A NEW APPROACH TO EFFECTIVE BIOPULPING: TREATING LOGS WITH *PHLEBIOPSIS GIGANTEA*

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ABSTRACT

Phlebiopsis gigantea is an aggressive white-rot fungus that colonizes conifer wood. It is ideally suited for bioprocessing because of its fast growth, prolific asexual spore production and benefits from controlling sapstain, reducing pitch and potential improvements fix pulp and paper production. Field investigations in Alabama and Minnesota using red and loblolly pine logs showcd the fungus was able to successfully colonize fresh sapwood and inhibit detrimental sapstain fungi Treated logs had significantly reduced amounts of pitch (61% less) and bark removal was facilitated. Mechcanically refined fibers from treated logs had increased cell wall porosity and reduced energy consumption (9 to 27% less) as compared to control logs. Paper made from pretreated wood also resulted in increases in burst, tear, and tensile strength properties.

INTRODUCTION

Inoculating logs with *Phlebiopsis gigantea* immeadiately after cutting allows the fungus to colonize the wood and begin the biopulping process during transport and storage of logs. Treating logs with the fungus has been shown to successfully inhibit detrimental sapstain fungi from colonizing the wood and prevents blue stain (1). Inoculating logs has also been shown to decrease wood extractives by as much as 61% compared to non-inoculated logs (1). The results presented at this meeting indicate that pretreating logs with *P. gigantea* reduces energy consumption and increases paper strength properties during biomechanical pulping processes.

MATERIALS AND METHODS

Alabama experiment. Loblolly pine trees 30 to 40 years old with an average diameter of 20 cm were felled at the Solon Dixon Forestry Center in March of 1996 and cut into 61 or 244 cm lengths. Prior to inoculation the logs were slit (through the bark exposing sapwood) four times around the outer circumference on the log every 20 cm down the log length. Both log ends and slits were sprayed Two piles per treatment were set up in a pyramid shape on the forest floor with 10 and 16 logs per pile for the 61 and 244 cm length logs, respectively. Treatments included inoculation with *P. gigantea* and a non-inoculated water control. A spore suspension, prepared and inoculated as previously described (1). Logs were examined 10 and 20 weeks after inoculation by sampling 2 logs per pile (4 logs / treatment) for the 61 cm logs and 3 logs per pile (6 logs / treatment) for 244 an logs. Two sub samples, from the end and middle, were cut from each of the 244 cm logs

Minnesota experiment. Red pine trees 40 to 50 years old with an average diameter of 20 cm were felled at the Cloquet Forestry Center in May of 1995 and 1996 cutting the logs into 61 and 122 cm lengths, respectively. Logs in the 1995 study received 2.5 an holes, to expose the sapwood, 4 times around the outer circumference of the log every 20 cm down the length of the log. Logs in the 1996 study were slit with a chainsaw through the bark to expose the sapwood 2 times around the outer circumference of the log every 15 cm down the length of the log. The log ends, holes, and slits were inoculated. Ten logs were piled in a pyramid shape on the forest floor in 1995 with 3 piles per treatment, while 16 logs were piled in a 4 X 4 log square with 2 piles per treatment in 1996. Treatments included inoculation of logs with *P. gigantea* and a non-inoculated water control. Spore concentrations and inoculation methods were previously described (1). Logs were sampled at 8, 11, and 17 weeks after inoculation in 1995, and 9 and 22 weeks after inoculation in 1996. Two logs per pile were sampled (6 logs per treatment) in 1995, by removing a 30 cm section from the log end. Three logs were sampled (6 logs per treatment) in 1996, taking segments from the end and middle. Fiber modification was analyzed after mechanical refining with the use of Simons' stain. Pulp sampled from a Sprout Waldron refiner after 1 pass was stained and analyzed according to previous procedures (14).

Energy consumed during the mechanical refining process and changes in the physical properties of handsheets, made from inoculated and non-inoculated logs were conducted at the Forest Products Laboratory, USDA, Forest Service, Madison, WI. Wood chips were passed through the refiner, calculating the percent of energy consumed, until a freeness of 100 (Canadian Standard of Freeness, CSF) was reached. All of the data was regressed to a CSF of 100 to make comparisons between treatments Handsheets were made from refined wood (pulp) with a CSF of approximately 100. One sample was analyzed from each treatment per study. Procedures similar to those previously published by Akhtar were followed (12). The amount of energy consumed during refining and changes in the physical properties of wood inoculated with *P. gigantea* and non-inoculated was compared to fresh control logs.

RESULTS AND DISCUSSION

Red and loblolly pine logs inoculated with *P. gigantea* had 90 to 100% of the sapwood colonized after 8 weeks, indicating that *P. gigantea* is a superb pioneer colonist capable of rapidly colonizing non-sterilized logs. Recently Behrendt and Blanchette (1) demonstrated the ability of *P. gigantea* to colonize freshly cut red pine logs, and prevent subsequent colonization by sapstain fungi. The ability of *P. gigantea* to tolerate resinous extractives present on freshly cut wood

allows the fungus to quickly colonize the sapwood, preventing other fungi from becoming established. Inoculating the fungus into cut exposed slits on the sides of the log enables *P. gigantea* to completely colonize any length of log in a short period of time (1). Application of fungal inoculants to logs immediately after cutting would allow beneficial biopulping effects to take place during transport and storage.

Application of *P. gigantea* to red and loblolly pine logs resulted in as much as a 61% reduction in wood extractives compared to non-inoculated logs. Thirty to 60% more wood extractives were removed from logs inoculated with *P. gigantea* than non-inoculated logs 9 to 12 weeks after inoculation. As time progressed, wood extractives were continually removed from the wood by the fungus. Previous research has shown a 33 to 35% reduction in extractives from loblolly pine wood chips inoculated with *Ceriporiopsis subvermispora* after 4 weeks (8). In our studies, wood extractives were reduced by as much as 61% in loblolly pine logs 8 to 12 weeks after inoculation with *P. gigantea.* Variation in the percent of wood extractives removed from loblolly logs (10% to 61%) is most likely due to either differences in log lengths (61 cm versus 244 cm) and time needed to colonize entire logs or variation in resin content among trees used for the study.

Staining of refined wood fibers from logs inoculated with *P. gigantea* after 17 weeks in the field revealed a larger percent (55 - 77%) of fibers staining orange-yellow than fibers from non-inoculated logs (24 - 58%) (Table I). Fibers from control logs after 0 weeks had only 8 to 9% of fibers staining orange-yellow. The staining observed in the aged non-inoculated logs was apparently due to colonization by naturally occurring *P. gigantea.* Although colonization was delayed compared to inoculated logs, the amount of *P. gigantea* increased with time.

Mechanical refining of logs inoculated with *P. gigantea* requited less energy to reach a freeness of 100 (Canadian Standard of Freeness, CSF) than non-inoculated logs (Table 1). Red pine logs inoculated with *P. gigantea* showed reductions of 9 to 27% in energy consumption after 17 weeks, while non-inoculated logs in the field had a 1 to 19% reduction in energy consumption (Table II). Although aged non-inoculated logs had some reduction in energy consumption when compared to freshly cut wood, logs inoculated with *P. gigantea* at the time of cutting yielded an additional 6 to 8% decrease in energy consumption. Up to 100% of inoculated logs were colonized by *P. gigantea,* wide non-inoculated logs had up to 100% and 56% colonization in loblolly and red pine, respectively. Thus, the reduction in energy consumption in non-inoculated logs was a result of natural colonization by *P. gigantea.* The cool, moist conditions at our field site and large amount of native *P. gigantea* resulted in unusually high levels of colonization in the non-inoculated logs overtime. In contrast to these results, when *P. gigantea* was used in a biocontrol study to inoculate logs in a dry, sunny location, non-inoculated logs remained free of *P. gigantea* colonization for the duration of the study (1). Inoculated logs, however, had 100% colonization of *P. gigantea* after 8 weeks. At most harvesting sites, the limited natural inocula and adverse environmental conditions for fungal growth would necessitate applying the fungus to insure successful colonization and optimum bioprocessing potential.

Since mechanical refining consumes large quantities of electrical energy during processing (2), savings of up to 27% in energy consumption would be significant to the pulp and paper industry. Previous laboratory investigations have shown a reduction in energy consumption during mechanical refining of wood chips inoculated with either *C. subvermispora* or *P. chrysosporium* (3,4,5,6). While energy use was not determined for the loblolly pine wood used in out refining study, a drop in freeness of 100 or more after either the first or second pass has been associated with a 20% energy savings Therefore, we predict that the loblolly pine logs inoculated with *P. gigantea* that had a drop in freeness of 122 (CSF) after 2 passes could also reduce energy consumption by approximately 20%. In the studies reported here, even the 8 foot red pine logs inoculated with *P. gigantea* revealed energy savings of up to 27%. Inoculating whole logs instead of wood chips appears to be an effective way to treat pulpwood for biological processing. The large initial set up costs required for some fungi for successful biopulping of wood chips, such as steam sterilization and aeration of chip piles during incubation, would not be needed.

Successful colonization of red pine logs by *P. gigantea* led to an increase in the physical strength properties of handsheets made from treated logs. An increase in burst, tear, and tensile strength properties of 17, 20, and 13%, respectively, was observed 22 weeks after inoculation with *P. gigantea* when compared to physical strength properties of paper made from the freshly cut control logs (Table III). The small increase in burst, tear, and tensile strength properties observed after 22 weeks in handsheets made from aged non-inoculated logs was undoubtedly due to the natural colonization of *P. gigantea.* Results from this study also showed a decrease in pulp brightness after 22 weeks in *P. gigantea* treated logs when compared to freshly cut logs. Previous research has shown that the loss of pulp brightness from fungal treatment can he easily regained by bleaching with either alkaline hydrogen peroxide a sodium hydrosulfite (7).

CONCLUSIONS

Biological pulping of logs with *Phlebiopsis gigantea* resulted in the degradation of large quantities of wood extractives, reduction in energy consumption during mechanical refining and improved paper strength properties. The application procedure for this process is relatively easy to carryout and allows colonization and bioprocessing to occur during transport and storage of logs. As with any application of white-rot basidiomycetes, extended storage of 6 months or longer should be avoided so losses of biomass to decay does not occur.

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TABLE I. Percent of relined wood fibers, from logs inoculated with *Phlebiopsis gigantea*, staining orange-yellow to green in cola

	Loblolly Pine (61 cm)			Loblolly Pine (244 cm)			Red Pine (61 cm)				Red Pine (122 cm)		
Treatment[y]	0	10	20	0	10	20	0	8	11	17	0	9	22
Control	9	13	24	9	14	25	8	18	23	58	8	2	38
Treated	-[z]	12	60	-[z]	36	55	-[z]	21	54	77	-[z]	50	69

[y] weeks
[z] At 0 weeks only control logs were analyzed.

TABLE II. Percent energy consumed during mechanical refining of wood from pine logs inoculated with *Phlebiopsis gigantea*

Data Collected	Treatment	Loblolly Pine (244 cm)		Red Pine (61 cm)			Red Pine (122 cm)				
		0 wk	20 wk	0 wk	17 wk	%[v]	0 wk	9 wk	%[v]	22 wk	%[v]
Freeness (CSF)[w]	Control	430	400	98	108	—[x]	98	105	—[z]	109	—[z]
	Treated	—[x]	278	—[x]	98	—[x]	—[x]	110	—z	80	—[z]
Energy Consumption[y]	Control	—[z]	—[z]	3118	3085	1	3118	2741	12	2532	19
	Treated	—[z]	—[z]	—[z]	2825	9	—[x]	2558	18	2279	27

[V] Percent reduction in energy consumption, compared to 0 week control.
[W] Canadian Standard of Freeness (CSF) for loblolly pine after 2 passes through mechanical refiner and CSF for red pine after 5 passes through the refiner, refining red pine logs to a freeness of approximately 100.
[X] At 0 weeks only control logs were analyzed.
[Y] Energy consumed in WH/kg (watt hours/kg) after standardizing to a CSF of 100.
[Z] ND, not determined

TABLE III. Physical properties of handsheets made from pine logs treated with *Phlebiopsis gigantea* for 9 or 22 weeks

	Burst Index (KN/g)			Tear Index ($mN\text{-}m^2/g$)			Tensile Index (Nm/g)			Brightness (%)			Opacity (%)		
Treatment[y]	0	9	22	0[y]	9	22	0[y]	9	22	0[y]	9	22	0[y]	9	22
Control	.85	.89	.98	2.74	2.86	3.10	22.1	23.5	23.5	56.8	54.3	50.4	96.9	94.6	94.3
% change[z]	--	4	13	--	4	12	--	6	6	--	-4	-11	--	2	3
Treated	--	.97	1.03	--	3.06	3.42	--	24.3	25.5	--	39.9	38.6	--	94.8	94.2
% change[z]	--	12	17	--	10	20	--	9	13	--	-30	-32	--	2	3

[y] At 0 weeks only control logs were analyzed.
[z] Percent reduction in energy consumption, compared to 0 week control.

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