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EFFECTS OF WOOD ON THE GROWTH OF LIGNICOLOUS FRESHWATER FUNGI

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Abstract

The addition of cottonwood and ash sapwood powder to YPSS/2 medium influenced the mycelial growth of 13 species of freshwater lignicolous Ascomycetes and Hyphomycetes. These species fall into three groups with respect to their responses to the presence of leached or non-leached wood powder. Neither wood type showed strong inhibitory effect on the mycelial growth of the species. Leaching did not affect the response of the species to the presence of wood powder.

Introduction

Terrestrial woody debris is an important component of freshwater ecosystems. It serves as a relatively stable source of energy for aquatic organisms [1]. In streams, woody debris serves as a potentially rich source of fixed carbon available to many species of freshwater fungi and other heterotrophs. Many studies have indicated the degradative ability of freshwater Hyphomycetes [2-5]. Degradative ability of fungi varies with: 1) nutritional status (both quality and quantity) of the resource (e.g. wood); 2) the physico-chemical characteristics of the habitat; 3) the chemical characteristics of the water; and 4) interactions within the decomposer community.

Wood quality is determined by both the chemical and anatomical composition of wood, and affects the degradative ability of lignicolous fungi [4-7]. Wood is composed of carbohydrates (such as cellulose, hemicellulose and starch), lignin, extractives and some minerals.

The inhibitory effects of pine and oak wood on the growth of aquatic and aeroaquatic Hyphomycetes have been studied [8]. Heartwood has more growth inhibitors than sapwood [9]. Since in previous studies on the enzymatic and degradative abilities of freshwater Ascomycetes and Hyphomycetes sapwood of ash and cottonwood was used [4], this study was carried out to determine whether sapwood stimulated or inhibited the growth of selected species of freshwater Ascomycetes and Hyphomycetes.

Keywords: Freshwater fungi; Hyphomycetes; Ascomycetes

Materials and Methods

Seven Ascomycetes, five Deutermycetes (Hyphomycetes) and one Oomycete were selected for the study. These fungi were: *Discomycete* sp. C-75-1; *Leptosphaeria* sp., J-52-1 ATCC 44622; *Nais inornata* Kohlm., CS-83-1 ATCC 32104; *Nectria haematococca* Berk. & Br., J-135-1; *N. lucidum* Hohen., J-51-1; *Pseudohalonectria lignicola* Minoura & Muroi, J-13-1 ATCC 44723; *Pseudohalonectria* sp. 1, CS-656-1 ATCC 52674; *Anavirga dendromorpha* Descal & Sutton, CS-702-1; *Clavariopsis aquatica Filosporella annelidica* (Shearer & Crane) Crane & Shearer, S-77-1 ATCC 32834; *Heliscus lugdunensis* Sacc. & Therry, S-193-1 ATCC 34624; *Pyramidospora* sp. CS-674-1 ATCC 34700; *Trichocladium lignincola* Schmidt, J-39-1; and *Pythium* sp. CS-807-1.

All of these species were selected from submerged wood except for *C. aquatica, F. Annelidica* and *H. lugdunensis* which were isolated from submerged leaves. The fungi were maintained at 4°C on slants of half-strength Emerson's yeast soluble starch agar (YPSS) [10] overlaid with a strip of balsa wood in the dark.

To determine whether sapwood influenced the growth of species, a medium containing various concentrations of leached and non-leached sapwood powder was used. Wood powder was prepared by grinding in a Wiley mill (40 mesh screen) sapwood of 25-30 year old newly fallen trees of ash (*Fraxinus pennsylvanica* L.) and cottonwood (*Populus deltoides* L.). Leached wood powder was prepared by running tap water through the wood powder for 96 hours

and deionized water for 12 hours. Petri dishes containing 25ml of YPSS medium were supplemented with 0.2, 0.4, or 0.8 g each of leached or non-leached sapwood powder of ash or cottonwood. After sterilization for 15 minutes at 120°C, 15lb medium was poured into glass petri dishes and the dishes were swirled to spread the wood powder evenly.

Duplicate petri dishes were inoculated centrally with 4mm. diam. disks of fungal mycelium cut from margins of colonies growing actively on corn meal agar (CMA) (Difco). Colony diameters were measured along three intersecting diameters every two days for 12 days. Controls were fungal disks in YPSS medium without wood powder.

Results

All species grew in medium containing sapwood powder. For six and seven of the 13 species tested, colony diameters were greater on media containing ground wood of cottonwood and ash than on the control medium, respectively. Four and five of the species, however, showed reduced colony diameters on ash and cottonwood, respectively. The growth of *T. lignincola* on media with non-leached and leached cottonwood powder was similar to that on its control medium. Colony diameters of more than half of the species increased with increased concentration of wood powder in the medium. In more than half of the species, colony diameters were greater in media with leached wood powder than on media with non-leached wood powder (Tables 1 and 2). Wood type had minimal effect on colony diameter.

Discussion

Species fall into three groups with respect to the presence of leached or non-leached cottonwood and ash wood powder in the medium. The first group displayed reduced radial growth rate and includes *N. inornata, N. haematococca*, and *Pythium* sp.; the second group displayed substantially increased radial growth rate and includes *Pseudohalonectria* sp. 1, *H. Lugdunensis*, and *Pyramidospora* sp.; and the third group displayed a slight or no change in their radial growth rates and includes the remaining seven species tested. The fact that the growth of more species was not inhibited may be because the wood powder used was obtained from sapwood of newly-fallen trees. Sapwood does not contain as much or as many of the inhibitory chemicals found in heartwood, but contains instead numerous substrates that support the growth of a wide variety of wood-degrading fungi [9]. These substrates include starch, soluble monosaccharides, lipids, proteins, peptides, amino acids, nucleic acids, and growth factors such as thiamine [9]. Zare-Maivan [4] and Zare-Maivan and Shearer [5,6] have shown the ability of these fungi to use many of the above substrates as their carbon source. Since all of the species tested positive for starch degradation [4,5], and considering the presence of starch in the sapwood of ash and cottonwood, increased radial growth is to be expected.

Table 1

Colony Diameters (mm) of Species on Control and Various Concentrations of Non-Leached and Leached Ash Sawdust after 12 Days, at 25°C in the Dark.

Species	CONTROL	NON-LEACHED			LEACHED		
	0.0	0.2	0.4	0.8	0.2	0.4	0.8
Discomycete sp. C-75-1	6.0[2]	4.0	5.0	5.0	4.5	5.0	5.0
Leptosphaeria sp. J-52-1	12.0	9.0	7.0	8.0	10.0	11.0	11.0
Nais Inornata	9.0	8.0	7.0	5.0	5.0	6.0	5.0
Nectria Haematocooc a[b]	49.0	50.5	52.0	53.0	48.0	43.5	57.5
N. Lucidum	32.0	31.0	31.0	26.5	33.0	30.0	33.5
Pseudohalonectria Lignicola	8.0	13.0	15.0	15.0	13.0	13.0	14.5
Pseudohalonectria sp.1	14.0	19.0	23.5	27.5	24.0	23.0	26.0
Anavirga Dendromorpha	20.0	18.0	21.0	23.0	21.0	23.0	24.0
Filosporella Annelidica	8.0	11.0	9.0	11.0	10.0	11.0	13.0
Heliscus Lugdunensis[b]	22.0	38.0	40.5	41.0	41.5	29.0	41.5
Pyramidospora sp. CS-704-1	5.0	12.5	15.0	21.0	16.5	16.0	20.5
Trichocladium Lignincola	3.0	3.0	2.5	1.0	2.2	2.0	2.0
Pythium sp. Cs-807- 1[c]	40.0	40.5	40.5	22.5	41.0	36.0	38.0

(a) = Values are average of three colony diameters in each of two replicates; (b) = Colony diameter after six days; (c) = Colony diameter after four days

Table 2

Colony Diameters (mm) of Species on Control and Various Concentrations of Non-Leached and Leached Conttonwood Sawdust after 12 Days, at 25°C in the Dark.

Species	CONTROL	NON-LEACHED			LEACHED		
	0.0	0.2	0.4	0.8	0.2	0.4	0.8
Discomycete sp. C-75-1	6.0	4.0	5.0	5.0	4.5	5.0	6.0
Leptosphaeria sp. J-52-1	12.0	10.5	11.0	10.5	10.5	11.0	10.5
Nais Inornata	9.0	10.0	5.0	5.0	5.0	10.0	LD
Nectria Haematocooc a[b]	49.0	48.5	51.0	53.0	50.0	48.0	51.5
N. Lucidum	32.0	32.0	28.5	28.5	33.5	29.5	33.5
Pseudohalonectria Lignicola	8.0	11.0	11.0	11.5	10.5	11.0	13.0
Pseudohalonectria sp.1	14.0	23.0	19.5	22.0	18.5	19.5	23.0
Anavirga Dendromorpha	20.0	19.5	18.0	21.0	20.0	16.0	19.0
Filosporella Annelidica	8.0	10.0	11.0	10.0	10.0	12.0	11.0
Heliscus Lugdunensis[b]	22.0	37.5	30.0	40.0	40.5	41.0	46.0
Pyramidospora sp. CS-704-1	5.0	16.0	13.0	21.0	13.0	14.5	26.0
Trichocladium Lignincola	3.0	2.0	3.0	4.0	3.0	2.0	3.0
Pythium sp. Cs-807- 1[c]	40.0	36.0	41.0	38.5	37.0	39.0	41.0

(a) = Values are average of three colony diameters in each of two replicates; (b) = Colony diameter after six days; (c) = Colony diameter after four days; (LD)= Lost data

In studying the inhibitory effects of pine and oak wood powder on the growth of aquatic lignicolous Hyphomycetes and aeroaquatic Hyphomycetes, Gunasekera and Webster [8] reported inhibition of growth in many species, especially by non-leached wood. Although they do not indicate which part of the wood they used, from the degree of inhibition it is deduced that it was heartwood.

Even though a few species showed greater growth rates in dishes supplemented with more or leached wood powder, the differences were not great enough to be conclusive. However, the fact that these fungi live in water, and considering the presence of lower concentrations of leached growth substances in water, may indicate the possible adaptive ability of these fungi to grow in conditions of minimal nutrients. This ability, along with the enzymatic and degradative abilities of these fungi, as well as their ability to grow in a wide range of temperatures [4], enables them to competitively occupy a resource and hold on to resources already occupied. Furthermore, many of the fungi tested displayed antagonistic activity against other saprophytic fungi [11].

References

1. Anderson, N.H. and Sedell, J.R. Detritus Processing by Microinvertebrates in Stream Ecosystem. *Ann. Rev. Entemol* **24**: pp. 351-377; (1979).
2. Leightley, L.E. and Eaton, R.A. Mechanisms of Decay of Timber by Aquatic Microorganisms. Br. Wood Preserv. Asso. Ann. Conv. pp. 1-26. (1977).
3. Jones, E. B. G. Observations on the Ecology of the Lignicolous Aquatic Hyphomycetes. pp. 731-742. In: The Fungal Community: Its Organization and Role in the Ecosystem. Eds., D.T. Wicklow and J.C. Carroll: Marcell Dekker. New York. (1981).
4. Zare-Maivan, H. Extracellular Enzyme Production and Wood Degradation by Freshwater Lignicolous Fungi. Ph.D. Thesis. University of Illinois. Urbana, II. USA. (1987).
5. Zare-Maivan, H. and Shearer, C.A. Extracellular Enzyme Production and Cell Wall Degradation by Freshwater Lignicolous Fungi. *Mycologia*: **80**: pp. 365-375. (1988).
6. Zare-Maivan, H. and Shearer, C.A. Wood Decay Activity and Cellulase Production by Freshwater Lignicolous Fungi. *Intern. Bioderioration*. **24**: pp. 459-474; (1988 b.)
7. Merrill, W. and Cowling, E.B. Role of Nitrogen in Wood Deterioration: Amounts and Distribution in Tree Stems. *Can. J. Bot*. **44**: pp. 555-580; (1966).
8. Gunasekera, S.A. and Webster, J. Inhibitors of Aquatic and Aeroaquatic Hyphomycetes in Pine and Oak Wood. *Trans. Br. Mycol. Soc*. **80**: pp. 121-125; (1983).
9. Scheffer, T.C. and Cowling. Natural Resistance of Wood to Microbial Deterioration. *Ann. Rev. Phytopathol*. **4**: pp. 147-170, (1966).
10. Fuller, M.S. *Media*. pp: 196-208. In: Lower Fungi in the Laboratory. Ed., M.S. Fuller. Palfrey Contributions in Botany, No. 1. Dept. of Botany. University of Georgia Press. Athens. GA. USA. (1978).
11. Shearer, C.A. and Zare-Maivan, H. *In vitro* Hyphal Interactions among Wood-and Leaf-Inhabiting Ascomycetes and Fungi Imperfecti from Freshwater Habitats. *Mycologia*. **80**: pp. 31-37; (1988).