THERMOPHILIC AND THERMOTOLERANT FUNGI INDUCED BY SAWDUST MIXTURES (DECAYED LAUREL AND CEDAR TREES) AT THE AMAZONIAN REGION

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ABSTRACT.

Following a sucessive dilution method for screening, 634 strains were isolated from which **Aspergillus** (37.5%), **Penicillium** (32.5%), **Gliocladium**(14.0%), **Trichoderma** (8.0%), **Paecilomyces** (3.3%) and **Fusarium** (1.9%) were characterized. **A. fumigatus** and **A. niger** were the most commun among **Aspergillus**. **P. janthinellum**, **P. verruculosum**, **P. restrictum** and **P. wasksnamii** were the most commun among **Penicillium**. For the simplified perfusion method 702 strains were isolated and 457 strains were identified. The strains distribution were the following: **Aspergillus** (32.4%), **Fusarium** (29.5%), **Gliocladium** (18.8%) and **Penicillium** (4.6%). **A. fumigatus** (82.0%) was the fungi with high frequency among the **Aspergillus** and **F. solani** (80.4%) among **Fusarium**. Both methods led to monitoring the population of different kind of fungi growing in sawdust from Amazonian woods.

INTRODUCTION

Thermophilic fungi are those species that have a maximum temperature for growth at or above 50°C, a thermotolerant fungi share this ability to grow at 50°C but also grow at 20°C. The thermophilic species are most frequently found in self-heating environments such as sawdust (e.g. Brazil) (1). The thermophilic grows on sawdust, and their ability to degrade cellulose confirm the destructive role on woods. However, the fungi could be useful, specially in the pulp and paper industry. There has been an increase interest for xylanase, cellulases and ligninases in the latter industry. In a previous study a significant modulation of enzymes production by the carbon source was found in the extracted fungi from self-heated industrial chips pile at the Amazonian region (2).
The present study documents the occurrence of thermolphilic and thermotolerant fungi in self-heating wood sawdust of laurel and cedar trees in Manaus, and its isolation and characterization were performed. The fungi were collected following the

Sucessive Dilution (SD) and Simplified Perfusion (SP) methods (2,3).

MATERIALS AND METHODS

Microorganisms.
Thermotolerant and thermophilics fungi were isolated from a sawdust accumulation of Louro (**Ocotea cimbarum**) and Cedro (**Cedrella odorata**) from Manaus Region, Amazonia. The Dilution (4) and Simplified Perfusion (3) Techniques wee selected for the screening procedure.
Identification.
The fungi identification was carried out following especialized literature (5-9).
Growth.
Aspergillus, Penicillium, were cultured in yeast-starcha-agar (YPSS), yeast-glucosr-agar (YG) and agar-Czapek (CZ).
Chemical Analyses.
Sawdust was analyzed to humidity, pH, total nitrogen, metabolizable phosphorous, magnesium ions content, iron content by standar methods (10). Cellulose (32.6%), lignin (38.3%), hemicellulose (9.8%) and extractives (2.0%) were analyzed by standard methods (2,11).

RESULTS AND DISCUSSION

A) DILUTION TECHNIQUE.

Related to the substrate temperature there were variation of 28.7°C-30.5°C and 2.8 to 5.6, respectively. Humidity registered in the collection periods (july-september-November) were around 75.4%, 50.5% and 73.9%, respectively. At these conditions fungal growing in the sawdust pile led the substrate to degrade similarly as previously observed (12). From 36 sawdust samples extracted from different depths led us to select 4.721 colonies and to isolate 634 strains (TABLE 1).

TABLE 1. PURIFIED SAMPLES IN SELECTIVE CULTURE MEDIUM (a)

COLLECTION	JULY			SEPTEMBER			NOVEMBER			TOTAL
CULTURE MEDIUM	YPSS	YG	CZ	YPSS	YG	CZ	YPSS	YG	CZ	
28°	33	37	32	46	34	33	43	47	45	350
40°	21	22	06	40	29	34	36	33	25	246
50°	02	-	-	03	06	05	02	14	06	38
STRAIN NUMBERS	56	59	38	89	69	72	81	94	76	634

a) YPSS: yeast-starch agar; YG: Yeast-glucose agar; CZ: agar czapek.

At room temeprature more colonies in November than the other months (52.5%) were collected. Colony Forming Units (CFU) was

high in the three periods in YG medium. The YPSS and CZ media demonstrated also viability for these kind of fungi but in low frequencies.
Comparing the total isolated strains the fungi distribution were: **Aspergillus** (37.5%), **Penicillium** (32.5%), **Gliocladium** (14.0%), **Trichoderma** (8.0%), **Paecilomyces** (3.3%) and **Fusarium** (1.9%). Within the identified species **Aspergillus fumigatus** e **Aspergillus niger** were the most abundant and with higher frequencies than other species. **P. verruculosum**, **P. janthinelum**, **P. restrictum** and **P. waksmanii**. **Gliocladium virens** and **Paecilomyces varioti** exhibited high frequency among the filametous fungi.

B) PERFUSION TECHNIQUE.

Through the modified perfusion technique (13) were isolated a representative mycotic flora as described above. These samples were colleted in August, October and December. The pH varied from 3.2 to 6.1 and temperature from 27.0° to 33.1°. In this experiment 702 fngal samples were collected from which 457 were identified. (TABLE 2).

TABLE 2. PURIFIED SAMPLES FROM SELECTIVE MEDIUM (a)

COLLECTION	AUGUST			OCTOBER			DECEMBER			TOTAL
CULTURE MEDIUM	YPSS	YG	CZ	YPSS	YG	CZ	YPSS	YG	CZ	
28°	40	53	45	32	23	38	26	10	16	283
40°	25	39	14	15	18	19	10	07	17	164
50°	-	02	01	-	01	-	-	03	03	10
STRAIN NUMBERS	65	94	60	47	42	57	36	20	36	457

a) YPSS: yeast-starch agar; YG: Yeast-glucose agar; CZ: agar czapek (cellulose).

From the identified strains **Aspergillus** (32.4%), **Fusarium** (29.5%), **Gliocladium** (18.8%) and **Penicillium** 94.6%) were the most important genera. **A. fumigatus** (81%) was the more abundant species within the **Aspergillus**, **F. solani** (80.4%) between **Fusarium**, **G. viride** (53.5%) from **Gliocladium** and **P. waksmanii** (19%) from the **Penicillium**.
In summary, 1091 strains were isolated and idenfified in this fungal screening from sawdust pile in the Manaus region. Laurel as Cedar sawdust appeared as a good source for thermophilic and thermotolerant fungi with great potentiality in industrial processes. Cellulases, ligninases and xylanases activities measurements are under progress.

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