Industrial Crops and Products 55 (2014) 102–108

Contents lists available at ScienceDirect

Industrial Crops and Products

journal homepage: www.elsevier.com/locate/indcrop

Enzyme-assisted isolation of microfibrillated cellulose from date palm fruit stalks

Mohammad L. Hassan[a,b,*], Julien Bras[c], Enas A. Hassan[b], Cecile Silard[c], Evelyne Mauret[c]

[a] Cellulose and Paper Department, National Research Centre, Dokki, Giza 12622, Egypt
[b] Centre of Excellence for Advanced, Advanced Materials and Nanotechnology Group, National Research Centre, Dokki, Giza 12622, Egypt
[c] Laboratoire de Génie des Procédés Papetiers (LGP2), UMR CNRS 5518, Grenoble INP-Pagora – 461, rue de la papeterie, 38402 Saint-Martin-d'Hères, France

ARTICLE INFO

Article history:
Received 14 September 2013
Received in revised form 26 January 2014
Accepted 31 January 2014
Available online 7 March 2014

Keywords:
Date palm
Microfibrillated cellulose
Xylanase
Tensile strength
Air permeability
Water absorption

ABSTRACT

Isolation of microfibrillated cellulose (MFC) from xylanase-pretreated date palm fruit stalks pulp by ultra-high friction grinding was studied. Xylanase pretreatment resulted in decreasing hemicelluloses content and increasing degree of polymerization (DP) of the fibers. Progress of isolation of MFC from fibers was followed by optical microscopy and *MorFi* fiber analysis after passing the fibers through the grinder for different times. *MorFi* analysis results showed that xylanase pretreatment of the fibers facilitate isolation of MFC. The width of MFC isolated from untreated pulp was 21 ± 9 nm whereas that of MFC isolated from xylanase-treated pulps ranged from 81 ± 25 to 60 ± 20 nm. In addition, surface charge of MFC isolated from xylanase-pretreated fibers was lower than that of MFC isolated from untreated fibers. MFC films made from xylanase pretreated fibers showed higher density and tensile strength properties, lower water absorption and air permeability than those made from MFC isolated from untreated fibers.

1. Introduction

Microfibrillated cellulose, or so called cellulose nanofibers, has been isolated from different lignocellulosic materials using different technologies such as high-pressure homogenizers (Turbak et al., 1983; Herrick et al., 1983; Nakagaito and Yano, 2004; Andresen et al., 2006; Andresen and Stenius, 2007; Erkisen et al., 2008; Stenstad et al., 2008; Syverud and Stenius, 2009; Zhang et al., 2012; Rezayati Charani et al., 2013; Winuprasith and Suphantharika, 2013; Alila et al., 2013; Djafari Petroudy et al., 2014), micro-fluidizers (Bendahou et al., 2010), ultrafine grinders (Iwamoto et al., 2005; Abe et al., 2007; Iwamoto et al., 2008; Subramanian et al., 2008; Hassan et al., 2012; Jang et al., 2013), cryo-crushing (Chakraborty et al., 2005; Janardhnan and Sain, 2006), and ultrasonic method (Zhao et al., 2007). The main action of the different methods to isolate microfibrillated cellulose from the fibers is the high shear force that makes separation of the fibrils which form the fibers possible. Isolation of the nanofibers is energy intensive and therefore pretreatments of the fibers prior to isolation of the nanofibers have been studied. In this context, it has been shown that chemical pretreatment using alkali facilitates isolation of microfibrillated cellulose from fibers (Hassan et al., 2010). TEMPO (2,2,6,6-tetramethylpiperidine-1-oxyl) oxidation was also used as a pretreatment before mechanical treatment for nanofibers isolation (Saito et al., 2007).

The use of enzymes as a clean and environmentally friendly treatment prior to isolation of microfibrillated cellulose has been also investigated with the aim to reduce energy and chemicals during the isolation process (Janardhnan and Sain, 2006; Henriksson et al., 2007; Pääkkö et al., 2007; Hassan et al., 2010; Henriksson et al., 2008; Siddiqui et al., 2010; Janardhnan and Sain, 2011). Hassan et al. studied cellulase and xylanase enzymes for isolation of MFC from bagasse pulp using ultrafine grinding and showed that the effect of xylanase enzymes was better than cellulase in terms of wet and dry tensile strength properties of the isolated MFC. In other studies, cellulase enzymes were tested in order to decrease the degree of polymerization of cellulose and then the treated fibers were passed through high pressure homogenizer for MFC isolation (Henriksson et al., 2007; Pääkkö et al., 2007; Janardhnan and Sain, 2011; Siddiqui et al., 2010; Zhu et al., 2011). Recently, combination of two enzymatic preparations (hemicellulases/pectinase and endoglucanase) was used as pretreatment for bagasse and Curaua fibers before isolation of MFC using ultrasonic method (De Campos et al., 2013).

Date palm (*Phoenix dactylifera*) is abundant in several areas of the world. About 105 million date palms are currently being grown around the world (Agoudjil et al., 2011). Different residues are left from date palm cultivation and fruit harvesting such as

* Corresponding author at: Cellulose and Paper Department, National Research Centre, Dokki, Giza 12622, Egypt. Tel.: +20 2 33335926; fax: +20 2 33370931.
E-mail addresses: mlhassan2012@yahoo.com, mlhassan2013@yahoo.com (M.L. Hassan).

0926-6690/$ – see front matter © 2014 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.indcrop.2014.01.055

rachis, leaves, and fruit stalks. Recent works studied the use of palm leaves, rachis, and trunk in wood products and plastic composites (El-Morsy, 1980; Ghosh et al., 2007; Sbiai et al., 2008; Ghosh and Nag, 2009; Amirou et al., 2013; Mahdavi et al., 2010). In addition, it has been showed that fibers isolated from palm rachis and leaves are good source for paper making (El-Morsy et al., 1981; Khristova et al., 2005; Khiari et al., 2011; Ghosh and Nag, 2010, 2009). Surprisingly, for the best of our knowledge, no studies have been published so far regarding the use of date fruit stalks residues in paper, cellulose, or wood products. In Egypt, after harvesting dates, the fruit stalks residues find no use except for making low value products such as brooms.

Similarly, in the area of cellulosic nanomaterials, few studies have been published regarding isolation of microfibrillated cellulose from date palm residues (Bendahou et al., 2010). In that work, micro-fluidizer was used for isolation of MFC. The lowest lateral size of the isolated MFC was from 5 to 10 nm in addition to larger fibril bundles of about 100 nm thickness. Some short MFC with less than 2 μm was also observed as a result of mechanical action during the isolation. No further characterization for the isolated MFC was carried out and the isolated MFC were used in rubber nanocomposites.

The aim of the current work is to isolate and characterize MFC from date palm fruit stalks using ultrafine-grinding or so called supermasscolloider. Enzymatic pretreatment of the fibers were carried out to facilitate the isolation of MFC and to improve the strength properties of the isolated MFC.

2. Experimental

2.1. Materials

Date palm fruit stalks were collected from local fields in Giza, Egypt after separating dates from the stalks. The stalks were then cleaned with water and cut into 2–3 cm long. Date palm fruit stalks pulp was prepared by alkali treatment using 15% NaOH (w/w, based on oven-dried stalks) at 150 °C for 3 h. The produced pulp was bleached using sodium chlorite/acetic acid mixture at 80 °C for 1 h (Wise et al., 1946). Chemical composition of the bleached pulp was determined according to the previously published methods (Browning, 1967) and was: α-cellulose 71.5%, pentosans 18.4%, degree of polymerization (DP) 1264, and 0.64% ash; the analyses were carried on two specimens and results were averaged. Analytical grade sodium citrate and citric acid were used for preparation of citrate buffer (pH 5.3). Commercial xylanases from *Thermomyces lanuginosus* was purchased from Sigma–Aldrich and used as received. PolyDADMAC (Poly(diallyldimethyl ammonium chloride)) with average M_w 400,000–500,000 as a 20 wt.% solution in water was purchased from sigma–Aldrich.

2.2. Enzymatic pretreatment of bleached date palm pulp

Enzymatic pretreatment of date palm pulp with xylanase enzymes was carried out as follows (Hassan et al., 2010): twenty grams of bleached pulp were treated with xylanase enzymes in citrate buffer (pH 5.3) in 500 ml conical flask at 10% consistency (w/w). The concentration of xylanase enzymes was 0.01, 0.02, and 0.04 g/g of pulp. Samples were given the codes0.0E, 0.01E, 0.02E, and 0.04E in the following sections where E stands for enzymatic treatment and the number stands for the concentration of the enzyme used (g/g pulp). The reaction mixture was kept under gentle stirring at 50 °C for 4 h. At the end of reaction, the temperature was raised to 90 °C to de-activate the enzymes then the pulp was filtered and washed thoroughly with distilled water.

2.3. Isolation of microfibrillated cellulose

The pulp suspension (2% consistency) was first disintegrated by using a high-shear mixer. The fibers suspension was then treated in a high-shear ultrafine friction grinder or a so-called supermasscolloider (MKCA6-2, Masuko Sanguo, Japan) by passing them through the device up to 60 times. The gap between the disks of the grinder was adjusted to 9 μm. Finally, MFC was centrifuged at 10,000 rpm to reduce its water content and kept wet in the fridge till used.

2.4. Characterization of microfibrillated cellulose

Atomic force microscopy (AFM) was carried out using a Multimodal AFM (DI, Veeco, Instrumentation Group) with both tapping and conductive mode (C-AFM). The tips were Multi130 for tapping and MESP for C-AFM. Before the analysis, a drop of highly diluted microfibrillated cellulose suspension was deposited on a fresh mica substrate and left to air dry for at about 2 h.

Diffraction patterns were obtained from a Phillips X-ray diffractometer using Cu-Ka radiation at 40 kV and 25 mA. Crystallinity index (CrI) was calculated from the X-ray diffraction patterns according to the following equation (Segal et al., 1959):

$$\mathrm{CrI} = \frac{I_{002} - I_{\mathrm{am}}}{I_{002}}$$

where I_{002} and I_{am} are the intensities of the peaks at $2-\theta$ of about 22 and 18, respectively.

For surface charge determination, a particle charge detector PCD 02 (Mütek, Germany) equipped with an automatic titrator was used. The anionic charges of the suspensions were measured by adding slowly a cationic polyelectrolyte (PolyDADMAC) until obtaining a zero potential (equivalence point). For this purpose, 10 ml of MFC suspensions containing between 0.04 and 0.05 g were titrated with a 2.97×10^{-4} M polyDADMAC solution. The concentration of the anionic groups, C (mole/g), was calculated as follows:

$$C = \frac{C_{\mathrm{polydadmac}} V_{\mathrm{polydadmac}}}{V}$$

where $C_{\mathrm{polydadmac}}$ is the concentration of the polyDADMAC solution, $V_{\mathrm{polydadmac}}$ the volume (mL) of the polyDADMAC solution and V (mL) the volume of the MFC suspension. Three specimens of each sample were measured and the results averaged.

2.5. MorFi analysis of pulp and MFC

To follow progress of fibrillation of fibers as a result of passing through the supermasscollider, the morphological properties of the pulp and MFC suspensions were measured using a *MorFi* analyzer (TECHPAP LB 01 *MorFi* equipment). The main parameters (fiber length, width, and content of fine elements) were assessed by image analysis of a diluted suspension flowing in a transparent flat channel observed by a CCD video-camera. Fine elements are defined as particles with length less than 200 μm and their corresponding content is calculated as the ratio of the total length of fines to the total length of the elements present in the suspension. The weighted length is calculated as following:

$$\bar{l}_W = \frac{\sum_i n_i l_i^2}{\sum_i n_i l_i}$$

Samples of fiber suspension were collected after 20, 40, and 60 passes through the supermasscolloider, diluted to about 0.300 g/L, and 1 L of this suspension was poured into the *MorFi* analyzer and measured. The test was duplicated and the obtained results averaged.