Fungal pretreatment of aspen chips improves strength of refiner mechanical pulp

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ABSTRACT *Pretreatment of wood chips with lignin-degrading fungi can improve the strength properties of unbleached refiner mechanical pulps. We studied the effect of two white-rot fungi (Phanerochaete chrysosporium and Dichomitus squalens) prior to refiner mechanical pulping. Strength and optical properties were compared in handsheets prepared from mechanical pulp of treated and untreated wood chips. Both fungal pretreatments resulted in handsheet strength properties better than the control when compared at an equivalent freeness. Fungal pretreatments decreased brightness and light-scattering coefficients but did not adversely affect opacity. Brightness of fungus-pretreated pulps was restored to that of the control by use of hydrogen peroxide bleaching. Lignin content of the chips was reduced by the fungal pretreatments.*

KEYWORDS
Biotechnology
Fungi
Mechanical pulps
Optical properties
Stock preparation
Strength properties

Mechanical pulps are much used in products where optical properties are of importance (*1–4*). Compared with chemical pulps, these pulps produce papers with high opacity and light scattering (*2, 3, 5*), and they have an important advantage in giving higher yields (85–95%) than chemical pulps (42–55%) (*1, 4, 5*). However, mechanical pulps have deficiencies that limit their usefulness. First, only certain softwoods and low-density hardwoods generally yield acceptable paper strength properties (*1, 2, 4, 6*). Second, low-strength properties often necessitate blending with low-yield, low-opacity chemical pulps (*1, 3, 4*). Third, large amounts of electrical energy are required to produce an acceptable mechanical pulp (*7–10*).

Chemical treatment of wood chips prior to mechanical pulping increases the utility of mechanical pulp (*8, 11–13*). Such pretreatment improves the strength properties of paper produced from mechanical pulps (*5, 11, 12, 14*) and increases the number of hardwood and softwood species yielding acceptable pulps (*6*). Chemical pretreatment of wood chips does have a serious disadvantage. Because pulping chemicals are used at low concentrations (*13, 14*), chemical recovery is usually not economical, requiring a costly cleanup of the waste liquors.

White-rot fungi have been proposed as agents for pretreating wood chips prior to mechanical pulping or for post-treating mechanical pulps (*15–17*). These treatments have been suggested as routes both to reduce fiberizing and refining energy requirements and to increase strength properties of the resulting paper. An additional benefit is anticipated in a decrease of the pollution loads resulting from destruction of extractive and other lignocellulosic components of pulping waste liquors (*18*). As yet, however, fungal treatments have not been comprehensively studied.

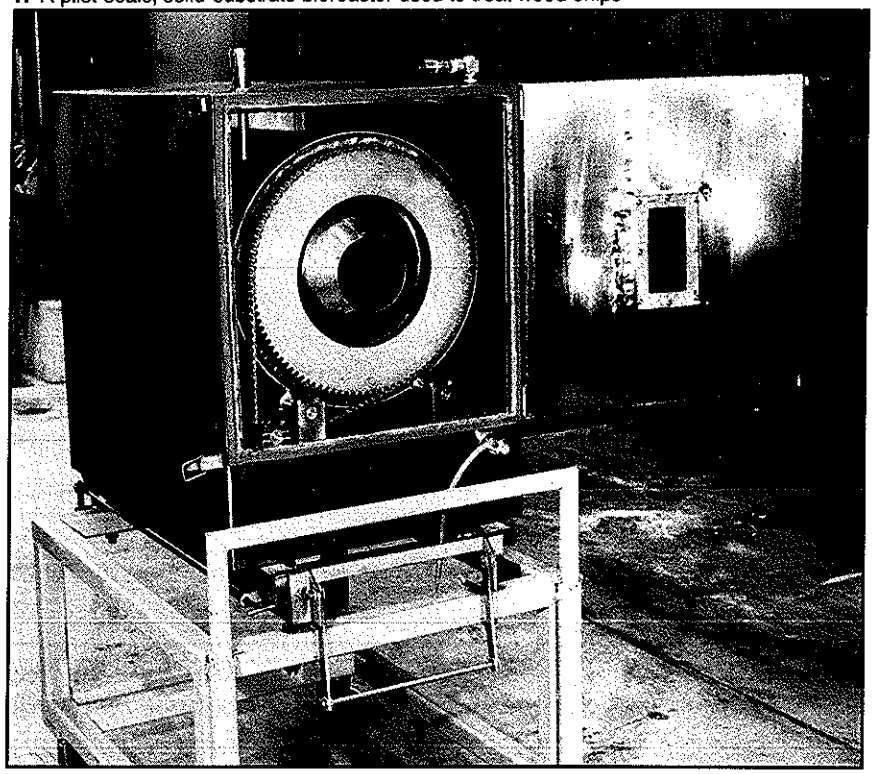
Only a few studies have been reported, and these show mixed results (*19–23*). Energy requirements in fiberizing and refining have been reported variously as being decreased and being increased (*19–21*). Fungal pretreatments of wood chips are reported to have increased and to have decreased paper strength properties (*19–22*). Fungal post-treatment of refiner mechanical pulp improves breaking length but decreases zero-span breaking length (*23*). Also, the biochemical mechanisms involved, assumed to be lignin removal and/or degradation, are not known, nor have the effects of isolated enzymes been demonstrated.

Despite the uncertainty and mixed results reported, the studies that have been done are encouraging. They show that at least under certain conditions, fungal pretreatments can reduce energy requirements (*19, 20*) and increase paper strength properties (*19, 20, 22*).

In the study reported here, our objective was to study the effects of pretreating aspen wood chips with two white-rot fungi—*Phanerochaete chrysosporium* and *Dichomitus squalens*—in a solid-substrate reactor prior to refiner mechanical pulping. Strength and optical properties of the handsheets prepared from pulps of pretreated and of untreated chips (as the experimental controls) were compared. The fungi were chosen for their degradation of lignin shown in previous studies (*24–26*); *P. chrysosporium* (*Sporotrichum pulverulentum*) has been used in previous studies of biopulping (*18–22, 27*).

Aspen wood chips were treated with fungi in a specially constructed, rotating, solid-substrate reactor (bioreactor) (**Fig. 1**). Treatment times, use of additives, and choice of environmental conditions were preselected, based upon previous experience, and were not optimized. Following treatment, the chips were fiberized and refined in a single-disk atmospheric refiner, and the pulps made into handsheets for evaluation.

1. A pilot-scale, solid-substrate bioreactor used to treat wood chips



1. Chemical analysis of unbleached pulp samples and hydrogen peroxide bleaching

	Cellulose	Hemicellulose, %				Handsheet brightness,%	
Chip pretreatment	Glucose, %	Xylose	Galactose	Mannose	Lignin, %	Before bleaching	After bleaching
Untreated control	52.30	18.09	...	2.21	19.60	51.4	...
D. squalens	58.12	17.94	...	2.01	16.60	39.7	55.2
P. chrysosporium	50.70	16.70	1.30	2.45	19.28	40.7	58.1

Results

Yield and pulp composition

Yield losses after fungal pretreatment were approximately 2% by weight. The amount of lignin and hemicellulose in the pretreated aspen wood chips was lower than in the untreated control (**Table I**). Based on total glucose (glucan) content, cellulose was relatively unaffected by either fungus. The slightly higher glucose content in one treated pulp may reflect the presence of glucan in the fungal cell walls, but it is most likely the result of the selective removal of lignin and xylan. The pulp pretreated with *D. squalens* had the lowest lignin content, whereas pulp pretreated with *P. chrysosporium* had the lowest xylan (xylose) content.

Strength and optical properties

Pretreatment with either fungus improved the strength properties of handsheets produced from the unbleached refiner mechanical pulps (**Table II**). Burst, tear, tensile, and zero span tensile indices were markedly increased, especially by *D. squalens*. At a comparable freeness of 85–95 mL CSF, the fungal pretreatments improved burst index by factors of 2.1 and 3.5, tear index by 1.3 and 2.5, tensile index by 1.4 and 1.7, and zero-span tensile by 1.2 and 1.2 for *P. chrysosporium* and *D. squalens*, respectively.

The effects on sheet optical properties are similar for fungal and chemical pretreatments of the wood chips. Fungal pretreatments did not adversely affect opacity but decreased brightness and scattering coefficient (Table II). As with chemical pretreatment, the brightness of the two pulps could be restored to that of the control by use of hydrogen peroxide bleaching (Table I).

Fiber size distribution

Pretreatment with *D. squalens* greatly altered the fiber size distribution of the subsequently produced unbleached pulp, whereas pretreatment with *P. chrysosporium* did not (**Table III**). The *D. squalens* pretreatment decreased the pulp fines content [those fibers passing the 0.149-mm (100-mesh) screen] and maintained the longer fiber content (those fiber retained by the 0.149-mm screen). At a nominal 90-mL CSF, the *D. squalens* pretreatment decreased fines content from about 53% to 34%, and the fiber length index was higher with *D. squalens*. The pulp pretreated with *P. chrysosporium* was comparable to the control.

Comparison of the two fungi

Although the *D. squalens* pretreatment gave better handsheet proper-

II. Physical properties of 60-g/m² handsheets from unbleached refiner mechanical pulps

Property measured	*Untreated control*				*D. squalens*			*P. chrysosporium*			
Freeness, mL CSF	65	75	90	130	55	65	85	60	95	115	150
Burst index, kPa·m²/g	0.64	0.36	0.35	0.26	1.30	1.24	1.21	0.71	0.75	0.59	0.50
Tear index, mN·m²/g	1.66	1.62	1.69	1.74	4.10	4.24	4.30	2.00	2.14	2.21	2.52
Tensile index, Nm/g	25.4	21.6	21.3	19.1	39.3	36.0	36.6	30.0	30.6	28.4	27.2
Zero-span breaking length, kPa/m	0.189	0.171	0.170	0.164	0.221	0.211	0.201	0.183	0.201	0.202	0.183
Density, kg/m³	422	414	408	368	401	388	382	419	405	384	370
Brightness, %	51.0	51.1	51.4	50.4	40.2	39.8	39.7	41.0	40.7	40.4	40.0
Opacity, %	97.7	97.3	96.8	97.1	95.8	95.2	97.1	97.7	98.5	98.5	97.8
Scattering coefficient, m²/kg	62.8	59.9	59.0	57.6	41.9	40.7	45.7	53.3	54.2	52.1	50.6
Drainage time, s	54.2	25.7	16.5	10.9	17.4	14.4	13.0	17.0	13.3	10.0	8.1

III. Bauer-McNett screen analysis of unbleached pulps

Property measured	*Untreated control*				*D. squalens*			*P. chrysosporium*			
Freeness, mL CSF	65	75	90	130	55	65	85	60	95	115	150
Retained on 0.595-mm screen	0.35	0.45	0.85	1.30	15.78	16.85	18.70	0.20	0.50	0.20	0.65
Retained on 0.297-mm screen	6.30	9.35	12.50	16.65	28.23	27.40	28.35	8.65	12.65	1.90	2.10
Retained on 0.149-mm screen	30.65	30.60	33.55	33.50	21.05	19.95	20.25	31.80	36.35	53.50	52.60
Retained on 0.075-mm screen	23.50	24.15	23.00	17.75	11.80	10.50	11.50	19.45	19.00	16.75	14.60
Passing 0.075-mm screen	39.20	35.45	30.10	30.80	23.15	25.30	21.20	39.90	31.50	27.65	30.05
Fiber length index, mm	0.0696	0.0742	0.0825	0.0838	0.1092	0.1044	0.1162	0.0700	0.0815	0.0867	0.0836

ties than the *P. chrysosporium* pretreatment, direct comparisons of the two fungal treatments cannot be made. In these initial tests, the conditions used were not optimized for either fungus and were not the same. Studies are under way to evaluate the influence of treatment time, inoculum amount, and various culture and environmental parameters for these and other selected fungi.

Conclusions and further work

Pretreatment of aspen wood chips with the white-rot fungus *Phanerochaete chrysosporium* or *Dichomitus squalens* improved the strength properties of handsheets produced from unbleached refiner mechanical pulp. At equivalent pulp freeness, strength properties were as much as 3.5 times higher than those of handsheets produced from an untreated, unbleached control pulp. The fungal pretreatments decreased brightness and scattering coefficient but did not adversely affect opacity. Hydrogen peroxide bleaching restored the brightness of treated samples to that of the control.

Further studies are planned to evaluate other fungi, optimize the fungal pretreatments, evaluate other wood species, assess refining energy requirements, and attempt to elucidate the biochemical basis for the beneficial effect. Understanding of the basic processes should provide a rational basis for improving the fungal strains through mutation and genetic engineering.

Experimental procedures

Fungal pretreatment bioreactor

Aspen wood chips were pretreated in a specially designed bioreactor that held approximately 4 kg of chips (oven-dry basis) (Fig. 1). The bioreactor is constructed of 316 stainless steel consisting of two major parts: (a) an outer case (565 × 762 × 873 mm) with a large access door and ports for aeration and nutrient addition and (b) an inner drum (406 mm in diameter × 762 mm long) for holding the wood chips.

Rotation of the inner drum is done by a chain and sprockets and can be varied from 1 rev/min to 1 rev/day. The drum shell is made from a perforated plate having 3-mm-diameter holes occupying 40% of the area. This design allows aeration of the wood chips and drainage of metabolically produced water. The reactor fits into an autoclave for sterilization. During operation, the reactor resides within a temperature-controlled incubator adjustable to ±1°C, which also has ample room for isothermal incubation of possible support devices such as an inlet gas humidifier.

Fungal pretreatment procedure

Aspen (*Populus tremuloides*) pulpwood obtained from midwestern sources was barked and chipped to a nominal 6-mm chip size. Four kg of wood chips at 60% moisture content were loaded into the reactor drum. A glucose-containing, chemically defined medium (*28*) with 1/10 the reported nitrogen source (*L*-glutamate) was added to the chips. The chemically defined medium was used to increase fungal growth and suppress cellulose degradation.

The entire reactor was autoclaved at 120°C for 90 min. Upon removal from the autoclave, the hot reactor was sealed, allowed to cool to room temperature, and inoculated. Inoculation took place after the outer reactor jacket was sterilized with 70% ethanol, and this inoculaton was done in a sterile, laminar, air-flow hood. The inoculum was prepared by growing the fungus on 200 g of sterilized 6-mm aspen wood chips for 6 weeks. The reactor was maintained at the optimal growth temperature for each of the fungi used—37°C for *P. chrysosporium* (SC 26) and 25°C for *D. squalens* (TON-429). After mixing the inoculum with the sterilized wood chips for 3 h at 1 rev/h, the rotation schedules presented

in Fig. 2 were followed.

Fiberizing, refining, and evaluation

At the end of the pretreatment time, the wood chips were removed from the bioreactor and fiberized in a single rotating disk atmospheric refiner, 305 mm in diameter, at about 5% consistency and 88°C. We used 5–7 passes to refine the pulps to 55–140 mL CSF. After each pass, the pulp was dewatered to about 25% solids content before the next pass. Care was taken to prevent loss of fines by collecting the pulp in a closed container and dewatering using a vacuum crock and canvas bag. Untreated control pulp was produced from nonautoclaved chips, using the same refining and dewatering procedures.

For all pulps, 60-g/m² handsheets were made at each freeness according to TAPPI method T205. Pulp and handsheet properties were determined by standard procedures. Burst, tear, and tensile indices were measured according to TAPPI methods T403, T414, and T404, respectively. Zero-span breaking length values were determined using a Pulmac testing apparatus. Brightness, opacity, and light-scattering coefficients were determined using an Elrepho photometer. Screen classification was done according to TAPPI method T233, using a Bauer-McNett screen classifier (Tyler series 0.595, 0.297, 0.149, and 0.075 mm). Lignin content was determined by the method of Effland (*29*), and sugar content was measured by high-pressure liquid chromatography (HPLC) (*30, 31*).

Peroxide bleaching procedure

Refiner mechanical pulps prepared from the pretreated chips were bleached with hydrogen peroxide (*32*). Identical procedures were followed for both fungus-pretreated pulps. Before H_2O_2 treatment, the pulps were treated with 0.5% diethylenetriaminepentaacetic acid (DPTA) solution (percentage based on oven-dry pulp weight, pH 7) to chelate any metal ions that were present. Pulp samples (5-g oven-dry basis) were added to the chelating solution and mixed for 10 min with a magnetic stirrer. Pulp samples were washed with distilled water and were filtered to remove excess liquid.

The peroxide bleaching liquor consisted of 2% H_2O_2, 5% $NaSiO_2$, 0.1% $MgSO_4$, and 1.7% NaOH (percentages based on oven-dry pulp weight). A 5-g (oven dry basis) pulp sample and 20 mL of bleaching liquor were mixed in a plastic bag, submerged in a 50°C constant temperature water bath, and reacted for 90 min. The bleaching liquor had an initial pH of 11.5 and a final pH of 9.2. The pulp had a final pH of 10.9. Bleached pulp was neutralized with a 9.1% sodium bisulfite solution (percentage based on oven-dry pulp weight). It was rinsed and then filtered to remove excess liquid. Bleached pulp samples were made into handsheets and were evaluated for brightness only (Table I).

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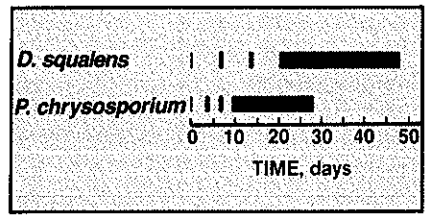
2. Bioreactor drum rotation schedules, showing periods of continuous rotation for fungal pretreatments



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