



Characteristics of pectin from black cherry tomato waste modified by dynamic high-pressure microfluidization



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ABSTRACT

The present work aimed at better understanding of the effects of dynamic high pressure microfluidization (DHPM) on physicochemical properties and rheological property of pectin extracted from black-cherry tomato pomace. It was found that DHPM effectively increased the average particle size of pectin, as the result of foamed filiform parts formation in micro-structure. Galacturonic acid with esterified structures were observed according the nuclear magnetic resonance (NMR) spectroscopy and relatively independent of the pressure of DHPM, while degree of esterification (DE) was slightly increased when the pressure was higher than 40 MPa. Solutions of pectin treated by DHPM were consistent with the power's law model at low shear rate (shear rate $< 1 \text{ s}^{-1}$) and sweep frequency ($f < 1 \text{ Hz}$). DHPM treatment significantly decreased ($p < 0.05$) the apparent viscosity (η) and consistency index (K) of pectin. Moreover, phase angle (δ) of pectin treated by DHPM decreased, that indicated better fluidity and higher consistency for the solutions. Additionally, effects of DHPM at 120 MPa were noticed, DPPM resulted in particle size decreasing and there were no significant changes ($p < 0.05$) of η . However, flow behavior index (n) and $\tan\delta$ indicated that character of pectin solutions changed compared to pectin without DHPM treatment.

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1. Introduction

Black cherry tomato, which originate in South America, is a subspecies of *Solanum lycopersicum* and abundant in nutrients (amino acids, 3581mg/100 g) and bioactive compounds (lycopene 54.20 mg per 100 g dry weight, β -Carotene 3.75 mg per dry weight). The content of phenolic compounds, catotene pigments, especially β -carotene and lycopene in black cherry tomatoes are higher than general red colored tomatoes (Choi et al., 2014). These chemicals were reported to reduce the risk of chronic disease such as cardiovascular disease (Mills et al., 2012). For these healthy benefits, black cherry tomatoes are consumed as fresh vegetable and after-processing liquid-based products in form of tomato juice, ketchup, sauce, puree, etc. During the processing, tomato's skin, vascular tissue and seeds (Capanoglu et al., 2008; Lenucci et al., 2013) are usually abandoned as waste. Tomato waste, are also called tomato pomace, are mainly cell wall tissue, seeds and middle lamellas that are constructed by cellulose and pectin substances,

thus they are good source of pectin (Grassino et al., 2016), natural colorants (Laufenberg et al., 2003) and lycopene (Saldana et al., 2010).

Pectin is safe for human body and already successfully applied in food industries to modify properties of food systems as thickener, emulsifier, stabilizer, binding agent, natural prophylactic (Grassino et al., 2016). Repeats of methoxylated (C6) and/or acetylated (O-2 or O-3) D-galacturonic acid, D-galactose and L-arabinose constitute (Caffall and Mohnen, 2009) the main chains of pectin, interaction among branched chains form complex spatial structures. Thus, Methoxy content (MeO), anhydrouronic acid (AUA) and degree of esterification (DE) are important parameters for purity and characteristics of pectin (Anese et al., 2013; Barba et al., 2015). For example, a floor level of galacturonic acid content for food grade pectins was set at 65% by EU regulation No.231/2012 (EU regulation No. 231/2012, 2012). Also, pectin is known to be sensitive to process condition, especially heating, pH and pressure (Shpigelman et al., 2015). There was literature evidence that pectin molecules in tomato pomace cross-linked by long-chain aliphatic and cyclic components, which were main component of tomato peel cuticle (Caffall and Mohnen, 2009; Johnson et al., 2007) and pectin obtained by ultrasonic assisted extraction gave higher pectin yield in

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comparison with conventional extraction (Grassino et al., 2016).

Dynamic high pressure microfluidization (DHPM) technology which emerge as alternative technology of thermal treatments to reduce the microbial activity draws more and more attentions. It combines forces of high-velocity impact, high-frequency vibration, instantaneous pressure drop, cavitation, intense shear and ultra-high pressures up to 200 MPa with a short treatment time (less than 5s) (Augusto et al., 2012; Liu et al., 2009). The energy produced by DHPM was used to modified properties of carbohydrates, such as starch (Liu et al., 2016), polysaccharides (Tu et al., 2013; Zhang et al., 2015), fibers (Tu et al., 2014; Wan et al., 2015). The depolymerization of pectin caused by DHPM has an inversely-proportional relationship with the presence of neutral sugar side chains which are dependent on pectin's source. The depolymerization, particle size, molecular weight and amount of reducing sugars change with the increase of HDPM pressure only when pectin above a specific threshold (Chen et al., 2012; Shpigelman et al., 2015). DHPM, as a potential homogenization technology with non-thermal processing, not only can inactivate microorganisms and enzymes, but also can modify the structure of pectin. Changes of physicochemical characteristics will induce rheological behavior, such as viscosity, consistency and gel properties (Moelants et al., 2013; Walkenstrom et al., 2003).

The main objective of this study is to investigate the effects of DHPM treatment on physicochemical properties of pectin from black cherry tomatoes waste (PBCTW). The main parameters of DHPM-pressures (40 MPa, 80 MPa, 120 MPa, 160 MPa) were chosen to study the characterization. To our best knowledge, there was no similar literature explore the effect of DHPM on the physicochemical properties of pectin from PBCTW.

2. Materials and methods

2.1. Materials

Black cherry tomatoes were obtained from farm located in Qibao campus, Shanghai Jiao Tong University, cleaned and frozen at $-70\text{ }^{\circ}\text{C}$ for further use. The materials were milled by juicer after unfrozen, and then filtered by screen mesh with 0.5 mm bore diameter. The filter residue was freeze-dried (Freeze dryer, Shanghai bilon Instruments Manufacturing Co. Ltd., China), crushed packed in polyethylene bags as BCTW, and the filtrate was collected for manufacturing black cherry tomatoes product, such as ketchup and juice.

All the chemicals and reagents were of analytical grade.

2.2. Pectin extraction

Pectin was isolated from BCTW according to the method described by Grassino et al. (2016) with some modifies. In brief, the extraction was carried out in ammonium oxalate/oxalic acid as solvent at temperature of $60\text{ }^{\circ}\text{C}$ and frequency of 37 kHz (ultrasonic bath, Kunshan Ultrasonic Instrument Co., Ltd., China) for 60 mins twice. At the end of extraction, the mixture was filtered through Whatman No. 3 filter paper and then the filtrates were collected, 3 vol of 96% ethanol were added in to settle pectin down. The precipitated were acquired by filtering, washing with ethanol (70% and 96%) and acetone, and finally freeze dried.

Pectin extracted by ultrasonic got higher yield at $60\text{ }^{\circ}\text{C}$ in comparison with conventional extraction (without ultrasonic), while temperature raised to $80\text{ }^{\circ}\text{C}$, ultrasonic assisted extraction promoted the dissolution of total sugars from tomato waste and pectin obtained got lower quality according to according to pe-experiments. Pectin yield (%) was 24.36 ± 0.08 , which was expressed as ratio of dried pectin to the initial of dried tomato

waste, and much lower than pectin yield from tomato waste which was 31%–36% (Grassino et al., 2016).

2.3. DHPM treatment

Pectin was dispersed in distilled water with the concentration 5 mg/mL which is similar to black tomato sauce, and then shocked in ultrasonic bath for 5 mins to achieve complete dissolved. Then the suspension was treated with high pressure homogenizer (APV-2000, Denmark). Regulating homogeneous valve to obtain different pressures (0 MPa, 40 MPa, 80 MPa, 120 MPa, 160 MPa) for pectin solutions passing through. It took 30s for each sample with volume of 120 mL to finish the process and two passes for each sample. Three repetitions were acquired at each pressure. PD-0, PD-4, PD-8, PD-12, PD-16 represented the pectin solution treated with DHPM at 0 MPa, 40 MPa, 80 MPa, 120 MPa and 160 MPa, respectively. Each sample was collected by freeze drying.

2.4. Determination of particle size distribution

Particle size distribution was determined by Zetasizer Nano S (Malvern Instruments Ltd., UK) according to the description of Sun et al. (2016) with slight modifications. PD-0, PD-4, PD-8, PD-12, PD-16 were diluted by ultra-pure water with the concentration 20 mg/mL at room temperature, quartz cuvette with a path length 1 cm was used. All measurements were carried out at $25\text{ }^{\circ}\text{C}$ and analyzed in triplicate for each sample.

2.5. Scanning microscopy analysis

PDs were spreaded out and stucked to one side of a double-sided adhesive tape linked to a circular specimen stub. Field-emission Scanning Electron Microscopy (SEM) (Sirion 200, FEI, USA) was used to view the PDs after the samples were gold-plated at 5 kV voltages and 3.0 spot size, Low vacuum mode was executed while operating.

2.6. Determination of pectin content

Titration was performed to determine methoxyl (MeO), anhydrouronic acid (AUA) contents and degree of esterification (DE) according to the method described by Nazaruddin et al. (2013). Briefly, 0.5 g pectin was moistening in 5 mL 96% ethanol, 1 g sodium chloride and 100 mL deionized water were added, stirred to fully dissolve, the solution was titrated with 0.1 M NaOH (Titration A). Then, 25 mL of 0.25 M NaOH was added and stand for 30 mins, 0.25 M HCl was added neutralize NaOH, and the mixing was titrated with 0.1 M NaOH again (Titration B). Calculations of MeO, AUA and DE preformed as following equation.

$$\text{MeO}\% = \frac{\text{meq Titration B} \times 31 \times 100}{\text{Weight of sample}(\text{mg})} \quad (1)$$

$$z = \frac{\text{weight of sample}(\text{mg})}{\text{meq Titration A} + \text{meq Titration B}} \quad (2)$$

$$\text{AUA}\% = \frac{176 \times 100}{z} \quad (3)$$

$$\text{DE}\% = \frac{176 \times \text{MeO}\% \times 100}{31 \times \text{AUA}\%} \quad (4)$$

where 31 and 176 are the molecular weights of the MeO and AUA.

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