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Effect of Dose of Xylanase on Bleachability of Sugarcane Bagasse Ethanol/Water Pulps

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Abstract

Pulps obtained from the ethanol/water cooking of sugarcane bagasse were bleached with the xylanase enzyme obtained from the fungus *Thermomyces lanuginosus* IOC-4145 and with the commercial enzyme Cartazyme HS from Sandoz. By changing the enzyme dose from 4.3 to 36 IU/g of pulp, kappa number and viscosity were maintained when the xylanase from *T. lanuginosus* was used. On the other hand, by using Cartazyme HS, kappa number decreased by 17%, reaching 35.5. This pulp was further extracted with NaOH without a decrease in viscosity (10 cP), and pulp with a kappa number of 13 was obtained. Xylanases had no significant effect on the ethanol/water pulps.

Index Entries: Organosolv pulping; sugarcane bagasse; ethanol/water pulp; xylanase bleaching.

Introduction

Because of the increasing environmental need to eliminate the use of chlorine in pulp-bleaching plants, the development of bleaching processes that maintain the economic advantages of pulps has been intensively studied. An alternative method used in the bleaching process is the enzyme treatment already studied for kraft pulps *(1–3)*.

The use of enzymes (e.g., xylanases) for pulp treatment offers the benefits, of environmental protection and improved pulp quality. Among the various enzymes of interest to the paper industry, the hemicellulolytic xylanases have been found to be commercially feasible for pulp bleaching *(4)*.

Xylanase pretreatment facilitates chemical extraction of lignin from pulp, reducing consumption of bleaching chemicals and the discharge of toxic compounds into the environment *(4)*. Xylanases catalyze the hydrolysis of xylose-xylose bonds within the xylan chain and solubilize only a fraction of the total xylan present *(5)*.

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Studies using hemicellulases (xylanases in particular) have been carried out for the prebleaching of nonwoody fibers, such as bamboo and bagasse *(4–8)*.

In the present work, ethanol/water pulps obtained from sugarcane bagasse were bleached with the xylanase enzyme obtained from the fungus *Thermomyces lanuginosus* IOC-4145, and the results were compared with the commercial enzyme Cartazyme HS from Sandoz.

Materials and Methods

Pulping

Pulping of depithed sugarcane bagasse with a 1:1 (v/v) ethanol:water mixture was carried out in a closed and pressurized vessel at 185°C for 3 h. The product was filtered and the obtained pulp dried to determine yield. Pulp samples were analyzed by kappa number and viscosity following standard TAPPI methods *(9,10)*.

Bleaching

Samples of bagasse ethanol/water pulp (1.5 g) were suspended under agitation in 50 mL of water (3% consistency) at 30°C for 10 min. Cartazyme HS and xylanase enzyme obtained from *T. lanuginosus* IOC-4145 were added at a dose from 4.3 to 90 IU/g of dry pulp. The samples were maintained in a shaker at 30°C for 4 h, followed by filtration and washing with 300 mL of distilled water at 30°C for removal of the enzyme. One set of enzymatic bleached pulps was further submitted to alkaline extraction. Samples obtained at different bleaching times (3 g of dry pulp) and original pulp were extracted with 150 mL of 1 mol/L of NaOH at 65°C for 1 h under magnetic stirring. After filtration, the pulps were washed with 150 mL of 1 mol/L of NaOH at 65°C and further with distilled water at 65°C until reaching pH 6.0.

Dry pulps (3 g) were suspended in 150 mL of water (2% consistency) and heated to 70°C. Sodium chlorite (3.9 mL of 40% aqueous solution) and glacial acetic acid (0.6 mL) were added. The solution was further heated at 70°C for 5 min, and the bleached pulp obtained was exhaustively washed with water. Pulps were oven-dried at 60°C for 15 min and analyzed with respect to kappa number and viscosity by standard methods *(9,10)*.

Hydrolysis of Pulp

One gram of dry pulp was treated with 10 mL of 72% H_2SO_4 with stirring at 45°C for 7 min. The reaction was interrupted by adding 50 mL of distilled water, and the mixture was then transferred to a 500-mL Erlenmeyer flask, and the volume reached 275 mL. The flask was autoclaved for 30 min at 1.05 bar and 120°C for complete hydrolysis of oligomers *(11)*. The mixture was filtered and the volume of the hydrolysate was made up to 500 mL with distilled water. The sample (40 mL) of the hydrolysate was diluted to 50 mL and the pH was adjusted to 2.0 with 2 mol/L NaOH. After filtration

in a Sep-Pak C_{18} cartridge to remove aromatic compounds, the hydrolysate was analyzed in an Aminex HPX-87H column (300 × 7.8 mm) (Bio-Rad) at 45°C using a Shimadzu chromatograph and refraction-index detector. The mobile phase was 0.005 mol/L H_2SO_4 at 0.6 mL/min. Sugar concentrations reported as xylan and glucan were determined using calibration curves of pure compounds.

Results and Discussion

Ethanol/water pulps of sugarcane bagasse were bleached by xylanase of two sources: *T. lanuginosus* and Cartazyme HS. Pulps were treated with 4.3–90 IU/g of xylanase at 30°C.

Values of the viscosity and kappa number of the ethanol/water pulps obtained in the xylanase treatment (X) and in the sequence xylanase-alkaline extraction (XE) are given in Table 1. The kappa number of the ethanol/water original pulp (42.8) was greater than that of Acetosolv pulps prepared for a previous study *(8)* (kappa number of the original pulp = 28), showing that the delignification with ethanol/water was not as efficient.

The results of the viscosity and kappa number of the pulps bleached with the enzyme were compared with the control and the bleached pulp with sodium chlorite. It was found that the results of enzymatic bleached pulps were poor, since there was no effect of xylanase treatment.

By using the endoxylanase from *T. lanuginosus* alone a slight decrease in kappa number of 8% occurred (42.8–39.3). However, when the second alkaline extraction step was added, the decrease in kappa number was 67% (42.8–14.1). There was no increase in the viscosity with the enzymatic bleaching followed by the alkaline extraction in comparison with the procedure with extraction alone. The cumulative effect of the alkaline extraction with the enzymatic treatment was not observed as it was found in the bleaching of Acetosolv pulps *(8)*.

The concentration of the endoxylanase obtained from *T. lanuginosus* did not improve the efficiency of the treatment; however, for Cartazyme HS the concentration of 36 U/g presented the best results in terms of kappa number reduction (17%, 42.8–35.5). Viscosity ranged from 10 to 12 cP in all experiments. Apparently, kappa number 13 was the limit reached in the alkaline extraction or xylanase sequence of ethanol/water pulps. The results in Table 1 indicate that xylanase treatment had a small effect on kappa number reduction and the xylanase-alkaline extraction sequence had no effect when compared to alkaline extraction alone.

Yields for the pulps obtained after bleaching with the enzyme obtained from *T. lanuginosus* IOC-4145 and with Cartazyme HS were about 88–92% and for the bleaching with sodium hydroxide and sodium chlorite were about 75%.

The content of xylan was preserved after the treatment with endoxylanase from *T. lanuginosus*. Only after alkaline extraction was a reduction in xylan detected, which means that xylanase acts over xylans (*see* refs. *4* and *5*), but the fragments formed did not seem to be easily released.

Table 1
Viscosity, kappa Number, Yield, and Chemical Composition of Different Treated Ethanol/Water Pulps[a]

	Endoxylanase from *T. lanuginosus*								Cartazyme							
Bleaching sequence[b]	Viscosity (cP)	kappa no.	Yield (%)	Total lignin (%)	Glucan (%)	Xylan (%)	Xylan/ glucan ratio	Acetyl (%)	Viscosity (cP)	kappa no.	Yield (%)	Total lignin (%)	Glucan (%)	Xylan (%)	Xylan/ glucan ratio	Acetyl (%)
C	10.8	5.6	74.6	5.9	73.8	11.6	0.15	0	10.8	5.6	74.6	5.9	73.8	11.6	0.15	0
E	11.9	14.1	76.6	4.5	81.9	5.9	0.07	0	11.9	14.1	76.6	4.5	81.9	5.9	0.07	0
4.3 IU/g																
X	10.1	39.3	90.0	9.7	72.0	10.8	0.15	2.6	9.9	36.4	88.4	7.0	49.1	7.4	0.15	0
XE	12.1	14.0	77.0	5.2	82.3	6.5	0.08	0	11.2	13.7	78.3	3.7	67.7	5.2	0.08	0
18 IU/g																
X	10.2	39.3	92.0	9.8	71.7	10.7	0.15	2.6	10.1	38.5	91.6	8.8	55.2	8.1	0.15	0
XE	12.0	14.6	77.6	5.3	81.6	6.3	0.08	0	11.3	13.8	78.7	3.7	68.2	5.0	0.07	0
36 IU/g																
X	10.1	39.6	90.0	11.3	66.9	10.7	0.15	4.2	9.8	35.5	87.6	11.5	68.7	10.5	0.15	5.9
XE	11.5	13.1	75.1	5.4	79.4	6.5	0.08	3.9	9.9	13.2	75.2	6.0	71.1	5.9	0.08	0
90 IU/g																
X	10.1	39.8	87.7	11.3	69.4	10.7	0.15	3.9	10.5	38.1	90.0	10.9	68.8	10.5	0.15	4.7
XE	9.6	14.0	76.1	5.0	79.0	6.5	0.08	3.7	11.1	12.9	80.0	5.0	80.1	6.3	0.08	4.3

[a] Control: viscosity = 11.1 cP; kappa no. = 36.9. Original pulp: viscosity = 9.8 cP; kappa no. = 42.8; 54.4% yield; 9.9% total lignin; 69.8% glucan; 10.7% xylan; xylan/glucan ratio = 0.15; 2.5% acetyl.

[b] C, chlorite bleaching; E, alkaline extraction; X, xylanase; XE, xylanase followed by alkaline extraction.

Alkaline extraction makes solubilization of lignin and both xylans and xylan fragments feasible. Application of Cartazyme HS at 4.3–18 IU/g degraded 24–31% of the xylan.

Little differences in the amount of xylans were detected when the results of the original pulp were compared with those of the bleached pulp with the xylanase obtained from *T. lanuginosus*. However, for Cartazyme HS, higher xylanase preservation was obtained with a greater concentration of the enzyme (36 and 90 U/g). Comparison of the results of enzyme bleaching with those of chlorite bleaching revealed that xylans were also preserved.

Comparison of the original and enzyme-treated pulps revealed that the xylan/glucan ratio was maintained. Xylan/glucan ratio decreased only after the treatment with enzyme followed by the alkaline extraction. The decrease in the xylan/glucan ratio is owing mainly to the alkaline extraction, and probably the main function of the enzyme is modification of xylan structure.

The acetyl groups were preserved even after the use of endoxylanase. After alkaline extraction, acetyl groups were totally removed. On the other hand, Cartazyme HS had a greater bleaching action. High enzyme doses seem to promote modification on xylan, maintaining acetyl groups that are not removed after alkaline extraction. With sodium chlorite, the xylan/glucan ratio was also preserved and the acetyl groups were removed.

Conclusion

The kappa number, viscosity, and hydrolysis values obtained were close for the two enzymes. For the ethanol/water pulps, a cumulative effect of the alkaline extraction with the enzymatic treatment was not observed. A kappa number of 13 was the limit reached with the ethanol/water pulp after alkaline extraction with or without xylanase treatment. In future work to elucidate the action of xylanase on ethanol/water pulps, the parameters will be better evaluated at lower severity.

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