Amenability of Acacia and Eucalyptus Hardwood Pulps to Elemental Chlorine-Free Bleaching: Application and Efficacy of Microbial Xylanase

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This study outlines the results of a biobleaching study of acacia (*A. mangium*) and eucalyptus (*E. globulus*) hardwood kraft pulps with commercial xylanase (Optimase CX 72 L). The comparative study was carried out using an elemental chlorine-free (ECF) bleaching sequence ($D_0E_PD_1D_2$) after the enzyme (X) stage. The enzyme treatment resulted in improved optical properties with a reduction in bleach chemical consumption. At an equivalent bleach chemical consumption, a brightness gain of 2.1 and 1.7 units and a whiteness gain of 2.7 and 2.3 units were observed with xylanase treatment in acacia and eucalyptus pulps, respectively. In ECF bleaching using the $D_0E_PD_1D_2$ sequence, a final brightness was achieved to the extent of 90% ISO and 89% ISO for acacia and eucalyptus, respectively, at an equivalent charge of bleach chemicals. The post-color (PC) number was also reduced by up to 45% for both hardwood pulps compared with the control. The bleachability of acacia was observed to be significantly higher than that of eucalyptus. In addition, a 17.0% and 23.0% reduction in chlorine dioxide and sodium hydroxide, respectively, were obtained for both hardwood pulps after xylanase pre-bleaching, thus indicating an environmentally friendly approach to the process.

Keywords: Chlorine dioxide; ECF bleaching; Post-color number; Pulp properties; Xylanase

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INTRODUCTION

Several pulp and paper mills are currently adopting enzymatic bleaching as an alternative to chlorine-based chemical bleaching, as such an approach can reduce or eliminate the use of chlorine and its compounds and therefore address environmental concerns and governmental regulations (Selvam and Arungandhi 2013; Sharma *et al.* 2014). In recent years, there has been increasing interest in applying green technology to the bleaching process to reduce pollution as well as to improve the quality of the pulp produced (Torres *et al.* 2000; Zhao *et al.* 2006). Chlorine dioxide-based elemental chlorine-free (ECF) bleaching for the pulp and paper industry offers a number of obvious benefits over the traditional methods. ECF bleaching results in significantly superior pulp quality in addition to providing an environmentally beneficial process. The enzymatic pretreatment of pulps before ECF bleaching can be of further advantage in terms of reducing the consumption of bleach chemicals, and therefore reducing the amount of pollutants generated (Bajpai 1999).

The concept of the addition of enzymes for improving the delignification of pulp is well known, and xylanase is the most common type of enzyme used for this purpose.

Biobleaching, *i.e.*, the enzymatic pre-treatment of pulp with xylanases (endo-1,4-xylanase activity, EC 3.2.1.8.) before chemical bleaching is an alternative and cost-effective process for reducing the consumption of bleach chemicals and for minimizing the generation of toxic chlorinated organic substances in the effluents of bleach mills (Viikari *et al.* 1994; Call and Mycke 1997; Suurnakki *et al.* 1997; Bajpai *et al.* 2006; Shatalov and Pereira 2008). The use of xylanases results in better optical properties and reduces the use of bleaching chemicals (Valls and Roncero 2009). Xylanase treatment reduces the xylan concentration in the secondary wall of the fiber surface, particularly in hardwood pulps, thus enhancing pulp bleachability and increasing the permeability of the fiber surface (Viikari *et al.* 1996; Kim and Paik 2000; Roncero *et al.* 2000). Xylan concentration is present in the range of 11 to 12.5% and 16 to 20% in *Acacia mangium* and *Eucalyptus globulus*, respectively (Pinto *et al.* 2005; Evtuguin and Neto 2007).

Xylanase enhances the effects of bleaching chemicals rather than removing lignin directly. The enzyme does not attack lignin-based chromophores, but rather the xylan network by which residual lignin particles are trapped in the pulp. Limited hydrolysis of the xylan network is often sufficient to facilitate the subsequent chemical removal of lignin without sacrificing yield. The viscosity of the pulp is also improved as a result of xylanase treatment. However, the viscosity of the pulp is adversely affected when cellulase activity is present. Therefore, cellulase activity by the enzyme preparation is undesirable in enzymatic bleaching (Bajpai and Bajpai 1997).

Studies have demonstrated that factors such as type of raw material, sequence of bleaching, and type of xylanase could significantly affect enzymatic treatment (Gallardo *et al.* 2010). Presently, xylanases are being modified to work at alkaline pH values and higher temperatures (Gangwar *et al.* 2014). Keeping in view the limited reports available on the bleach-boosting properties of xylanase on acacia (*A. mangium*) kraft pulp and its comparative study with eucalyptus (*E. globulus*) kraft pulps, the present study aimed at examining the influence of xylanase on the optical and physical properties of both pulps and bleach chemical consumption over the conventional bleaching process.

EXPERIMENTAL

The plantation of acacia and eucalyptus is usually done in several parts of India viz; West Bengal, Tamil Nadu, Uttar Pradesh, Madhya Pradesh, Maharashtra and Karnataka. Acacia (*A. mangium*) and eucalyptus (*E. globulus*) wood chips were procured from different pulp and paper mills in India. The cooking was done in a stationary digester using the kraft process. The cooking experiments were performed at different temperatures, durations, and active alkali doses to optimize the pulping conditions and obtain pulps with a kappa number in the range of 18 to 20 for both types of wood pulp.

Optimase CX 72L sourced from Dupont Genencor Sciences was used in the present study. It has low or negligible cellulase activity and is designed for a bleach-boosting effect. This enzyme is alkali and thermostable in nature, which are favorable conditions for the enzymatic bleaching process. The optimum temperature, pH, and activity of Optimase CX 72L were 55 °C, 8.0, and 46,876 IU, respectively.

The activity of xylanase and CMCase were determined according to the methods outlined by Bailey *et al.* (1992) and Ghose (1987), respectively. The activities were assayed by measuring the release of reducing sugars (xylose and glucose) from birch wood xylan and carboxymethyl cellulose (CMC). One international unit of enzyme was defined as the amount of enzyme that released 1 µmole of xylose or glucose sugars per minute per mL of enzyme.

The kneading mechanism was followed for the proper dilutions and mix of enzymes to be used in the experiments. The control pulp was also treated in the same way as the xylanase-treated pulp in each experiment, where the enzyme was replaced with water. The efficacy of the xylanase enzyme was determined for acacia and eucalyptus hardwood pulps based on pulp yield, kappa number reduction, and brightness gain of the pulp. After the enzyme treatment (X stage), the pulps were washed and subjected to ECF bleaching.

Enzyme-treated and untreated wood pulps were bleached using the ECF bleaching sequence ($D_0E_PD_1D_2$) to explore the potential savings on chlorine dioxide and sodium hydroxide and to determine the gain in final brightness and whiteness of the bleached pulps. The processing conditions (pH, temperature, time, and consistency) used at various bleaching stages were maintained as indicated in Table 1.

Table 1. Processing Conditions Used during ECF Bleaching ($D_0E_PD_1D_2$)

Parameter	Chlorine Dioxide (D_0) Stage	Alkali Extraction (E_P) Stage	Chlorine Dioxide (D_1) Stage	Chlorine Dioxide (D_2) Stage
Consistency (%)	5	10	10	10
Time (min)	45	120	180	180
Temperature (°C)	60	80	75	75
pH	1.8 to 2.0	10.5 to 11.0	3.0 to 4.0	3.0 to 4.0

Analytical Methods

Kappa number was determined according to TAPPI test method T236 om-99 (1999). Kappa number is the volume (in milliliters) of 0.1 N potassium permanganate solution consumed by one gram of moisture-free pulp under the specified conditions. The results were corrected to 50% consumption of the permanganate added. The viscosity of the pulp was determined by the capillary viscometer method according to TAPPI test method T230 om-99 (1999). The viscosity of the 0.5% cellulose solution was determined, using 0.5 M cupriethylenediamine as a solvent and a capillary viscometer. Viscosity gives an indication of the average degree of polymerization of the cellulose. The brightness (% ISO) and CIE whiteness of the pulps were measured using a Konica Minolta instrument according to TAPPI test methods T452 om-02 (2002) and T560 om-96 (1996), respectively. The post-color (PC) number was measured according to TAPPI test method T260 om-85 (1985). It is a measurement of reversion in brightness of the paper at a specified time and temperature.

Canadian Standard Freeness (CSF) was measured according to TAPPI test method T227 om-94 (1994), in which the freeness of the pulp was designed to give a measure of the rate at which a dilute suspension of pulp (3 g of pulp in 1 L of water) may be drained. Handsheets were prepared with a smooth and reproducible surface for reflectance measurements according to T205 sp-02 (2002). Bursting strength was checked using TAPPI test method T403 om-97 (1997), and is defined as the maximum hydrostatic pressure required to produce a rupture of the material when a controlled and constantly increasing pressure is applied through a rubber diaphragm. Tearing strength was also measured according to T414 om-98 (1998), in which multiple sheets of the sample material were torn together through a fixed distance using the pendulum.

RESULTS AND DISCUSSION

Enzyme-treated pulps showed a reduction in kappa number of 4.6%, 8.0%, and 10.1% for acacia pulp and 4.1%, 7.8%, and 9.8% for eucalyptus pulp at xylanase doses of 0.1, 0.3, and 0.5 kg/tp, respectively, compared with the control (Table 2). These observations on the reduction in kappa number by xylanase were also supported by Thakur *et al.* (2012), wherein Pulpzyme HC was used and a 4.2% reduction in kappa number of eucalyptus pulp was achieved. Brightness gains of 2.4, 2.7, and 2.9 units were obtained in acacia wood pulp, while brightness gains of 2.3, 2.4, and 2.8 units were observed in eucalyptus pulp at enzyme doses of 0.1, 0.3, and 0.5 kg/tp, respectively, compared with the control (Table 2). Shatalov and Pereira (2008) obtained 1.2 to 1.6 units of brightness gain after xylanase treatment on eucalyptus hardwood pulp, which is relatively lower than the observations obtained in the present study. A minor loss in yield was also observed in both acacia and eucalyptus wood pulps in the range of 0.2% to 0.6% with xylanase doses of 0.1, 0.3, and 0.5 kg/tp compared with the control (Table 2).

Table 2. Characterization of Enzyme-treated and Untreated Pulp after Enzymatic Treatment

Particulars	Set	Xylanase Dose (kg/tp)	Yield (%)	Kappa Number		Brightness	
				Value	Reduction (%)	Value (%ISO)	Gain (unit)
Acacia Kraft pulp	Control	---	99.8 ± 0.23	19.7 ± 0.50	---	28.5 ± 0.58	---
	Xylanase-treated	0.1	99.5 ± 0.31	18.8 ± 0.42	4.6	30.9 ± 0.35	2.4
		0.3	99.4 ± 0.35	18.1 ± 0.42	8.1	31.2 ± 0.38	2.7
		0.5	99.2 ± 0.45	17.7 ± 0.25	10.2	31.4 ± 0.36	2.9
Eucalyptus Kraft pulp	Control	---	99.7 ± 0.40	19.3 ± 0.42	---	28.7 ± 0.40	---
	Xylanase-treated	0.1	99.5 ± 0.32	18.5 ± 0.35	4.1	31.0 ± 0.31	2.3
		0.3	99.3 ± 0.25	17.8 ± 0.40	7.8	31.1 ± 0.44	2.4
		0.5	99.1 ± 0.38	17.4 ± 0.35	9.8	31.5 ± 0.15	2.8

Xylanase indirectly affects the lignin degradation in the pulps as it breaks the linkage between xylan and the hexenuronic acid component, facilitating better penetration of bleach chemicals into the fibre. Increased penetration of bleach chemicals invariably reduced their consumption in the process. In addition, a key reduction was seen in the consumption of chlorine dioxide and sodium hydroxide in ECF bleaching, as shown in Table 3. A reduction in chlorine dioxide of 14.3% was also reported in the eucalyptus pulp during enzymatic bleaching (Thakur *et al.* 2012).

Bleaching experiments, performed at both an equivalent kappa factor (KF=0.22) and reduced kappa factor (KF=0.187), resulted in decreasing consumption of 14.9% to 17.8% chlorine dioxide, along with a reduction of sodium hydroxide in the range of 18.9% to 23.5% for acacia pulp compared with the control (Table 3: AcK pulp) at a reduced kappa number. A similar reduction was also observed for eucalyptus wood pulp (Table 3: EuK pulp), wherein a reduced use of chlorine dioxide and sodium hydroxide to 14.6% to 17.6% and 18.4% to 23.1%, respectively, was required at the reduced kappa factor compared with the control.

Table 3. Bleach Chemical Consumption in ECF Bleaching of Acacia (AcK) and Eucalyptus (EuK) Pulps

Particulars		Control	At Equivalent KF (0.22)			At Reduced KF (0.187)		
			0.1 (kg/t)	0.3 (kg/t)	0.5 (kg/t)	0.1 (kg/t)	0.3 (kg/t)	0.5 (kg/t)
AcK Pulp	Applied ClO_2 (kg/tp)	27.6	26.8	26.3	25.9	23.5	23.0	22.7
	ClO_2 reduction (%)	---	2.90	4.71	6.16	14.86	16.67	17.75
	NaOH (kg/tp)	21.7	20.7	19.9	19.5	17.6	16.9	16.6
	NaOH reduction (%)	---	4.61	8.29	10.14	18.89	22.12	23.50
EuK Pulp	Applied ClO_2 (kg/tp)	27.3	26.6	26.0	25.7	23.3	22.8	22.5
	ClO_2 reduction (%)	---	2.56	4.76	5.86	14.65	16.48	17.58
	NaOH (kg/tp)	21.2	20.4	19.6	19.1	17.3	16.6	16.3
	NaOH reduction (%)	---	3.77	7.55	9.91	18.40	21.70	23.11

Handsheets of the final pulp were prepared and evaluated for the desired optical properties. The results indicated that the bleachability of acacia was significantly higher than that of eucalyptus pulp. The final brightness and efficacy of the enzyme were higher in acacia than in eucalyptus pulp. The final pulp brightness and whiteness of the enzyme-treated pulps was superior to untreated pulp at both equivalent and reduced amounts of bleach chemicals. Brightness gains of approximately 2.1 and 0.9 units were observed at equivalent and reduced bleach chemical consumption in the bleaching of acacia pulp, respectively (Fig. 1(a)). A similar trend was observed in whiteness improvement (up to 2.7 and 1.6 units) at equivalent and reduced bleach chemical consumption in the enzymatic treatment of acacia pulp (Fig. 1(b)). Correspondingly, 1.7 and 0.8 units of brightness gain and whiteness gains of 2.3 and 1.5 units were obtained in the bleaching process of eucalyptus pulp at equivalent and reduced bleach chemical consumption, respectively (Figs. 1a and 1b). The observations of Gallardo *et al.* (2010) on hardwood pulp showed a brightness gain in the range of 0.7 to 1.0 units more than the control pulp with the xylanase enzyme.

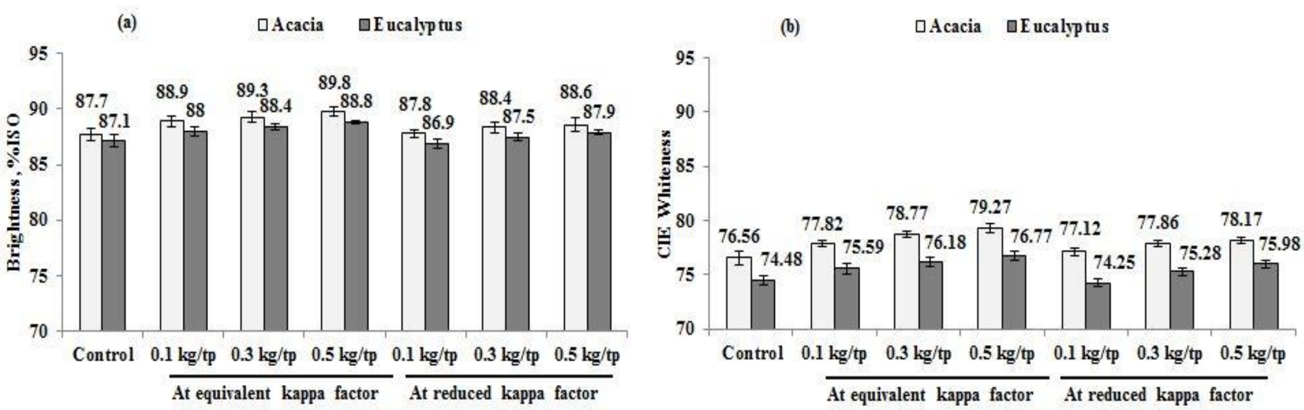

Fig. 1. (a) Brightness (% ISO) and (b) CIE whiteness of the final bleached pulp samples at equivalent and reduced kappa factors

The viscosity of the final acacia pulp was also determined and found to be similar to that of untreated pulp (Fig. 2a). The reduction in post-color (PC) number increased with increasing doses of xylanase and was higher when the pulp was treated at the same chemical dosages (Fig. 2b).

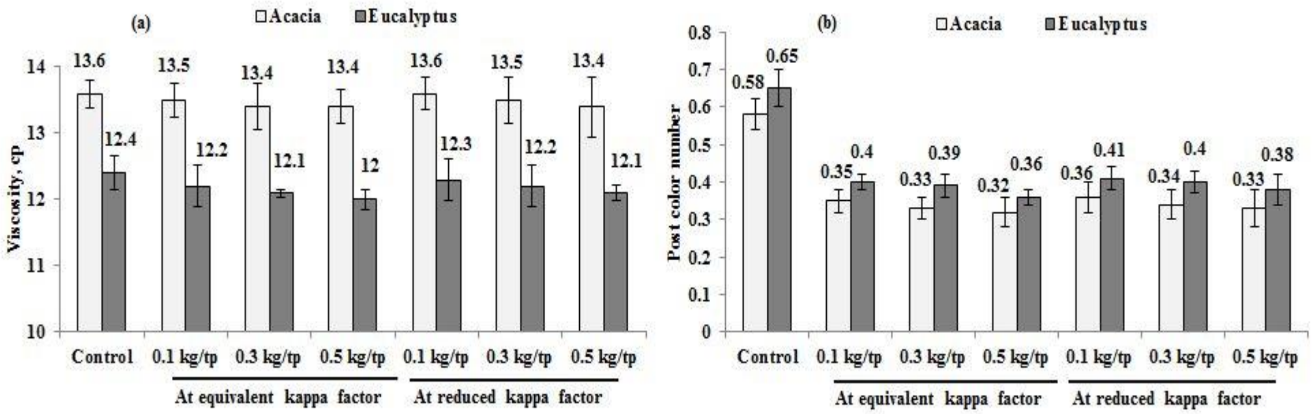

Fig. 2. (a) Viscosity and (b) Post-color number of the final bleached pulp samples at equivalent and reduced chemical charges

Here as well, the same trend of final bleached pulp viscosity was obtained for eucalyptus wood pulp (Fig. 2a), and it was nearly similar to that of untreated pulp. As in acacia pulp, a reduction in PC number was observed with eucalyptus pulp.

Table 4. Final Pulp Properties of Enzyme-treated and Untreated Acacia (AcK) and Eucalyptus (EuK) Pulps

Particulars		Control	At Equivalent KF (0.22)			At Reduced KF (0.187)		
			0.1 (kg/t)	0.3 (kg/t)	0.5 (kg/t)	0.1 (kg/t)	0.3 (kg/t)	0.5 (kg/t)
AcK Pulp	Shrinkage (%)	5.1 ± 0.32	5.3 ± 0.15	5.6 ± 0.35	5.9 ± 0.40	5.2 ± 0.15	5.5 ± 0.20	5.7 ± 0.26
	Strength properties							
	PFI Revolutions (PFI)	3600	3600	3600	3600	3600	3600	3600
	Initial CSF	619	625	628	630	628	631	634
	End CSF	431	436	445	452	443	454	461
	Grammage (g/m^2)	70.81	70.25	70.19	70.67	70.89	71.17	71.03
	Burst index (kN/g)	2.6 ± 0.31	2.6 ± 0.29	2.5 ± 0.15	2.6 ± 0.45	2.5 ± 0.38	2.6 ± 0.25	2.6 ± 0.17
	Tear index (mN.m^2/g)	6.2 ± 0.45	6.2 ± 0.35	6.2 ± 0.29	6.0 ± 0.25	6.1 ± 0.26	6.0 ± 0.35	5.9 ± 0.31
EuK Pulp	Shrinkage (%)	5.5 ± 0.30	5.8 ± 0.32	6.2 ± 0.10	6.4 ± 0.15	5.7 ± 0.45	6.1 ± 0.21	6.2 ± 0.21
	Strength properties							
	PFI Revolutions (PFI)	2000	2000	2000	2000	2000	2000	2000
	Initial CSF	584	587	591	593	592	594	597
	End CSF	422	438	447	455	441	456	465
	Grammage (g/m^2)	70.22	70.59	70.79	70.46	71.01	70.76	70.43
	Burst index (kN/g)	4.2 ± 0.23	4.2 ± 0.10	4.2 ± 0.15	4.3 ± 0.17	4.2 ± 0.06	4.1 ± 0.15	4.1 ± 0.25
	Tear index (mN.m^2/g)	7.3 ± 0.44	7.2 ± 0.21	7.1 ± 0.31	7.1 ± 0.25	7.2 ± 0.38	7.2 ± 0.42	7.0 ± 0.30

However, it was higher when the pulp was treated at reduced chemical doses, as shown in Figs. 2a and b. According to Kim and Paik (2000), xylanase treatment was responsible for the removal of carboxylic acids and their counter ions, which resulted in color reversion. By the partial removal of xylan in the pulps, xylanase treatment increased brightness stability.

The strength properties of eucalyptus were found to be higher than in acacia pulp. Enzyme-treated and untreated acacia and eucalyptus hardwood pulps were refined at 3600 and 2000 PFI revolutions, respectively. To achieve the same CSF level, more PFI revolutions were required for acacia wood fibers. Comparable results were observed in terms of the bursting strength of enzyme-treated and untreated acacia (Table 4: AcK) and eucalyptus (Table 4: EuK) wood pulps. The enzyme treatment showed a slightly negative impact on the tearing strength of acacia and eucalyptus pulps as the action of the xylanases reduced the intrinsic fibrillar strength due to the removal of superficial hemicelluloses (Bajpai 1999). This is presumably the reason underlying the reduced tear index after the enzymatic pretreatment of wood pulps.

CONCLUSIONS

1. A comparative study of acacia and eucalyptus kraft pulps was carried out in an ECF bleaching sequence after xylanase treatment with commercial xylanase (Optimase CX 72L), in which the optical and physical properties of the final bleached pulp and paper were tested.
2. The results showed enzymatic bleaching exhibited higher selectivity in the case of acacia kraft pulp.
3. The reduction of bleach chemicals consumption was also observed to be higher in acacia than eucalyptus wood pulps with xylanase enzyme treatment before the ECF bleaching sequence. A reduction in chlorine dioxide (ClO_2) and sodium hydroxide (NaOH) by more than 17.0% and 23.0%, respectively, was noticed for both hardwood pulps, with an improvement in the optical properties of the pulps.
4. The strength properties of eucalyptus pulp were observably better than acacia pulp.
5. Marginal increment in pulp shrinkage was observed in both hardwood pulps as compared to their respective control with shrinkage lesser in acacia than eucalyptus pulp.
6. Reduction in post color number by more than 44% was observed in both hardwood pulps.
7. The present results provide a clear indication of the advantage of xylanase treatment, and a better selectively of acacia over eucalyptus for use in the paper-making process, as these approaches resulted in the decreasing consumption of bleach chemicals, leading to a reduced cost in the treatment of hazardous chemicals and improved qualities of the final paper.
8. Many commercial xylanase products are available in market now and there may be significant variations in the bleaching response with different xylanase products towards final bleached pulp qualities.

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REFERENCES CITED

Bailey, M. J., Biely, P., and Poutanen, K. (1992). "Inter laboratory testing of methods for assay of xylanase activity," *Journal of Biotechnology* 23(3), 257-270. DOI: 10.1016/0168-1656(92)90074-J

Bajpai, P. (1999). "Application of enzymes in the pulp and paper industry," *Biotechnology Progress* 15(2), 147-157. DOI: 10.1021/bp990013k

Bajpai, P., Anand, A., Sharma, N., Mishra, S. P., Bajpai, P. K., and Lachenal, D. (2006). "Enzymes improve ECF bleaching of pulp," *BioResources* 1(1), 34-44. DOI: 10.15376/biores.1.1.34-44

Bajpai, P., and Bajpai, P. K. (1997). "Microbial xylanolytic enzyme system: Properties and applications," in: *Advances in Applied Microbiology*, Neildleman, S., Laskin, A., (eds.), Vol. 43, Academic Press, New York, NY, pp. 141-194. DOI: 10.1016/s0065-2164(08)70225-9

Call, H. P., and Mycke, I. (1997). "History, overview and applications of mediated lignolytic systems, especially laccase-mediator-systems (Lignozym®-process)," *Journal of Biotechnology* 53(2-3), 163-202. DOI: 10.1016/S0168-1656(97)01683-0

Evtuguin, D. V., and Neto, P. C. (2007). "Recent advances in eucalyptus wood chemistry: Structural features through the prism of technological response," *3° International Colloquium on Eucalyptus Pulp*, Belo Horizonte, Brazil.

Gallardo, O., Fernandez, M. F., Vallis, C., Valenzuela, S. V., Roncero, M. B., Vidal, T., Diaz, P., and Pastor, F. I. J. (2010). "Characterization of a family GH5 xylanase with activity on neutral oligosaccharides and evaluation as a pulp bleaching aid," *Applied and Environmental Microbiology* 76(18), 6290-6294. DOI: 10.1128/AEM.00871-10

Gangwar, A. K., Prakash, N. T., and Prakash, R. (2014). "Applicability of microbial xylanases in paper pulp bleaching: A review," *BioResources* 9(2), 3733-3754. DOI: 10.15376/biores.9.2.3733-3754

Ghose, T. K. (1987). "Measurement of cellulase activities," *Pure and Applied Chemistry* 59(2), 257-268. DOI: 10.1351/pac198759020257

Kim, D. H., and Paik, K. H. (2000). "Effect of xylanase pre and post treatment on oxygen bleaching of oak kraft pulp," *J. Industrial and Engineering Chemistry* 6(3), 194-200.

Pinto, P. C., Evtuguin, D. V., and Neto, C. P. (2005). "Effect of structural features of macromolecular components of hardwood cell walls on wood pulping and bleaching performance," *Journal of Industrial & Engineering Chemistry Research* 44(26), 9777-9784. DOI: 10.1021/ie050760o

Roncero, M. B., Torres, A. L., Colom, J. F., and Vidal, T. (2000). "Effects of xylanase treatment on fibre morphology in totally chlorine free bleaching (TCF) of eucalyptus pulp," *Process Biochemistry* 36(1), 45-50. DOI: 10.1016/S0032-9592(00)00178-3

Selvam, K., and Arungandhi, K. (2013). "Biobleaching and delignification of hard wood kraft pulp (HWKP) by *Trametes sp.*, *Ganoderma sp.* and *Poria sp.*," *International*

Journal of Plant, Animal and Environmental Sciences 3(3), 96-100.
Sharma, A., Thakur, V. V., Shrivastava, A., Jain, R. K., Mathur, R. M., Gupta, R., and Kuhad, R. C. (2014). "Xylanase and laccase based enzymatic kraft pulp bleaching reduces adsorbable organic halogen (AOX) in bleach effluents: A pilot scale study," *Bioresource Technology* 169, 96-102. DOI: 10.1016/j.biortech.2014.06.066
Shatalov, A. A., and Pereira, H. (2008). "Effect of xylanases on peroxide bleachability of eucalypt (*E. globulus*) kraft pulp," *Biochemical Engineering Journal* 40(1), 19-26. DOI: 10.1016/j.bej.2007.11.012
Suurnakki, A., Tenkanen, M., Buchert, J., and Viikari, L. (1997). "Hemicellulases in the bleaching of chemical pulps," in: *Biotechnology in the Pulp and Paper Industry*, Eriksson, K. (ed.), Springer-Verlag, Berlin, pp. 261-287. DOI: 10.1007/bfb0102077
T205 sp-02 (2002). "Forming handsheets for physical tests of pulp," TAPPI Press, Atlanta, GA.
T227 om-94 (1994). "Freeness of pulp," TAPPI Press, Atlanta, GA.
T230 om-99 (1999). "Viscosity of pulp," TAPPI Press, Atlanta, GA.
T236 om-99 (1999). "Kappa number of pulp," TAPPI Press, Atlanta, GA.
T260 om-85 (1985). "Test to evaluate the ageing properties of bleached chemical pulps," TAPPI Press, Atlanta, GA.
T403 om-97 (1997). "Bursting strength of paper," TAPPI Press, Atlanta, GA.
T414 om-98 (1998). "Internal tearing resistance of paper (Elmendorf type method)," TAPPI Press, Atlanta, GA.
T452 om-02 (2002). "Brightness of pulp, paper, and paperboard (directional reflectance at 457 nm)," TAPPI Press, Atlanta, GA.
T560 om-96 (1996). "CIE whiteness and tint of paper and paperboard (using d/0, diffuse illumination and normal viewing)," TAPPI Press, Atlanta, GA.
Thakur, V. V., Jain, R. K., and Mathur, R. M. (2012). "Studies on xylanase and laccase enzymatic prebleaching to reduce chlorine based-chemicals during CEH and ECF bleaching," *BioResources* 7(2), 2220-2235. DOI: 10.15376/biores.7.2.2220-2235
Torres, A. L., Roncero, M. B., Colom, J. F., Pastor, F. I. J., Blanco, A., and Vidal, T. (2000). "Effect of a novel enzyme on fibre morphology during ECF bleaching of oxygen delignified *Eucalyptus* kraft pulps," *Bioresource Technology* 74(2), 135-140. DOI: 10.1016/S0960-8524(99)00178-9
Valls, C., and Roncero, M. B. (2009). "Using both xylanase and laccase enzymes for pulp bleaching," *Bioresource Technology* 100(6), 2032-2039. DOI: 10.1016/j.biortech.2008.10.009
Viikari, L., Kantelinen, A., Sundquist, J., and Linko, M. (1994). "Xylanases in bleaching: From an idea to the industry," *FEMS Microbiology Letters* 13(2-3), 335-350. DOI: 10.1111/j.1574-6976.1994.tb00053.x
Viikari, L., Suurnakki, A., and Buchert, J. (1996). "Enzyme-aided bleaching of kraft pulps: Fundamental mechanisms and practical applications," *Enzymes for Pulp and Paper Processing* 655, 15-24. DOI: 10.1021/bk-1996-0655.ch002
Zhao, J., Li, X., and Qu, Y. (2006). "Application of enzymes in producing bleached pulp from wheat straw," *Bioresource Technology* 97(13), 1470-1476. DOI: 10.1016/j.biortech.2005.07.012

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