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MICROBIOLOGY

Thermophilic fungi in Araucaria Forest, Atlantic Forest Biome, Brazil

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Abstract: Thermophilic fungi constitute an ecologically well-defined group, commonly found in environments wherever decomposition of organic matter takes place, making them self-heating. The importance of thermophilic fungus in ecosystems contrasts with the incompleteness of our understanding of the group's biogeography patterns, phylogenies and coevolution relationships. Actually, the lack of data about thermophilic fungi from the Brazil is a limiting factor that also contributes for this scenario. In order to reduce this gap of knowledge, we aimed to characterize thermophilic filamentous fungi in Araucaria Forest, Atlantic Forest biome. Species identification was achieved by using internal transcribed spacers (ITS) as molecular ribosomal markers. In total, 240 heat-tolerant fungal strains were isolated and identified as *Thermothielavioides terrestris*, *Thielavia* sp., *Thermoascus crustaceus, Aspergillus fumigatus*, *Rhizomucor miehei*, *Rhizomucor pusillus*, and *Rhizopus microsporus*. All thermophilic strains exhibited optimal growth at 45 °C. *T. crustaceus*, *T. miehei* e *R. pusillus* were the dominant species, with the frequencies of occurrence of 35.00%, 28.33% and 23.33%, respectively. Our data reveals the apparent diversity of the Neotropical realm and may serve as reference to future studies that will try to elucidate important aspects of group.

Key words: Brazilian biome, diversity, filamentous fungi, thermophilia.

INTRODUCTION

Fungi are eukaryotic microorganisms that play ecological roles as decomposers, mutualists, and pathogens of animals and plants. They fundamentally drive carbon cycling in forest soil, mediate mineral nutrition of plants, and relieve carbon limitations of other soil organisms (Blackwell 2011, Bruns 2019). Thermophilic fungi are a particular group of fungi which show interesting features, such as growing at high temperatures through structural and physiological modifications which are unusual to the others eukaryotic forms (Maheshwari et al. 2000, Oliveira & Rodrigues 2019).

As one of the richest biodiversity hotspots in America, the Atlantic Forest biome is included in the global list of priority conservation regions (Faoro et al. 2010). The exceptional levels of species endemism, species richness and the loss of large areas of the original forest cover make this biome one of the five biodiversity hotspots (Myers et al. 2000, Mittermeier et al. 2011). Climatic as well as edaphic factors contribute to this diversity (Serna-Chavez et al. 2013). Among 5,719 fungal species recorded in Brazil, 3,017 were isolated from the Atlantic Rainforest, which remains the best known and most investigated biome of the country. Regardless of its importance, a large fraction of microbial diversity in the Atlantic Forest remains unexplored (Lima-Perim et al. 2016).

Several studies investigated the Brazilian thermophilic fungi isolated from decomposing

organic materials, trunks of trees, and domestic and industrial waste piles for biotechnological purposes (Ferrarezi et al. 2014, Pereira et al. 2015, Contato et al. 2021). Recently, Oliveira et al. (2016) assessed the heat-tolerant fungi present in composting pressmud. With regard to Brazilian soils, some reports have focused on individual isolates having potential applications (Martin et al. 2010, Moretti et al. 2012), which did not bring significant contributions to the measurement of the existing diversity, as well as to the understanding of their functions in these niches.

Several molecular data on fungal soils communities have been accumulated in public sequence databases that provide interesting analyses when combined (Egidi et al. 2019). Morgenstern et al. (2012) have presented empirical studies that report a robust phylogeny for thermophilic fungi. However, among the 115 specimens analyzed, none of them was sampled from Brazilian biomes. Thus, the attempts of reconstructing phylogenies at a global scale obviously suffer from the lack of Brazilian reference data, especially because tropical areas are the nest of high species diversity. This lack of data affects significant aspects such as hypothesized phylogenies, coevolution relationships, and correct interpretation of biogeographic patterns (Mueller & Schmit 2007).

Understanding fungal diversity allows us to predict new approaches for the management and conservation of biodiversity, especially in habitats with high devastation rates. Therefore, we focused on describe thermophilic fungal species from Atlantic Forest biome.

MATERIALS & METHODS

Sampling area and sample collection

A total of 30 soil and 30 leaf litter samples were collected from the following three Araucaria Forest sites (Atlantic Forest Biome), in Paraná, Brazil: Fazenda Canarinho (site 1), Parque Natural Municipal das Araucárias (site 2), and Parque Municipal São Francisco da Esperança (site 3). These sites exhibited heterogeneity with regard to the species in native climax, predominance of *Araucaria*, and species of trees, shrubs, and herbs. Soil samples and organic layer were collected in each site from depths of 0-20 cm using a soil liner sampler and from the surface, respectively. The samples were then transferred to sterilized plastic bags, mixed thoroughly, stored at 4 °C, and processed within a few days.

Fungal isolation and maintenance

Sabouraud culture medium was used to isolate fungi from soil and organic layer samples. To inhibit bacterial growth, 100 mg of chloramphenicol per liter was added to the medium. The isolation method was based on a series of dilutions (Clark 1965). Soil sample (1 g) was blended with sterile distilled water (SDW, 10 mL), followed by three serial dilutions with sterilized water. Organic layer sample (10 g) was crushed with 100 mL SDW in a food processor, followed by serial dilution. Next, the plates were maintained at 45 °C and monitored at regular time intervals for the emergence of fungal growth. Fungi were checked for purity, and impure isolates were repeatedly cultured. After ensuring purity, fungal strains were cultivated on Vogel agar slants, during seven days at 45 °C, and after stored at 4 °C according to Castellani's method.

Fungal growth at different temperatures

Growth performance of the eight fungal thermophilic strains was examined at different temperatures according to Morgenstern et al. (2012). Cultures were grown on Sabouraud agar plates adjusted to pH 5.5. The agar plates were inoculated with 2 µL of 10^7 spores in solution per

µL. Cultures were grown at 22 °C, 34 °C, 45 °C, and 55 °C until differential growth was clearly visible. The relative growth performance was recorded for each strain by estimating the relative surface area of the agar plates covered with fungal mycelium at the different temperatures. A simple ranking from strongest to weakest (or absent) growth was then obtained for each strain.

DNA extraction, PCR amplification, and sequencing

The strains were grown at 45 °C in Czapek medium in Erlenmeyer flasks and shaken at 200 rpm to form pellets. The mycelium was collected and washed with distilled water, frozen with liquid nitrogen, and ground to a fine powder with a mortar and pestle. Genomic DNA was extracted from 15–20 mg of fungal pellets using the cetyltrimethylammonium bromide (CTAB) method (Doyle & Doyle 1987).

DNA amplification was performed in a thermocycler (Mastercycler®, Eppendorf, USA). PCR amplification of the ITS1–5.8S–ITS2 DNA region was achieved in one fragment using ITS5 forward (5'-GGAAGTAAAAGTCGTAACAAGG-3') and ITS4 reverse (5'-TCCTCCGCTTATTGATATGC-3') primers as described by White et al. (1990). PCR amplification mix of the ITS5-ITS4 comprised approximately 20 ng genomic DNA, 1 × Biolase™ buffer with 1.5 mM $MgCl_2$ (Bioline, London, UK), 10 µM of each primer, 0.2 mM each dNTPs, and 1.25 unit of Biolase™ DNA polymerase (Bioline). The reaction was adjusted with ddH_2O to the final volume of 20 µL. The amplification profiles included an initial denaturation at 94 °C for 5 min, 30 cycles of 30 s at 95 °C (denaturation), 60 s at 50 °C (annealing), and 60 s at 72 °C extension, with a final extension at 72 °C for 7 min.

PCR products were purified using QIAquick PCR purification spin columns (Qiagen). Purified PCR products were quantified using NanoDrop 2000 spectrophotometer with the software NanoDrop 2000/2000c (Thermo Fisher Scientific, Inc.).

Sequencing was performed in 10 µL reactions using BigDye Terminator sequencing reagents and protocols (Applied Biosystems, Foster City, California, USA), and data were collected on an ABI-Prism 3500 automated sequencer (Applied Biosystems) by ACTGene Molecular Analyses at Federal University of Rio Grande do Sul. The ITS1–5.8S–ITS2 was sequenced in both directions using the primers described above. All sequences were deposited in GenBank.

The ITS sequences were aligned to each other as well as the other sequences of thermophilic fungi deposited in GenBank NCBI database, using the basic local alignment tool BLAST (www.blast.ddbj.nig.ac.jp/). We included 40 accessions (22 taxa). All the groups comprised only thermophilic fungi that have been used by recent and old phylogenetic studies (Maheshwari et al. 2000; Pan et al. 2010; Morgenstern et al. 2012). The outgroup taxa belonged to the genus *Batrachochytrium* and was selected based on data reported by Morgenstern et al. (2012). The phylogenetic tree was inferred using Bayesian analysis (MrBayes v.3.1.2). There were a total of 942 positions in the final dataset.

RESULTS

From 60 collected samples of soil and leaf litter of Araucaria Forest, 240 heat-tolerant fungal strains were isolated from Araucaria Forest fragments. Based on ITS nucleotide sequence analyses, the isolated strains were identified as *Thermothielavioides terrestris* (GenBank accession number, MG694572), *Thielavia* sp. (MG694573), *Thermoascus crustaceus* (MG694569 and MG694570), *Aspergillus fumigatus* (MG694571), *Rhizomucor miehei* (MG694574), *Rhizomucor pusillus* (MG694575), and *Rhizopus*

microsporus (MG694576). *T. crustaceus*, R. *miehei* and *R. pusillus* were the most prevalent species, being founded in all three sampling sites, with frequencies of occurrence of 35.00%, 28.33% and 23.33%, respectively (Table I).

The fungal strains described in this study were subjected to a temperature-dependent growth at different temperature ranges (55 °C, 45 °C, 34 °C, and 22 °C). Optimal growth temperature was determined to be 45 °C, except for *R. miehei* that displayed no difference in growth rate between 45 °C and 34 °C (Table II). Only two species were capable to grow at 55 °C (*T. crustaceus* and *A. fumigatus*). The last one was capable to grow over the entire temperature range tested. The thermotolerant *R. microsporus*, as well *A. fumigatus*, grew at 22 °C. This was evidence of the pronounced cell plasticity that enables survival over a broad range of temperatures.

The ITS marker identified almost all isolated fungal strains. However, one fungal isolate of the genus *Thielavia* did not present species level definition after amplification of ITS region. In order to evaluate the genetical relationships between our isolates and other thermophilic fungi reported previously, phylogenetical analysis was done. The dendrogram based on ITS sequence analysis showed that all isolates were included in two well-supported clades (Fig. 1). The first comprised *Rhizopus* spp. and *Rhizomucor* spp. (phylum *Mucoromycota*), whereas the second formed clade comprises species from phylum *Ascomycota*, and order *Sordariales* and *Eurotiales*.

Table I. Thermophilic fungi from Atlantic Forest biome with their frequencies of occurrence and relative abundance.

Species	Numbers	Sources	Sampling sites	Frequencies of occurrence (%)	Relative abundance (%)
Thermothielavioides terrestris	26	7	1, 3	11.66	10.83
Thielavia sp.	32	13	2, 3	21.66	13.33
Thermoascus crustaceus	58	21	1, 2, 3	35.00	24.16
Aspergillus fumigatus	28	9	1, 2, 3	15.00	11.67
Rhizomucor miehei	39	17	1, 2, 3	28.33	16.25
Rhizomucor pusillus	34	14	1, 2, 3	23.33	14.17
Rhizopus microsporus	23	6	2, 3	10.00	9.59

Numbers represent the number of strains; Sources represent the number of soil and leaf litter samples in which each fungal species was registered; Frequencies of occurrence represent the percentage of the total samples in which a particular species was registered; Relative abundance represents the number of particular species isolates in the sample series over the total number of fungal strains in the sample series.

Table II. Temperature dependence growth of Araucaria Forest thermophilic fungal strains.

GenBank accession number	Fungal species	Growth time	Growth temperature (°C)			
			55	45	34	22
			Relative growth*			
MG694576	*Rhizopus microsporus*	1d	0	3	2	1
MG694575	*Rhizomucor pusillus*	2d	0	3	2	0
MG694569	*Thermoascus crustaceus*	2d	1	4	2	0
MG694570	*Thermoascus crustaceus*	3d	0	4	3	0
MG694571	*Aspergillus fumigatus*	3d	1	4	4	2
MG694573	*Thielavia* sp.	4d	0	3	2	0
MG694572	*Thermothielavioides terrestris*	4d	0	3	2	0
MG694574	*Rhizomucor miehei*	4d	0	2	2	0

***The relative growth performance of each organism is indicated by numbers: 0, no growth; 1-4, increasing rate growth.**

DISCUSSION

Among fungi, only a few species have a unique mechanism of growing at high temperatures between 45 °C and 55 °C. According to Cooney & Emerson (1964), such fungi are arbitrarily distinguished in two groups based on their temperatures of growth. The thermophilic fungi exhibit a minimum temperature of growth at or above 20 °C and a maximum growth temperature at or above 50 °C, while thermotolerant fungi have a temperature range of growth from below 20 to ~50 °C.

Ours results corroborate those reported by Morgenstern et al. (2012) who reported that few thermophilic fungus species have been described, corresponded to only 40 of the 120,000 currently accepted fungal species (Hawksworth & Lücking 2017). According to Oliveira & Rodrigues (2019), 46 fungal thermophilic species have been described until now. Thus, many thermophilic fungal species were recorded in Araucaria Forest. They comprised five true thermophilic and two thermotolerant fungal species.

T. crustaceus was the most prevalent specie. *Thermoascus* comprises many saprobic strains commonly isolated from soil, but their species can occur in a wide range of substrates, such as plants, animals, food products and air (Luangsa-ard et al. 2004).

Soil is an excellent ecological niche for colonization of thermophilic fungi. It is interesting that these microorganisms are ubiquitous in soils where the sun can heat them up, reaching temperatures that are suitable for their germination and growth (Rajasekaran & Maheshwari 1993, Ahirwar et al. 2017). However, variation in the abundance of individual species can depend on the type of soil, depth, season of the year and organic matter content (Subrahmanyam 1999). Until now, only a few studies have reported the occurrence of thermophilic species of fungi from tropical and temperate soils (Redman et al. 1999, Córdova et al. 2003, Salar & Aneja 2007, Pan et al. 2010, Powell et al. 2012).

The species shown in Table I exhibited strong growth rate at 45 °C. *A. fumigatus* was capable to grow over the entire temperature range tested,

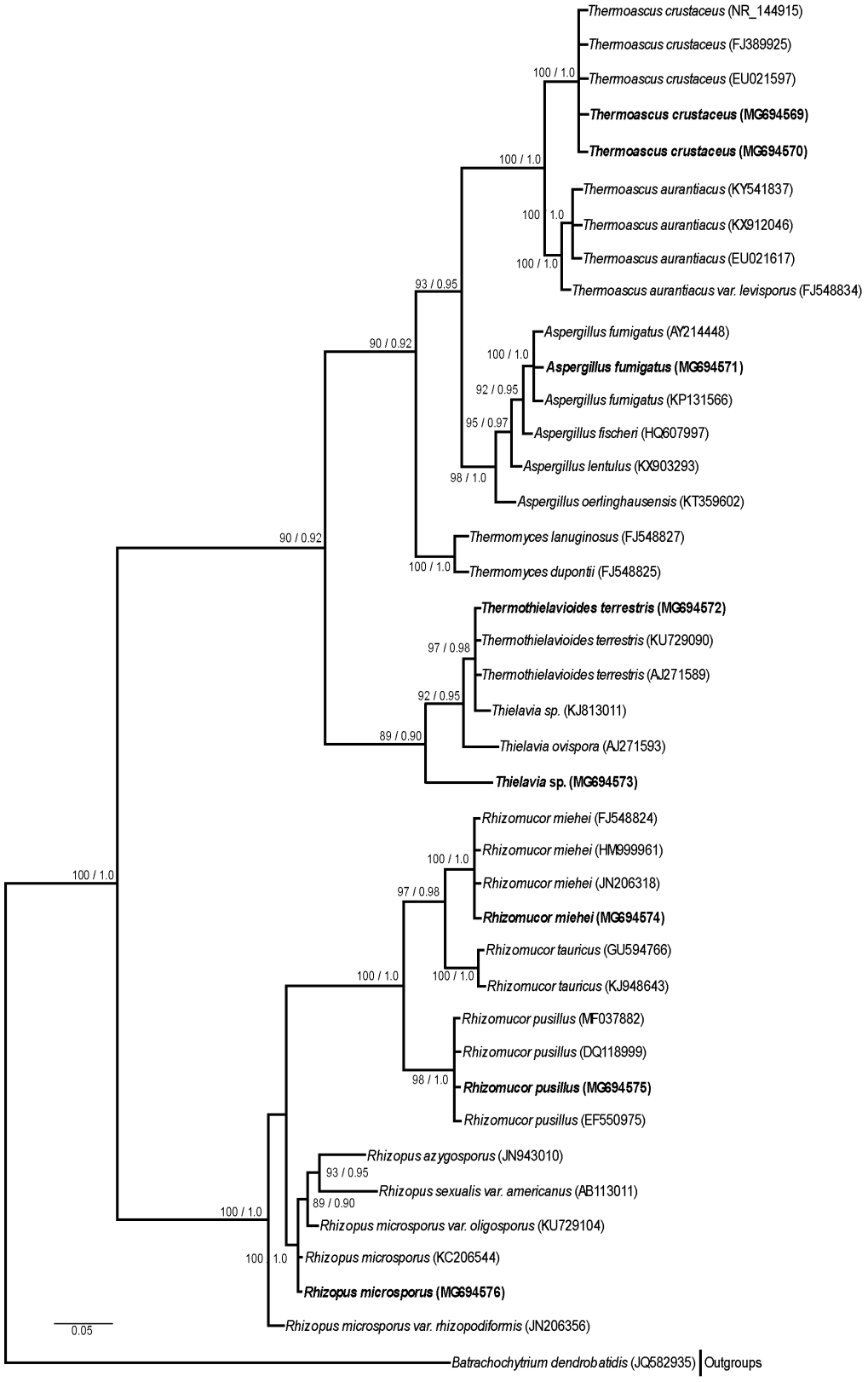

Figure 1. Phylogenetic tree of the ITS region nucleotide sequences of Atlantic Biome fungal isolates (new accesses) and related thermophilic fungi. The tree was built with Bayesian analysis. The Bootstrap values are based on 1000 replicate runs, shown as percent. *Batrachochytrium dendrobatidis* was used as the outgroup. The GenBank accession number follows the name of fungal species.

in accordance with the study conducted by Cooney & Emerson (1964). Curiously, *R. pusillus*, *R. miehei*, and *T. terrestris* did not show growth rate at 55 ºC, contrary to what was reported by Morgenstern et al. (2012) in their study.

One fungal isolate of the genus *Thielavia* was not identified to the species level. According to Schoch et al. (2012), ITS exhibits the highest probability of successful identification of a broad range of fungi. *Thielavia* is a common genus of environmental ascomycetes belonging to the family *Chaetomiaceae* in the order *Sordariales*. Its taxonomy and phylogeny have been the subject of some ambiguity, since optimal markers for species distinction have not been established yet. The ITS region is highly conserved in *Sordariales*. As a result, it is not very useful to establish phylogenetical relationships at the species level (Stchigel et al. 2002). Thus, the amplification of other molecular markers should be performed, as it may provide discrimination of this fungal strain at the species level. Contrastingly, since most unknown species are found in the Neotropical realm, the least explored major region in the world, this fungal strain should be investigated more accurately, because it could represent a new species. According to Oliveira et al. (2015), the amount of thermophilic and thermotolerant fungi described tends to increase as new habitats are studied.

In the ITS analysis, the thermophilic fungus were placed into two well supported clades. The first comprised *Rhizomucor* spp. and *Rhizopus* spp. The ability of thermophilic fungi to develop at high temperature was displayed by a few Mucoromycota, including the presently-isolated *R. miehei*, *R. pusillus*, and *R. microsporus* (Zhou et al. 2014).

The second clade comprised species from phylum *Ascomycota*, and order *Sordariales* and *Eurotiales*. All of them have been known as thermophilic molds found mainly in lignocellulosic degrading biomass, with a widespread distribution around the world (Hibbett et al. 2007, Berka et al. 2011, Morgenstern et al. 2012, van den Brink 2015, Wijayawardene et al. 2018). In the *Ascomycota*, thermophilic fungi are restricted to *Sordariales*, *Eurotiales*, *Hypocreales* and *Microascales* (Oliveira & Rodrigues 2019). For the *Sordariales*, we included the sequences from *Thielavia* sp. and *T. terrestris* (*Sordariomycetes* class, *Chaetomiaceae* family).

Species of the genus *Rasamsonia*, *Thermoascus* and *Thermomyces* of *Eurotiales* are recognized as thermophiles (Oliveira & Rodrigues 2019). *T. crustaceus* was one of the thermophilic species described in the present investigation. Also described in our study, *A. fumigatus* (*Eurotiales*) is not a truly thermophile. However, some species of genera such as *Aspergillus phoenicis*, *Aspergillus niger* and *A. fumigatus* are thermotolerant, as regarded by Cooney & Emerson (1964).

The lack of samples of thermophilic fungus from the Brazil environments is a limiting factor for the understanding of distribution ranges, phylogeny, and systematic of the group, because of its exceptional levels of species endemism and richness. In the present study, we described seven thermophilic fungal species in Atlantic Forest Biome, supporting the apparent diversity of the Neotropical realm. However, more efforts are needed to obtain a better understating of thermophilic fungal species in the present environmental scenario of global deforestation.

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REFERENCES

AHIRWAR S, SONI H, PRAJAPATI BP & KANGO N. 2017. Isolation and screening of thermophilic and thermotolerant fungi for production of hemicellulase from heated environments. Mycology 8: 255-266.

BERKA RM ET AL. 2011. Comparative genomic analysis of the thermophilic biomass-degrading fungi *Myceliophthora thermophila* and *Thielavia terrestris*. Nat Biotechnol 29: 922-927.

BLACKWELL M. 2011. The fungi: 1,2,3... 5.1 million species? Am J Bot 98: 426-438.

BRUNS TD. 2019. The developing relationship between the study of fungal communities and community ecology theory. Fungal Ecol 39: 393-402.

CLARK FE. 1965. Agar-plate method for total microbial count. In: Black CA, Evans D, White JL, Ensminger LE, Clark FE & Dinauer RC (Eds), Methods of soil analysis. Part 2. Chemical and microbiological properties, New York: Madson Inc, p. 1460-1466.

CONTATO AG ET AL. 2021. Prospection of fungal lignocellulolytic enzymes produced from jatoba (*Hymenaea courbaril*) and tamarind (*Tamarindus indica*) seeds: scaling for bioreactor and saccharification profile of sugarcane bagasse. Microorganisms 9: 533.

COONEY DG & EMERSON R. 1964. Thermophilic fungi: an account of their biology, activities and classification. San Francisco: WH Freeman & Co, 188 p.

CÓRDOVA SR, BARATTI J, NUNGARAY J & LOERA O. 2003. Identification of mexican thermophilic and thermotolerant fungal isolates. Micol Aplicada Int 15: 37-44.

DOYLE JJ & DOYLE JL. 1987. A rapid DNA isolation procedure for small quantities of fresh leaf tissue. Phytochemical Bull 19: 11-15.

EGIDI E ET AL. 2019. A few Ascomycota taxa dominate soil fungal communities worldwide. Nature Comm 10: 2369.

FAORO H, ALVES AC, SOUZA EM, RIGO LU, CRUZ LM, AL-JANABI SM, MONTEIRO RA, BAURA VA & PEDROSA FO. 2010. Influence of soil characteristics on the diversity of bacteria in the Southern Brazilian Atlantic Forest. Appl Environ Microbiol 76: 4744-4749.

FERRAREZI AL ET AL. 2014. Production and characterization of lipases and immobilization of whole cell of the thermophilic *Thermomucor indicae seudaticae* N31 for transesterification reaction. J Mol Catal B Enzym 107: 106-113.

HAWKSWORTH DL & LÜCKING R. 2017. Fungal diversity revisited: 2.2 to 3.8 million species. Microbiol Spectr 5: 1-17.

HIBBETT DS ET AL. 2007. A high level phylogenetic classification of the fungi. Mycol Res 3: 509-547.

LIMA-PERIM JE, ROMAGNOLI EM, DINI-ANDREOTE F, DURRER A, DIAS ACF & ANDREOTE FD. 2016. Linking the composition of bacterial and archaeal communities to characteristics of soil and flora composition in the Atlantic Rainforest. Plos One 11: e0146566.

LUANGSA-ARD JJ, HYWEL-JONES NL & SAMSON RA. 2004. The polyphyletic nature of *Paecilomyces* sensu lato based on 18S-generated rDNA phylogeny. Mycologia 96: 773-780.

MAHESHWARI R, BHARADWAJ G & BHAT MK. 2000. Thermophilic fungi: their physiology and enzymes. Microbiol Mol Biol Rev. 64: 461-488.

MARTIN ET AL. 2010. Pectinase production by a Brazilian thermophilic fungus *Thermomucor indicae-seudaticae* N31 in solid-state and submerged fermentation. Microbiology 79: 306-313.

MITTERMEIER RA, TURNER WR, LARSEN FW, BROOKS TM & GASCON C. 2011. Global biodiversity conservation: the critical role of hotspots. In: Zachos F & Habel JC (Eds), Biodiversity hotspots: distribution and protection of conservation priority areas, Berlin: Springer-Verlag Berlin Heidelberg, p 3-22.

MORGENSTERN I, POWLOWSKI J, ISHMAEL N, DARMOND C, MARQUETEAU S, MOISAN MC, QUENNEVILLE G & TSANG A. 2012. A molecular phylogeny of thermophilic fungi. Fungal Biol 2: 489-502.

MORETTI MMS, BOCCHINI-MARTINS DA, DA SILVA R, RODRIGUES A, SETTE LD & GOMES E. 2012. Selection of thermophilic and thermotolerant fungi for the production of cellulases and xylanases under solid-state fermentation. Braz J Microbiol 43: 1062-1071.

MUELLER GM & SCHMIT JP. 2007. Fungal biodiversity: what do we know? what can we predict? Biodivers Conserv 16: 1-5.

MYERS N, MITTERMEIER RA, FONSECA GAB & KENT J. 2000. Biodiversity hotspots for conservation priorities. Nature 403: 853-858.

OLIVEIRA TB, GOMES E & RODRIGUES A. 2015. Thermophilic fungi in the new age of fungal taxonomy. Extremophiles 19: 31-37.

OLIVEIRA TB, LOPES VCP, BARBOSA FN, FERRO M, MEIRELLES LA, SETTE LD, GOMES E & RODRIGUES A. 2016. Fungal communities in pressmud composting harbour beneficial and

detrimental fungi for human welfare. Microbiology 162: 1146-1156.

OLIVEIRA TB & RODRIGUES A. 2019. Ecology of thermophilic fungi. In: Tiquia-Arashiro SM & Grube M (Eds), Fungi in extreme environments: ecological role and biotechnological significance, Switzerland: Springer Nature, p. 39-57.

PAN WZ, HUANG XW, WEI KB, ZHANG CM, YANG DM, DING JM & ZHANG KQ. 2010. Diversity of thermophilic fungi in Tengchong Rehai National Park revealed by ITS nucleotide sequence analyses. J Microbiol 48: 146-152.

PEREIRA, JC, MARQUES NP, RODRIGUES A, OLIVEIRA TB, BOSCOLO M, DA SILVA R, GOMES E & MARTINS DAB. 2015. Thermophilic fungi as new sources for production of cellulases and xylanases with potential use in sugarcane bagasse saccharification. J Appl Microbiol 18: 928-939.

POWELL AJ, PARCHERT KJ, BUSTAMANTE JM, RICKEN JB, HUTCHINSON MI & NATVIG DO. 2012. Thermophilic fungi in an ariland ecosystem. Mycologia 104: 813-825.

RAJASEKARAN AK & MAHESHWARI R. 1993. Thermophilic fungi: an assessment of their potential for growth in soil. J Biosc 18: 345-354.

REDMAN RS, KITVINTSEVA A, SHEEHAN KB, HENSON JM & RODRIGUEZ R. 1999. Fungi from geothermal soils in Yellowstone National Park. Appl Environ Microbiol 65: 5193-5197.

SALAR RK & ANEJA KR. 2007. Thermophilic fungi. Taxonomy and biogeography. J Agric Technol 3: 77-107.

SCHOCH CL ET AL. 2012. Nuclear ribosomal internal transcribed spacer (ITS) region as a universal DNA barcode marker for fungi. Proc Natl Acad Sci U.S.A 9: 6241-6246.

SERNA-CHAVEZ HM, FIERER N & BODEGON PM. 2013. Global drivers and patterns of microbial abundance in soil. Glob Ecol Biogeogr 22: 1162-1172.

STCHIGEL AM, FIGUERA L, CANO J & GUARRO J. 2002. New species of *Thielavia*, with molecular study of representative species of the genus. Micology Res 106: 975-983.

SUBRAHMANYAM A. 1999. Ecology and Distribution. In: Johri BN, Satyanarayana T & Olsen J (Eds), Thermophilic Moulds in Biotechnology, London: Kluwer Academic Publishers, p. 13-42.

VAN DEN BRINK J. 2015. Thermophilic growth and enzymatic thermostability are polyphyletic traits within *Chaetomiaceae*. Fungal Biol 119: 1255-1266.

WHITE TJ, BRUNS TD, LEE SB & TAYLOR JW. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ & White TJ (Eds), PCR Protocols, San Diego: Academic Press, p. 315-322.

WIJAYAWARDENE NN, HYDE KD, LUMBSCH HT & LIU JK, MAHARACHCHIKUMBURA SSN, EKANAYAKA AH, TIAN Q & PHOOKAMSAK R. 2018. Outline of *Ascomycota*: 2017. Fungal Divers 88: 167-263.

ZHOU P ET AL. 2014. Genome sequence and transcriptome analysis of the thermophilic zygomycete fungus *Rhizomucur miehei*. BMC Genom 15: 294.

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