



IMPROVING PULP AND PAPER INDUSTRY PROCESSES USING BIOTECHNOLOGY

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Introduction. The use of hydrolytic enzymes and oxidoreductases reduces manufacturing costs to bleach paper pulp, provides novel high quality products with process environmentally friendly. Xylanases and cellulases act on the surface and inner layers of cellulose fibres, its hydrolytic action increases the flexibility, breakability and detachability of fibre outer walls, thereby resulting in external fibrillation. Specifically, Cel9B alters the morphology of cellulose fibres and leads to increased pulp properties as a consequence of a major water absorption capacity in the inner fibre matrix. The use of oxidoreductase enzymes in delignification processes has exposed their added value with a view to developing effective sequences to obtain pulp with a high brightness. Laccase low redox potential allows only oxidization of phenolic groups of lignin, and in the presence of a mediator which acts as intermediary redox, oxidation of non-phenolic units is possible. Researches performed with the laccase mediator system are aimed to optimize the application conditions in order to get greater selectivity, reduce toxicity and costs generated by the use of synthetic mediators. In the present days, natural mediators are becoming a powerful alternative to obtain environment-friendly processes.

Methods. Cellulases treatment: ECF kraft pulp from *E. globulus* was subjected to enzymatic hydrolysis using the 1,4- β -*endoglucanase* Cel9B from *Paenibacillus* sp. BP-23 (2.5 IU/g Cel9B based on the oven dry weight pulp - odp). Laccase mediator system (L): was applied to ECF and TCF kraft pulp from *E. globulus* previously acid-washed at pH 4.0 during 30 min. The treatment was realized during 4 (L 4h) using 0.05% (w/v) *Tween* 80, 5% consistency pulp, pressure of 6 kg/cm², pH 4.0, HBT mediator 2% odp and 20 IU/g odp of a commercially available laccase from *Trametes villosa* (*Novozymes*). Enzyme treatment was followed by an alkaline extraction stage (E) and reductive stage (R) by using 2% NaBH₄ at room temperature (1). Pulp samples provided by each treatment were subjected to the following determinations: hexenuronic acids (HexAs), kappa number; brightness, specific viscosity, functional (carbonyl and carboxyl) groups, and electrokinetic properties such as ζ potential and cationic demand -streaming potential and colloid titration- (2). Pulp samples were refined using a PFI mill Mark VI (ISO 5264), pulp and paper properties were measured.

Results. The application of Cel9B on ECF pulp increased the %WRV and °SR with respect to control pulp -IP. The improvement was especially marked at 3000 and 4500 rev. Enzyme Cel9B alters the morphology of cellulose fibres leading to energy saving. At 3000 rev. Cel9B

reaches 56 °SR whereas in the conventional process the same value demands a superior refining (up to 4500 rev.) (Table 1). The effect of Cel9B on drainability can be ascribed to improvement of water absorption within the cellulose matrix, and internal - external fibrillation, which increase the specific surface area of refined fibres.

Table 1. Mechanical properties of pulp and handsheets.

PFI/rev.	Cel9B				Control pulp			
	0	1500	3000	4500	0	1500	3000	4500
Water retention, %WRV	106	137	155	169	103	128	142	150
Drainage resistance, °SR	21	34	56	74	20	25	35	50
Specific volume, cm ³ g ⁻¹	1.78	1.52	1.40	1.32	1.81	1.57	1.46	1.39
Permeability, mm (Pa.s) ⁻¹	49.5	21.8	5.8	1.8	48.2	28.4	16.3	7.4
Tear index, mN m ² g ⁻¹	8.53	10.92	9.33	8.94	5.91	10.19	11.36	11.13
Folding endurance	1.11	3.22	3.63	3.61	0.81	2.48	3.49	3.65
Burst index, kPa m ² g ⁻¹	1.25	3.25	3.95	4.46	0.94	2.47	3.29	3.95
Tensile index, Nm g ⁻¹	23.78	50.44	60.57	65.72	23.27	41.90	54.30	61.69

Hydrolysis by the enzyme reduces spacings between fibres by effect of the increased amount of fibre/fibre bonds. Refining Cel9B at 3000 rev. produces paper of similar density and permeability to those obtained by beating untreated pulp at 4500 rev. Cel9B facilitates the formation of paper and increases the uniformity of its fibre structure and resistance (tensile and burst index).

The application of the laccase mediator system on *eucalyptus* kraft pulp altered its kappa number (KN), hexenuronic acid content (HexAs), ISO brightness, improves brightness stability and reduces the content in carboxyl groups of TCF pulp, in addition reduces the ζ potential of fibre suspensions by acting on ionizable groups. The biomodification of the chemical composition of TCF pulp by the L stage causes no substantial changes in the properties of the refined pulp (°SR, %WRV).

Conclusions. Enzymatic effects on fibre morphology can lead to saving bleaching reagents and improve fibre-fibre bonding. The results lead to confirm the action of the cellulases such as "refining adjuvants", and laccase mediator system causes no biodegradation or cleavage of the cellulose chain. A reductive step (LR) confirmed that L oxidizes carbohydrates to form carbonyl groups, In the presence of HBT, the enzyme laccase alters the kappa number and hexenuronic acid content.

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