Carbohydrate Polymers 195 (2018) 114–119

Contents lists available at ScienceDirect

Carbohydrate Polymers

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

Effect of ultrasonic time on the structural and physico-chemical properties of hemicelluloses from *Eucalyptus grandis*

Ji-Yuan Xu[a], Tong-Qi Yuan[a], Lin Xiao[b], Run-Cang Sun[a,*]

[a] Beijing Key Laboratory of Lignocellulosic Chemistry, Beijing Forestry University, Beijing 100083, China
[b] Shandong Longlive Bio-Technology Co., Ltd., Shandong 251200, China

ARTICLE INFO

Chemical compounds studied in this article:
Xylose (PubChem CID: 135191)
Rhamnose (PubChem CID: 25310)
Arabinose (PubChem CID: 439195)
Galactose (PubChem CID: 6036)
Glucose (PubChem CID: 5793)

Keywords:
Eucalyptus grandis
Yield
Ultrasound-assisted extraction
Structural properties

ABSTRACT

Effect of sonication on the extractability and physico-chemical properties of hemicelluloses from *Eucalyptus grandis* using 5% KOH solution at 50 °C for 3 h has been comparatively studied. The results showed that the yield of hemicelluloses increased from 2.6 to 19.6% as the ultrasonic time was extended from 5 to 35 min. The highest yield of hemicelluloses (95.2%) was achieved at 30 min ultrasonic time. Xylose was the dominant sugar (82.94–84.96%) of all the hemicellulosic fractions. Furthermore, the hemicelluloses obtained by ultrasound-assisted extractions had slightly lower molecular weights (74,510–66,770 g/mol) and thermal stabilities, but higher contents of xylose (83.95–84.96%). The increased yield of ultrasonically extracted hemicelluloses, which have preserved their main structural properties, confirmed the great potential of ultrasound-assisted extraction to separate hemicelluloses from *Eucalyptus grandis* at an industrial level.

1. Introduction

Lignocellulosic biomass is thought to be one of the most abundant resources for replacing petroleum to produce chemicals, fuels, and polymeric materials. Fast-growing hardwood species are currently the most important source for pulp production, including the poplar and the *Eucalyptus grandis*. In addition, much attention has been paid to utilizing *Eucalyptus grandis* wood because of its potential for pulp and paper making (Brandt et al., 2011; Evtuguin et al., 2001). *Eucalyptus grandis* wood is primarily composed of cellulose, hemicelluloses, and lignin. These components are closely combined with each other to constitute the cellular complex (Chandel, Chandrasekhar, Radhika, Ravinder, & Ravindra, 2011). Hemicelluloses have recently become more popular due to their unique applications. They can be used as film-former substances, thickeners, emulsifiers, stabilizers and binders in the food, pharmaceutical and cosmetic industries (Peng & She, 2014; Spiridon & Popa, 2008). In addition, hemicelluloses can be converted into chemicals and other materials (Gao et al., 2016; Saha, 2003). Certainly, extracting hemicelluloses from the plant cell walls is necessary to utilize the substance efficiently. However, lignin and carbohydrates (mainly hemicelluloses) are linked by chemical bonds, forming a special compound—lignin carbohydrate complex (LCC) (Balakshin, Capanema, Gracz, Chang, & Jameel, 2011). The main covalent bonds between hemicelluloses and lignin are *α*-benzyl ether linkages. In addition, hemicelluloses form hydrogen bonds with cellulose. Theses bonds restrict the discharge of hemicelluloses from the plant cell walls (Peng et al., 2009). Therefore, developing a beneficial extraction method to isolate hemicelluloses from the plant cell walls with high purity and yield is required.

Although various methods are available to isolate hemicelluloses from the plant cell walls, hemicelluloses isolated with alkalis are widely used since it can be easily performed at a lower cost. In recent years, there has been an increased interest in using ultrasonic irradiation in the extraction and refining processes (Abbot, Nielsen, & Kleiman, 1995; Mason, Paniwnyk, & Lorimer, 1996; Mason, 1999; Paniwnyk, Beaufoy, Lorimer, & Mason, 2001; Sališová, Toma, & Mason, 1997). The mechanisms of ultrasonic irradiation could be attributed to the phenomenon of cavitation, which is induced by the propagation of ultrasound pressure waves. When the pressure waves pass through the surface of lignocellulosic materials, the cavitation bubble implodes to cause micro-fractures, which accelerate the extraction of organic compounds from plant cell walls (Él'piner, 1964; Mason, 1992; Vinatoru, 2001; Vinatoru et al., 1997). Furthermore, ultrasonic irradiation has been found to significantly improve the extraction yield of polysaccharides during the separation of plant components (Gan, Manaf, & Latiff, 2010; Wang, Liu, & Hu, 2014; Zhao, Zhang, Li, Dong, & Liu, 2015). As compared with the experiment performed without ultrasonic assistance, higher yields of hemicelluloses can be obtained using lower

* Corresponding author.
E-mail address: rcsun3@bjfu.edu.cn (R.-C. Sun).

https://doi.org/10.1016/j.carbpol.2018.04.067
Received 23 December 2017; Received in revised form 10 April 2018; Accepted 16 April 2018
Available online 21 April 2018
0144-8617/

temperatures and shorter times during the ultrasonic irradiation processes (Ebringerová, Hromadkova, & Hřibalová, 1995; Sun, Sun, & Ma, 2002).

Although much attention has been paid to utilizing the ultrasonic technology during the isolation of plant materials, there is little information regarding the extraction of hemicellulosic polymers from *Eucalyptus grandis* wood by applying a combined aqueous alkalis and ultrasound irradiation process. Therefore, the objective of this study was to examine the effect of ultrasound on the yield and physico-chemical properties of the hemicelluloses from *Eucalyptus grandis* in an aqueous alkali solution. In addition, the optimal conditions for maximizing extraction yield of hemicelluloses were determined by treating the *Eucalyptus grandis* in 5% KOH solution at 50 °C for 3 h under various ultrasonic times (5–35 min). The hemicellulosic fractions obtained were characterized by high-performance anion-exchange chromatography (HPAEC), gel permeation chromatography (GPC), Fourier transform infrared (FT-IR), 1D and 2D nuclear magnetic resonance (NMR) spectroscopy, and thermogravimetric analysis (TGA).

2. Materials and methods

2.1. Materials

Eucalyptus grandis wood was obtained from the Guangxi Province, China. The *Eucalyptus grandis* wood was first cut into small pieces and then dried in an oven at 60 °C. The wood sample was then ground in a mill to obtain a 40–60 mesh fraction. After that, the powder was stored in a desiccator until needed. The main composition of the dried dewaxed *Eucalyptus grandis* wood was cellulose 40.1%, hemicelluloses 18.2%, and lignin 29.0% (acid-insoluble lignin 23.8% and acid-soluble lignin 5.2%).

2.2. Ultrasound-assisted extraction and isolation of hemicelluloses

A scheme for ultrasound-assisted extraction and isolation of hemicelluloses is illustrated in Fig. 1. The dried *Eucalyptus grandis* powder was first extracted with toluene-ethanol (2:1, v/v) in a Soxhlet apparatus for 6 h. Then, the dewaxed sample was delignified with sodium chlorite in an acidic solution (pH 3.8-4.0, adjusted by acetic acid) at 75 °C for 2 h. After the treatment, the residue (holocellulose) was filtered off, washed completely with distilled water, and dried further in an oven at 50 °C for 16 h. Subsequently, the holocellulose was extracted in 5% KOH solution with a solid to liquor ratio of 1:30 (g mL^{-1}) at 50 °C under various ultrasonic times (0, 5, 10, 15, 20, 25, 30, and 35 min) in a 400 mL glass beaker. The sonication was performed with the Sonic system JY99-IIDN (Nanjing, 19–21 kHz) provided with a horn at sonic power of 180 W. The mixture was then sequentially treated with the remaining 5% KOH solution under continous stir at 50 °C for a total time of 3 h, respectively. The filtrates were neutralized to pH 5.8 with glacial acetic acid, where they were then concentrated, dialyzed and concentrated again. The alkali-soluble hemicelluloses were collected by precipitation of the concentrated filtrates with 2 volumes of ethanol. After centrifugation, the isolated hemicelluloses were freeze-dried and the hemicellulosic fractions were noted as H_0, H_5, H_{10}, H_{15}, H_{20}, H_{25}, H_{30}, and H_{35}, respectively.

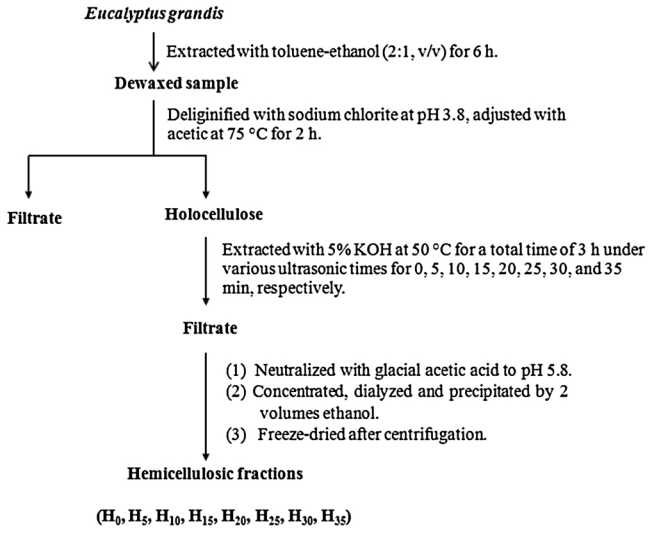

Fig. 1. Scheme for extraction of hemicelluloses from ultrasound-treated *Eucalyptus grandis*.

2.3. Sugar analysis

The composition of the neutral sugars and uronic acids in the eight hemicellulosic fractions were analyzed by HPAEC. The hemicellulosic fractions (4–6 mg) were hydrolyzed with 1.475 mL 6% H_2SO_4 for 2.5 h at 105 °C. After hydrolysis, the hydrolysates were filtered and diluted 50-fold with ultrapure water. The samples were analyzed by high-performance anion-exchange chromatography (HPAEC) system (Dionex ICS3000, USA) with a pulsed amperometric detector, a Carbopac™ PA-20 column (3 mm × 150 mm, Dionex, Sunnyvale, USA), and a guard PA-20 column (3 × 30 mm, Dionex, Sun-nyvale, USA). The neutral sugars and uronic acids were separated in a 5 mM NaOH isocratic (carbonate free and purged with nitrogen) for 20 min, followed by a 0.75 mM NaAc gradient in 5 mM NaOH for 15 min. The columns were then washed with 100 mM NaOH for 10 min to remove carbonate, followed by a 5 min elution with 5 mM NaOH to re-equilibrate the column before the next injection. The total analysis time was 50 min, and the flow rate was 0.4 mL/min. The standard solutions of L-rhamnose, L-arabinose, D-glucose, D-xylose, D-mannose, D-galactose, glucuronic acid, and galacturonic acid were used as calibration. The measurements were run in duplicate and the average values were calculated for the eight hemicellulosic fractions.

2.4. Molecular weight determination

The molecular weights of the eight hemicellulosic fractions were estimated by gel permeation chromatography (GPC) on a PL aquagel-OH 50 column (300 × 7.7 mm, Polymer Laboratories Ltd.). They were calibrated with PL pullulan polysaccharide standards (peak average molecular weights 180, 9600, 107,000, 708,000, Polymer Laboratories Ltd.). The eluent was 0.02 M NaCl in 0.005 M sodium phosphate buffer (pH 7.5), and the flow rate was maintained at 0.5 mL/min. Detection was achieved with a Knauer differential refractometer. The column oven was maintained at 30 °C. All hemicellulosic fractions were dissolved with 0.02 M NaCl in 0.005 M sodium phosphate buffer, pH 7.5, at a concentration of 0.1%.

2.5. Infrared analysis

FT-IR spectra were measured with a FT-IR microscope (Thermo Nicolet Corporation, Madison, WI, USA) equipped with a liquid nitrogen cooled MCT detector. The hemicellulosic fractions were dried, and the averages of 128 scans recorded at a resolution of 4 cm^{-1} in the range of 4000–600 cm^{-1} were denoted of the spectra.

2.6. 1D and 2D NMR analysis

The solution-state ^1H NMR, ^{13}C NMR, and HSQC spectra of the samples were obtained with a Bruker AVIII 400 MHz at 25 °C. The ^1H NMR and ^{13}C NMR spectra were obtained on a Bruker spectrometer at 400.13 and 100.6 MHz, respectively. The hemicelluloses (40 mg for ^1H, 80 mg for ^{13}C) were dissolved in 1.0 mL D_2O, and two drops of NaOH solution were added to the solution to make the hemicelluloses dissolve