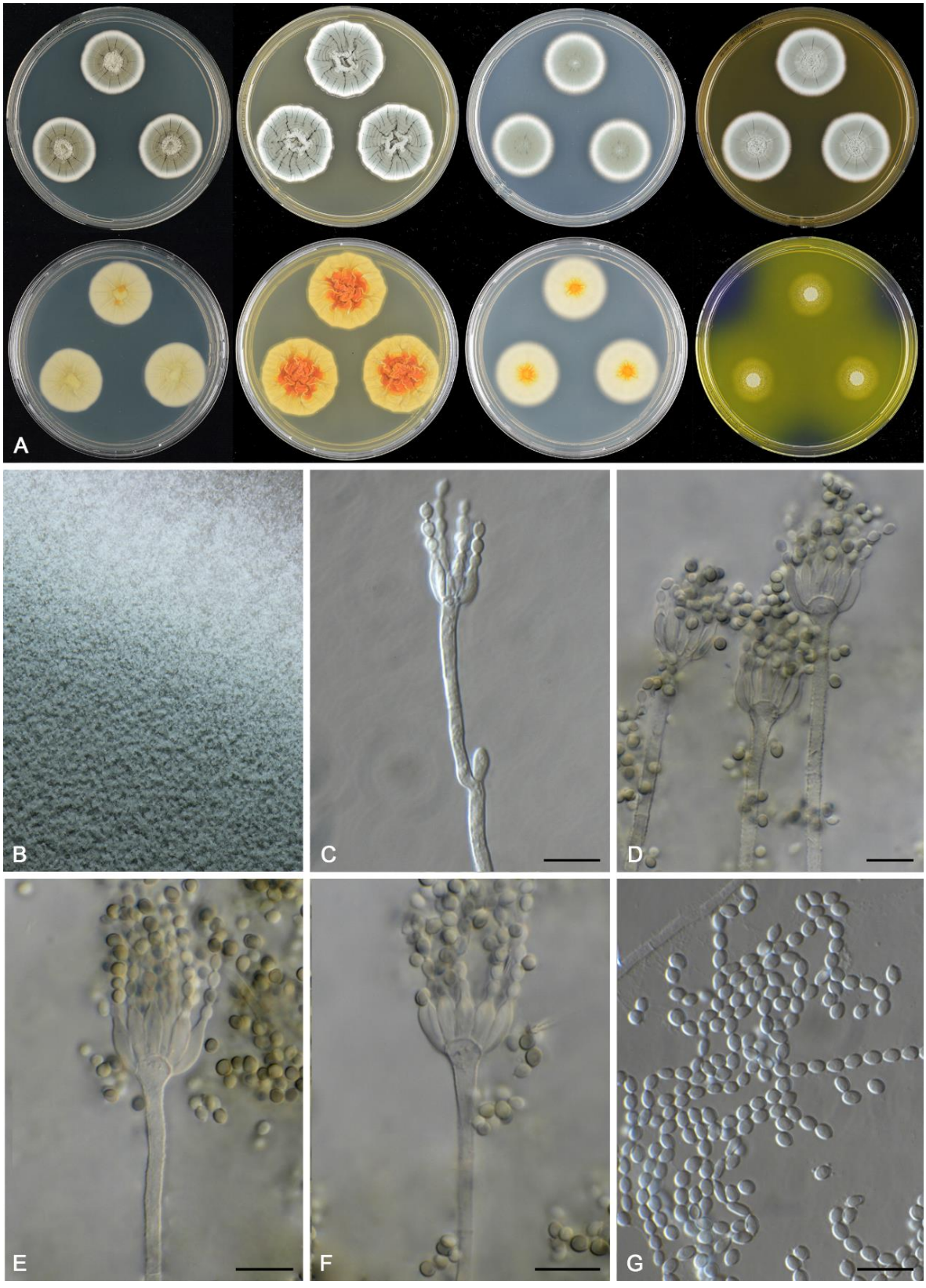


Figure 18. *Penicillium daejeonium* (DTO 514-E8). A. Colonies on CYA, YES, DG18 and MEA (top row = obverse) and CYA, YES, DG18 (bottom row = reverse) and CREA (bottom row = obverse) from left to right; b. texture on MEA; c–f. conidiophores; g. conidia. — Scale bar in = 10 µm.

Colony morphology (7 d, in mm CREA 12; CYA 24; CYA 30°C 19-20; CYA 37°C no growth; CYAS 12-13; DG18 14-16; MEA 24; OA 20; YES 21.

Colony characters: CYA 25 °C, 7 d: Colonies moderately deep, radially and concentrically sulcate, crateriform; margins low, narrow, entire; mycelia white; texture velutinous; sporulation poor, conidia en masse white (3A1); soluble pigments absent; exudates clear; reverse yellowish white (3A2); *MEA* 25 °C, 7 d: Colonies moderately deep, radially sulcate, margins low, narrow, entire; mycelium white; colony texture velutinous; sporulation moderate; conidia en masse greyish turquoise (24B3); exudates clear; soluble pigments absent; reverse greyish yellow (4B6); *YES* 25 °C, 7 d: Colonies raised at center, radially sulcate, margin entire; mycelium white; colony texture velutinous; sporulation poor; conidia en masse grey (3B1); exudates absent; soluble pigments absent; reverse yellow (3A6); *DG18* 25 °C, 7 d: Colonies moderately deep, radially sulcate; margins wide, entire; mycelium white; colony texture velutinous; sporulation moderate; conidia en masse dull green (26E3); exudates absent; soluble pigments absent; pale yellow (2A3); *CREA* 25 °C, 7 d: poor growth, acid production absent, base formation absent.

Micromorphology: conidiophores monoverticillate, occasionally biverticillate; stipes smooth, occasionally rough-walled, 27–136 (–235) × 2.2–3.3 µm, vesicle 5–7 µm (6.1 ± 0.4); phialides ampulliform, 3–19 per stipe, 6–12.5 × 2.5–3.6 µm (8.9 ± 1.2 × 3.0 ± 0.3); conidia smooth walled,

ellipsoidal to subglobose, hyaline to greyish, 2.7–3.5 × 2–2.6 µm (3.0 ± 0.2 × 2.4 ± 0.1), n = 30.

4. DISCUSSION

This study represents one of the few investigations that describe new *Penicillium* species from the Brazilian Cerrado. Species from the phyla *Ascomycota* and *Basidiomycota* are dominant in this biome, representing more than 70% of the total fungal community in its soil (Ferreita et al., 2017). Among the members of the genus *Ascomycota*, the genus *Penicillim* is frequently found and considered one of the most abundant in the brazilian Cerrado (Monteiro et al., 2016; Silva et al., 2021).

The genus *Penicillium* is known for its worldwide distribution and is found in different substrates, including soil (Visagie et al., 2014). In this substrate, *Penicillium* species act mainly in the decomposition of organic matter. The Cerrado has a seasonal tropical climate, with dry winters and an average annual temperature between 22 and 23 °C, with highs

exceeding 40 °C. The soils are characterized as acidic and poor in nutrients, such as phosphorus (Lira-Martins et al., 2022; Correia Filho et al., 2023). Species of the genus *Penicillium* produce a large number of enzymes and have a high capacity to adapt to diverse abiotic conditions, such as those found in the Cerrado soil. Among the enzymes produced, many species of this genus have already been reported as producers of phosphatases, which act to break down insoluble phosphorus into simpler, more soluble forms of phosphorus (Asea, Kucey & Stewart, 1998; Qiao et al., 2019; Mercl et al., 2020; Suraby et al., 2023). Thus, these enzymes increase the availability of phosphorus for plants and other microorganisms present in the soil.

Other *Penicillium* species are known to be acid-tolerant, members of the section *Lanata-divaricata*, described in 2021, were isolated from acidic soils (pH 4–6.5) of tropical and subtropical regions of China (Diao et al., 2018). These acidic conditions are similar to those of the Brazilian Cerrado soils, where five species of the section *Lanata-divaricata* were found in this study, two of which represent new species, *P. beraoi* and *P. moreirae.*

The species *P. guizhouense*, section *Guizhouorum*, and *P. dabashanicum*, section *Fasciculata*, recently described and isolated from soils in regions of China, were also found in native Cerrado soils in Brazil, expanding the knowledge of the geographic distribution of these taxa (Wang, Zhang & Zhuang, 2023; Zhang et al., 2024). Other species recorded in the Brazilian Cerrado include *P. menonorum* and *P. daejeonium*, belonging to the sections *Exilicaulis* and *Sclerotiorum*, respectively. Sect. *Exilicaulis* is a taxonomically diverse group currently containing 76 species (Visagie et al., 2016). *P. menonorum* was isolated from garden soil in California, USA (Peterson, Orchard & Menon, 2011)

The section *Sclerotiorum* was introduced in 2011 and currently comprises 39 species. The species of this section are known for being frequently found in soils, decomposing organic matter, and fruits (Houbraken & Samson, 2011). The type isolate of *P. daejeonium* was isolated from grape fruits, whereas DTO514-E8 was isolated from soil of native Cerrado. Other species of genus *Penicillium*, section *Sclerotiorum*, including new taxa, have already been isolated from different substrates and biomes in Brazil, including soils from restinga and Caatinga, highlighting the broad ecological and adaptive diversity of this fungal group. (Crous et al. 2014, Barbosa et al. 2016, Barbosa et al. 2018, Crous et al. 2018, Ramos et al. 2021, Barbosa et al. 2022).

Morphological differences were observed between fungal strains isolated from the Brazilian Cerrado and the type isolates of the species *P. guizhouense*, *P. dabashanicum*, *P. menonorum* and *P. daejeonium*. Although the type isolates of these species were also obtained

from soil, as were the isolates from the Brazilian Cerrado, they originate from different countries, China, the United States and Korea, respectively. The climate and edaphic factors of these regions are distinct from the conditions found in the Brazilian Cerrado. Studies have shown that environmental factors, including variations in climate and edaphic characteristics, can significantly influence morphological traits within the same species, resulting in intraspecific variations (ZHANG et al. 2024).

Of the total of 156 isolates identified in this study (Table S1), 38 fungal strains originate from the native Cerrado, 11 of these isolates represent new species and six isolates represent occurrence records described in this work. Based on a megablast search of NCBIs GenBank nucleotide database, used *BenA* sequences, the remains of isolates were identified like occurrence record of species: *Aspergillus spelaeus, Talaromyces amestolkiae*, *Penicillium ubiquetum*, *Penicillium tanzanicum, Penicillium virgatum*, *Penicillium sizovae, Penicillium arabicum*, *Penicillium brevicompactum, Penicillium nothofagi*, *Penicillium dokdoense, Penicillium coprophilum, Penicillium westlingii* and *Penicillium koreense*. This species of genus *Penicillium* belong to sections *Brevicompacta, Citrina, Exilicaulis, Lanata-Divricata, Ramosum* and *Robsamsonia.* The *BenA* gene is considered the secondary marker for the genus *Penicillium*, since this locus presents sufficient variability to distinguish phylogenetically related species (Visagie et al. 2014).

Only one of the new species described in this study originates from regenerative coffee cultivation soil, *P. patrocinensis*. This study highlights the potential of the native Cerrado as a reservoir for novel taxa and its divesrity filametous fungi, underscoring the importance of preserving the native forests of this biome.

ACKNOWLEDGMENTS

This work was carried out with the support of the Coordination for the Improvement of Higher Education Personnel – Brazil (CAPES) – Funding Code 001. In addition, this work received financial support from the INCT Soil Biodiversity (CNPq #406658/2022-6 and FAPEMIG #APQ-04011-24) and the FAPEMIG Networks (#RED-00181-23 and RED-00330-16).

REFERENCES

Alves A, Santos A, et al. Diversity of filamentous fungi communities in the soils of agroecological crop polycultures and the Atlantic Rain Forest. Archives of Agronomy and Soil Science v. 69, n. 3, p. 374–386, 2021.

Asea, P.E.A., Kucey, R.M.N., Stewart, J.W.B. Inorganic phosphate solubilization by two *Penicillium* species in solution culture and soil. Soil Biology and Biochemistry, v. 20, n. 4, p. 459-464, 1988.

Barbosa RN, Bezerra JDP, Costa PMO et al (2016) Aspergillus and Penicillium (Eurotiales: Trichocomaceae) in soils of the Brazilian tropical dry forest: diversity in an area of environmental preservation. Rev Biol Trop 64:45–53

Barbosa, RN., Bezerra, JDP., Souza-Motta, CM., Frisvad, JC., Samson, RA., Oliveira, NT, Houbraken, J. New *Penicillium* and *Talaromyces* species from honey, pollen and nests of stingless bees. Antonie van Leeuwenhoek, Volume 111, pages 1883–1912, (2018).

Barbosa, RN; Felinto dos Santos, JE; Bezerra, JDP; ISTEL, L; Houbraken, J; Oliveira, NT; SOUZA-MOTTA, CM. Brazilian Atlantic Forest and Pampa Biomes in the spotlight: an overview of *Aspergillus, Penicillium*, and *Talaromyces* (Eurotiales) species and the description of *Penicillium nordestinense* sp. Nov. *Acta Botanica Brasilica*, v. 36, p. e2021abb0390, 2022. DOI: 10.1590/0102-33062021abb0390

Berrin J G, Navarro D, Couturier M, Olivé C, Grisel S, Haon M, Taussac S, Lechat C, Courtecuisse R, Favel A, Coutinho P M, Meessen L L. 2012. Exploring the Natural Fungal Biodiversity of Tropical and Temperate Forests toward Improvement of Biomass Conversion. Applied and Environmental Microbiology, v. 78, n. 18, p. 6483-6490, 2012.

Bhattarai A, Bhattarai B, Pandey S. 2015. Variation of soil microbial population in different soil horizons. J Microbiol Exp. 2(2):75–78.

Cleere M M, Novodvorska M, Geib E, Whittaker j, Dalton h, Salih n, Hewitt s, Kokolski m, Brock m, Dyer p s. 2024. New colours for old in the blue-cheese fungus *Penicillium roqueforti*. Npj Science of Food 8, 3 (2024). https://doi.org/10.1038/s41538-023-00244-9.

Crous PW, Shivas RG, Quaedvlieg W et al (2014) Fungal Planet description sheets: 214-280. Persoonia 32:184–306.

Crous PW, Wingfield MJ, Lombard L et al (2019) Fungal Planet description sheets: 951-1041. Persoonia 43:223–425.

Correia Filho, W.L.F., de Oliveira-Júnior, J.F., Santiago, Abdo, H.G., Almohamad, H., Dughairi, A.A, Junior, C.A. The assessment of climatic, environmental, and socioeconomic aspects of the Brazilian Cerrado. Ecological Processes, v. 12, n. 19, 2023. DOI https://doi.org/10.1186/s13717-023-00433-0

Magistà D.; Ferrara, M.A, Nobile, D.; Gammariello, D.; Conte, A. Perrone, G. *Penicillium 129ertil* strain ITEM 15302: A new promising fungal starter for salami production.

International Journal of Food Microbiology, Volume 231, 16 August 2016, Pages 33-41.

De Castro, A.P., Quirino, B.F., Pappas, G. et al. Diversity of soil fungal communities of Cerrado and its closely surrounding agriculture fields. Arch Microbiol 190, 129–139 (2008). https://doi.org/10.1007/s00203-008-0374-6.

Diao, Y.-Z., Chen, Q., Jiang, X.-Z., Houbraken, J., Barbosa, R.N., Cai, L., Wu, W.-P. *Penicillium* section *Lanata-divaricata* from acidic soil. Cladistics, v. 35, n. 5, p. 514-549, 2019.

Colli G, Vieira C, Dianese J. Biodiversity and conservation of the Cerrado: recent advances and old challenges. Biodiversity and Conservation, Volume 29, pages 1465–1475, (2020).

Ferreira de Araujo AS, Bezerra WM, dos Santos VM, et al. Fungal diversity in soils across a gradient of preserved Brazilian Cerrado. Microbial Ecology and Environmental Microbiology, v. 55, p. 273–279, 2017. Doi: 10.1007/s12275-017-6350-6.

Frisvad J C, R.A. Samson. Polyphasic taxonomy of Penicillium subgenus Penicillium. A guide to identification of food and air-borne terverticillate Penicillia and their mycotoxins. Stud. Mycol., 49 (2004), pp. 1-174.

Glass N L, Donaldson G C. Development of primer sets designed for use with the PCR to amplify conserved genes from flamentous ascomycetes. Appl Environ Microbiol. 1995, v. 61, n. 4 p. 1323-30.

Gonçalves M F M, Santos L, Silva B M V, Abreu A C, Vicente T F L, Esteves A C, Alves A. 2019. Biodiversity of *Penicillium* species from marine environments in Portugal and description of Penicillium lusitanum sp. Nov., a novel species isolated from sea water. Systematic and evolutionary microbiology 69:3014-3021. DOI: 10.1099/ijsem.0.003535

Hong S B, Cho H S, Shin H D, Frisvad J C, Samson R A. Novel Neosartorya species isolated from soil in Korea. International Journal Systematics and Evolutionary Microbiology, v. 56, p.477–86, 2006.

Houbraken J, J.C. Frisvad, R.A. Samson Fleming's penicillin producing strain is not Penicillium chrysogenum but P. rubens.IMA Fungus, 2 (2011), pp. 87-95.

Houbraken J, Samson RA. Phylogeny of *Penicillium* and the segregation of *Trichocomaceae* into three families. Studies in Mycology, v. 15, n. 70, p. 1-51, 2011. Doi: 10.3114/sim.2011.70.01. PMID: 22308045; PMCID: PMC3233907.

Houbraken J, S Kocsubé S, Visagie C, et al. 2020. Classification of Aspergillus, Penicillium, Talaromyces and related genera (Eurotiales): An overview of families, genera, subgenera, sections, series and species. Stud Mycol. 2020 Jun 27; 95:5–169.

Iftikhar, A., Farooq, R., Akhtar, M., Khalid, H., Hussain, N., Ali, Q., Malook, S., Ali, D. Ecological and sustainable implications of phosphorous-solubilizing microorganisms in soil. Discover Applied Sciences, v. 6, n. 33, 2024. DOI: https://doi.org/10.1007/s42452-024-05683-x

Jr N M, Sánchez-García M. 2019. Brazilian fungal diversity represented by DNA markers generated over 20 years. Brazilian Journal of Microbiology, v.51 (2): 729-749.

Kornerup, A. and Wanscher, J.H., 1963. Methuen Handbook of Colour.

Kong HZ, Liang ZQ. A new Penicillium species isolated from Jiangxi, China. Mycosystema, v. 22, p. 4–5, 2003.

Lira-Martins, D, Nascimento, D.L, Abrahão, A, Costa, P.B, D'Angioli, A.M, Valézio, E, Rowland, L., Oliveira, R.S. Soil properties and geomorphic processes influence vegetation composition, structure, and function in the Cerrado Domain. Plant Soil, v. 476, p. 549–588, 2022. DOI https://doi.org/10.1007/s11104-022-05517-y

Mercl F., García-Sánchez M., Kulhánek M., Kosnár Z., Száková J., Tlustos P., et al. Improved phosphorus 131ertilization efficiency of wood ash by fungal strains *Penicillium* sp. PK112 and *Trichoderma harzianum* OMG08 on acidic soil. Applied Soil Ecology, v. 147, 2020. DOI:103360. 10.1016/j.apsoil.2019.09.010

Monteiro, MC., Passamani, FR, Terra, MF, Silva, DM, Cirillo, MA, Batista, LR. Soilborne Fungi of the *Aspergillus* and *Penicillium* Genera in a Preserved Region of the Brazilian Cerrado Biome. Journal of Microbiology Research, v. 6, n. 1, p. 14-22, 2016. Doi:10.5923/j.microbiology.20160601.03

Oliveira PTS, et al. NEXUS of water-food-energy-ecosystem services in the Brazilian Cerrado. In: da Silva RCV, Tucci CEM, Scott CA, et al. editors. Water and Climate: Modeling in Large Basins. ABRHidro; 2019. Pp. 7–35.

Park M S, Lee S, Oh S-Y, Cho G Y, Lim Y W. Diversity and enzyme activity of Penicillium species associated with macroalgae in Jeju Island. Microbial Systematics and Evolutionary Microbiology, v. 54, p. 646–654, (2016).

Park M S, Lee J W, Kim S H, Park J H, You Y H, Lim Y W. Penicillium from Rhizosphere Soil in Terrestrial and Coastal Environments in South Korea. Myobiology, 16;48 (6):431–442, 2020.

Peterson, SW, Orchard, SS, Menon, S. *Penicillium menonorum*, a new species related to *P. pimiteouiense.* IMA fungus, v. 29, n. 2, p. 121–125, 2011. Doi: 10.5598/imafungus.2011.02.02.02

Qiao H, Sun XR, Wu XQ, Li GE, Wang Z, Li DW. The phosphate-solubilizing ability of *Penicillium guanacastense* and its effects on the growth of *Pinus massoniana* in phosphate-limiting conditions. Biology Open, v. 8, n. 11, 2020. Doi: 10.1242/bio.046797

Ramos, SMS, Cruz, R., Barbosa, RN., Houbraken, J., Machado, AR., Souza-Motta, CM., Oliveira, NT. Two new Penicillium section Sclerotiorum species from sugarcane soil in Brazil. Mycological Progress, v. 20, p. 823–835, 2021. DOI: https://doi.org/10.1007/s11557-021-01705-9

Ropars J, Didiot E, Vega R C R I, Bennetot B, Coton M, Poirier E, Coton E, Snirc A, Prieur SL, Giraud T.2020. Domestication of the Emblematic White Cheese-Making Fungus

Penicillium camemberti and Its Diversification into Two Varieties. Current Biology, Volume 30, Issue 22, 16 November 2020, Pages 4441-4453.e4.

Rodrigues A, Macedo M, Silvério D, Maracahipes L et al. Cerrado deforestation threatens regional climate and water availability for agriculture and ecosystems. Global Change Biology. Volume28, Issue22, November 2022, Pages 6807-6822.

Sang, H., An, TJ., Kim, C.S. et al. *Penicillium daejeonium* sp. Nov., a new species isolated from a grape and schisandra fruit in Korea. Journal of Microbioly, v. 51, p. 536–539, 2013. https://doi.org/10.1007/s12275-013-3291-6

Silva, C., Viana, I., Souza, D., Silva, D., Portella, A., Giongo, M. Effect of fire on abundance and fungal diversity in the Cerrado soil. Ciência Florestal, v. 31, n. 4, 2021. DOI https://doi.org/10.5902/1980509854717

Stallman J, Haelewaters D, Bach R, et al. The contribution of tropical long-term studies to mycology. IMA Fungus volume 15, Article number: 35 (2024).

Strassburg B. B. N., Brooks T., Feltran-Barbieri R., Iribarrem A., Crouzeilles R., Loyola R., Latawiec A. E., Oliveira F. J. B., Filho, Scaramuzza C. A. M., Scarano F. R., Soares-Filho B., Balmford A. Moment of truth for the Cerrado hotspot. Nature Ecology & Evolution. 2017;1(4):0099. Doi: 10.1038/s41559-017-0099;

Sullivan M J P, Talbot J, Lewis S L, Phillips O L, Qie L, Begne S K, Chave J, et al. 2017. Diversity and carbon storage across the tropical forest biome. Scientific Reports, v. 7, n. 39102.

Suraby, E.J., Agisha, V.N., Dhandapani, S., Sng, Y.H., Lim, S.H., Naqvi, N.I., Sarojam, R., Yin, Z., Park, B.S. Plant growth promotion under phosphate deficiency and improved phosphate acquisition by new fungal strain, *Penicillium olsonii* TLL1. Frontiers Microbiology, v. 14, 2023.

Tan, Y., P.; Bishop-Hurley, S., L.; Shivas, R., G.; Cowan, D., A.; Maggs-Kölling, G.; Maharachchikumbura, S., S., N.; Pinruan, U. et al. Fungal Planet description sheets: 1436-1477. Persoonia, v. 49, p. 261-350. Doi: 10.3767/persoonia.2022.49.08.

White T J, Bruns T, Lee S, Taylor J. 1990. Amplification and direct identification of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ. (eds), PCR Protocols: a guide to methods and applications: 315–322 Academic Press, USA.

Visagie, C. M., Houbraken, J., Frisvad, J. C., Hong, S.-B., Klaassen, C. H. W., Perrone, G., Seifert, K. A., Varga, J., Yaguchi, T., & Samson, R. A. (2014). Identification and nomenclature of the genus *Penicillium*. Studies in Mycology, 78, 343–371. https://doi.org/10.1016/j.simyco.2014.09.001

Visagie, C.M., Seifert, K.A., Houbraken, J., Samson, R.A, Jacobs, K. A phylogenetic revision of *Penicillium* sect. *Exilicaulis*, including nine new species from fynbos in South Africa. IMA Fungus v. 7, p. 75–117, 2016. https://doi.org/10.5598/imafungus.2016.07.01.06

Visagie C, Yilmaz N, Kocsubé S, Frisvad J, Hubka V, Samson R, Houbraken J. A review of

recently introduced Aspergillus, Penicillium, Talaromyces and other Eurotiales species. Stud Mycol. 2024 Jan 10;107:1–66.

Tedersoo L, Mikryukov V, Zizka A, Bahram M, et al. 2022. Global patterns in endemicity and vulnerability of soil fungi. Global change biology, v. 28, I. 22, p. 6696-66710. https://doi.org/10.1111/gcb.16398

Wang XC, Zhang ZK, Zhuang WY. Species Diversity of *Penicillium* in Southwest China with Discovery of Forty-Three New Species. Journal of Fungi, v. 9, n. 12, 2023. Doi: 10.3390/jof9121150.

Zhang ZY, Pan H, Tao G, Li X, Han YF, Feng Y, Tong SQ, Ding CY 2024 – Culturable Mycobiota from Guizhou Wildlife Park in China. Mycosphere, v. 15, n. 1, p. 654–763, 2024. Doi 10.5943/mycosphere/15/1/5

Zhang L, Bao DF, Shen HW, Luo ZL. Diversity of Lignicolous Freshwater Fungi from Yuanjiang River in Yunnan (China), with the Description of Four New Species. J Fungi (Basel). 2024 Dec 18;10(12):881. Doi: 10.3390/jof10120881.

Supplemental Information for:

Description of new species and occurrence record genus *Penicillium* in the Brazilian Cerrado

C.N. Figueiredo[1], F.R. Passamani[1], H.S. Guimarães[1], Y.B. Zhou[2], B. Krak[2], T.S. Carvalho[1], J.T. de Souza[1], L.R. Batista[1], P.W. Crous[2], J. Houbraken[2], V.S. Pylro[1,*]

Geographic coordinates

Area/Identification of the Transect	Variety	Planting Date	Management Practice	Latitude	Longitude
AREA 1					
NC 1	IBC 12/IAC 125	2015	Organic with irrigation	18°46’13.56”	46°56’02.76”
C1	IBC 12/IAC 125	2017	Conventional with irrigation	18°41’36.03”	46°58’14.67”
NC2	Catuaí 62	2010	Organic without irrigation	18°46’56.89”	46°56’49.78”
C1	Catuaí 62	2014	Conventional without irrigation	18°47’24.95”	46°57’01.12”
Ce1	Cerrado nativo			18°46’35.21”	46°56’53.92”
Ce2	Cerrado nativo			18°47’20.55”	46°57’07.30”
AREA 2					
NC3	Catuaí 144	2008	Regenerative with irrigation	18°52’45.33”	47° 05’51.91”
C3	Catuaí 144	2007	Conventional with irrigation	18°51’41.91”	47°07’05.51”
NC4	Topázio	2008	Regenerative with irrigation	18°52’47.00”	47°05’45.53”
C4	Topázio	2007	Conventional with irrigation	18°51’48.11”	47°07’08.96”
Ce3	Cerrado nativo			18°53’08.77”	47°06’14.29”
Ce4	Cerrado nativo			18°51’28.32”	47°07’18.95”

TBLE S1. Strains identified at the species level from partial sequences of the gene *BenA*.

Strains	DTO culture collection deposit number	URMICRO culture collection deposit number	Name of the species	Strains	DTO culture collection deposit number	URMICRO culture collection deposit number	Name of the species
MM67	DTO 510-E4	URMICRO 11737	*Penicillium ubiquetum*	MC206	DTO 515-G1	URMICRO 11825	*Penicillium ubiquetum*
56	DTO 510-E5	URMICRO 11738	*Penicillium koreense*	MC182	DTO 514-H1	URMICRO 11826	*Penicillium koreense*
22	DTO 510-E6	URMICRO 11739	*Penicillium moreirae* sp. Nov.	MC212	DTO 514-E9	URMICRO 11827	*Penicillium koreense*
29	DTO 513-B1	URMICRO 11740	*Purpureocillium lilacinum*	MC146	DTO 514-F1	URMICRO 11828	*Penicillium ubiquetum*
42	DTO 510-E7	URMICRO 11741	*Penicillium koreense*	MC145	DTO 514-F2	URMICRO 11829	*Aspergillus pseudoustus*
53	DTO 510-E8	URMICRO 11742	*Penicillium moreirae* sp. Nov.	MC19	DTO 514-F3	URMICRO 11830	*Aspergillus pseudoustus*
2[a]	DTO 510-E9	URMICRO 11743	*Penicillium moreirae* sp. Nov.	MC181	DTO 514-F4	URMICRO 11831	*Penicillium moreirae* sp. Nov.
CR41	DTO 510-F1	URMICRO 11744	*Penicillium ubiquetum*	MC162	DTO 514-F5	URMICRO 11832	*Purpureocillium lilacinum*
Ipa51	DTO 510-F2	URMICRO 11745	*Penicillium menonorum*	RM59	DTO 517-F9	URMICRO 11833	*Penicillium westlingii*
15d	DTO 510-F3	URMICRO 11746	*Penicillium beraoi* sp. Nov.	MC2	DTO 514-F6	URMICRO 11834	*Penicillium koreense*
34	DTO 513-I7	URMICRO 11747	*Penicillium ubiquetum*	MC24	DTO 514-F7	URMICRO 11835	*Penicillium shaerii*
MM13	DTO 510-F4	URMICRO 11748	*Penicillium pequii*	Cpe26	DTO 514-F8	URMICRO 11836	*Penicillium raperi*
Ipa67	DTO 513-F3	URMICRO 11749	*Aspergillus niger*	MC120	DTO 514-F9	URMICRO 11837	*Penicillium koreense*
Ipa120	DTO 510-F5	URMICRO 11750	*Fusarium oxysporum*	MC230	DTO 514-H3	URMICRO 11838	*Penicillium ubiquetum*
9	DTO 513-F4	URMICRO 11752	*Penicillium moreirae* sp. Nov.	ML110	DTO 517-A3	URMICRO 11839	*Penicillium guizhouense*
5b	DTO 510-F6	URMICRO 11753	*Penicillium koreense*	MM183	DTO 517-G1	URMICRO 11840	*Penicillium nothofagi*
MC235	DTO 510-I7	URMICRO 11754	*Penicillium moreirae* sp. Nov.	MC144	DTO 515-G3	URMICRO 11841	*Penicillium koreense*
MM218	DTO 513-F5	URMICRO 11755	*Penicillium tanzanicum*	MC186	DTO 515-G4	URMICRO 11842	*Penicillium patrocinensis* sp. Nov.
33	DTO 510-I8	URMICRO 11756	*Penicillium koreense*	MC240	DTO 514-H4	URMICRO 11843	*Aspergillus westerdjki*
101	DTO 510-I9	URMICRO 11757	*Penicillium moreirae* sp. Nov.	MC232	DTO 514-H5	URMICRO 11844	*Penicillium koreense*
118	DTO 511-A1	URMICRO 11758	*Penicillium beraoi* sp. Nov.	MM189	DTO 517-G2	URMICRO 11845	*Penicillium ubiquetum*
CR57	DTO 511-A2	URMICRO 11759	*Penicillium ubiquetum*	Cpe116	DTO 514-H6	URMICRO 11846	*Penicillium brevicompactum*
112	DTO 510-I6	URMICRO 11760	*Penicillium moreirae* sp. Nov.	MC91	DTO 514-H7	URMICRO 11847	*Aspergillus insuetus*
14	DTO 511-A3	URMICRO 11761	*Penicillium ubiquetum*	MC88	DTO 515-G5	URMICRO 11848	*Penicillium ubiquetum*

38	DTO 511-A4	URMICRO 11762	*Penicillium koreense*
17	DTO 511-A5	URMICRO 11763	*Penicillium moreirae* sp. Nov.
39	DTO 511-A6	URMICRO 11764	*Penicillium moreirae* sp. Nov.
45.1	DTO 511-A7	URMICRO 11766	*Penicillium sizovae*
61	DTO 511-A8	URMICRO 11767	*Penicillium ubiquetum*
Cpe102a	DTO 513-B2	URMICRO 11769	*Penicillium moreirae* sp. Nov.
184	DTO 513-B3	URMICRO 11770	*Penicillium moreirae* sp. Nov.
188	DTO 513-B4	URMICRO 11771	*Penicillium koreense*
178	DTO 513-B5	URMICRO 11772	*Penicillium moreirae* sp. Nov.
262	DTO 513-B6	URMICRO 11773	*Aspergillus pseudoustus*
197	DTO 513-B7	URMICRO 11774	*Penicillium citrinum*
202	DTO 513-B8	URMICRO 11775	*Penicillium moreirae* sp. Nov.
256	DTO 513-B9	URMICRO 11776	*Penicillium moreirae* sp. Nov.
246	DTO 513-C1	URMICRO 11777	*Penicillium sizovae*
220	DTO 513-C2	URMICRO 11778	*Penicillium beraoi* sp. Nov.
297.1	DTO 513-F6	URMICRO 11779	*Aspergillus westerdjki*
CR100	DTO 513-C3	URMICRO 11781	*Penicillium moreirae* sp. Nov.
ML273	DTO 513-C4	URMICRO 11782	*Penicillium steckii*
ML222	DTO 513-C5	URMICRO 11783	*Talaromyces amestolkiae*
ML169	DTO 513-C7	URMICRO 11786	*Penicillium virgatum*
ML224	DTO 513-C8	URMICRO 11787	*Penicillium sizovae*
ML198	DTO 517-A1	URMICRO 11788	*Penicillium virgatum*
Ipa48	DTO 513-C9	URMICRO 11789	*Penicillium brevicompactum*
MLX	DTO 513-D1	URMICRO 11790	*Penicillium arabicum*
217[a]	DTO 513-D2	URMICRO 11791	*Aspergillus lebretii*
ML293	DTO 513-D3	URMICRO 11792	*Penicillium brevicompactum*
Ipa190	DTO 513-D4	URMICRO 11793	*Aspergillus sydowii*
ML215	DTO 513-I8	URMICRO 11794	*Penicillium ubiquetum*
ML162	DTO 513-D5	URMICRO 11795	*Penicillium virgatum*
MC53.1	DTO 514-H8	URMICRO 11849	*Penicillium koreense*
MC168	DTO 514-H9	URMICRO 11850	*Penicillium donggangicum*
MC55	DTO 514-I1	URMICRO 11851	*Aspergillus pseudoustus*
MC81	DTO 514-I2	URMICRO 11852	*Aspergillus westerdjki*
MC142	DTO 514-I3	URMICRO 11853	*Aspergillus westerdjki*
MC235	DTO 515-G7	URMICRO 11854	*Penicillium koreense*
Cpe27	DTO 514-I4	URMICRO 11855	*Penicillium koreense*
MC149	DTO 514-I5	URMICRO 11856	*Aspergillus pseudoustus*
MC233	DTO 514-I6	URMICRO 11857	*Aspergillus westerdjki*
ML261	DTO 518-D3	URMICRO 11858	*Penicillium guarae* sp. Nov.
MC183	DTO 514-I7	URMICRO 11859	*Penicillium koreense*
MC98	DTO 514-I8	URMICRO 11860	*Penicillium moreirae* sp. Nov.
Cpe104	DTO 514-I9	URMICRO 11861	*Penicillium ubiquetum*
Ipa166	DTO 515-A2	URMICRO 11862	*Penicillium dabashanicum*
MTb444	DTO 515-G9	URMICRO 11863	*Penicillium moreirae* sp. Nov.
MM414	DTO 517-G4	URMICRO 11864	*Penicillium pluvialis* sp. Nov.
MTb443	DTO 515-A3	URMICRO 11865	*Penicillium koreense*
IR168	DTO 515-H2	URMICRO 11867	*Penicillium ubiquetum*
MTb389	DTO 517-A4	URMICRO 11868	*Penicillium beraoi* sp. Nov.
MM400	DTO 517-A5	URMICRO 11869	*Penicillium pequii*
MTb396	DTO 517-G5	URMICRO 11870	*Penicillium adametzioides*
MTb243	DTO 516-A7	URMICRO 11871	*Penicillium citrinum*
MM214	DTO 517-A7	URMICRO 11872	*Penicillium pluvialis* sp. Nov.
Mpe257	DTO 515-H3	URMICRO 11873	*Penicillium seriemae* sp. Nov.
MTb157	DTO 515-H4	URMICRO 11874	*Penicillium donggangicum*
MTb258	DTO 515-H5	URMICRO 11875	*Penicillium sizovae*
MTb328	DTO 515-H6	URMICRO 11876	*Penicillium raperi*
MTb245	DTO 515-H7	URMICRO 11877	*Aspergillus insuetus*
MTb180	DTO 515-H8	URMICRO 11878	*Penicillium ubiquetum*

ML163	DTO 514-G3	URMICRO 11796	*Penicillium virgatum*	MTb405	DTO 515-H9	URMICRO 11879	*Penicillium donggangicum*
ML204	DTO 514-G2	URMICRO 11798	*Penicillium ubiquetum*	Ipa 193	DTO 517-A9	URMICRO 11880	*Penicillium madriti*
ML221	DTO 513-F9	URMICRO 11799	*Penicillium guizhouense*	MTb321	DTO 515-I1	URMICRO 11881	*Penicillium nothofagi*
Ipa195	DTO 513-G2	URMICRO 11801	*Aspergillus gorakhpurensis*	MTb436	DTO 515-I2	URMICRO 11882	*Penicillium ubiquetum*
ML297a	DTO 513-G4	URMICRO 11802	*Penicillium virgatum*	MTb442	DTO 517-B1	URMICRO 11883	*Penicillium ubiquetum*
ML284	DTO 513-G4	URMICRO 11804	*Penicillium virgatum*	Ipa50	DTO 515-I3	URMICRO 11884	*Aspergillus versicolor*
ML180	DTO 513-G5	URMICRO 11805	*Aspergillus spelaeus*	MTb357	DTO 515-I4	URMICRO 11885	*Penicillium donggangicum*
ML285	DTO 513-G7	URMICRO 11806	*Penicillium nothofagi*	MTb230	DTO 515-I5	URMICRO 11888	*Purpureocillium lilacinum*
Cpe28	DTO 513-G8	URMICRO 11808	*Penicillium ubiquetum*	MTb335	DTO 515-I6	URMICRO 11889	*Penicillium raperi*
ML74a	DTO 513-G3	URMICRO 11809	*Penicillium ubiquetum*	Cpe30	DTO 515-I7	URMICRO 11890	*Aspergillus insuetus*
ML83a	DTO 513-G9	URMICRO 11810	*Penicillium dokdoense*	MTb227a	DTO 515-I8	URMICRO 11891	*Aspergillus ninger*
Cpe40	DTO 514-D5	URMICRO 11811	*Penicillium wotroi*	Cpe103	DTO 515-I9	URMICRO 11892	*Penicillium koreense*
IR143	DTO 513-H1	URMICRO 11812	*Aspergillus pseudoustus*	MTb247	DTO 516-A1	URMICRO 11893	*Penicillium citrinum*
ML135	DTO 514-G4	URMICRO 11813	*Penicillium wotroi*	MTb400	DTO 516-A2	URMICRO 11894	*Penicillium raperi*
ML226	DTO 514-D4	URMICRO 11814	*Penicillium guizhouense*	MTb322	DTO 517-B2	URMICRO 11895	*Aspergillus koreense*
ML171	DTO 513-I9	URMICRO 11815	*Penicillium ubiquetum*	Ipa125	DTO 517-G6	URMICRO 11896	*Aspergillus dimorphicus*
IR55	DTO 513-H2	URMICRO 11816	*Aspergillus ninger*	Cpe38	DTO 517-B4	URMICRO 11897	*Aspergillus dimorphicus*
ML165	DTO 514-G6	URMICRO 11817	*Bartalinia pini*	MTb181	DTO 516-A3	URMICRO 11898	*Aspergillus westerdjki*
ML170a	DTO 514-G7	URMICRO 11818	*Penicillium nothofagi*	Ipa122	DTO 516-A4	URMICRO 11899	*Penicillium brevicompactum*
MC101	DTO 514-G8	URMICRO 11819	*Penicillium ubiquetum*	MTb249	DTO 517-G7	URMICRO 11900	*Penicillium raperi*
MR110	DTO 517-A2	URMICRO 11820	*Penicillium coprophilum*	MTb182	DTO 516-A5	URMICRO 11901	*Penicillium koreense*
MC95	DTO 514-E5	URMICRO 11821	*Penicillium raperi*	IR18	DTO 516-A6	URMICRO 11902	*Aspergillus tamarii*
MC104	DTO 514-E6	URMICRO 11822	*Penicillium koreense*	MTb184	DTO 517-G8	URMICRO 11903	*Aspergillus versicolor*
MC178	DTO 514-E7	URMICRO 11823	*Penicillium ubiquetum*	IR131	DTO 517-B6	URMICRO 11904	*Penicillium sizovae*
Mpe262	DTO 514-E8	URMICRO 11824	*Penicillium daejonium*	MTb394	DTO 517-B7	URMICRO 11905	*Penicillium raperi*

4. CONSIDERAÇÕES FINAIS

Neste trabalho, comparamos solos de cerrado nativo e solos oriundos de áreas agrícolas de café regenerativo e convencional. Os gêneros leveduriformes *Saitozyma* e *Apiotrichum* pertencentes ao Filo Basidiomycota se destacaram nas áreas do cerrado nativo, enquanto solos sob plantações de café possuem a comunidade fúngica composta majoritariamente por gêneros do Filo Ascomycota, independente da forma de manejo, convencional ou regenerativo, e da variedade do café. Esses dados demonstram que a mudança do uso da terra, altera a composição das comunidades fúngicas do cerrado.

Ao analisar os dados da produção enzimática, os fungos que apresentaram maior produção de quitinase foram uma cepa oriunda do solo do café regenerativo e cepas do Cerrado nativo, demonstrando que o Cerrado possui grande potencial enzimático. A descrição de novas espécies do gênero *Penicillium*, isoladas do solo do Cerrado nativo e uma do café regenerativo, corrobora que as áreas remanescentes do Cerrado abrigam uma grande diversidade de fungo ainda inexplorada.

Esses resultados enfatizam a importância de preservar a vegetação nativa desse bioma, bem como, a realização de estudos taxonômicos adicionais, além de explorar o potencial enzimático do Cerrado com intuito de sustentabilidade e preservação do meio ambiente.