

Polyporoid fungi (Agaricomycetes, Basidiomycota) in the Estação Científica Ferreira Penna (State of Pará, Brazilian Amazonia): diversity and ecological aspects

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Four field trips to the Estação Científica Ferreira Penna, an area of pristine Amazonian Forest in the State of Pará in Brazil, were undertaken on July 2006, March and August 2007, and February 2008, including the dry and rainy seasons. Six transects (called plot and TAB) of 20×1000 m were surveyed in each field trip, totalising 24 visits. In the first 500 m, all fallen, dead trees longer than 2 m and larger than 0,1 m were labelled and their measures, decay stage and species were taken. Ninety-six species of polyporoid fungi were identified, five of them new to science, 10 new to Brazil, and 39 new to the Brazilian Amazonia.

Despite the high number of species, the cumulative species curve was not stabilised. The similarity among transects varied from 40.9 to 62.1%, being plot 2 the most dissimilar to the other plots, and it was mostly affected by the seasons. The highest number of occurrences and of species was observed in less decayed and larger dead trees.

1 Introduction

According to [1], the Kingdom *Fungi* is divided in four phyla: *Chytridiomycota*, *Zygomycota*, *Ascomycota* and *Basidiomycota*. The latter is subdivided in three classes: *Basidiomycetes*, *Urediniomycetes* and *Ustilaginomycetes*. The former groups two subclasses, *Tremellomycetidae* and *Agaricomycetidae*, in which the polyporoid fungi can be found. According to [2], the polyporoid fungi or polypores are those belonging to the family *Polyporaceae* of the ancient order *Aphylliphorales*, and are mostly characterised by the poroid hymenial surface. However, other fungi may also show this kind of hymenial surface and may be also included in the polyporoid group, an assembly with no evolutionary relation since the poroid condition seems to have developed many independent times during basidiomycete evolution [3]. Currently, the polypores are spread among different orders of *Agaricomycetes* such as *Corticiales*, *Gloeophyllales*, *Hymenochaetales*, *Polyporales*, *Russulales*, and *Trechisporales* [4].

The polypores are generally characterised by the production of holobasidia, i.e. basidia without septa, in a well-defined, non lamellate hymenium. The basidioma (also called basidiocarp or fruitbody), where basidia are produced, have a strongly variable colour, consistency and morphology [2, 5, 6]. The hymenium is a continuous layer where the fertile structures, such as basidia and basidiospores, and also sterile structures, as cystidia and hyphal pegs, are produced. In the polypores, the hymenium is in general tubular, opening as pores in the hymenial surface, although lamellae and elongated pores may also occur [2, 5, 6]. The basidiospores are not repetitive, are forcibly discharged and therefore called ballistospores and their morphology may be extremely variable [2, 5, 6]. The hyphal system can be monomitic, when only generative hyphae are present; dimitic, when generative and skeletal hyphae, or seldom generative and binding hyphae are present; or trimitic, when the all three types of hyphae are present in the basidioma. The generative hyphae may also be clamped or simple septate [2, 5, 6].

Species of polyporoid fungi are mainly saprobes on dead wood, but many are dangerous parasites, especially in different tree species [7, 8]. They are considered the major wood decomposers, playing crucial role in nutrient cycling, releasing among other things carbon originally removed from the atmosphere by autotrophic organisms in arboreous and shrubby ecosystems [5, 6, 7, 8]. The saprotrophic species are generally divided in two major groups, those which cause white rot and those causing brown (cubical) rot.

Species in the first group are the majority and are the only organisms known to be able to degrade lignin. They also remove cellulose and hemicelluloses from wood, leaving the substrate with a whitish and spongy aspect. The brown-rot fungi are able only to degrade cellulose and hemicelluloses, reducing the substrate to brownish residuum of lignin. Since the decay of the wood is irregular, the wood cracks in cubic pieces [5, 6, 8, 9]. Some species are able to penetrate lesions or similar openings caused by broken tops, lighting etc. in living trees and then attack the heartwood and in the end causing tree weakening or death [7, 9].

Many species can be used as indicators of disturbance in native forests because they are more frequent in less affected areas and prefer to colonize larger and more decomposed logs, which become less frequent with selective logging [10, 11, 12]. This aspect is very important for evaluation of fungal diversity because loss of plant diversity can lead to loss of fungal diversity. Other aspects that can affect polypore diversity in ecosystems are the diversity, size and quality of plant species [12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22], and the rainfall gradient [12, 23].

Due to the flexibility of the enzymatic apparatus, many species of polyporoid fungi are being tested in bioremediation processes of dyes [24, 25, 25, 27, 28], olive oil mill wastewaters [29, 30], pesticides [31, 32, 33], polycyclic aromatic hydrocarbons [34], among others, making evident their biotechnological potential. Extracts of polypores species have been and are still tested for anti-viral activity against HIV-1 [35, 36], and also for anti-bacterial and anti-fungal activity [37, 38, 39, 40], with promising results. Some others are used as improvers of the immunological system and in cancer therapy [41, 42, 43].

2 Materials and methods

After a preliminary visit to the ECFPn on January 2006, six transects delimited as pre-existent footpaths in “terra firme” ecosystem, with approximately 20 m width \times 1000 m length, were investigated on July 2006, March and August 2007, and February 2008 (plot 1: 51°27'34.35" W and 1°42'24.09" S; plot 2 51°29'0.73" W and 1°43'43.24" S; plot 3 51°30'38.62" W and 1°43'59.21" S; plot 4 51°31'14.72" W and 1°45'12.84" S; “TAB-D” and “TAB-E”: 51°27'17.75" W and 1°44'12.96" S, both behind the buildings of the scientific base). In the first 500 m of the transects, all trees longer than 2 m and wider than 0,1 m were labelled and had their vulgar name, length and diameter, and decay stage registered. When possible, living material equivalent to the dead substrate was collected and identified by the parataxonomists of the Museu Paraense Emílio Goeldi (MPEG). The decay stage was evaluated in accordance to the scale of 1 to 3 defined by [44, modified from 45], in which the stage 1 the wood is recently fallen and thus still rigid, and a knife penetrates 2 mm maximum using hand strength; in the stage 2 the knife penetrates easily 2-20 mm, and in the 3 the wood is fragile and the knife penetrates easily through the log. Additional sites, in general in the way to the transects such as areas of secondary forest (“capoeira”) and the area of the Esecافلor experiment, were also investigated.

The basidiomata were put in stove at 45-50°C during approximately five days [46]. Later on, they were macroscopically analysed, according to size (length, width, thickness) and colour of abhymenial and hymenial surfaces, tubes, context, and margin, when present [47].

Free-hand slices of the abhymenial and hymenial surfaces, and context were made with razor blade for microscopic observation of the material. The slices were placed between glass slides and cover slips with potassium hydroxide 3-5% and stained with phloxine 1%, Amann Blue, and Melzer reagent [5]. These biochemical characteristics, as well as the micro- and macroscopic characters were used for identification and elaboration of keys for families, genera and species.

For the identification of the material, the following bibliography was used: [5, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57]. The terminology followed [1] and the classification the CABI (www.indexfungorum.org) and CBS (www.cbs.knaw.nl/databases) databases.

After identification, all specimens were deposited in the herbarium MG (Museu Paraense Emílio Goeldi). When possible, duplicates were incorporated to the herbaria URM (Departamento de Micologia, Universidade Federal de Pernambuco), and O (University of Oslo).

The Index of Bray-Curtis was used for evaluation of the similarity among areas in accordance to polypore richness. The Analysis of Similarities (ANOSIM) was used to test if rainy and dry seasons present significant differences considering polypore associations [58]. The χ^2 test evaluated if the occurrence of polypores was related to quality (plant species and plant families, and decay stage) and quantity (radius, diameter, area, and volume) of wood [59]. All analysis used 0.05 as significance level.

3 Results and discussion

Nine families *sensu* [1], 39 genera and 96 species of polyporoid fungi were identified. Even though the area had been surveyed before [60, 61], five new species, 39 new records to the Brazilian Amazonia and 10 to Brazil were found. This fact could be due to the lack of experts on this group of fungi in the area and the lack of standardised collections. Despite the high number of species, the species accumulation curve has not reached the asymptote, as observed by [13, 23, 62].

The most diverse family was *Polyporaceae*, with higher number of genera and species (22 e 48, respectively), followed by *Ganodermataceae* (four and 16) and *Hymenochaetaceae* (four and 14). *Steccherinaceae* was represented by four genera and six species, and *Meripilaceae* by one genus and seven species. *Bondazeriaceae*, *Grammothelaceae*, *Hapalopilaceae*, *Meruliaceae* and *Schizoporaceae* were represented by one genus and one species each. The higher number of species observed for *Polyporaceae*, *Hymenochaetaceae* and *Ganodermataceae* was expected, because they present higher specific diversity compared to the other families of poroid Basidiomycetes [1], and the data obtained agree with studies that include polypores in Brazil [63, 64, 65].

Among the registered genera, 26 had just one species, being five them monospecific genera, and four of them represented, in the neotropics, by one species each. *Amauroderma* and *Phellinus* were the genera with higher number of species (11 each), followed by *Perenniporia* (nine) and *Rigidoporus* (seven), *Polyporus* (six), *Trichaptum* (five) and *Trametes* (four), which was expected for large genera in the Americas or in the tropics [48, 49, 50, 53].

The number of occurrences and species was related to quality (decay stage) and quantity (radius, diameter, area, and volume) of wood. Occurrences were observed more than expected in the decay stage 1 and less than expected in decay stage 3 ($\chi^2 = 11.075$, $df = 2$, $p < 0.05$). The same pattern was observed to the number of species, but it was not statistically significant ($\chi^2 = 1.582$, $df = 2$). Less decayed wood has more of the easily decomposing sapwood, and could support more fungi [23, 62].

Occurrences were also observed more than expected in the radius classes 5 (0.175 to 0.208 m) and 8 (0.278 to 0.6 m) ($\chi^2 = 21.38$, $df = 7$), in the diameter classes 5 (0.326 to 0.391 m) and 9 (0.586 to 1.2 m) ($\chi^2 = 26.535$, $df = 8$), in the length class 10 (17.391 to 20.870 m) ($\chi^2 = 33.322$, $df = 11$), in the area classes 4 (16.087 to 22.609 m²) and 5 (22.608 to 29.13 m²) ($\chi^2 = 22.342$, $df = 5$), and in the volume class 5 (4.521 to 24.808 m³) ($\chi^2 = 21.148$, $df = 4$). The number of species showed similar pattern, being observed more than expected in the length class 10 ($\chi^2 = 36.38$), area classes 4 and 5 ($\chi^2 = 19.754$), and in the volume classes 4 (3.217 to 4.521 m³) and 5 (4.521 to 24.808 m³) ($\chi^2 = 20.870$). The number of species was more observed than expected also in the radius class 5 and diameter class 9, but the values of χ^2 were not significant ($\chi^2 = 13.633$, and $\chi^2 = 15.331$, respectively). The number of occurrences and species was observed less than expected in the area class 1 (0.958 to 3.043 m²) ($\chi^2 = 22.342$, and $\chi^2 = 19.754$, respectively), and volume area 1 (0.024 to 0.608 m³) ($\chi^2 = 21.148$, and $\chi^2 = 20.870$, respectively). The number of species was observed less than expected also in the length class 2 (3.478 to 5.217 m) ($\chi^2 = 36.38$). The same pattern was observed by [23], which could also be related to the amount of resource of the substrate.

The number of occurrences of polypores was higher than expected on the plant species *Guatteria* sp. (Annonaceae), *Licania* sp. 3 (Chrysobalanaceae), *Pseudopiptadenia* sp. (Fabaceae) ($\chi^2 = 56.68$, $df = 29$), and on the plant family Chrysobalanaceae ($\chi^2 = 34.710$, $df = 29$), and less than expected on *Vouacapoua americana* (Fabaceae) ($\chi^2 = 56.68$, $df = 29$) and on Fabaceae ($\chi^2 = 34.710$, $df = 29$). The number of

species was higher than expected on the plant species *Guatteria* sp. (Annonaceae), and *Pseudopiptadenia* sp. (Fabaceae) ($\chi^2 = 56.68$, $df = 29$), and less than expected on *Vouacapoua americana* (Fabaceae) ($\chi^2 = 58.02$, $df = 29$), which could indicate more or less susceptibility of the wood to fungi colonization, but not host-specificity, which could not be tested since none of the species had more than 20 occurrences on the labelled trees. However, the lack of host-specificity is expected for tropical wet forests where plant communities are highly diverse and natural selection can act against specificity that limits colonization of widely spaced hosts [13, 66, 67].

Similarity between transects (plots) varied from 40,9 to 62,1%, being plot 2 the most dissimilar to the other plots. Similarity was considered to be high, and it was mostly affected by the season ($R_{global} = 0.563$, number of permutations = 729, $p = 0.008$), as observed by [12, 23, 62].

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