



Physical properties of bacterial cellulose aqueous suspensions treated by high pressure homogenizer



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ARTICLE INFO

Article history:

Received 15 July 2014

Accepted 21 October 2014

Available online 29 October 2014

Keywords:

Bacterial cellulose aqueous suspension

High pressure homogenization

Morphology

Rheology

Texture

ABSTRACT

In the present study, purified wet bacterial cellulose was disrupted into cellulose aqueous suspensions (0.5%, w/v) and then homogenized by 10 passes at 0, 200, 400 and 600 bar respectively. In order to evaluate the effects of high pressure homogenization (HPH) on bacterial cellulose aqueous suspensions, morphology, rheology, stability, texture, water holding capacity (WHC), water swelling ability (WSA) and water release rate (WRR) were investigated. Morphological analysis by atomic force microscopy revealed changes in microstructure and dispersion of cellulose ribbons after HPH treatment. The diameter of micro-fibril ribbons decreased from 95.6 nm to 60.3 nm with increasing pressure up to 600 bar. The rheology results suggested that all the suspensions displayed a shear thinning behaviour, moreover the suspensions treated by HPH showed 3 regions (shear thinning region, plateau region and shear thinning region again) and the flow curves followed the Herschel–Bulkley model. Furthermore, the stability and textural properties of the suspensions were enhanced after HPH treatment. Additionally, WHC and WSA of cellulose suspensions displayed high stability and the WRR increased after HPH treatment during storage.

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1. Introduction

Bacterial cellulose (BC), an environmentally–friendly polymeric material, is now receiving increased attention in the world (Klemm et al., 2011). BC has high purity, high crystallinity, high degree of polymerization, high water absorbing and holding capacity, high tensile strength, and strong biological adaptability, which make it more valuable than plant derived cellulose (Iguchi, Yamanaka, & Budhiono, 2000; Klemm, Heublein, Fink, & Bohn, 2005). Due to its unusual physicochemical and mechanical properties, BC presents a potential alternative to plant-derived cellulose for specific applications in bio-medicine, cosmetics, high-end acoustic diaphragms, paper-making, and other applications (Gatenholm & Klemm, 2010; Klemm, Schumann, Udhardt, & Marsch, 2001; Petersen & Gatenholm, 2011; Svensson et al., 2005).

BC can form hydrogels and is well known as traditional gel delicacy (nata) in the Southeast Asia (Phisalaphong & Chiaoprakobkij, 2012). Due to its unique suspending, thickening, water-holding, stabilizing, bulking and fluid properties, it has been demonstrated that bacterial cellulose could be a promising low-calorie bulking ingredient for the development of novel rich functional foods of different forms such as powder, gelatinous, or shred forms (Chau, Yang, Yu, & Yen, 2008; Lin, Chen, & Chen, 2011; Okiyama, Motoki, & Yamanaka, 1992, 1993). In many cases, bio-polymers have been used in food industry after drying, because it would be better to have micro-fibrillated cellulose in the dried form to facilitate their storage and carriage (Agoda-Tandjawa et al., 2010). However, in the case of bacterial cellulose, re-dispersion of micro-fibrillated cellulose in water, after drying, would not allow the recovery of the rheological properties of the initial suspension. Moreover it would not allow the recovery of the water absorbing capacity of cellulose (Agoda-Tandjawa et al., 2010; Lowys, Desbrieres, & Rinaudo, 2001). Taking all of these considerations into account, it would be interesting to have bacterial cellulose in the undried state as an ingredient in food industry.

Well-dispersed suspensions are a prerequisite when using food additives, however native bacterial cellulose suspensions present pronounced particle aggregation due to the hydrogen bonds and

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Van der Waals attraction (Kuijk et al., 2013). Recently, different technological approaches have been developed to enhance the properties of the colloidal suspensions and dispersions of cellulose fibres in food industry (Koh et al., 2014; Rabetafika, Bchir, Aguedo, Paquot, & Blecker, 2014). High pressure homogenization (HPH) is one of the most encouraging alternatives to traditional non-thermal treatment in food industry for emulsion stabilization and to improve viscosity, texture, taste, and colour uniformity (Kubo, Augusto, & Cristianini, 2013; Ye & Harte, 2014; Zamora, Ferragut, Juan, Guamis, & Trujillo, 2011). HPH leads to a reduction in size and an increase in number of particles in the dispersed phase (Augusto, Ibarz, & Cristianini, 2012; Lopez Sanchez et al., 2011). During this process, the fluid is subjected to several simultaneous force-induced mechanisms such as cavitation, turbulence, shear, friction, heat, compression, acceleration, and rapid pressure drop (Koh et al., 2014). In the published literature, Agoda-Tandjawa et al. (2010) used a high pressure homogenizer to treat micro-fibrillated cellulose suspension from sugar-beet and several investigations reported high pressure homogenization for plant cellulose pulp (Donsi, Esposito, Lenza, Senatore, & Ferrari, 2009; Saito, Nishiyama, Putaux, Vignon, & Isogai, 2006). However there is limited number of works using high-pressure homogenization to treat bacterial cellulose aqueous suspensions, which is considered as a thickening system for food applications.

In the present study, purified wet bacterial cellulose was disrupted in a blender operated at 20,000 rpm, 20 °C for 10 min and then a concentration of 0.5% (w/v) cellulose aqueous suspension was homogenized by 10 passes (in order to ensure homogeneity) at each homogenization pressure of 0, 200, 400 and 600 bar using a high-pressure homogenizer. The effect of high pressure homogenization on the physical properties was investigated. Firstly, the dispersions were evaluated by atomic force microscopy. Then rheological properties were determined using a dynamic rheometer. Furthermore, textural properties of the suspensions were determined using a textural analyser (TA) and their stability was also evaluated during 3 weeks of storage at 4 °C. Additionally, we determined water holding capacity (WHC), water release rate (WRR) and water swelling ability (WSA) of bacterial cellulose aqueous suspensions stored at 4 °C for 3 weeks.

2. Materials and methods

2.1. Microorganism

The bacterial cellulose producing strain used in this study was isolated from homemade persimmon vinegar and identified as *Gluconoacetobacter xylinus* based on biochemical characterization and 16S rRNA sequence information. It was deposited as strain CICC 10529 at China Centre of Industrial Culture Collection (CICC), Beijing, China, and it was maintained on glucose agar: glucose 2% (w/v), yeast extract 0.5% (w/v), K₂HPO₄ 0.1% (w/v), MgSO₄ 1.5% (w/v), ethanol 2% (v/v), agar 1.7% (w/v). It was stored at 4 °C in a refrigerator and sub-cultured every 2 months for inoculum development or stored at –80 °C using 20% (v/v) glycerol instead of agar for long-time storage (Ge et al., 2011).

For seed inoculum, a loop of *G. xylinus* CICC 10529 was transferred from a slant culture into an Erlenmeyer flask (250 mL) containing 100 mL of seed medium with the same components as glucose agar slants but without agar. Then it was cultivated at 30 °C in a rotary shaker incubator at 150 rpm for 12–18 h until it reached the logarithmic growth phase (Ge et al., 2011). The prepared seed inoculum (10%, v/v) was transferred into a glass vessel (500 mL) containing 100 mL of fermentation medium with the same components as the seed medium. The starting pH of the medium was

adjusted to 5.0. The glass vessel was covered with 8 layers of gauzes, and then statically cultured at 30 °C for 14 days.

After cultivation, a cellulose yield of 6.86 g L^{–1} was obtained and the cellulose membrane was rinsed with running water overnight, soaked in 0.1 M NaOH solution at 80 °C for 2 h, and then washed with deionized water several times to completely remove alkali. The purified cellulose was kept in the sterilized water and stored at 4 °C in the fridge (Wu et al., 2010).

2.2. Homogenization treatment

The purified wet bacterial cellulose was disrupted in a blender (CWFJ-15, Changzhou, China) operated at 20,000 rpm, 20 °C for 10 min at a concentration of 0.5%, then bacterial cellulose suspensions were homogenized by 10 passes at 0, 200, 400 and 600 bar respectively using a high-pressure homogenization (SRH 60-70, Shanghai shen deer homogenizer co., LTD, China). At each level of homogenization pressure, cellulose aqueous suspensions were treated by 10 passes to make it fully homogeneous. After each homogenization pass, the cellulose aqueous suspensions were cooled down to 20 °C due to the increasing temperature of samples with increasing homogenization pressure. All the BC aqueous suspensions were stored at 4 °C in fridge.

2.3. Atomic force microscopy (AFM)

The topography and surface roughness of the cellulose films were determined using a NanoScope V & Multimode (Veeco instrument Inc., USA) operating in air. The BC aqueous suspensions were diluted 20 times with deionized water and one droplet was placed on a ~1 cm² piece of freshly cleaved mica for 1 min and the excess was absorbed with blotting paper. After drying at room temperature, the mica was attached to an AFM specimen disk and analysed.

2.4. Rheological properties

The rheological properties of BC aqueous suspensions were determined using a dynamic rheometer (ZX7M-AR1000, TA Instruments, USA). The dynamic rheometer was equipped with rough surface parallel-plate geometry (20 mm diameter) to prevent sample slippage. Gap and strain were set at 1.0 mm and 1.0%, respectively. Three replicates were conducted at 20 °C and experimental flow curves were compared with the Herschel–Bulkley (HB) model (The choice of the model was determined by the software after considering the highest regression value of $R^2 \geq 0.982$): (Long, Zhao, Zhao, Yang, & Liu, 2012)

$$\tau - \tau_0 = K \cdot \dot{\gamma}^n$$

where τ is shear stress (Pa); τ_0 is the yield stress (Pa); K is the consistency index (Pa·s^{*n*}) and n is the flow index ($n < 1$ for a shear-thinning fluid and $n = 1$ for a Newtonian fluid).

2.5. Texture analysis

The textural properties (firmness, consistency, cohesiveness and index of viscosity) of BC aqueous suspensions were determined using a texture analyser (TA. XT PLUS/50, Stable Micro systems Ltd) with a 5 mm cylinder probe (A/BE35). The test conditions for the TA were set at 1 mm s^{–1} for the pretest speed, 2 mm s^{–1} for the test speed and 1 mm s^{–1} for the post test speed. The calculations of those textural properties were performed by TA software.

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