Properties of Enzyme Pretreated *Wikstroemia sikokiana* and *Broussonetia papyrifera* Bast Fiber Pulps

Lang-Dong Lin,[a] Fang-Chih Chang,[b,]* Chun-Han Ko,[b,c,]* C. Y. Wang,[c] and Ya-Nan Wang [c]

Xylanase, pectinase complex, and BL11 pectinase were employed for the pretreatment of gampi and paper mulberry bast fiber pulps prior to chlorine dioxide bleaching. The bleaching efficiencies of the pulps with different enzymatic pretreatments were investigated. Accelerated aging by heat-humidity treatment was also conducted to evaluate yellowing phenomena and to estimate the prevention of brightness reversion (brightness retention) by enzymatic pretreatment. The order of active chlorine required with respect to pretreatment was pectinase complex > xylanase > BL11 pectinase for soda and soda/oxalate gampi pulps and pectinase complex > BL11 pectinase > xylanase for soda and soda/oxalate paper mulberry pulps. Higher brightness retention values were observed for soda/oxalate pulps compared to soda pulps. The brightness retention levels for gampi pulps and mulberry pulps after ClO_2 bleaching with enzymatic pretreatment were higher than the levels of ClO_2 and NaClO bleaching pulps. Enzymatic treatments were thus able to reduce the usage of ClO_2 and to assist in producing photo-stable paper materials for art and artifact-repairing applications. Thus, enzymatic pretreatment of the pulp has the potential to meet world trends and environmental sustainability for pulp and paper industries.

Keywords: Bast fiber; Brightness; Broussonetia papyrifera (L.) Hert. ex Vent; Pectinase; Polygalacturonase; Wikstroemia sikokiana Franch. et Sav.; Xylanase

Contact information: a: Department of Cultural Heritage Conservation, National Yunlin University of Science and Technology, Yunlin 640, Taiwan; b: The Experimental Forest, National Taiwan University, No. 12, Sec. 1, Chien-Shan Rd., Chu-Shan, Nan-Tou 55750, Taiwan; c: School of Forestry and Resource Conservation, National Taiwan University, Taipei 10617, Taiwan;
** Corresponding author: d90541003@ntu.edu.tw; chunhank@ntu.edu.tw*

INTRODUCTION

Pulping chemistry has been thoroughly studied for more than 100 years. The pulping and commercial processes have been refined and optimized to produce high-quality paper at low prices. Changes in wood resources have prompted researchers to explore whether fibers other than softwood and hardwood can be used for papermaking (Han and Rymsza 1999). Non-wood resources are important raw materials in many countries where wood is not available in sufficient quantities to meet the demand for pulping and papermaking (Salmela *et al.* 2008).

Fibers can be grouped into three categories: wood, non-wood (grass, bast, leaf, or fruit fibers), and non-plant (Saijonkari-Pahkala 2001). Bast fibers have been utilized to obtain high-quality paper (Marques *et al.* 2010; Shi *et al.* 2011; Tahir *et al.* 2011; Jin *et al.* 2012). Gampi (*Wikstroemia sikokiana*) and paper mulberry (*Broussonetia papyrifera*) are bast fibers that have a long history of use in hand papermaking (Hubbe and Bowden

2009). They are sustainable and reproducible raw materials and have been applied in the making of conventional oriental art paper. In comparison to wood pulps, non-wood pulps produced from specific plants are more heterogeneous, and the ability to process these materials into paper products would have the additional benefit of mitigating disposal costs and the associated environmental pollution (Spiridon 2007). It is also expected that non-wood fibers will play an increasingly important role in the pulp and paper industry due to diminishing forest resources, environmental issues, growing paper consumption, and progress in advanced technologies of non-wood utilization (Hammett *et al.* 2001; Salmela *et al.* 2008).

Enzymes including lipases, esterases, pectinases, hemicellulases, cellulases, and ligninolytic enzymes have been applied to improve the properties of woody and nonwoody fibers (Spiridon 2007; Fillat *et al.* 2010; Pathak *et al.* 2011; Maijala *et al.* 2012; Martín-Sampedro *et al.* 2012a,b; Virk *et al.* 2012). Biobleaching and bioprocessing of pulps using enzymes is the most suitable biological alternative to environmentally unfriendly chemicals (Ninawe and Kuhad 2006; Dhiman *et al.* 2008; Kumar and Satyanarayana 2012; Iqbal *et al.* 2013; Jegannathan and Nielsen 2013). Enzymes are green chemicals that can improve pulp and paper operations because they require less energy in refining, less consumption of bleaching agents, and lower toxic pollution loading (Spiridon 2007; Bajpai 2010; Lian *et al.* 2012). To meet environmental and legislative requirements in the market, the pulp and paper industry is modifying delignification, bleaching, and effluent technologies to reduce the environmental impact. Thus, elimination of chlorine in the bleaching process and the application of chlorine-free bleaching processes has become increasingly important (Spiridon 2007; Reinstaller 2008; Singh *et al.* 2011).

The main aim of this study is to investigate the effects of enzymatic pretreatment (xylanase, pectinase complex, and BL11 pectinase) on the pulp properties of gampi and paper mulberry bast fibers, which are suitable for art paper. The bleaching agent efficiency on pulp after the enzymatic pretreatments was analyzed. Accelerated aging by heat-humidity treatment were also conducted to evaluate yellowing phenomena and brightness retention.

EXPERIMENTAL

Materials

Gampi (*Wikstroemia sikokiana*) is a protophyte found in the Philippines whose bark was imported for the experiments. The inner bark of a one-year-old paper mulberry (*Broussonetia papyrifera*) tree was imported from Thailand. Gampi and paper mulberry bark were cut into pieces approximately 5 to 7 cm in length. Certain bark was dried in an oven to measure moisture content. Both gampi and paper mulberry bark were treated with either 15% NaOH or 15% NaOH with 1.35% $Na_2C_2O_4$ for 90 min before being transferred to the digester. The chemical compositions of gampi and paper mulberry pulp are shown in Table 1. A significant decrease in the xylose content was observed, which shows that much of the xylose was solubilized during the pulping processes. However, most of the lignin was still retained in the pulp. The high lignin content yielded pulps that are prone to yellowing during storage (Baty *et al.* 2010).

Table 1. Chemical Compositions of Gampi and Paper Mulberry Pulp

Sample (g/100 g oven-dried meal)	Gampi			Paper mulberry		
	Raw	Soda	Soda with oxalate	Raw	Soda	Soda with oxalate
Alcohol-benzene solubility	7.20± 0.54	3.24±0.13	4.84±0.23	6.60±0.54	2.14±0.33	3.16±0.38
Ash	4.33±0.24	2.19±0.24	2.81±0.21	4.12±0.84	1.33±0.13	0.87±0.03
Lignin	12.00±1.43	7.40±0.33	8.80±0.56	6.00±1.23	4.50±0.77	4.63±0.65
Holocellulose	61.25±5.22	32.15±2.45	26.55±0.43	74.55±3.98	33.75±5.46	38.25±3.89
α-Cellulose	50.19±6.34	26.43±3.65	24.44±1.25	55.96±4.12	23.12±0.50	29.43±2.69
Pentosan	16.32±2.88	9.13±1.23	8.88±0.87	7.88±1.33	3.32±0.53	3.08±1.10
Glucose	41.94±0.54	23.80±0.43	18.08±0.36	31.96±0.43	24.40±0.09	27.58±0.23
Xylose	8.06±0.23	2.76±0.14	2.06±0.10	3.78±0.11	0.62±0.10	0.54±0.06
Galacturonic acid	3.72±0.12	0.41±0.01	0.44±0.03	2.94±0.04	0.43±0.01	0.59±0.01
Arabinose	2.93±0.08	2.00±0.11	1.88±0.23	1.99±0.02	1.78±0.12	1.81±0.08
Total sugar	56.65±0.55	28.98±0.24	22.46±0.28	40.67±0.35	27.24±0.14	30.52±0.20
Kappa number	-	44.80±1.00	45.78±0.30	-	12.04±1.10	14.80±0.30
Pulp yield	-	52.79±3.54	56.57±3.94	-	44.77±6.71	43.67±3.87

Data provided as the mean of triplicate ± the standard deviation

Xylanase used in this study was Pulpzyme HC, provided by Novozymes (Denmark). Pulpzyme HC is produced by a genetically modified *Bacillus* microorganism under submerged fermentation and was used in experiments to catalyze the hydrolysis of deacetylated xylan substrates. Pectinase used in this study was Novozym 863, provided by Novozymes. Novozym 863 is a highly active pectolytic enzyme preparation produced by a selected strain of *Aspergillus aculeatus*. The BL11 pectinase was produced by the selected strain BL11 *Paenibacillus campinanesis*. The bacterial strains were screened and isolated from thermo-alkaline black liquor (58 °C, pH 9.83) from Hsinying Mill of the Taiwan Pulp and Paper Company (Tainan, Taiwan).

Methods

Bleaching stages

Four types of pulp were placed in polyethylene bags and then treated with chlorine dioxide for bleaching. The control pulp was treated with ClO_2 at Kappa factors of 0.05, 0.1, 0.15, 0.2, and 0.3. Applied chlorine dioxide charge is defined as active chlorine value divided 2.63. The chlorine dioxide dose of D_0 is usually expressed using the Kappa factor, where a greater Kappa factor is used as more lignin is removed. DE (D: Chlorine dioxide, E: Alkaline extraction) bleaching materials were obtained from an industrial source, with bleaching stage conditions of pH 2 adjusted by HCl, 0.25% active chlorine as Cl_2, and 5% consistency at 50 °C for 1 h. Alkaline extraction was carried out at 10% consistency, at 70 °C for 1 h. Pulp with a Kappa factor of 0.3 was then treated with a DD bleaching succession. Pulp brightening was carried out at 10% consistency with either 0.35% or 0.15% ClO_2 at 70 °C for 3.5 h. After each bleaching stage, the pulp was washed with deionized water and dewatered using a 75-μm mesh. The NaClO bleaching was at 5% consistency, 70 °C, and pH 11 for 1 hr, then compared with chlorine dioxide bleaching.

For gampi soda pulp, gampi soda pulp with oxalate, paper mulberry soda pulp, and paper mulberry soda pulp with oxalate, enzymatic pretreatments were carried out in polyethylene bags with 60 g of air-dried pulp at 10% consistency in a water bath at 50 °C for xylanase and BL11 pectinase and at 35 °C for pectinase complex. Xylanase, pectinase complex, and BL11 pectinase dosages ranged from 0 to 3000 IU/g oven-dried pulp (g o. d. p.) for 3 h at pH 3.5, 7, and 10, respectively. To ensure complete mixing, the enzyme was mixed into the pulp by hand kneading. Prior to full DE bleaching, the enzymatic treated pulps were thoroughly washed and dewatered using a 75-μm mesh. After pretreatment with enzymes, pulps were bleached with the DE sequence and treated in parallel with the control group.

Analytical methods

All tests were performed according to standard methods as follows: forming handsheets for physical tests of pulp (TAPPI T205 2002); physical testing of pulp handsheets (TAPPI T220 2010); tensile properties (TAPPI T494 2006); tearing strength (TAPPI T414 2012); low bursting strength (CNS 1353 2008); and folding endurance (TAPPI T511 2013). The basis weight of each physically tested handsheet was 60 g/m^2. Kappa number, a measure of lignin content, was determined by reaction of pulp samples with acidified potassium permanganate (TAPPI T236 2013). Brightness was determined by measuring the reflectance at 457 nm on handsheets using the Technidyne brightness meter (TAPPI T452 2008). Brightness retention was determined using the Technidyne brightness meter according to the TAPPI test method T452. Samples were treated at 85 °C and 87% RH for 16 h. The calculation of brightness retention was performed according to Chang *et al.* (1998). The surface structure of unbleached and bleached pulp were analyzed by scanning electron microscope (SEM) provided with Vega TC software.

RESULTS AND DISCUSSION

Pulp Properties of Enzymatic Pretreatment after Chlorine Dioxide Bleaching

Compared with the results found by Ko *et al.* (2010), xylanase pretreatment at 2.5 IU/g.o.d.p. increased brightness by 4.6% for hardwood kraft pulp using a Kappa factor of 0.1 in the D_0 stage of DED bleaching, and xylanase pretreatment at 6 IU/g.o.d.p. increased the brightness of gampi soda pulp by 6.93%, oxalate gampi soda pulp by 6.00%, paper mulberry soda pulp by 5.33%, and oxalate paper mulberry soda pulp by 3.60%.

Increasing the dosage of xylanase treatment on pulp had the expected result of decreasing Kappa number and increasing brightness (Table 2). In terms of both gampi and paper mulberry Kappa number and brightness, adding oxalate increased Kappa number and decreased brightness. The low bleaching efficiency of soda pulp with oxalate addition may be attributed to lower xylose content compared to soda pulp with no addition. The xylanolytic enzyme system has less glycan to convert into its constituent sugars. From the experimental results, we suggest that adequate dosages of xylanase are 300 IU/g.o.d.p. for gampi soda pulp, 60 IU/g.o.d.p. for gampi soda pulp with oxalate, 1200 IU/g.o.d.p. for paper mulberry soda pulp, and 1200 IU/g.o.d.p. for paper mulberry soda pulp with oxalate.

Increasing the dosage of pectinase complex decreased the Kappa number and increased brightness obviously (Table 2). The bleaching efficiency was higher with pectinase complex pretreatment compared to xylanase pretreatment. In terms of both gampi and paper mulberry, adding oxalate increased the value of the Kappa number. Oxalate addition had a negative impact on the brightness of gampi pulp only for the 3000 IU/g.o.d.p. pectinase complex dosage. Oxalate addition had a negative impact on the brightness of paper mulberry at most pectinase complex dosages. Based on the experimental results, it is suggested that adequate dosages of pectinase complex are 600 IU/g.o.d.p. for gampi soda pulp, 3000 IU/g.o.d.p. for gampi soda pulp with oxalate, 600 IU/g.o.d.p. for paper mulberry soda pulp, and 240 IU/g.o.d.p. for paper mulberry soda pulp with oxalate.

Table 2. Impact of Enzymatic Addition on Brightness of 0.15 Kappa Factor Bleached Pulp

Dosage (IU/g.o.d.p)	Xylanase				Pectinase complex			
	Gampi soda	Gampi soda with oxalate	Paper mulberry soda	Paper mulberry soda with oxalate	Gampi soda	Gampi soda with oxalate	Paper mulberry soda	Paper mulberry soda with oxalate
0	46.94±0.82	46.46±0.46	63.57±1.00	56.23±0.57	46.94±0.82	46.94±0.82	63.50±0.20	56.23±0.57
6	48.33±0.20	46.70±0.44	63.70±0.15	56.50±0.33	47.18±0.44	47.32±0.54	63.53±0.80	56.25±0.30
12	48.40±0.23	47.00±0.26	63.90±0.28	56.90±0.53	47.25±0.35	47.37±0.51	63.55±1.00	56.25±0.57
18	48.60±0.20	47.60±0.53	64.00±0.36	57.10±0.57	47.35±0.27	47.42±0.46	63.57±0.51	56.40±0.31
60	48.82±0.52	48.60±0.46	64.14±0.47	57.60±0.53	49.29±0.90	50.77±0.78	63.80±1.50	56.47±0.30
300	51.10±0.36	48.86±0.53	65.00±0.33	57.80±0.43	51.20±0.50	52.88±0.60	64.92±0.22	61.06±0.40
600	51.18±0.36	49.74±0.78	65.46±0.53	57.85±0.15	52.98±0.22	55.00±0.43	67.00±0.40	62.77±0.82
1200	51.50±0.20	49.88±0.26	65.82±0.57	58.00±0.28	53.52±0.38	56.42±0.25	67.94±0.45	63.67±0.95
1800	52.70±0.43	49.90±0.53	66.10±0.53	58.55±0.36	53.80±0.30	57.58±0.59	68.55±0.65	64.00±0.35
3000	52.90±0.82	49.94±0.46	66.12±0.43	58.59±0.47	54.32±0.44	58.50±0.54	69.50±0.20	64.77±0.87

Data provided as the mean of triplicate ± the standard deviation

If the objective is to obtain a target brightness of 47.5% GE, then xylanase pretreatment can decrease the usage of chlorine dioxide. A xylanase dosage of 3000 IU/g.o.d.p. can decrease required active chlorine *per* g.o.d.p. for gampi soda pulp by 33.3%, and a xylanase dosage of 1200 IU/g.o.d.p. can decrease required active chlorine *per* g.o.d.p. for gampi soda pulp with oxalate by 14.29%. If the objective is to obtain a target brightness of 59% GE, a xylanase dosage of 1200 IU/g.o.d.p. can decrease required active chlorine *per* g.o.d.p. for paper mulberry soda pulp by 33.33% and a xylanase dosage of 3000 IU/g.o.d.p. can decrease required active chlorine *per* g.o.d.p. for paper mulberry soda pulp with oxalate by 25.00% (Fig. 1a). Pectinase complex can decrease the usage of chlorine dioxide for gampi and paper mulberry pulp. If the objective is to obtain a target brightness of 47.5% GE, then a pectinase complex dosage of 600 IU/g.o.d.p. can decrease required active chlorine *per* g.o.d.p. for gampi soda pulp with oxalate by 66.67% and a pectinase complex dosage of 60 IU/g.o.d.p. can decrease the required active chlorine *per* g.o.d.p. for gampi soda pulp with oxalate by 33.73%. If the objective is to obtain a target brightness of 59%, then a pectinase complex dosage of 600 IU/g.o.d.p. can decrease the required active chlorine *per* g.o.d.p. for paper mulberry soda pulp by 50.00% and a pectinase complex dosage of 240 IU/g.o.d.p. can decrease the required active chlorine *per* g.o.d.p. for paper mulberry soda pulp with oxalate by 60.11% (Fig. 1b).

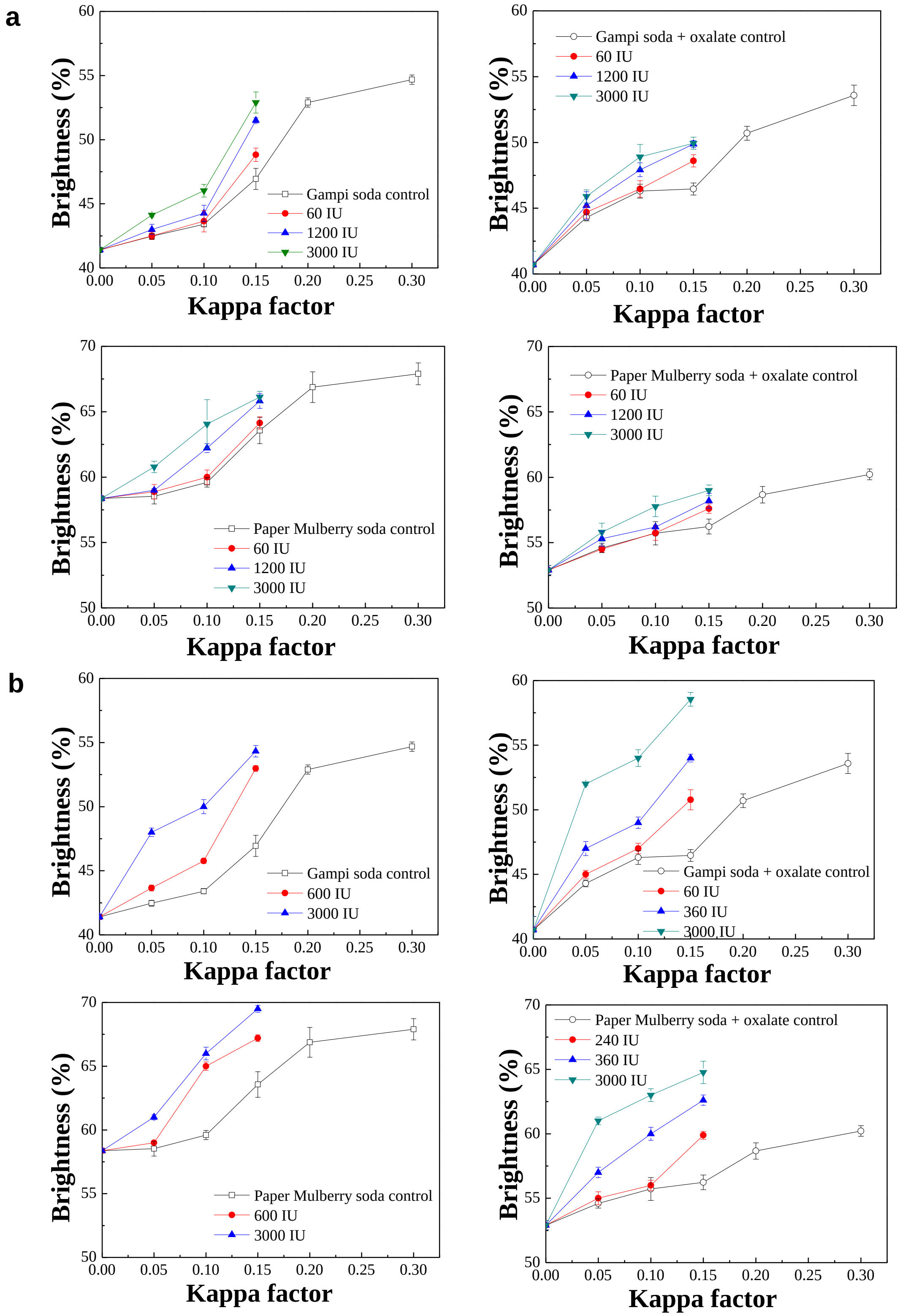

Fig. 1. Impact of different dosages of (a) xylanase and (b) pectinase on the brightness of pulp. Data provided as the mean of triplicate ± the standard deviation

To obtain a target brightness of 47.5% GE, a pectinase complex dosage of 170 IU/g.o.d.p. can decrease the required active chlorine *per* g.o.d.p. for gampi soda pulp with oxalate by 14.29%. To obtain a target brightness of 59% GE, a pectinase complex dosage of 260 IU/g.o.d.p. can decrease the required active chlorine *per* g.o.d.p. for gampi soda pulp with oxalate by 33.33%. Even if brightness was not as high as in the case of a pectinase complex, they still exceeded the results for xylanase (Fig. 2).

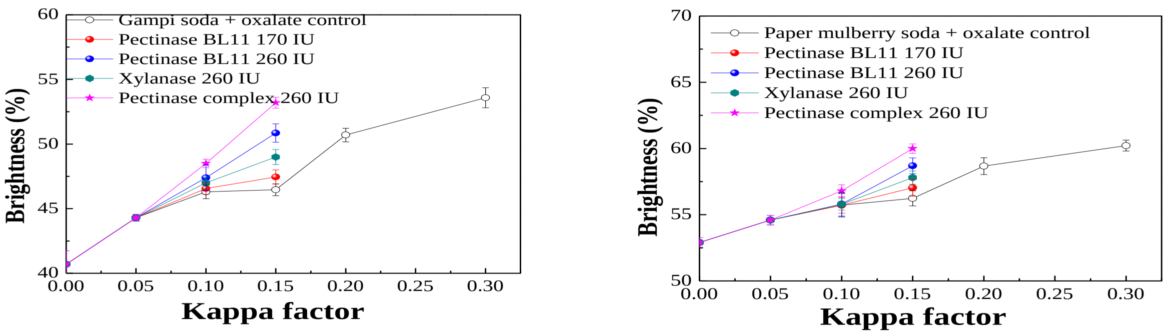

Fig. 2. Effect of enzymes on the brightness of pulp. Data provided as the mean of triplicate ± the standard deviation

The surface structure of unbleached pulp is shown in Fig. 3a. Gampi and paper mulberry fibers are slender and remain intact after pulping, although gampi pulp has more impurities. The SEM micrographs of ClO_2 bleached pulp are shown in Fig. 3b.

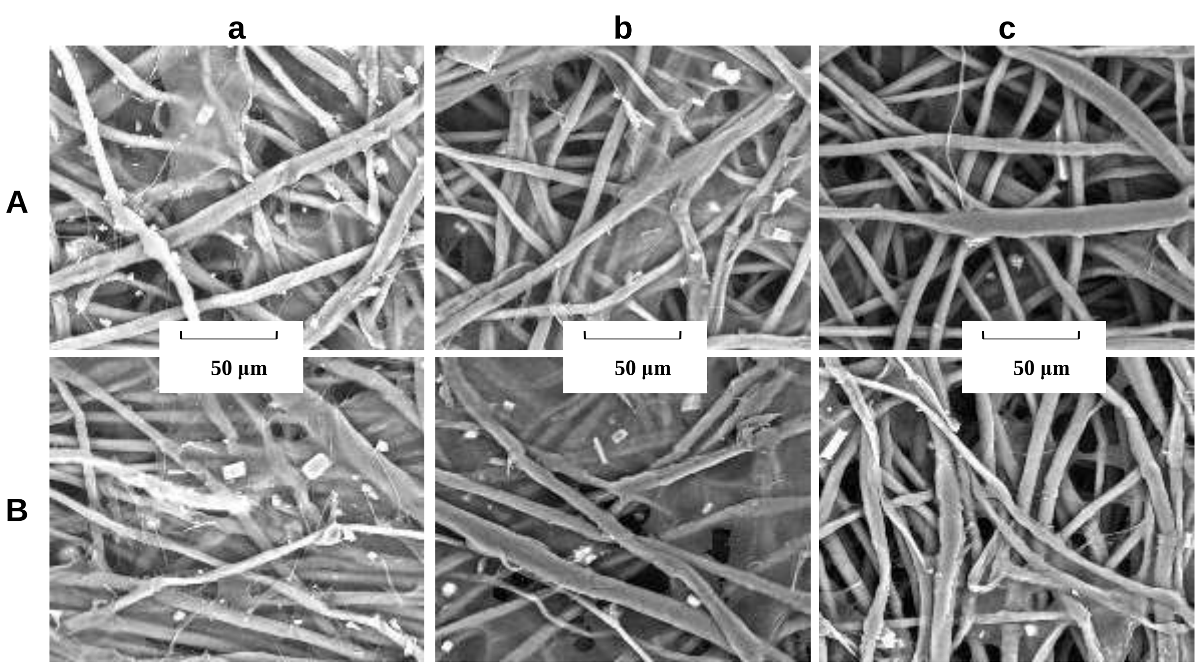

Fig. 3. SEM micrographs obtained for (A) gampi soda pulp and (B) gampi soda pulp with oxalate for (a) unbleached pulps, (b) 0.15 Kappa factor ClO_2 bleached pulps, and (c) after 0.15 Kappa factor ClO_2 bleaching with pectinase BL11 260 IU/g.o.d.p pretreatment

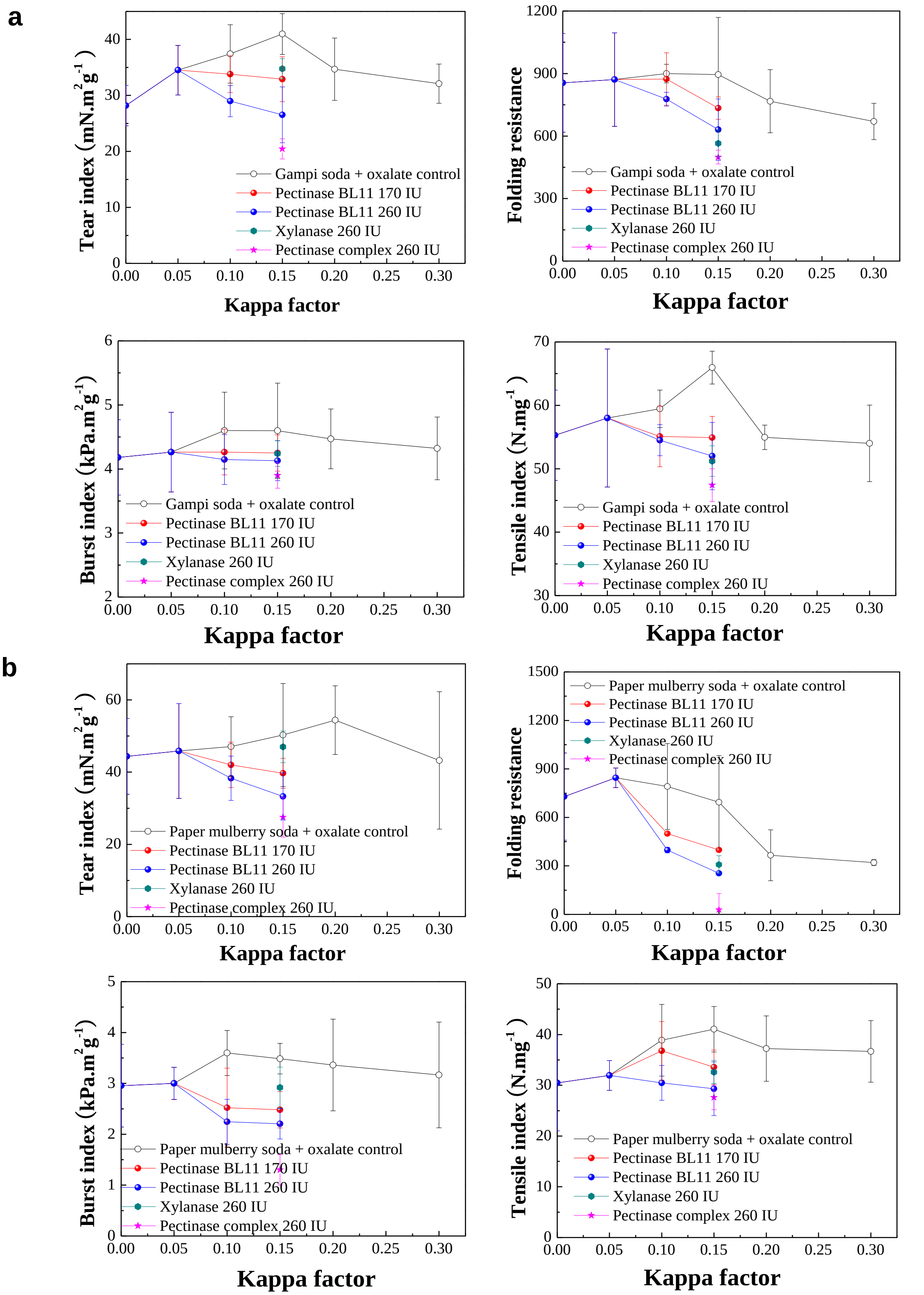

Fig. 4. Effect of enzymes on handsheet properties of (a) gampi soda pulp with oxalate and (b) paper mulberry soda pulp with oxalate. Data provided as the mean of triplicate ± the standard deviation.

As the chemical dosage of the bleaching sequence $D_0ED_1D_2$ was increased, the impurities of all pulps decreased. At a dosage of 0.15 Kappa factor on D_0E, gampi and paper mulberry pulp had a lot of impurities (Fig. 3b). However, there were fewer pulp impurities with BL11 pectinase pretreatment (Fig. 3c). Only gampi soda pulp with oxalate had few impurities.

In this study, oxalate addition had no significant effect on the tear index, folding resistance, burst index, or tensile index of gampi pulp or paper mulberry pulp (at level of $P < 0.05$ according to the Scheffe test). Thus, oxalate addition did not improve the handsheet properties. The handsheet properties of pulps decreased as enzymatic dosage increased. The handsheet properties of pectinase complex pretreated pulp were lower than those of pulp pretreated with xylanase or BL11 pectinase. The BL11 pectinase pretreatment yielded better handsheet properties than pectinase complex pretreatment (Fig. 4).

Prevention of Brightness Reversion

The brightness retention (B.R.) of NaClO bleaching decreased to 60 to 70% after aging. Only paper mulberry soda pulp obtained high brightness retention. Oxalate addition had a positive effect on the brightness retention of 0.15 NaClO Kappa factor bleached gampi pulp. While the bleaching dosage increased, the brightness retention of gampi pulp decreased (Table 3). Compared with the NaClO bleaching process, ClO_2 bleaching can improve brightness retention, but paper mulberry soda pulp had a lower initial brightness retention. Oxalate addition had positive effects on all ClO_2 bleached pulps (Table 3).

Oxalate addition had a significant effect on the brightness retention of xylanase pretreated 0.15 Kappa factor bleached pulps. The use of xylanase in the post-treatment of bleached hardwood kraft pulp resulted in an obvious reduction of yellowing (Gangwar *et al.* 2014). The best xylanase dosages for increasing brightness retention are 60 IU/g.o.d.p. for gampi soda pulp, 6 IU/g.o.d.p. for gampi soda pulp with oxalate, 60 IU/g.o.d.p. for paper mulberry soda pulp, and 6 IU/g.o.d.p. for paper mulberry soda pulp with oxalate (Table 4). Paper mulberry soda pulp can give higher brightness retention than paper mulberry soda pulp with oxalate (Table 3). Oxalate addition had no obvious effect on pectinase complex pretreated 0.15 ClO_2 Kappa factor bleached gampi pulp.

Table 3. Brightness Retention (%/%) of NaClO or ClO_2 Bleached Gampi and Paper Mulberry Pulp

Bleached with	Kappa factor	Gampi Soda	Gampi Soda with oxalate	Paper mulberry soda	Paper mulberry soda with oxalate
None	0	94.49±0.29	96.49±0.34	81.00±0.15	88.36±1.85
NaClO	0.15	75.80±0.60	79.42±1.19	88.61±0.74	77.03±1.18
	0.3	74.58±0.15	75.65±1.01	80.58±0.43	79.57±0.69
ClO_2	0.05	95.72±0.02	99.38±1.00	71.47±0.06	91.61±0.11
	0.1	94.54±0.42	98.79±1.23	75.20±0.35	95.43±0.45
	0.15	96.75±0.38	99.08±0.98	73.56±0.65	95.88±0.75
	0.2	86.91±0.11	98.88±0.94	73.03±0.44	95.63±0.11
	0.3	87.85±0.10	98.96±1.03	78.21±0.54	95.87±0.48

Data provided as the mean of triplicate ± the standard deviation

Table 4. Brightness Retention (%/%) of Gampi and Paper Mulberry at 0.15 Kappa Factor and ClO_2 Bleached Pulp Pretreated with Xylanase, Pectinase Complex, and BL11 Pectinase

Enzymatic dosage		Gampi Soda	Gampi soda with oxalate	Paper mulberry soda	Paper mulberry soda with oxalate
Raw	0	94.49±0.29	96.49±0.34	81.00±0.15	88.36±1.85
Xylanase	60	96.40±0.45	100.95±0.55	101.56±1.24	103.14±0.56
	300	95.74±0.32	98.79±0.65	97.96±0.55	103.70±0.94
	600	98.37±0.99	98.79±0.32	104.69±0.13	102.69±1.20
	1200	99.81±1.01	102.02±0.48	100.26±0.45	103.60±1.54
	1800	99.50±1.05	98.79±0.56	102.32±0.89	103.38±1.41
	3000	99.81±0.89	98.79±0.99	99.51±0.85	104.47±0.54
Pectinase complex	60	109.01±0.82	108.30±1.44	102.82±0.20	98.00±0.89
	120	109.80±0.78	104.97±0.56	101.63±1.50	95.74±1.04
	240	104.47±0.43	102.06±1.24	102.37±1.00	98.58±1.56
	360	109.80±0.54	104.41±1.00	104.97±0.80	99.94±0.67
	600	101.30±0.30	108.07±0.55	101.40±0.40	97.28±1.34
	3000	106.75±0.43	102.55±0.65	100.33±0.40	96.78±1.65
BL11 Pectinase	170	104.99±0.84	102.41±1.65	109.18±1.67	105.24±1.96
	260	100.05±0.55	102.41±0.65	103.54±1.54	99.86±0.78

Data provided as the mean of triplicate ± the standard deviation

It is suggested that pectinase complex dosage should be increased to 60 IU/g.o.d.p. of each pulp for the best brightness retention. From Tables 3 and 4 it can be seen that the enzyme pretreatment can improve brightness retention. Pulp has the least resistance to brightness reversion under the high temperature and humidity. The enzymes used in this study have a potential industrial use.

CONCLUSIONS

1. The enzymatic treatments were able to reduce the active chlorine usage of chlorine dioxide on the achievement of the same Kappa value, where pectinase complex pretreatments saved the largest values of active chlorine. The BL11 pectinase was the most effective biological alternative to conventional bleaching chemicals.
2. The most appropriate doses of enzymes were found to be 300 IU/g.o.d.p. of xylanase for gampi soda pulp, 60 IU/g.o.d.p. for gampi soda pulp with oxalate, 1200 IU/g.o.d.p. for paper mulberry soda pulp, and 1200 IU/g.o.d.p. for paper mulberry soda pulp with oxalate; 600 IU/g.o.d.p. of pectinase complex for gampi soda pulp, 3000 IU/g.o.d.p. for gampi soda pulp with oxalate, 600 IU/g.o.d.p. for paper mulberry soda pulp, and 240 IU/g.o.d.p. for paper mulberry soda pulp with oxalate; and 170 IU/g.o.d.p. of BL11 pectinase for both gampi and paper mulberry pulps.
3. Xylanase assisted bleaching was achieved through the cleavage of xylan and its associated lignin-carbohydrate complexes. Hence, resulting pulp after enzyme pretreatment will have low residual xylan and galacturonic acid contents.

4. Pretreatment by enzymes can decrease brightness reversion under high temperature and high humidity and assist in producing photo-stable paper materials for art and artifact-repairing applications.

REFERENCES CITED

Bajpai, P. K. (2010). "Solving the problems of recycled fiber processing with enzymes," *BioResources* 5(2), 1311-1325. DOI: 10.15376/biores.5.2.1311-1325

Baty, J. W., Maitland, C. L., Minter, W., Hubbe, M. A., and Jordan-Mowery, S. K. (2010). "Deacidification for the conservation and preservation of paper-based works: A review," *BioResources* 5(3), 1955-2023. DOI: 10.15376/biores.5.3.1955-2023

CNS 1353 (2008). "Method of test for low bursting strength of paper and board," *Chinese National Standards*, Taiwan.

Chang, S. T., Chen, H. T., Ku, Y. C., and Chang, H. L. (1998). "Effects of light on the properties of Chinese writing and painting paper," *Bull. Taiwan For. Res. Inst.* 3(2), 85-92.

Dhiman, S. S., Sharma, J., and Battan, B. (2008). "Industrial applications and future prospects of microbial xylanases: A review," *BioResources* 3(4), 1377-1402. DOI: 10.15376/biores.3.4.1377-1402

Fillat, U., Pepió, M., Vidal, T., and Roncero, M. B. (2010). "Flax fibers as a raw material: How to bleach efficiently a non-woody plant to obtain high-quality pulp," *Biomass Bioenerg.* 34(12), 1896-1905. DOI: 10.1016/j.biombioe.2010.07.022

Gangwar, A. K., Prakash, N. P., 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

Hammett, A. L., Youngs, R. L., Sun, X., and Chandra, M. (2001). "Non-wood fiber as an alternative to wood fiber in China's pulp and paper industry," *Holzforschung* 55(2), 219-224. DOI: 10.1515/HF.2001.036

Han, J. S., and Rymsza, T. A. (1999). "Determining the minimum conditions for soda-AQ pulping of kenaf bast, core, and whole stalk fibers," *Second Annual American Kenaf Society Conference*, San Antonio, TX.

Hubbe, M. A., and Bowden, C. (2009). "Handmade paper: A review of its history, craft, and science," *BioResources* 4(4), 1736-1792. DOI: 10.15376/biores.4.4.1736-1792

Iqbal, H. M. N., Kyazze, G., and Keshavarz, T. (2013). "Advances in the valorization of lignocellulosic materials by biotechnology: An overview," *BioResources* 8(2), 3157-3176. DOI: 10.15376/biores.8.2.3157-3176

Jegannathan, K. R., and Nielsen, P. H. (2013). "Environmental assessment of enzyme use in industrial production - A literature review," *J. Clean. Prod.* 42(3), 228-240. DOI: 10.1016/j.jclepro.2012.11.005

Jin, Z., Jin, G., Shao, S., and Katsumata, K. S. (2012). "Lignin characteristics of bast fiber and core in kenaf, bark and wood of paper mulberry and mulberry," *J. Wood Sci.* 58(2), 144-152. DOI: 10.1007/s10086-011-1228-4

Ko, C. H., Lin, Z. P., Tu, J., Tsai, C. H., Liu C. C., Chen, H. T., and Wang, T. P. (2010). "Xylanase production by *Paenibacillus campinasensis* BL11 and its pretreatment of

hardwood kraft pulp bleaching," *Int. Biodeter. Biodegr.* 64(1), 13-19. DOI: 10.1016/j.ibiod.2009.10.001
Kumar, V., and Satyanarayana, T. (2012). "Thermo-alkali-stable xylanase of a novel polyextremophilic *Bacillus halodurans* TSEV1 and its application in biobleaching," *Int. Biodeterior. Biodegr.* 75, 138-145. DOI: 10.1016/j.ibiod.2012.09.007
Lian, H. L., You, J. X., and Lian, Z. N. (2012). "Effect of prior mechanical refining on biobleaching of wheat straw pulp with laccase /xylanase treatment," *BioResources* 7(3), 3113-3124. DOI : 10.15376/biores.7.3.3113-3124
Marques, G., Rencoret, J., Gutiérrez, A., and del Río, J. C. (2010). "Evaluation of the chemical composition of different non-woody plant fibers used for pulp and paper manufacturing," *Open Agric. J.* 4, 93-101.
Martín-Sampedro, R., Eugenio, M. E., and Villar, J. C. (2012a). "Effect of steam explosion and enzymatic pre-treatments on pulping and bleaching of *Hesperaloe funifera*," *Bioresour. Technol.* 111, 460-467. DOI: 10.1016/j.biortech.2012.02.024
Martín-Sampedro, R., Rodríguez, A., Requejo, A., and Eugenio, M. E. (2012b). "Improvement of TCF bleaching of olive tree pruning residue pulp by addition of a laccase and/or xylanase pre-treatment," *BioResources* 7(2), 1488-1503. DOI: 10.15376/biores.7.2.1488-1503
Maijala, P., Mäkinen, M., Galkin, S., Fagerstedt, K., Härkäsalmi, T., and Viikari, L. (2012). "Enzymatic modification of flaxseed fibers," *J. Agric. Food Chem.* 60(44), 10903-10909. DOI: 10.1021/jf303965k
Ninawe, S., and Kuhad, R. C. (2006). "Bleaching of wheat straw-rich soda pulp with xylanase from a thermoalkalophilic *Streptomyces cyaneus* SN32," *Bioresour. Technol.* 97(18), 2291-2295. DOI: 10.1016/j.biortech.2005.10.035
Pathak, P., Bhardwaj, N. K., and Singh, A. K. (2011). "Optimization of chemical and enzymatic deinking of photocopier waste paper," *BioResources* 6(1), 447-463. DOI: 10.15376/biores.6.1.447-463
Reinstaller, A. (2008). "The technological transition to chlorine free pulp bleaching technologies: Lessons for transition policies," *J. Clean. Prod.* 16(1), 133-147. DOI: 10.1016/j.jclepro.2007.10.020
Saijonkari-Pahkala, K. (2001). "Non-wood plants as raw material for pulp and paper," *Agric. Food Sci. Finland* 10, 1-101.
Salmela, M., Alén, R., and Hong Vu, M. T. (2008). "Description of kraft cooking and oxygen-alkali delignification of bamboo by pulp and dissolving material analysis," *Ind. Crops Prod.* 28(1), 47-55. DOI: 10.1016/j.indcrop.2008.01.003
Shi, J., Shi, S. Q., Barnes, H. M., and Pittman Jr, C. U. (2011). "A chemical process for preparing cellulosic fibers hierarchically from kenaf bast fibers," *BioResources* 6(1), 879-890. DOI: 10.15376/biores.6.1.879-890
Singh, S., Dutt, D., and Tyagi, C. H. (2011). "Environmentally friendly totally chlorine free bleaching of wheat straw pulp using novel cellulase-poor xylanases of wild strains of *Coprinellus disseminatus*," *BioResources* 6(4), 3876-3882. DOI: 10.15376/biores.6.4.3876-3882
Spiridon, I. (2007) "Modifications of *Asclepias syriaca* fibers for paper production," *Ind. Crops Prod.* 26(3), 265-269. DOI: 10.1016/j.indcrop.2007.03.015
TAPPI T205. (2002). "Forming handsheets for physical tests of pulp," *TAPPI Press*, Atlanta, GA.

TAPPI T220. (2010). "Physical testing of pulp handsheets," *TAPPI Press*, Atlanta, GA.
TAPPI T236. (2013). "Kappa number of pulp," *TAPPI Press*, Atlanta, GA.
TAPPI T414. (2012). "Internal tearing resistance of paper," *TAPPI Press*, Atlanta, GA.
TAPPI T494. (2006). "Tensile breaking properties of paper and paperboard," *TAPPI Press*, Atlanta, GA.
TAPPI T452. (2008). "Brightness of pulp, paper, and paperboard," *TAPPI Press*, Atlanta, GA.
TAPPI T511. (2013). "Folding endurance of paper," *TAPPI Press*, Atlanta, GA.
Tahir, P. M., Ahmed, A. B., SaifulAzry, S. O. A., and Ahmed, Z. (2011). "Retting process of some bast plant fibres and its effect on fibre quality: A review," *BioResources* 6(4), 5260-5281. DOI: 10.15376/biores.6.4.5260-5281
Virk, A. P., Sharma, P., and Capalash, N. (2012). "Use of laccase in pulp and paper industry," *Biotechnol. Progr.* 28(1), 21-32. DOI: 10.1002/btpr.727

Article submitted: July 15, 2014; Peer review completed: February 5, 2015; Revisions received and accepted: April 6, 2015; Published: April 28, 2015.
DOI: 10.15376/biores.10.2.3625-3637