A COMPARISON OF DECOMPOSITION OF JACK PINE AND TREMBLING ASPEN WOOD BLOCKS BY EIGHT WOOD DECAY FUNGI

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Dr. Leonard Hutchison	Dr. Mathew Leitch
Major Advisor	Second Reader

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ABSTRACT

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Keywords: *Bjerkandera adusta*, conifer, decay, *Fomitopsis cajanderi*, *Fomitopsis pinicola*,*Ganoderma applanatum*, *Gloeophyllum sepiarium*, hardwood, Jack Pine, *Phellinus ignarius, Pinus banksiana*, *Populus tremuloides*, *Trametes pubescens*, trembling aspen, *Trichaptum abietinum*.

Decay is an important component of the carbon cycle, as it breaks down wood and releases the stored carbon back into the atmosphere. White rot, brown rot, and soft rot are all types of decay which initiate this process. To test if fungi are host specific, break down wood at different rates, and are dependant on host species x fungus interactions, a study was conducted using two species of wood (*Pinus banksiana* and *Populus tremuloides*) and eight fungi (*Trichaptum abietinum, Fomitopsis cajanderi, Gloeophyllum separium, Fomitopsis pinicola, Trametes pubescens, Bjerkandera adusta, Ganoderma appanatum,* and *Phellinus ignarius*). Small pre-weighed blocks of *Pinus banksiana* and *Populus tremuloides* were aseptically inoculated with mycelium of each of the eight fungi and final dry weights of the blocks were taken, after 3 months of incubation, to determine the percent rate of decay.

Both *Pinus banksiana* and *Populus tremuloides* experienced a decline in weight for all eight fungus species. However, Anova testing showed that there was no significant difference in the percent rate between the two wood types, but there was a difference between the fungi and the fungi x wood species interaction. The two *Fomitopsis* species caused the most statistically significant difference in the dry weight of trembling aspen blocks, while *Fomitopsis pinicola* caused the most statically significant difference in dry weight of jack pine blocks. Although both *Fomitopsis* species are primarily conifer-inhabiting, the ability to decay hardwood blocks suggests that ideal lab conditions may alter fungal species ecological strategies compared to less than idea natural conditions in the field.

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INTRODUCTION

Forests cover 40% of Canada's landmass (CCFM 2017). They provide habitat for a wide variety of birds, mammals, and amphibians (CCFM 2017). Equally as important, forests are an important component of the carbon cycle (CCFM 2017). Through the process of photosynthesis, trees absorb carbon dioxide and release oxygen (CCFM 2017). The carbon dioxide is stored in the tree until the wood breaks down resulting in the carbon being released back into the atmosphere (CCFM 2017). This process of breaking down wood is known as decomposition or decay (CCFM 2017).

All wood is threatened by the danger of decay (Nicholas 1982). Decay causes wood to weaken, it may alter wood's pigmentation, and it can bury itself deep into the grains of the wood (Nicholas 1982). Some types of decay cause wood to lose its shape by forming sunken patches to appear on its surface, while severe decay may cause wood shrinkage (Nicholas 1982).

Decay is frequently prevalent in mature forests where a high percentage of deadfall is present (Hammel 1997). Warm temperatures, high moisture content, oxygen availability, and the viability of fungi to cause decay, increase the decomposition rate of wood (Hammel 1997). However, factors such as high soil and/or litter pH and tree species that are naturally resistant to rot, will decrease the rate of decay (Hammel 1997).

TYPES OF DECAY

There are three main types of decay: brown rot, white rot, and soft rot (Dix and Webster 1995, Nicholas 1982). Brown rot is the most common type of decay that affects coniferous wood (Goodell *et al*. 2008). Wood which is decayed by brown rot generally becomes brown and crumbles in the process of disintegration (Goodell *et al*. 2008). This degraded wood is due to the removal of cellulose and hemicellulose in the wood, which leaves the wood with a high lignin content (Nicholas 1982). All fungi that cause brown rot belong to the Basidiomycota (Nicholas 1982).

White rot is generally found on deciduous wood, and degrades cellulose, hemicelluloses and lignin (Goodell *et al*. 2008). This can result in patchy pocket decay or a more uniform decay across the wood (Goodell *et al*. 2008). Pocket decay or pocket rot occurs during selective delignification, where cellulose and lignin are unevenly targeted, causing pockets to develop which are filled with white hyphae (Worrall *et al.* 1997). Simultaneous break down of all components at an equal rate in wood results in a spongy, moist, and bleached appearance (Worrall *et al.* 1997). White rot fungi occur among both the Basidiomycota and the Ascomycota (Goodell *et al*. 2008).

Soft rot mainly occurs to wood located in wet environments (Blanchette 1995). It also may be present in environments which are too harsh for brown or white rot to develop (Blanchette 1995). Decay is limited to the external surface

of the wood that is in direct contact with wet soil or other environments (Blanchette 1995). Soft rot fungi occur exclusively among the Ascomycota (Blanchette 1995).

COMPETITIVE STRATEGIES

All fungi use at least one of three strategies to compete for resources: ruderal, stress-tolerant, and competitive (Dix and Webster 1995, Jennings and Lysek 1999). Fungi that use the ruderal strategy reproduce quickly in an environment that is disturbed, yet productive (Dix and Webster 1995, Jennings and Lysek 1999). The stress-tolerant strategy fungi are able to adapt, to endure times of environmental stress from droughts, floods, high temperatures or limited resources (Dix and Webster 1995, Jennings and Lysek 1999). Lastly, competitive strategy fungi are able to out compete other fungi for resources, which allow them to maximize in growth and productivity (Dix and Webster 1995, Jennings and Lysek 1999). These fungi also are able to take over resources that were previously used by other fungi (Dix and Webster 1995).

Fortunately, wood does have properties that reduces colonization by decay fungi from occurring (Dix and Webster 1995). The cell walls of the wood are coated with lignin that makes it difficult for fungi to break down (Dix and Webster 1995). Many tree species have low nitrogen content in their wood, which limit the ability to be utilized by fungi (Dix and Webster 1995). Extractives with antifungal properties are found in some tree species such as tannins found in chestnut (*Castanea sativa*) and oak (*Quercus*) (Dix and Webster 1995).

These substances are able to prevent decay fungi from decaying wood, although some fungi have evolved enzymatic systems to detoxify these substances (Dix and Webster 1995). As a consequence, many wood decay fungi exhibit some degree of host specificity.

An experiment was set up to investigate such specificity utilizing four wood decay fungi found predominately on hardwoods [*Bjerkandera adusta* (Willd.) P. Karst., *Ganoderma applanatum* (Pers.) Pat., *Phellinus ignarius* (L.) Quel., and *Trametes pubescens* (Schmach.) Pilat] and four decay fungi predominately found on conifers *[Fomitopsis cajanderi* (P.Karst.) Kotl. & Pouzar, *Fomitopsis pinicola* (Sw.) P.Karst., *Gloeophyllum sepiarium* (Wulf.) P. Karst., and *Trichaptum abietinum* (Pers. : J.F. Gmel.) Ryvarden]. Isolates of these fungi were inoculated onto wood blocks of a representative hardwood (*Populus tremuliodes* Michx.) and a representative conifer (*Pinus banksisana* Lamb.)

NULL HYPOTHESES

1. There will be no difference in percent rate of decay between the conifer and hardwood blocks for each fungus used.
2. There will be no difference observed in percent rate of decay between the different fungi used.
3. There will be no difference in percent rate of decay between the different fungus species x tree species interactions.

METHODS AND MATERIALS

Isolates of *Bjerkandera adusta*, *Fomitopsis cajanderi*, *Fomitopsis pinicola*,*Ganoderma applanatum*, *Gloeophyllum sepiarium*, *Phellinus ignarius, Trametes pubescens*, and *Trichaptum abietinum*, were cultured from tissue samples extracted within the interior of fresh basidiomata or from spore prints. All isolates were maintained on modified 2% malt extract agar (20g malt extract, 1g yeast extract, 15g agar, 1000mL water). All cultures are maintained in the Lakehead University Mycological Herbarium Fungal Culture Collection (Appendix I).

Wood-decay tests were conducted as described in a standard method (Anonymous 1986). Forty-five small wood blocks of *Pinus banksiana* Lamb. (jack pine) and forty-five small wood blocks of *Populus tremuloides* Michx. (trembling aspen) (approximately 1.5 – 2.0cm^3), respectively, were labeled from 1 through 90 and placed in corresponding aluminum weigh boats. The blocks were then dried in an oven for 72 hours at 100^oC, before being weighed to the nearest milligram. Afterwards, the blocks were then all placed in jars of water to rehydrate.

One hundred and twenty millilitres of a 10:1 vermiculite:peat mixture was poured into 90 square bottles (Quorpak) (250mL). Seventy millilitres of a 2% malt extract broth was poured into the bottles, to promote fungal growth. The wooden blocks were placed into each bottle, buried under the moistened

vermiculite:peat mixture, and the bottles were loosely capped and labeled. The caps had a hole drilled in the middle with a filter placed inside to facilitate air exchange. Aluminum foil was placed on top of each bottle and labeled before the bottles were autoclaved for 60 minutes at 121^{o}C and 1.7kg/cm^{2} pressure. They were allowed to cool overnight.

Once the bottles were cool, a transfer hood was prepped by wiping it down with 70% alcohol. In the transfer hood, a 7mm diameter cork-borer was flamed, cooled, and utilized to cut 20 plugs from each fungus growing on 2% malt extract agar. One at a time, the bottles were flamed at the bottleneck opening with a Bunsen burner, then forceps were used to place two plugs of the inoculum beside the wood block. These plugs were covered with the vermiculite:peat mixture. Afterwards the bottles were recapped and covered with aluminum foil. Each of the eight fungi were inoculated onto on 5 jack pine and 5 trembling aspen wood blocks as shown in Figure 1. In addition, 5 of each wood species (10 bottles in total), did not receive fungi and acted as the control. The forceps were sterilized by flame between uses, and the transfer hood was wiped down between inoculation with each species of fungus. Next, the bottles were placed in a dark incubator at 20^{o}C for 82 days (Figure 2).

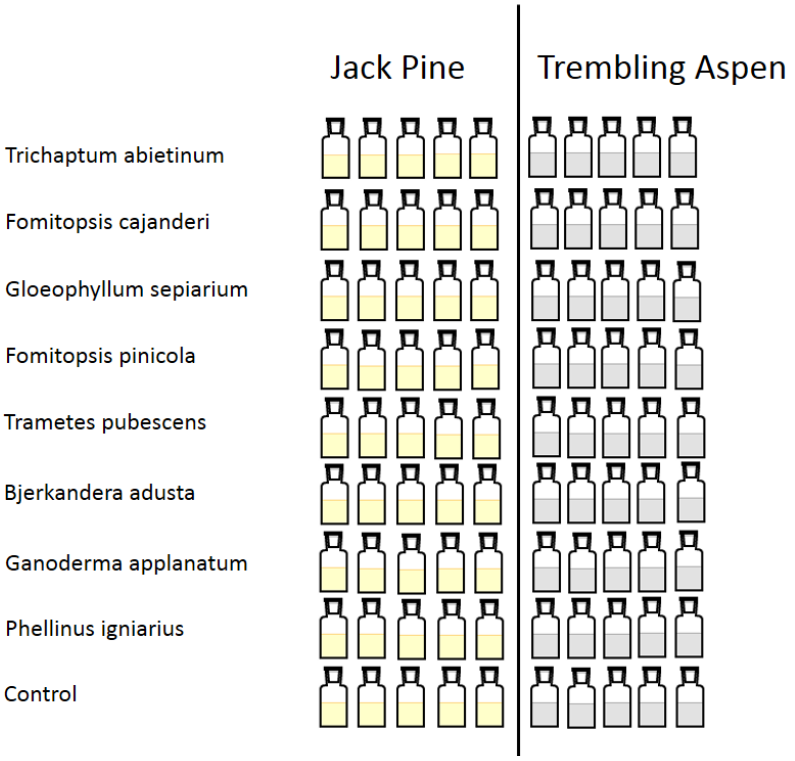

Figure 1. Experimental Design.

Figure 2. Inoculated wood blocks in incubator.

After three months, the wood blocks were removed from the bottles, scraped clean of mycelium and of the vermiculite:peat mixture, and placed on the aluminum weigh boats. The blocks were then placed in the oven at 100^{o}C for 72 hours. After this time, they were weighed to the nearest milligram.

RESULTS

Average weights were calculated for trembling aspen and jack pine using the raw data found in Appendix II. In both trembling aspen and jack pine, the average final weight (g) of the wood blocks was lower than the average initial weight (g) as shown in Figure 3 and 4. However, when accounting for standard error of the mean, only *Fomitopsis pinicola*, *Fomitopsis cajanderi,* and *Ganoderma appanatum* resulted in statistically significant weight loss in trembling aspen wood. As well, jack pine only showed statistically significant weight loss from *Fomitopsis pinicola*, *Fomitopsis cajanderi*, and *Gloeophyllum sepiarium*.

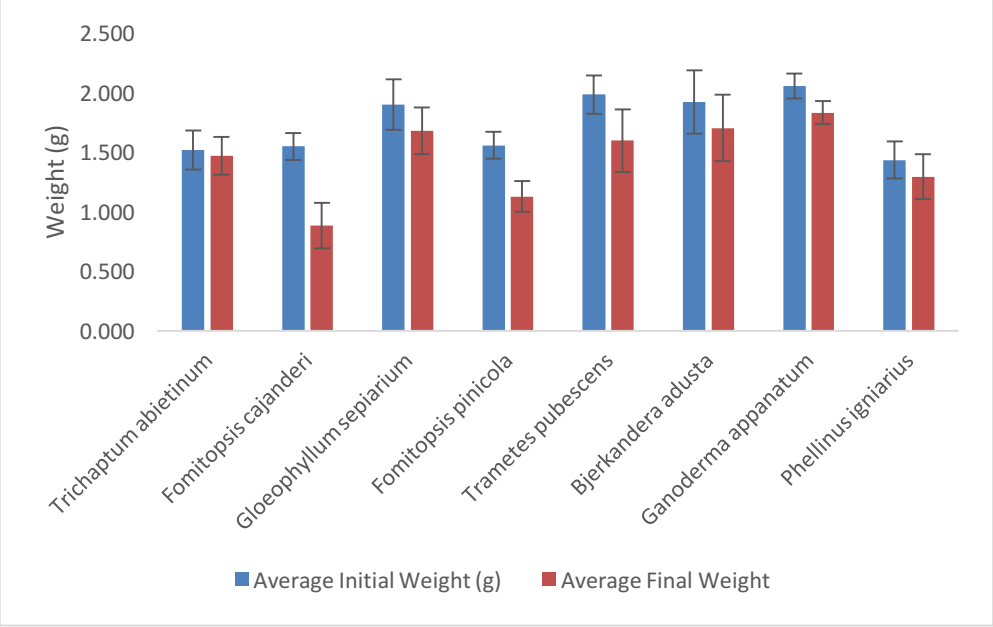

Figure 3. Average Weights for trembling aspen. Source: Appendix II

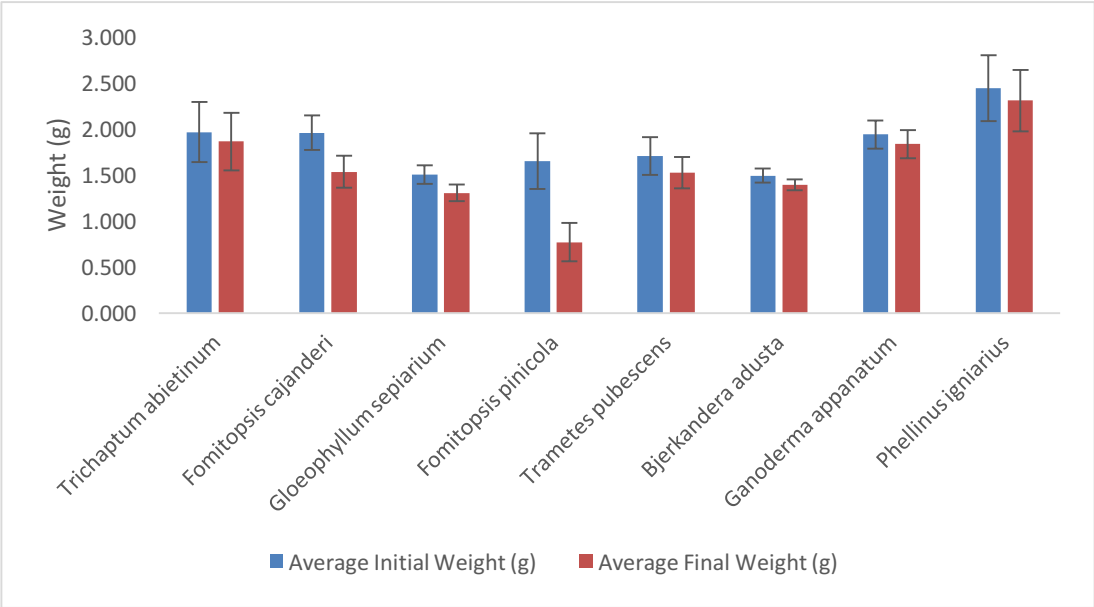

Figure 4. Average Weights for jack pine. Source: Appendix II

Percent rate of decay was calculated using the formula: 100-((Final Weight/Initial Weight)*100) as shown in Table 1. Jack pine had higher percent rates of decay than trembling aspen for all conifer specific fungi except for *Fomitopsis cajanderi*. All hardwood specific fungi had higher percent rates of decay in trembling aspen compared with jack pine.

Table 1. Percent Rate of Decay Source: Appendix II

Fungi Species		Jack Pine	Trembling Aspen
	Control	0.0	0.3
Conifer Specific Fungi	Trichaptum abietinum	5.1	3.3
	Fomitopsis cajanderi	21.6	42.9
	Gloeophyllum sepiarium	13.4	11.8
	Fomitopsis pinicola	53.6	27.6
Hardwood Specific Fungi	Trametes pubescens	10.6	19.5
	Bjerkandera adusta	6.5	11.4
	Ganoderma appanatum	5.3	10.8
	Phellinus igniarius	5.6	9.7

To determine if the null hypotheses were correct a two-way Anova was run to test the hypotheses (Table 2). The results of the Anova demonstrated that the second hypothesis was correct, while the first and third hypotheses were rejected as they had a significance of under 0.05.

Table 2. Anova test results. Source: Appendix III

Dependent Variable:

Source	Type III Sum of Squares	df	Mean Square	F	Sig.
Fungi	14971.188	8	1871.398	21.926	0.000
Species	106.013	1	106.013	1.242	0.269
Fungi * Species	3813.922	8	476.740	5.586	0.000
Error	6145.277	72	85.351		
Total	44896.379	90			

A Post Hoc test was conducted on hypotheses one and three, using data from Table 2 and Appendix III, to determine which fungus and fungus x wood species interactions had a significant result. The test showed that *Fomitopsis cajanderi* and *Fomitopsis pinicola* had significantly higher percent rates of decay across both wood block species (Figure 5). As well, *Fomitopsis cajanderi* x trembling aspen and *Fomitopsis pinicola* x jack pine had significantly higher percent rates of decay than all the other fungi x wood species interactions (Figure 6).

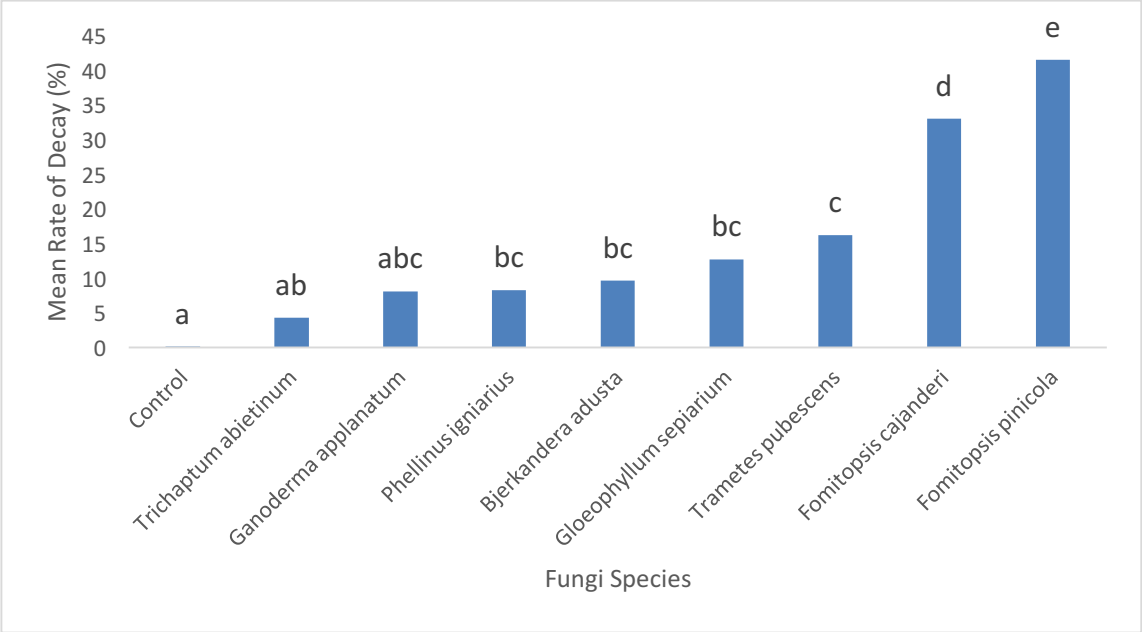

Figure 5. Post Hoc Test Results for Fungi (Hypothesis 1). Source: Appendix II

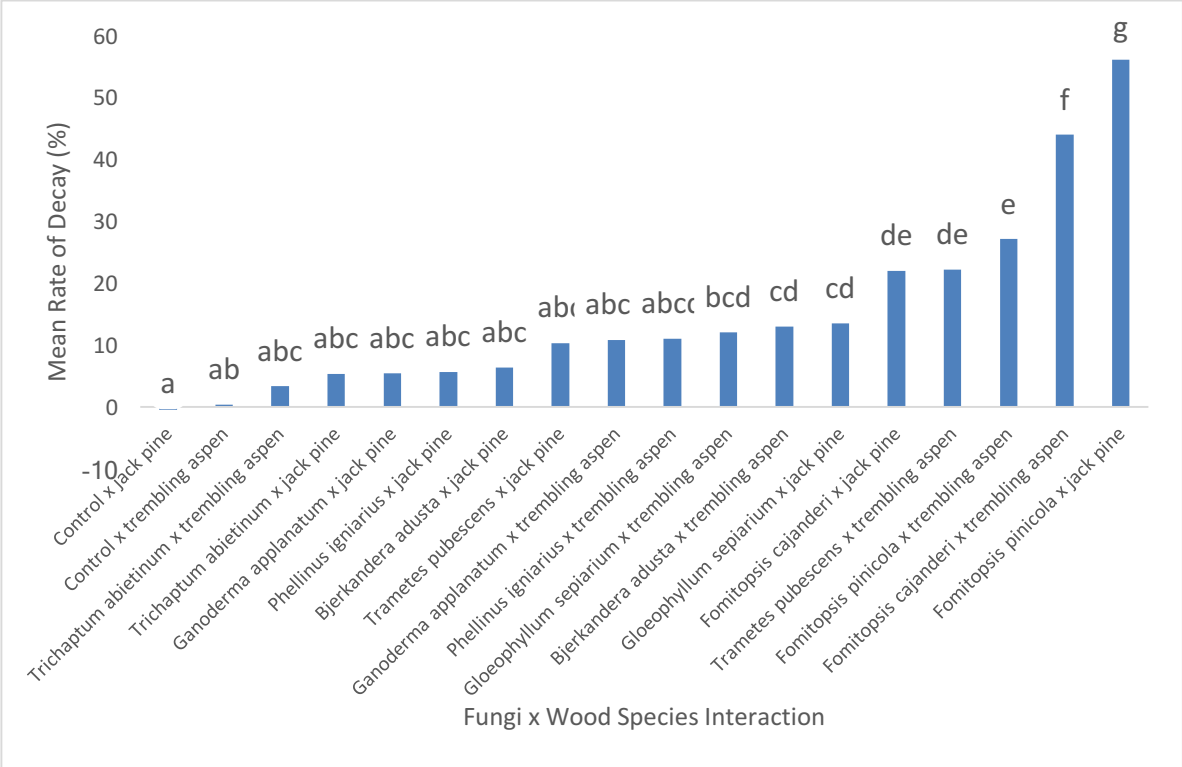

Figure 6. Post Hoc Test Results of Fungus x Wood Species Interaction (Hypothesis 3). Source: Appendix III

DISCUSSION

The results proved Hypothesis Two, as there were no differences in the percent rate of decay between jack pine and trembling aspen. However, Hypotheses One and Three were rejected. There was a difference between the various fungi as *Fomitopsis cajanderi* and *Fomitopsis pinicola* were statistical outliers when compared to the other fungus' percent rate of decay. A difference in percent rate of decay was also found between the fungus x wood species interactions in *Fomitopsis cajanderi* x trembling aspen and *Fomitopsis pinicola* x jack pine.

In nature, the fungi used in the experimental procedure tend to lean toward conifer or hardwood specific. *Trichaptum abietinum* is commonly found on the dead wood of conifer trees (Overholts 1953). It has been known to grow on species such as *Pinus* (pine), *Picea* (spruce), and *Larix* (larch) (Breitenbach and Kranzlin 1986). However, it has been found occasionally on *Populus* (poplar) and *Quercus* (oak) (Overholts 1953). *Fomitopsis cajanderi* exclusively establishes itself on dead conifer wood such as *Abies* (fir) and spruce, and it is rarely found on hardwoods (Overholts 1953). *Gloeophyllum sepiarium* is solely known to grow on dead conifer wood, particularly wood in service, such as fence posts and railway ties (Breitenbach and Kranzlin 1986). *Fomitopsis pinicola* meanwhile, can be found on the dead wood of both conifer and deciduous species (Breitenbach and Kranzlin 1986).

Trametes pubescens, *Bjerkandera adusta*, *Ganoderma appanatum,* and *Phellinus ignarius* are all fungi that are known to establish on hardwoods such as poplar, oak, *Betula* (birch), and *Acer* (maple) (Overholts 1953). All of them grow on dead wood with the exception of *Phellinus ignarius,* which is found on living trees (Overholts 1953). *Bjerkandera adusta* may also occasionally establish on conifer wood such as spruce and pine. *Phellinus ignarius* has been found rarely on pine under ideal conditions (Overholts 1953).

The results of this study show that the majority of the fungi tended to preferentially decay the same host species as they would in nature with the exceptions of *Fomitopsis cajanderi* and *Fomitopsis pinicola.* Both of these latter fungi had exceptionally higher percent rates of decay on both host species compared to the other fungi. This difference may be due to the ideal growing conditions that the fungi were growing in. The growing medium (vermiculite:peat mixture) moistened with 2% malt extract broth and temperature (20^{o}C) both promote the growth of fungi. As well, both fungi did not have to compete for resources with other fungi or bacteria, which may have led to the higher percent rate of decay.

Although *Fomitopsis cajanderi* is rarely found on hardwoods, there was a significant difference between the *Fomitopsis cajanderi* x trembling aspen compared to all the other fungi x wood species interactions (excluding *Fomitopsis pinicola* x jack pine). While the *Fomitopsis pinicola* x jack pine interaction was reasonable as *Fomitopsis pinicola* is found on conifers, *Fomitopsis cajanderi* x trembling aspen was unexpected as *Fomitopsis cajanderi*

is rarely found on hardwoods. This is mostly likely a result from trembling aspen's poor structural qualities, and lack of antifungal extractives which make it highly susceptible to decay (Peterson and Peterson 1992), when combined with the ideal growing conditions, which increased the chances of *Fomitopsis cajanderi* decaying the trembling aspen.

If the experiment would have run over a longer period of time, it is expected that there would have been more significant results as increased percent rates of decay would have become more evident. If the experiment were done under non-septic conditions, the impact of ideal growing conditions would be lessened, as it is expected that the fungi would adhere closer to their host specific strategy. As such, fungus x wood species interactions such as *Fomitopsis cajanderi* x trembling aspen might not end up being relevant.

CONCLUSION

The results of this study may be useful in determining which fungi are of most concern for wood in service. Although service wood is primarily made from softwood species, this study shows that under ideal conditions it is possible for any fungus to have a significant negative impact on the lifespan of the wood. As such, when wood preservatives are developed one must recognize that it is possible for any wood-decaying fungus to establish on a non-host species. Furthermore, these results show that all of the fungi contribute to the carbon cycle in hardwood (trembling aspen) and softwood stands (jack pine), although both do decay at different rates.

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APPENDICES

APPENDIX I – COLLECTION INFORMATION FOR CULTURES OF WOOD-DECAYING FUNGI

Bjerkandera adusta (297)

10 November 2004, Lakehead University campus, Thunder Bay, Ontario. Isolated by Owen Bott. Isolated from spore print from basidioma on deciduous log.

Fomitopsis cajanderi (621)

2009, Lakehead University campus, Thunder Bay, Ontario. Isolated by Joey Tanney. Isolated from basidioma.

Fomitopsis pinicola (310)

29 October 2004, Lakehead University campus, Thunder Bay, Ontario. Isolated by Tammy Rancourt. Isolated from basidioma on *Picea glauca* log.

Ganoderma applanatum (089)

18 November 2002, Lakehead University campus, Thunder Bay, Ontario. Isolated by Steve Hill. Isolated from basidioma.

Gloeophyllum sepiarium (615)

01 October 2009, Lakehead University campus, Thunder Bay, Ontario. Isolated by Joey Tanney. Isolated from basidioma on conifer stump.

Phellinus ignarius (culture not saved)

October 2016, Lakehead University campus, Thunder Bay, Ontario. Isolated by Erin Kielt. Isolated from basidioma.

Trametes pubescens (728)

4 October 2013, Lakehead University campus, Thunder Bay, Ontario. Isolated by Leonard Hutchison. Isolated from basidioma on *Populus tremuloides* log.

Trichaptum abietinum (732)

06 October 2016, Lakehead University campus, Thunder Bay, Ontario. Isolated by Leonard Hutchison. Isolated from spore print from a basidioma on fallen *Picea glauca*.

APPENDIX II – INITIAL AND FINAL WEIGHTS

Block Number	Species	Fungi	Initial Weight (g)	Final Weight (g)
1	Pb	Control	3.145	3.163
2	Pb	Control	1.380	1.400
3	Pb	Control	1.564	1.569
4	Pb	Control	1.700	1.705
5	Pb	Control	2.601	2.603
6	Pb	Trichaptum abietinum	1.284	1.189
7	Pb	Trichaptum abietinum	2.860	2.690
8	Pb	Trichaptum abietinum	2.110	2.051
9	Pb	Trichaptum abietinum	2.404	2.293
10	Pb	Trichaptum abietinum	1.160	1.092
11	Pb	Fomitopsis cajanderi	1.652	1.134
12	Pb	Fomitopsis cajanderi	1.570	1.299
13	Pb	Fomitopsis cajanderi	2.260	1.803
14	Pb	Fomitopsis cajanderi	2.540	2.061
15	Pb	Fomitopsis cajanderi	1.756	1.367
16	Pb	Gloeophyllum sepiarium	1.683	1.438
17	Pb	Gloeophyllum sepiarium	1.423	1.206
18	Pb	Gloeophyllum sepiarium	1.174	1.015
19	Pb	Gloeophyllum sepiarium	1.733	1.533
20	Pb	Gloeophyllum sepiarium	1.505	1.316
21	Pb	Fomitopsis pinicola	2.787	1.534
22	Pb	Fomitopsis pinicola	1.713	0.880
23	Pb	Fomitopsis pinicola	1.127	0.416
24	Pb	Fomitopsis pinicola	1.436	0.522
25	Pb	Fomitopsis pinicola	1.190	0.480
26	Pb	Trametes pubescens	1.684	1.524
27	Pb	Trametes pubescens	2.458	2.140
28	Pb	Trametes pubescens	1.404	1.248
29	Pb	Trametes pubescens	1.683	1.532
30	Pb	Trametes pubescens	1.287	1.172
31	Pb	Bjerkandera adusta	1.418	1.361
32	Pb	Bjerkandera adusta	1.277	1.240
33	Pb	Bjerkandera adusta	1.691	1.559

Block Number	Species	Fungi	Initial Weight (g)	Final Weight (g)
34	Pb	Bjerkandera adusta	1.444	1.313
35	Pb	Bjerkandera adusta	1.626	1.496
36	Pb	Ganoderma appanatum	2.081	1.982
37	Pb	Ganoderma appanatum	2.098	1.910
38	Pb	Ganoderma appanatum	1.749	1.678
39	Pb	Ganoderma appanatum	2.320	2.249
40	Pb	Ganoderma appanatum	1.437	1.348
41	Pb	Phellinus igniarius	1.857	1.820
42	Pb	Phellinus igniarius	3.201	3.026
43	Pb	Phellinus igniarius	2.768	2.598
44	Pb	Phellinus igniarius	3.035	2.845
45	Pb	Phellinus igniarius	1.350	1.238
46	At	Control	1.641	1.625
47	At	Control	1.059	1.058
48	At	Control	1.412	1.415
49	At	Control	1.149	1.138
50	At	Control	1.456	1.459
51	At	Trichaptum abietinum	1.677	1.594
52	At	Trichaptum abietinum	1.437	1.401
53	At	Trichaptum abietinum	1.291	1.261
54	At	Trichaptum abietinum	1.114	1.072
55	At	Trichaptum abietinum	2.061	2.002
56	At	Fomitopsis cajanderi	1.187	0.434
57	At	Fomitopsis cajanderi	1.478	1.178
58	At	Fomitopsis cajanderi	1.716	1.432
59	At	Fomitopsis cajanderi	1.857	0.848
60	At	Fomitopsis cajanderi	1.484	0.518
61	At	Gloeophyllum sepiarium	2.579	2.275
62	At	Gloeophyllum sepiarium	1.595	1.357
63	At	Gloeophyllum sepiarium	2.064	1.856
64	At	Gloeophyllum sepiarium	1.914	1.723
65	At	Gloeophyllum sepiarium	1.334	1.159
66	At	Fomitopsis pinicola	1.706	1.595
67	At	Fomitopsis pinicola	1.914	1.057
68	At	Fomitopsis pinicola	1.307	1.076
69	At	Fomitopsis pinicola	1.428	0.818

Block Number	Species	Fungi	Initial Weight (g)	Final Weight (g)
70	At	Fomitopsis pinicola	1.412	1.077
71	At	Trametes pubescens	1.352	0.572
72	At	Trametes pubescens	2.041	1.786
73	At	Trametes pubescens	2.110	2.039
74	At	Trametes pubescens	2.241	1.675
75	At	Trametes pubescens	2.158	1.899
76	At	Bjerkandera adusta	2.525	2.290
77	At	Bjerkandera adusta	1.304	1.097
78	At	Bjerkandera adusta	2.107	1.891
79	At	Bjerkandera adusta	2.386	2.245
80	At	Bjerkandera adusta	1.272	0.974
81	At	Ganoderma appanatum	1.710	1.480
82	At	Ganoderma appanatum	2.008	1.813
83	At	Ganoderma appanatum	2.269	2.033
84	At	Ganoderma appanatum	2.261	1.929
85	At	Ganoderma appanatum	2.002	1.886
86	At	Phellinus igniarius	1.972	1.933
87	At	Phellinus igniarius	1.231	0.978
88	At	Phellinus igniarius	1.039	0.876
89	At	Phellinus igniarius	1.510	1.390
90	At	Phellinus igniarius	1.399	1.280

APPENDIX III – ANOVA TEST RESULTS: DESCRIPTIVE STATISTICS

Descriptive Statistics				
Fungi		Mean	Std. Deviation	N
Ba	A	12.95	6.86	5
	P	6.36	2.72	5
	Total	9.66	6.03	10
C	A	0.32	0.60	5
	P	-0.54	0.54	5
	Total	-0.11	0.70	10
Fc	A	43.94	23.69	5
	P	21.97	5.55	5
	Total	32.96	19.93	10
Fp	A	27.08	16.44	5
	P	56.00	8.64	5
	Total	41.54	19.63	10
Ga	A	10.81	3.48	5
	P	5.41	2.29	5
	Total	8.11	3.98	10
Gs	A	11.98	2.10	5
	P	13.49	1.49	5
	Total	12.73	1.89	10
Pi	A	10.93	7.25	5
	P	5.63	2.29	5
	Total	8.28	5.79	10
Ta	A	3.28	1.09	5
	P	5.32	1.72	5
	Total	4.30	1.73	10
Tp	A	22.16	21.34	5
	P	10.29	1.72	5
	Total	16.23	15.59	10
Total	A	15.94	17.03	45
	P	13.77	16.64	45
	Total	14.85	16.77	90