Available online at www.sciencedirect.com
ScienceDirect
Bioresource Technology 99 (2008) 7433–7437
BIORESOURCE TECHNOLOGY

Comparative study on application of *T. lanuginosus* SSBP xylanase and commercial xylanase on biobleaching of non wood pulps

Y. Ziaie-Shirkolaee *, A. Talebizadeh, S. Soltanali

Department of Chemical Engineering, Faculty of Engineering, Tehran University, P.O. Box, 11365/4563 Tehran, Iran

Received 31 October 2007; received in revised form 18 February 2008; accepted 21 February 2008
Available online 1 April 2008

Abstract

Biobleaching of three non wood kraft pulps (bagasse, rice straw and wheat straw) by *Thermomyces lanuginosus* SSBP xylanase and commercial xylanase (cartazyme sandoz), was studied in order to investigate their potential and effect on their various properties (reduction sugars, chlorine dioxide, kappa number, brightness and chromophores). In generally, xylanases released chromophores and reducing sugars and decreased kappa number of pulps. These samples gained over six brightness points over controls. Biobleaching of rice straw pulp with xylanase cartazyme (Sandoz) produced chlorine dioxide savings of up to 25% or 3.5–4 kg chlorine dioxide/ton pulp.

Keywords: *T. lanuginosus* SSBP xylanase; Commercial xylanase; Biobleaching; Non wood; Brightness

1. Introduction

The paper and pulp industry is a potential source of major pollution, generating large volumes of intensely coloured effluent for each metric ton of paper produced. In the production of paper, residual lignin from cellulose pulp is chemically liberated by using chlorine bleaching. Plants as raw materials of pulp-making are usually cooked under high temperature and pressure in the conventional pulping process which is involved with many problems including amounts of energy consumed, requirement of volumes of chemicals and environmental polluting (Hongzhang et al., 2002). Therefore, the need to decrease pollution from cellulose pulp mills has promoted the search for alternative bleaching and especially ozone, hydrogen peroxide and biobleaching processes (Jiménez et al., 1999; Roncero et al., 2003; Khristova et al., 2006; Adachi and Chen, 2007; Shen Liu, 2007; Rajasekar, 2007; Han et al., 2007). Biobleaching with xylanase has already proven its potential as an environmentally friendly bleaching technology (Lavielle et al., 1992).

The biobleaching process is based on the action of the microorganisms and/or enzymes. Microbial xylanases that are thermostable and cellulose-free are generally preferred for biobleaching of paper pulp. The interest for xylan degrading enzyme and its applications in the pulp and paper industries has advanced significantly over the past few years (Bajpai et al., 1994; Garg et al., 1998; Christov et al., 1999; Srinivasan and Rele, 1999). In this process, the bond between lignin and hemicelluloses is primarily between lignin and xylan which can be removed by xylanase. Once this layer of hemicellulose is removed, the lignin layer is easily available for degradative action of the ligninolytic enzymes (Eriksson, 1993).

The use of xylanase as a biobleaching process allows the attainment of pulps with high brightness with savings of bleaching chemicals (Bajpai, 1999) and is widely used in the bleaching of non woody pulps (Bajpai and Bajpai, 1996; Roncero et al., 2003; Chauhan et al., 2006).

A locally isolated *Thermomyces lanuginosus* strain SSBP produced one of the highest levels of xylanase (3575 U/ml) reported so far (Singh et al., 2000a). Its high thermostability

* Corresponding author. Tel.: +98 9173376809; fax: +98 21 66957781.
E-mail address: ya_ziaee@yahoo.com (Y.Z. Shirkolaee).

0960-8524/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.biortech.2008.02.032

(up to 80 °C) and broad pH range (5.5–10.0) renders this enzyme attractive for application in industrial bioprocessing (Singh et al., 2000b).

Also, several commercially available xylanase preparations, most of which were active at slightly acidic or neutral pH, have been investigated in pulp bleaching. Pulpzyme HA (*Novo Nordisk A/S*) produced by *Trichoderma reesei* was the first commercially available xylanase to be used in the biobleaching of wood pulps and achieved a 20% decrease in kappa number of oxygen-delignified birch kraft pulp (Zamost et al., 1991). Cartazyme (*Clariant*) also improved the brightness of kraft pulps (Garg et al., 1998). The aim of this work was to evaluate and compare the bleaching potential of a crude xylanase produced by *T. lanuginosus* strain SSBP and a commercial xylanase cartazyme (*Sandoz*) preparation on three non woody (bagasse, wheat straw and rice straw) pulps obtained from our laboratory. In addition, SEM observation of pulps is reported.

2. Methods

2.1. Purified xylanase

Purified xylanase from *T. lanuginosus* SSBP was supplied by Lin et al. (1999). The enzyme displayed pH and temperature optima of 6.5 and 65 °C, respectively, with a molecular mass of 23.6 kDa and a pI value of 3.8.

2.2. Xylanase assay

Xylanase was assayed as described by Bailey et al. (1992) by incubating the diluted enzyme solution (citrate buffer, pH 6.5) at 50 °C for 5 min using a substrate solution of 1% (w/w) birchwood xylan (Roth, Karlsruhe, Germany). One unit of xylanase activity was defined as that amount of enzyme that catalyses the release of 1 μmol of xylose equivalents per minute of reaction.

2.3. Pulps

Three types of non woody kraft pulp (bagasse, wheat straw and rice straw) were made in a 21-l batch cylindrical mini digester (stainless steel 321) in our laboratory, that process and conditions, has been described by Ziaie-Shirkolaee et al., 2007, 2008; Ziaie-Shirkolaee and Soltanali, 2007).

2.4. Xylanase pretreatment of pulp

Pulp equivalent to 10 g dry weight (pH 6.5) was charged with 1, 5 and 10 U purified xylanase/g pulp. Samples of 10% pulp consistency were incubated in polyethylene plastic bags in a water bath at 60 °C for 3 h with intermittent kneading. Control samples were treated under the same conditions with inactivated (boiled) enzyme. After incubation, the pulp slurry was filtered on a Buchner funnel and the enzyme filtrates were retained for analyses while the pulp was washed thoroughly with distilled water.

2.5. Analyses of pulp properties and filtrates

The kappa number of wheat straw pulp was determined according to TAPPI Test Methods T236 cm-85 (TAPPI, 2000) and the brightness was determined with an Elrepho photoelectric reflectance photometer (Carl Zeiss, Germany).

The enzyme-mediated release of lignin-derived compounds (LDC's) and chromophoric material from pulp was monitored in filtrates by measuring the absorbance at 280 nm and 465 nm, respectively (Wong et al., 1997). The amount of reducing sugars (RS) released from pulp was determined spectrophotometrically at 540 nm according to the DNS method (Miller, 1959).

2.6. Chemical bleaching of pulps

Enzyme treated and untreated pulp samples were bleached in a multistage elemental chlorine-free (ECF) bleaching process using a chlorine dioxide (D), alkali extraction (E), chlorine dioxide (D) treatment sequence

Table 1
Conditions for DED bleaching of pulps[a]

Bleaching conditions	D_1	E	D_2
Pulp consistency (%, w/w)	10	10	10
Temperature (°C)	67	67	67
Reaction time (min)	113	67	180
Active chlorine (% on pulp, w/w)	2.6	–	1.3
Sodium hydroxide (% on pulp, w/w)	–	0.7	–

[a] D_1, chlorine dioxide; E, alkali extraction; D_2, chlorine dioxide.

Table 2
Conditions for DED bleaching of pulps using reduced charges of chlorine dioxide[a]

D_1 (% w/w)	Reduction at D_1 (%)	D_2 (% w/w)	Reduction at D_1 (%)	Total reduction of chlorine dioxide	
				(%)	(kg/t pulp)
2.63	0	1.31	0	0	0
2.63	0	1.31	0	0	0
2.49	5	1.31	0	3.3	0.5
2.36	10	1.31	0	6.7	1.0
2.36	10	1.18	10	10.0	1.5
2.23	15	1.18	10	13.3	2.0
2.23	15	1.05	20	16.7	2.5
2.10	20	1.05	20	20.0	3.0
2.10	20	0.92	30	23.3	3.5
1.97	25	0.92	30	26.7	4.0
1.84	30	0.92	30	30.0	4.5
1.70	35	0.92	30	33.3	5.0

[a] D_1, chlorine dioxide: 2.6% active chlorine on pulp (w/w), 67 °C, 113 min; E, alkali extraction: 0.7% NaOH on pulp (w/w), 67 °C, 67 min; D_2, chlorine dioxide: 1.3% active chlorine on pulp (w/w), 67 °C, 180 min. All treatments were carried out at pulp consistency of 10%. The percentage reduction of chlorine dioxide in D_1 and D_2 was converted to total reduction of chlorine dioxide and expressed as kg/t pulp.