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Soilborne Fungi of the *Aspergillus* and *Penicillium* Genera in a Preserved Region of the Brazilian Cerrado Biome

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Abstract Conducting studies on the fungi that are present in the soils of preserved biomes of natural ecosystems is important; such research leads to knowledge regarding the biodiversity of species and to the discovery of new products of importance to human health. However there is little information about the biodiversity of the filamentous fungi in the cerrado biome of Brazil. Therefore, the aim of this study was to investigate the fungi of the genera *Aspergillus* and *Penicillium* present in the preserved soils of the Brazilian cerrado. Thirty soil samples were collected during periods of high and low rainfall in three different regions. The filamentous fungi present in the soil were isolated using a serial dilution technique and were incubated in two culture media, Dichloran Glycerol (DG-18) and Corn Meal Agar (CMA). A total of 183 isolates belonging to the *Aspergillus* (82) and *Penicillium* (101) genera were identified. The following species were the most abundant: *Aspergillus flavus*, *Aspergillus ostianus* and *Penicillium citrinum*. Species of *Aspergillus* and *Penicillium* that are potentially toxigenic and of biotechnological importance are naturally present in preserved areas of the Brazilian cerrado.

Keywords Biodiversity, Soil, Cerrado, *Aspergillus*

1. Introduction

Brazil is among the most biologically rich countries, and it maintains substantial areas of high value for the preservation of biodiversity (Sparovek et al. 2012). Among these biologically important areas is the cerrado biome. “Cerrado” is the Portuguese word for the plateau of woodlands, savannas, grasslands, gallery and dry forests in central Brazil (Klink & Machado, 2005). The cerrado is the second largest biome in South America, where it is only surpassed in area by the Amazon forest. This ecosystem occupies approximately 20% of Brazil andincludes parts of the following Brazilian states: Bahia, Goias, Maranhao, Mato Grosso, Mato Grosso do Sul, Minas Gerais, Parana, Piaui, Sao Paulo, and Tocantins (Ratter et al. 1997; Sano et al. 2010).

The cerrado biome is considered a biodiversity hotspot and contains a large number of endemic species (Myers et al. 2000). However, the cerrado is one of the most endangered biomes worldwide. The indiscriminate advances caused by the expansion of agriculture and animal farming have endangered this biome (Klink and Machado, 2005). Large areas of monoculture cover savannah regions in the south of Minas Gerais (Myers et al. 2000; Giannetti et al. 2011).

Despite its importance, the biodiversity of the cerrado has been poorly studied, particularly the subject of microorganisms. Little is known about the soil microbiota. Soil microorganisms are fundamental for the maintenance of terrestrial ecosystems (Moreira and Siqueira, 2001), and fungi constitute an important part of the soil biomass (Ritz and Young, 2004). In the biosphere, the richest fungal habitat is the soil. Processes such as soil aggregation, the decomposition of organic residues, nutrient mineralization, the establishment of symbiotic relationships and the control of plagues and diseases rely on the effective participation of fungi (Pfenning and Abreu, 2006).

Fungi belonging to the *Aspergillus* and *Penicillium* genera play important roles in food contamination and deterioration, and some species produce mycotoxins that may have harmful effects on human and animal health (Pitt, 2000). *Aspergillus* and *Penicillium*, which include many economically interesting species (Peterson, 2012), are two of the most studied genera (Pitt, 2000; Klich, 2002; Varga et al. 2004).

These fungi are of great importance, not only in terms of biotechnological applications but also for economic reasons, due to their metabolic properties (Hoffmeister and Keller, 2007). Thus, the importance of these genera in ecosystems and in the daily life of the human species ranges from basic

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research to microbial ecology, environmental purification and biotechnology applications, which reinforces the urgent need to conserve natural areas (Tauk-Tornisielo et al. 2005). Considering the importance of the *Aspergillus* and *Penicillium* genera and due to the dearth of information about *Aspergillus* and *Penicillium* species in the savannah soils in the state of Minas Gerais, the aim of this work was to isolate and identify fungi of these genera in preserved soils of the savannah biome in Minas Gerais using morphologic methods in three different locations during periods of high and low rainfall.

2. Materials and Methods

2.1. Study Area

The study sites were located in the state of Minas Gerais in the municipalities of Arcos, Luminarias, and Passos, which have preserved cerrado regions. The Arcos region is located in the upper Sao Francisco zone, in the midwestern region of the state of Minas Gerais, at the following geographical coordinates: 20º17'29"Sand 45º32'23"W. The Luminarias region is in the southern Minas region at the following coordinates: 21°31'26"S and 44°54'11"W. The Passos region is also located in the south of the state, at the following coordinates: 20º43'08"S and 46º36'35"W. The climate is seasonal (wet from October to March and dry from April to September) and mild year-round, with temperatures ranging from 22 to 27° C. The average annual rainfall is 1500 mm (Klink & Machado, 2005).

2.2. Sampling

The soil sampling was conducted during periods of high and low rainfall in the regions of Arcos, Luminarias, and Passos, which are preserved savannah areas in the state of Minas Gerais. A total of 5 compound samples were collected in the 3 areas of study, in the 2 periods, January and August of 2010, totaling 30 soil samples. A total of 300 g of soil collected in these regions was used for the sample processing. For each sampling point, 12 subsamples were collected in two concentric circles with radii of 3 and 6 m from the center at a depth of 20 cm. The soil samples were extracted with the aid of an auger that had been washed in alcohol and flamed. Subsequently, the samples were stored in sterile plastic bags and were transported to the laboratory.

2.3. Isolation of *Aspergillus* and *Penicillium* from the Soil

For the isolation of these fungi from the soil samples, 10.0 g of soil was weighed and added to 90.0 mL of sterilized Peptone water (1.0 %). Each sample was homogenized in a shaker at 120 rpm for 30 minutes. Serial dilutions were performed in two different culture media. After successive dilutions up to 10^{-3}, an aliquot of 0.1 mL was added to DG18 culture medium (dichloran1.0 mL; bacteriological peptone 5.0 g; KH_2PO_4 1.0 g; $MgSO_4 \cdot 7H_2O$0.5 g; glycerol 220 g; chloramphenicol 1 mg; agar15.0 g; and distilled water 1000 mL) and CMA (filtered cooked cornmeal 30.0g; agar 15.0 g; distilled water 1000 mL; and chloramphenicol 1mg), and the aliquot was spread with a Drigalski spatula. The plates were incubated at 25ºC for seven days. The results were expressed as colony-forming units per gram of soil.

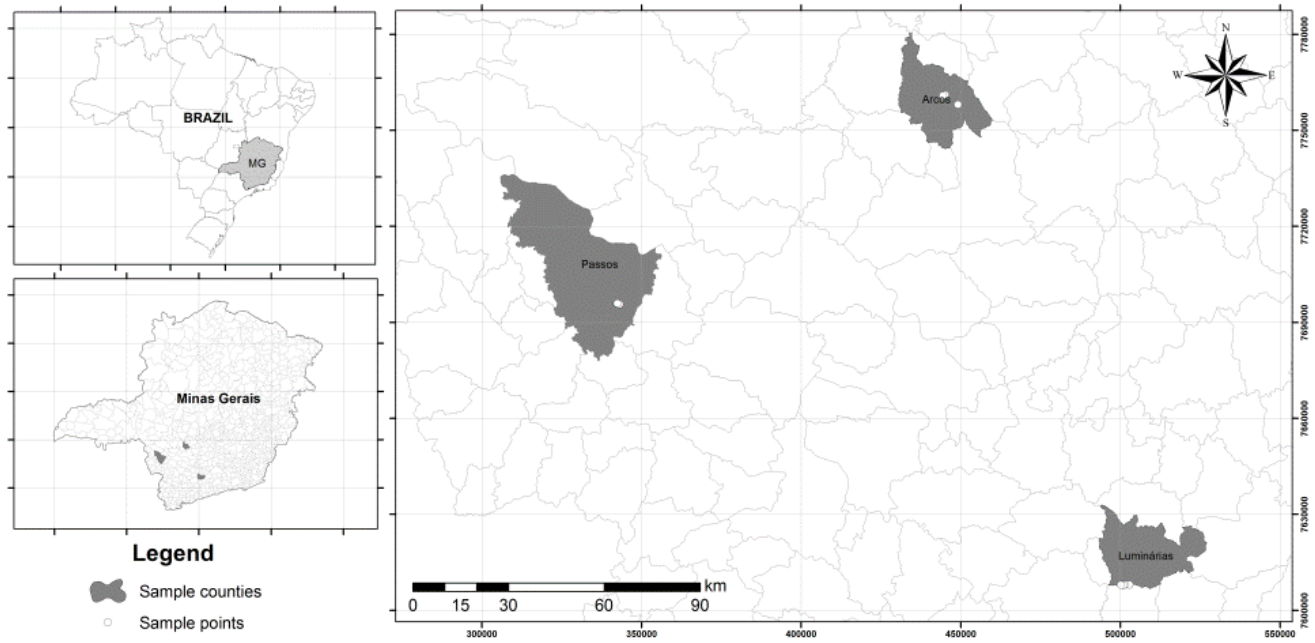

Figure 1. Location of the study area

2.4. Morphological Identification of Species Belonging to the *Aspergillus* and *Penicillium* Genera

After pure cultures were obtained, isolates belonging to the *Aspergillus* genus were inoculated into culture media with the following formulations: CYA (K_2HPO_4 1.0g; Czapek concentrate 10.0 mL; metal solution 1mL; yeast extract 5.0g; agar 15.0g; sucrose 30.0 g; and distilled water 1000 mL) and MEA (malt extract 20.0g; bacteriological peptone 1.0g; glucose 20.0g; agar 20;and distilled water 1000 mL). The cultures were incubated for seven days, with CYA at 25ºC and 37ºC and MEA at 25ºC.

For the morphological identification of *Aspergillus* species, macroscopic characteristics were analyzed, such as colony color, mycelia color, the presence of exudate, reverse color, colony diameter, the presence of soluble pigmentation, and cleistothecia/sclerotia, according to Klich (2002). The identification Manual were based on those of Pitt and Hocking (2009), Domsch, Gams, and Anderson (2007) Samson et al (2014) and others.

Isolates belonging to the *Penicillium* genus were inoculated into standard culture media as previously described, with CYA at 25ºC and 37 ºC, MEA at 25ºC and Creatine Sucrose AgarCREA (creatine 3.0 g; sucrose 30g; HCl 0.5 g; $MgSO_4$ 0.5 g; $K_2HPO_4 \cdot 3H_2O$ 0.5g; FeSO4·7H2O 0.01g; and distilled water 1000 mL).

The *Penicillium* species were identified based on the methods of Pitt (2000), Pitt and Hocking (2009), Samson et al. (2004), and Domsch, Gams, and Anderson (2007) and others.

2.5. Statistical Analyses

In this study, analyses of variance and comparison tests of averages were not used because these techniques assume that the samples are independent and normally distributed. However, these assumptions do not apply to incidence data or the identification of isolates; these are count data that are highly dispersed in relation to the evaluated regions. Thus, the data were organized into contingency tables, which enabled the application of a simple correspondence analysis (Greenake and Blasius 2006) and allowed the construction of percentage maps, with the results displayed as a graphic distribution of the analyzed variables.

3. Results and Discussion

3.1. Isolation of *Aspergillus* and *Penicillium* from the Soil

Thirty soil samples containing 813 isolates were analyzed. Isolates belonging to the genera *Aspergillus* and *Penicillium* were found in both culture media (Figure 2), and serial dilutions were performed. *Penicillium* was the dominant genus in all the samples, demonstrating that species of this genus are the best adapted to the soil in the area studied. The *Penicillium* colonies exhibited rapid growth and abundant sporulation in the DG18 culture medium (Figure 2A, 2B).

Many *Penicillium* species produce large numbers of spores with conidia that are easily dispersed through the atmosphere, resulting in the predominance of some species of this genus (Gomez et al. 2007). Depending on the limits of ecosystem productivity, precipitation and moisture changes may alter the composition of the fungal community (Castro et al. 2010).

3.2. Morphological Identification of Isolates Belonging to the *Aspergillus* and *Penicillium* Genera

Through the analysis of the morphological characteristics of the encountered fungi, 183 isolates belonging to the *Aspergillus* (82) and *Penicillium* (101) genera were identified. There were eight *Aspergillus* species (*A. flavus; A. japonicus; A. foetidus; A. niger; A. ochraceus; A. tubingensis; A. sulphureus;* and *A. ostianus*) and eighteen *Penicillium*s pecies (*P. citrinum; P. decumbens; P. solitum; P. glabrum; P. expansum; P. simplicissimum; P. canescens; P. waksmanii; P. citrinum; P. chrysogenum; P. raistrick; P. minioluteum; P.miczynskii; P. varibile; P. corylophilum; P. commune; P. brevicompactum;* and *P. pinophilum*), for a total of 26 identified species (Table 1).

Among the filamentous fungi, *Aspergillus* and *Penicillium* are common in soil and foods and are frequently mentioned in ecological studies (Pitt and Hocking, 2009).

The species with the largest number of isolates identified in this study were *Penicillium citrinum* and *Aspergillus flavus* (Table 1). Isolates belonging to the *Citrina* section are abundant and found worldwide, where they are very common in soils (Pitt, 2000).

A deficiency of available nutrients and the presence of acid soils are characteristic of the soils of the cerrado biome (Lopes, 1996). To survive in this hostile habitat, it is thought that the species living in this biome develop a more sophisticated type of metabolism. *Penicillium citrinum* is known to produce toxic secondary metabolites (Luca et al., 2007; Zhelifonova et al., 2010), which may be beneficial by providing a competitive advantage when colonizing a new substrate.

Furthermore, many *Penicillium* species are capable of solubilizing phosphate, which may also explain the incidence of this species in cerrado soils. A study conducted by Pandy et al. (2008) revealed that the genus *Penicillium* contains species with a capacity for phosphate solubilization.

Aspergillus flavus was another commonly isolated species in this study. *Aspergillus flavus* is an opportunistic pathogen with a worldwide distribution; it is extremely common in soils (Klich 2002). This species has been described as a producer of mycotoxins, particularly aflatoxins, which makes it important to conduct studies of *A.flavus* in cultivated soils, such as those by Horn, Greense and Dorner (1995) and Razzaghi-Abyaneh et al. (2006). Chen et al. (2011) detected *Aspergillus flavus* in soils and characterized the production of a fungistatic substance against the *Alternaria brassicicola* pathogen; this substance inhibited the germination of the conidia.

However, there is a great need to conduct studies to increase the knowledge of the diversity and distribution of the species of these fungi in soils and preserved regions, particularly in tropical biomes. Knowledge about soil microbiota, in addition to being fundamental for the taxonomic count of the populations that are found there, may lead to the discovery of metabolic processes used by these organisms.

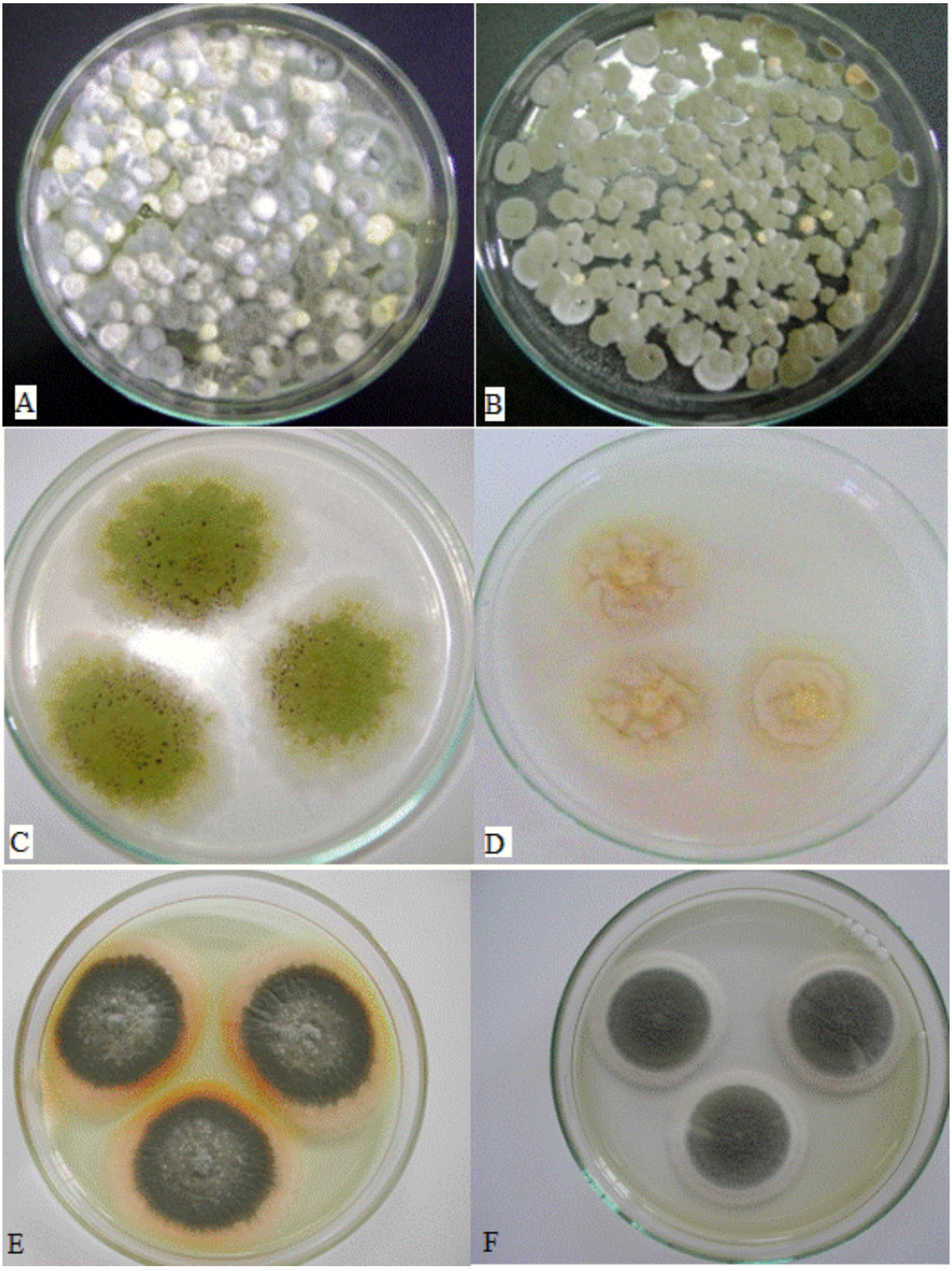

Figure 2. Fungi isolation from soils sample, (A and B) in the DG18 culture medium, (C and D) *Aspergillus* colonies in the CYA culture medium, (E and F) *Penicillium* colonies in the CYA culture medium

Table 1. Numbers of isolates and species identified during the wet and dry seasons in three different regions

	Wet season			Dry season		
	Preserved cerrado regions in the Minas Gerais state of Brazil					
Species	**Arcos**	**Luminarias**	**Passos**	**Arcos**	**Luminárias**	**Passos**
Aspergillus flavus	0	0	8	0	11	2
Aspergillus foetidus	0	6	4	1	4	5
Aspergillus japonicus	0	1	9	0	0	0
Aspergillus niger	0	0	2	0	1	0
Aspergillus ochraceus	0	1	0	1	0	0
Aspergillus ostianus	2	1	0	0	11	0
Aspergillus sulphureus	1	1	5	0	4	0
Aspergillus tubingensis	0	0	1	0	0	0
Penicillium brevicompactum	0	0	0	4	0	1
Penicillium canescens	0	0	1	0	0	0
Penicillium chrysogenum	0	0	0	0	3	1
Penicillium citreonigrum	0	0	0	0	1	0
Penicillium citrinum	5	8	0	1	8	6
Penicillium commune	0	0	0	2	0	0
Penicillium corylophilum	0	0	0	0	2	0
Penicillium decumbens	1	1	0	0	0	0
Penicillium expansum	1	0	0	0	0	0
Penicillium glabrum	3	5	1	1	2	2
Penicillium miczynskii	0	0	0	1	0	1
Penicillium minioluteum	0	0	0	0	2	0
Penicillium pinophilum	3	2	5	0	0	0
Penicillium raistrick	0	0	0	0	0	1
Penicillium simplicissimum	2	4	0	1	8	3
Penicillium solitum	0	0	0	2	1	0
Penicillium varibile	0	0	2	0	0	0
Penicillium waskmanii	0	0	0	3	0	0
Total: 183	**18**	**30**	**38**	**17**	**58**	**22**

3.3. Simple Correspondence Analyses (CA)

The results of the simple correspondence analyses (CA) of the fungal species of the *Aspergillus* and *Penicillium* genera in the three regions and in the two different collection periods indicated revealed a significant association between the *Aspergillus* and *Penicillium* species with the savannah regions in the different seasonal periods. The determination of these groups and the estimation of the components were based on the results of the contributions of the line and column profiles (Tables 2 and 3) in relation to the components used for the construction of the percentage map.

In the interpretation of these components, it was noted that the variability of the points decomposed into each component. Thus, the coefficients specified in Contr (contributed) in the Table 2, indicate the amount that each point contributed to the determination of the direction of the axes. Finally, the coordinates specified in Coord (Table 2) represent the scores obtainedfor each component.

Thus, analogous to the interpretation of the values described in Table 2, the contributions and the coordinates obtained for the components were interpreted in relation to the variables during different seasonal periods.

Table 2. Summary of the statistics for the variables classified as species

Species	**Component 1**		**Component 2**	
	Coord.	**Contr.**	**Coord.**	**Contr.**
A. flavus	0.389	0.084	-0.055	0.003
A. japonicus	0.222	0.014	-1.130	0.620
A. foetidus	-0.805	0.376	-0.053	0.003
A. sulphureus	0.306	0.016	0.880	0.225
A. ochraceus	-2.584	0.369	0.576	0.032
A. tubingensis	0.748	0.031	0.248	0.006
A. ostianus	0.460	0.111	0.347	0.111

Components 1 and 2 refer to the decomposition of the variability for the first and second components, respectively; Coord refers to the coordinates used for the profile plots; and Contr. refers to the contribution of each species to the formation of components 1 and 2.

Table 3. Summary of the statistics for the variables classified as regions

Region	Component 1		Component 2	
	Coord.	Contr.	Coord.	Contr.
Wet (Arc.)	0.733	0.059	0.897	0.156
Dry (Arc.)	-2.519	0.351	0.515	0.026
Wet (Lum.)	-0.956	0.252	0.070	0.002
Dry (Lum.)	0.364	0.110	0.428	0.267
Wet (Pas.)	0.272	0.055	-0.645	0.545
Dry (Pas.)	-0.943	0.172	-0.105	0.004

Components 1 and 2 refer to the decomposition of the variability for the first and second components, respectively; Coord refers to the coordinates used for the profile plots; and Contr. refers to the contribution of each species to the formation of components 1 and 2. Thus, points located in the same quadrant with greater contributions were determinants for the conduction of the groupings illustrated in circles in Figure 3.

Figure 3 shows that among the *Aspergillus* species, *Aspergillus ochraceus* was associated with the region of Arcos during the period of high rainfall, remaining distant from the other species. The incidence of the isolates of *Aspergillus japonicus* was more highly associated with the region of Passos during the period of high rainfall. The results indicate that there were no associations with the regions or the different periods of rainfall for the *Aspergillus foetidus*, *A. tubingensis*, *A. ostianus*, and *A.flavus*.

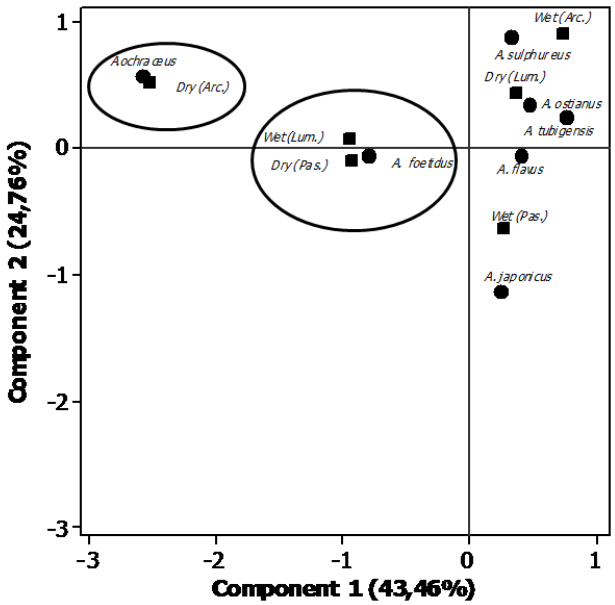

Figure 3. Percentage map for the fungal species of the *Aspergillus* genus based on region and seasonal period

In the percentage map (Figure 3), components 1 and 2 represent the distribution of the species and regions in a bidimensional graph, which explains 68.22% of the sample variability.

The percentage map illustrated in Figure 4 includes the following *Penicillium* species: *Penicillium glabrum*, *P. citrinum*, *P. pinophilum*, *P. simplicissimum*, *P. chrysogenum* and *P. minioluteum.*

Factors such as temperature and rainfall may influence the development of these fungi. The results that justify the groups are described in Tables 4 and 5.

Table 4. Summary of the statistics for the variables classified as fungal species of the *Penicillium* genus

Species	Comp 1		Comp 2	
	Coord	Contr	Coord	Contr
P. citrinum	0.246	0.047	0.187	0.103
P. glabrum	-0.131	0.007	0.371	0.202
P. simplicissimum	0.393	0.078	-0.113	0.024
P. chrysogenum	0.734	0.060	-0.805	0.273
P. minioluteum	0.795	0.035	-1.233	0.320
P. pinophilum	-1.667	0.773	-0.271	0.077

Components 1 and 2 refer to the decomposition of the variability for the first and second components, respectively; Coord refers to the coordinates used for the profile plots; and Contr. refers to the contribution of each species to the formation of components 1 and 2.

Table 5. Summary of the statistics for the variables classified as regions

Region	Comp 1		Comp 2	
	Coord	Contr	Coord	Contr
Wet (Arc.)	-0.378	0.052	0.220	0.066
Dry (Arc.)	0.247	0.005	0.419	0.056
Wet (Lum.)	-0.034	0.001	0.351	0.246
Dry (Lum.)	0.547	0.191	-0.436	0.460
Wet (Pas.)	-2.052	0.703	-0.464	0.136
Dry (Pas.)	0.379	0.048	0.170	0.036

Components 1 and 2 refer to the decomposition of the variability for the first and second components, respectively; Coord refers to the coordinates used for the profile plots; and Contr. refers to the contribution of each species to the formation of components 1 and 2.

The incidence of the *Penicillium pinophilum* species was associated with the region of Passos during the period of high rainfall. Furthermore, *Penicillium chrysogenum* and *Penicillium minioluteum* were associated in the region of Luminarias during the period of low rainfall. *Penicillium glabrum* was associated with the region of Arcos during the period of high rainfall. These groups are shown in Figure 4.

The analysis used in this study highlighted the separation of groups of fungi for some species of *Aspergillus* and *Penicillium* in the sampled areas during the different seasonal periods. However, it should be stressed that the formation of groups that appear heterogeneous may be related to the intrinsic characteristics of the habitat of each analyzed region.

Fungi have long been used in different biotechnology applications to develop new products such as medicines, proteins, hormones and disease-resistant cultivars (Tauk-Tornisielo et al., 2005).

The results indicate that some of the isolated strains are of great importance for human activities (table 6). *Penicillium* fungi are a large component of microbial diversity and offer exciting prospects for the search for biomolecules with

biological and pharmacological potential and other related properties.

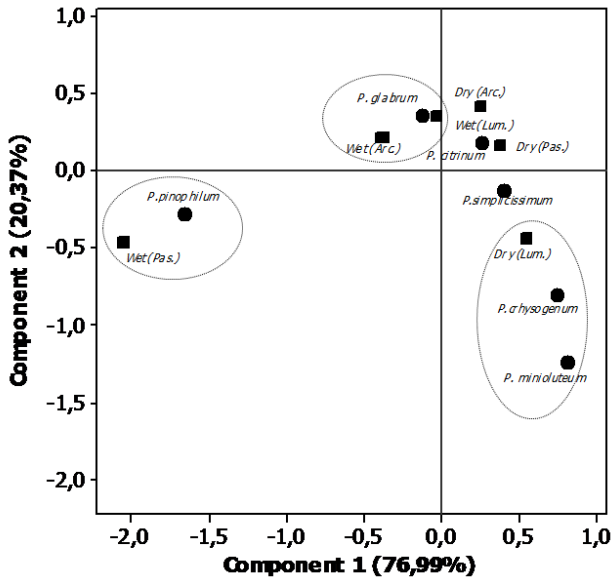

Figure 4. Percentage map for the fungal species of the *Penicillium* genus based on region and seasonal period

One of the most famous and economically important fungal products, penicillin, is produced by *Penicillium chrysogenum* (Frisvad et al., 2004). Compactin, an important cholesterol-lowering agent, is produced by *Penicillium solitum* (Frisvad et al., 2004) and *P. citrinum* (Hoffmeister and Keller, 2007). Mycophenolic acid mofetil, a valuable drug used in heart transplantation to avoid organ rejection, is produced by *P. brevicompactum* (Frisvad et al., 2004). 3-O-methylfunicone (OMF), a secondary metabolite produced by the soil fungus *Penicillium pinophilum*, is a selective inhibitor of breast cancer stem cells (Nicoletti et al., 2008). Brefeldin A, a macrolide antibiotic that was isolated from *Penicillium decumbens* and *P. simplicissimum* (Nicoletti et al., 2008), has a wide varietyof biological properties, including antitumor, antiviral, antifungal, and antimitotic effects (Anadu et al., 2006).

The nephrotoxin citrinin is produced by *Penicillium expansum* (Frisvad et al., 2004), *P. citrinum* (Pitt and Hocking, 2009) and *P. waskmanii* (Nicoletti et al., 2008). The first two species are commonly found on apples and other pomaceous fruits, taproot plants and cereals (Frisvad et al., 2004).

Table 6. Strains identified in this study and the production of important secondary metabolites for human activities

Species	Biotechnology application (BA) / Mycotoxin (M)	Reference
***Aspergillus* Genus**		
A. flavus	Aflatoxin B1 and B2 (M)	Pitt and Hocking (2009)
A. foetidus	Pyranonigrin (M) andantafumicin (M)	Samson el al (2004)
A. japonicus	Cycloclavine (BA) andfestuclavine (BA)	Somma et al. (2012)
A.niger	Ocratoxina (M) and citric acid (BA)	Nielsen et al. (2009)
A. ochraceus	Ochratoxin (M)	Pitt and Hocking (2009)
A. ostianus	Circumdatin D	Pitt and Hocking (2009)
A. sulphureus	Ochratoxin (M)	Pitt and Hocking (2009)
A. tubingensis	Asperazine (BA) and tensidol A (B)	Somma et al. (2012)
***Penicillium* Genus**		
P. brevicompactum	Mycophenolic acid (M)	Frisvad et al. (2004)
P. chrysogenum	Penicillin (BA)	Hoffmeister and Keller (2007)
P. citreonigrum	Citrioviridine (M)	Pitt and Hocking (2009)
P. citrinum	Citrinin (M) andCompactin (BA)	Hoffmeister and Keller (2007)
P. commune	Cyclopaldic acid (BA) andCyclopiazonic acid (M)	Frisvad et al. (2004)
P. corylophilum	Gliotoxin (M)	Nicoletti et al. (2008)
P. decumbens	Brefeldin A (BA)	Nicoletti et al. (2008)
P. expansum	Patulin (M) and Roquefortine C (BA)	Frisvad et al. (2004)
P. glabrum	Gliotoxin (M) andAsterric acid (BA)	Nicoletti et al. (2008)
P. miczynskii	Citreoviridin (M) and Cyclopiazonic acid (M)	Houbraken et al. (2011)
P. minioluteum	Secalonic acid (M)	Nicoletti et al. (2008)
P. pinophilum	3-O-methylfunicone	Nicoletti et al. (2008)
P. raistrick	Griseofulvin (BA)	Nicoletti et al. (2008)
P. simplicissimum	Vermistatin (BA) andBrefeldin A (BA)	Nicoletti et al. (2008)
P. solitum	Compactins (BA) and Solistatin (BA)	Frisvad et al. (2004)
P. varibile	Agrovlavine (M)	Nicoletti et al. (2008)
P. waskmanii	Pyrenocines (BA) and Citrinin (M)	Nicoletti et al. (2008)

The *Aspergillus* genus includes some of the most widespread food and feed contaminants; however, they are also widely used in the biotechnology industry (Nielsen et al., 2009).

Circumdatin D, produced by *A. japonicus*, has been investigated for its ability to inhibit clinical isolates of methicillin-resistant *Staphylococcus aureus* (MRSA) (Suryanarayanan, 2012).

The nephrotoxic and possibly carcinogenic ochratoxins are only produced by *A.niger*, *A. ochraceus* and *A. sulphureus* (Pitt and Hocking, 2009). Aflatoxin B1 is the most potent known natural carcinogen and is the major aflatoxin produced by most toxigenic strains of *A. flavus*.

Mycotoxins that are produced by fruits native to the region or secondary metabolites of pharmacological importance, such as compactin and cycloclavine, can be economically exploited. For this reason, it is important to study the biodiversity of soil microbes in natural areas of the Brazilian cerrado before using these microbes for agricultural production.

4. Conclusions

In conclusion, the results obtained in this study indicate that species of *Aspergillus* and *Penicillium* are naturally present in preserved areas of the Brazilian cerrado. The species *A. ochraceus*, *A. japonicus*, *P. pinophilum* and *P. glabrun* were associated with the rainy season, while *P. chrysogenum* and *P. minioluteum* were associated with drought. This results provide valuable information about the microbiota of *Aspergillus* and *Penicillium*, including significant evidence that these genera are present at relevant frequencies in Brazilian cerrado regions.

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REFERENCES

[1] Anadu NO, Davisson VJ, Cushman M (2006). Synthesis and anticancer activity of brefeldin a ester derivatives. J. Med. Chem. 49 (13): 3897-3905.

[2] Bennet JW (2010). An Overview of the Genus *Aspergillus.*

[3] Cantrell SA, Casillas-Martínez L, Molina M (2006). Characterization of fungi from hypersaline environments of solar salterns using morphological and molecular techniques. Mycol. Res. 110 (8), p. 962-970.

[4] Castro HF, Classen AT, Austin EE, Norby RJ, Schadt CW (2010). Soil microbial community responses to multiple experimental climate change drivers. Appl. Environ. Microbiol. 76(4): 999–1007.

[5] Chávez R, Bull P, Eyzaguirre J (2006). The xylanolytic enzyme system from the genus *Penicillium.* J. Biotechnol. 123(4): 413-433.

[6] Domsch KH, Gams W, Anderson TH (2007). Compendium of Soil Fungi. 2nd Ed. Eching, The Netherlands: IHW-Verlag. 860 p.

[7] Donner M, Atehnkeng J, Sikora RA, Bandyopadhyay R, Cotty PJ (2009). Distribution of *Aspergillus* section *Flavi* in soils of various maize fields among three agroecological zones of Nigeria. Soil Biol. Biochem. 41 (1): 37-44.

[8] Frisvad JC, Smedsgaard J, Larsen TO (2004). Mycotoxins, drugs and other extrolites produced by species in *Penicillium* subgenus *Penicillium*. Stud. Mycol. 49: 201–241

[9] Gams W (2007). Biodiversity of soil-inhabiting fungi. Biodivers. Conserv. 16: 69 – 72.

[10] Houbraken J, Frisvad JC, Samson RA (2011). Taxonomy of *Penicillium* section *Citrina*. Stud. Mycol. 70: 53–138.

[11] Hedayati MT, Pasqualotto AC, Warn PA, Bowyer P, Denning DW (2007). *Aspergillus flavus*: human pathogen, allergen and mycotoxin producer. Microbiol. 153 (Pt6): 1677-1692.

[12] Hoffmeister D, Keller NP (2007). Natural products of filamentous fungi: enzymes, genes, and their regulation. Nat. Prod. Rep. 24(2) :393-416.

[13] Horn BW (2007). Biodiversity of *Aspergillus* section Flavi in the United States: a review. Food Add. Contam. 24(10): 1088-1101.

[14] Houbraken J, Frisvad JC, Samson RA (2011). Taxonomy of *Penicillium* section Citrina. Stud. Mycol. 70(1): 53-138.

[15] Klink CA, Machado RB (2005). Conservation of the Brazilian Cerrado. Conserv. Biol. 19(3): 707-713.

[16] Klich MA (2002a). Biogeography of A*spergillu*s species in soil and litter. Mycol. 94(1): 21-27.

[17] Klich MA (2002b). Identification of Common *Aspergillus* species. Amsterdam: Centraalbureau voor Schimmelautures. 116 p.

[18] Lopes AS (1996). Soils under cerrado: a success story in soil management. Better Crops Internat. 10(2): 9-15.

[19] Moreira FMS, Siqueira J O (2002). Microbiologia e bioquímica do solo. Lavras: UFLA, 625 p.

[20] Nielsen KF, Mogensen JM, Larsen TO, Frisvad JC (2009). Review of secondary metabolites and mycotoxins from the *Aspergillus* group. Anal. Bioanal. Chem. 395: 1225−1242.

[21] Myers N, Mittermeier R, Mittermeier GC, Dafonseca GAB, Kent J (2000). Biodiversity hotspots for conservation priorities. Nature 403(6772): 853–858.

[22] Pandey A, Yadav LS, Singh SK, Singh P, Singh PN, Ravindra1R (2008). Phosphate solubilization by *Penicillium* spp. isolated from soil samples of Indian Himalayan region. World J. Microb. Biot. 24: 97-102.

[23] Peterson SW (2012) *Aspergillus* and *Penicillium* identification using DNA sequences: barcode or MLST? Applied Microbiology and Biotechnology, 95:339–344.

[24] Pfenning, L. H.; Abreu, L. M. Diversity of microfungi in

tropical soils. In: MOREIRA, F. .S.; SIQUEIRA, J. O.; BRUSSARD, L. (Ed.). Soil biodiversity in Amazonian and other Brazilian Ecosystems. Wallingford: CABI, 2006. v. 1, p. 184-205.

[25] Pitt, J. I. A laboratory guide to common *Penicillium*s pecies. Food Science Australia a Joint Venture of CSIRO and AFISC, 2000. 197 p.

[26] Pitt, J. I. et al. Mycotoxins and toxigenic fungi. Medical Mycology, Oxford, v. 38, n. 1, p. 41-46, 2000.

[27] RatteR, J. A.; Ribeiro, J. F.; Bridgewater, S. The Brazilian cerrado vegetation and threats to its biodiversity. Annals of Botany, London, v. 80, p. 223-230, 1997.

[28] RAzzaghi-abyaneh, M. et al. A survey on distribution of Aspergillus section Flavi in corn field soils in Iran: populations patterns based on aflatoxins, cyclopiazonic acid and sclerotia production. Mycophatologia, Heidelberg, v. 161, 183-192, 2006.

[29] Ritz K, Young IM, 2004. Interaction between soil structure and fungi. Mycologist 18: 52–59.

[30] Samson, R. A.; Frisvad, J. C. *Penicillium* subgenus *Penicillium:* new taxonomic schemes, mycotoxins and other extrolites. Studies in Mycology, Utrecht, v. 49, p. 1-251, Jan. 2004.

[31] Samson, R. A.; HOEKSTRA, E. S.; FRISVAD, J. C. Introduction to food and airborne fungi. 7th ed. Utrecht: Centraalbureauvoor Schimmelcultures, 2000. 389 p.

[32] Samson, R. A.; VARGA, J. Aspergillus systematic in the genomic era. Utrecht: CBS Fungal Biodiversity Centre, 2007. 207 p.

[33] Samson, R. A; VARGA, J. What is a species in Aspergillus? Medical Mycology, Oxford, v. 47, p. 13-20, 2009. Suppl.

[34] Samson RA, Houbraken JAMP, Kuijpers AFA, Frank JM, Frisvad JC (2004). New ochratoxin or sclerotium producing species in *Aspergillus* section *Nigri*. Studies in Mycology 50: 45–61.

[35] Samson RA, Yilmaz N, Houbraken J, Spierenburg H, Seifert KA, Peterson SW, Varga J, Frisvad JC (2011) Phylogeny and nomenclature of the genus Talaromyces and taxa accommodated in *Penicillium* subgenus *Biverticillium*. Stud Mycol 70:159–184.

[36] Samson, R. A., Visagie, C. M., Houbraken, J., Hong, S. B., Hubka, V., Klaassen, C. H., ... & Frisvad, J. C. (2014). Phylogeny, identification and nomenclature of the genus *Aspergillus*. Studies in Mycology, 78, 141-173.

[37] Somma, S.; Perrone, G.; Logrieco, A.F. 2012. Diversity of black *Aspergilli* and mycotoxin risks in grape, wine and dried vine fruits. Phytopathol. Mediterr. 51, 131–147.

[38] Tauk-Tornisielo, S.M., Garlipp, A., Ruegger, M., Attili, D.S., Malagutti, E., 2005. Soilborne filamentous fungi in Brazil. J. Basic Microbiol. 45, 72–82.

[39] Varga, J. et al. Molecular diversity of agriculturally important *Aspergillus* species. European Journal of Plant Pathology, Dordrecht, v. 110, p. 627–640, 2004.

[40] in soil. Journal of Soils and Sediments, Amherst, v. 8, p. 379-388, 2008.

[41] Visagie, C. M., Houbraken, J., Frisvad, J. C., Hong, S. B., Klaassen, C. H. W., Perrone, G., ... & Samson, R. A. (2014). Identification and nomenclature of the genus *Penicillium*. Studies in Mycology, 78, 343-371.

[42] ZHELIFONOVA, V.P. et al. Secondary Metabolites in Taxonomy of the *Penicillium* Fungi. Microbiology, New York, v. 79, n. 3, p. 291-300, 2010.