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Preparation of peach gum polysaccharides using hydrogen peroxide

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ABSTRACT

Most polysaccharides cannot dissolve in water but can be hydrolysed using hydrogen peroxide (H_2O_2) to yield a water-soluble product. This study presents a method of preparing water-soluble polysaccharides from peach gum by hydrolysis using H_2O_2. Extraction was monitored by the recovery rate. Factors affecting the hydrolysis of peach gum were investigated, and the optimum hydrolysis conditions were determined as follows: time, 8 h; temperature, 55 °C; H_2O_2 concentration, 4% (v/v); and NaOH concentration, 2.0 M. The hydrolysates were filtered, neutralised with HCl, concentrated to ~20% (w/v), precipitated with 5 volumes of ethanol, freeze-dried, and ground to yield a water soluble and white powder. The polysaccharide content of the product was 97.8%, and the yield was 83.6% (w/w).

1. Introduction

Peach tree belongs to the subfamily *Prunoideae* of the family *Rosaceae*. This species produces copious gum exudates caused by a disease (gummosis) on their fruit and trunk, especially after a mechanical injury followed by a microbial attack (Simas et al., 2008). Peach gum usually consists of polysaccharides with complex structures and a large number of monosaccharides, as well as glycosidic linkages that mostly have highly branched structures (Fernanda et al., 2008; Qian, Cui, Wang, Wang, & Zhou, 2011; Simas-Tosin et al., 2009; Tischer, Iacomini, & Gorin, 2002). Peach gum is widely used in food, medicine, and other industries in China (Wang & Huang, 2005).

Peach gum polysaccharides have low solubility in water owing to their high molecular weight and highly branched structure, thus limiting its wide application, particularly in medicine and the food industry. To improve its solubility as well as its biological, chemical, and physical properties, both enzymatic and acidic treatments are used to prepare a water-soluble low molecular weight peach gum polysaccharides (Kardošová, Rosík, & Kubala, 1978; Li & Huang, 2007). Hydrogen peroxide (H_2O_2) is used to hydrolyse polysaccharides such as cellulose, starch, hemicellulose, and peach gum polysaccharides because H_2O_2 is easy to handle, readily available, and environmentally friendly (Chang, Tai, & Cheng, 2001; Qin, Du, & Xiao, 2002; Shao, Yang, & Zhong, 2003; Wu, Cai, & Sun, 2012). This technique is based on the formation of free radicals, which can attack the glucosidic linkages of the polysaccharides. Whether H_2O_2 can attack the glucosidic linkages of peach gum polysaccharides remains undetermined.

Studies on the hydrolysis of peach gum polysaccharides by H_2O_2 are rarely reported. This study established an efficient method of degrading peach gum polysaccharides by hydrolysis using H_2O_2. After optimising the hydrolysis conditions, the product was partially characterised.

2. Materials and methods

2.1. Materials

Gum exudates of the peach (*Prunus persica*) tree trunk were collected at the Kong Wang Shan peach farm (Lianyungang, Jiangsu Province, China). H_2O_2 was purchased from the Laiyang Kant Chemical Co., Ltd. (Laiyang, China). All other chemicals were reagent-grade.

2.2. Extraction of peach gum polysaccharides with H_2O_2

Peach gum was soaked in distilled water at ~60 °C for 24 h and homogenised in a blender (Guangzhou Aoyu of Electronic Science and Technology Co., Ltd., Guangzhou Province, China) to yield a suspension with 1% concentration (w/v). Different volumes of H_2O_2 were poured into a reactor containing 500 mL of peach gum suspension, and the reactor was maintained in a thermostatic water bath at different temperatures for varying times. Aliquots of the reaction mixture were periodically collected and cooled below 10 °C to terminate the reaction.

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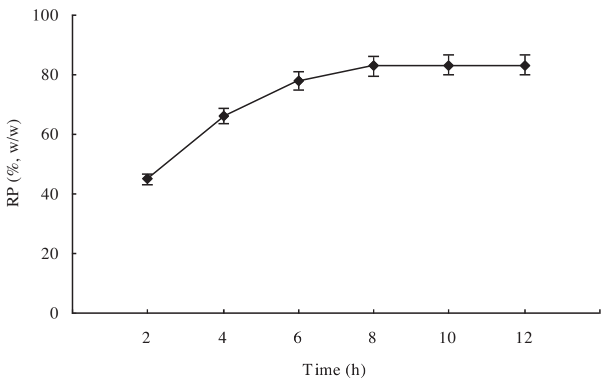

Fig. 1. Effect of time on extraction of peach gum polysaccharides with H_2O_2. Data are shown as mean ± SD ($n = 3$).

2.3. Recovery of peach gum polysaccharides

Aliquots of hydrolysates were filtered through a Whatman GF/A filter paper, neutralised with HCl, concentrated to ~20% (w/v), precipitated with 5 volumes of absolute ethanol, filtered through Whatman GF/A filter paper again and freeze-dried. The % recovery of peach gum polysaccharides (RP) was calculated using Eq. (1).

$$RP = 100\frac{W_2}{W_1} \quad (1)$$

where W_1 and W_2 represent the weights of the recovered peach gum polysaccharides and the original peach gum, respectively.

2.4. Analytical methods

Ash, moisture, total sugar and protein contents of the samples were determined according to standard methods (Hou, 2004). The reducing sugars were estimated by the Somogyi method and expressed as a dextrose equivalent (DE) value (Nelson, 1944). The DE values were used to evaluate the enzyme activities in the reaction mixture. The Fourier transform infrared (FTIR) spectra of representative hydrolysate samples were obtained in KBr pellets by using a Nicolet Nexus FTIR 470 spectrophotometer over a wavelength range of 400–4000 cm^{-1}.

3. Results and discussion

3.1. Effect of time on extraction of peach gum polysaccharides with H_2O_2

The effect of reaction time on the extraction of peach gum polysaccharides by H_2O_2 was determined over a period of 12 h (Fig. 1). The RP value rapidly increased within 6 h [from 45% (w/w) to 78% (w/w)], gradually increased from 6 h to 8 h [from 45% (w/w) to 78% (w/w)] and did not further increase after 8 h. Therefore, the optimal reaction time was 8 h.

3.2. Effect of temperature on extraction of peach gum polysaccharides with H_2O_2

The reaction temperature crucially affects peach gum polysaccharide extraction using H_2O_2. The temperatures studied were from 40 °C to 65 °C. The maximum RP was obtained at 55 °C (Fig. 2). The decrease in RP above 55 °C can be attributed to the excessive hydrolysis of H_2O_2 at high temperatures, which produced oligosaccharides with an extremely low degree of polymerisation that made precipitation with ethanol difficult. In contrast to our findings, several studies reported that the optimal temperatures for chitosan extraction using H_2O_2 are from 40 °C to 60 °C (Tian, Liu, Hu, & Zhao, 2004) and 70 °C (Huang, Wang, Huang, Zhuo, & Guo, 2007), whereas that for curdlan hydrolysis using H_2O_2 is 60 °C (Wu et al., 2012). The differences may be ascribed to the discrepancies in polysaccharide type, reaction time, and pH of reaction mixture.

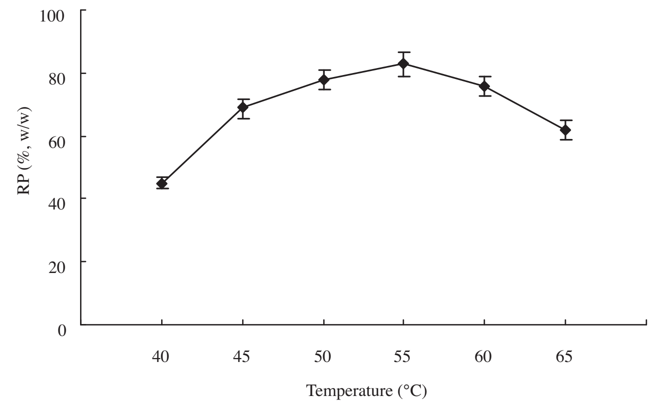

Fig. 2. Effect of temperature on extraction of peach gum polysaccharides with H_2O_2. Data are shown as mean ± SD ($n = 3$).

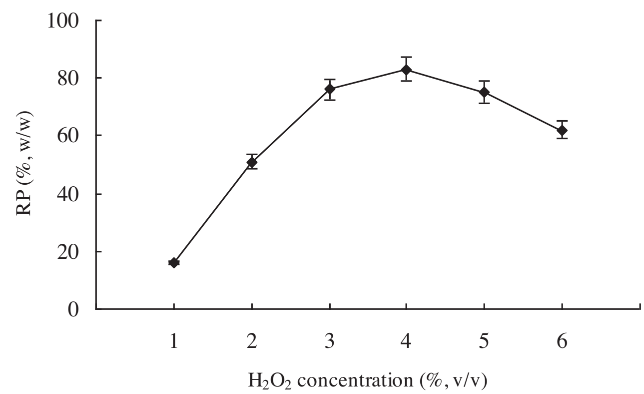

Fig. 3. Effect of H_2O_2 concentration on extraction of peach gum polysaccharides with H_2O_2. Data are shown as mean ± SD ($n = 3$).

3.3. Effect of H_2O_2 concentration on extraction of peach gum polysaccharides with H_2O_2

Fig. 3 shows the effect of the H_2O_2 concentration on peach gum polysaccharide extraction. The maximum RP was achieved at

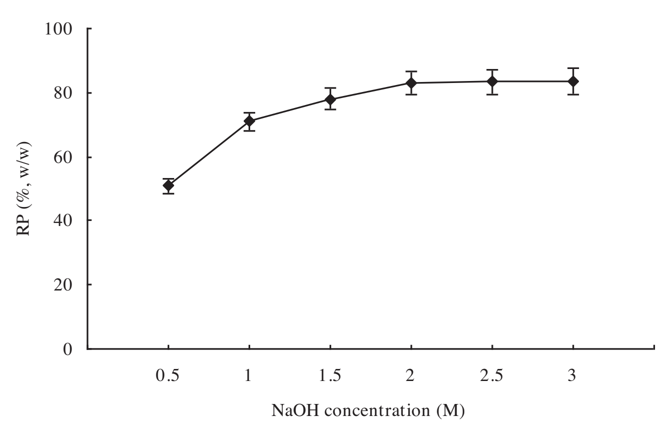

Fig. 4. Effect of NaOH concentration on extraction of peach gum polysaccharides with H_2O_2. Data are shown as mean ± SD ($n = 3$).

Fig. 5. Photograph of peach gum polysaccharides powder.

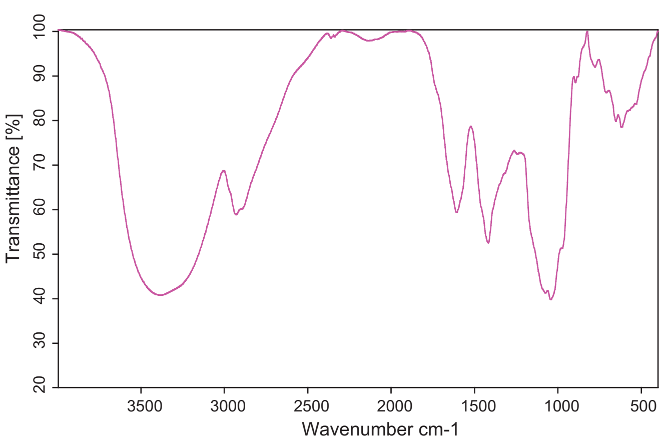

Fig. 6. FT-IR spectra of peach gum polysaccharides.

a H_2O_2 concentration of 4% (v/v). When the H_2O_2 concentration exceeded 4% (v/v), the increase in H_2O_2 concentration decreased RP; such decrease can also be attributed to the excessive hydrolysis of H_2O_2 in the presence of excess H_2O_2. Therefore, the optimum H_2O_2 concentration was 4% (v/v).

3.4. Effect of NaOH concentration on extraction of peach gum polysaccharides with H_2O_2

Fig. 4 shows the effect of NaOH concentration on peach gum polysaccharide extraction. The RP value sharply increased with the increase in NaOH concentration up to 1.5 M, gradually increased between 1.5 and 2 M NaOH and did not further increase beyond 2 M NaOH. Therefore, the optimum NaOH concentration was 2 M.

3.5. Product characterisation

Ash, moisture, and total sugar contents were 0.4%, 1.3% and 97.5% (w/w), respectively. The DE of the resulting products was 8.13, indicating that the average degree of polymerisation was ~12. All product samples consisted of water-soluble white powders (Fig. 5).

The FTIR spectra of the peach gum polysaccharides peaked at ~3400 cm^{-1} (O—H), ~1420 cm^{-1} (symmetrical deformation of $—CH_3$ and $—CH_2$), ~1040 cm^{-1} (stretching vibration of the C—O—C in glucose circle), and ~1705 cm^{-1} (special absorbance peaks of aldehyde in peach gum polysaccharides) (Fig. 6). These data also demonstrated that the peach gum was mainly composed of polysaccharides.

4. Conclusions

Peach gum polysaccharides can be effectively extracted using H_2O_2, and the maximum RP can be obtained under the optimum conditions of 55 °C, 4% (v/v) H_2O_2, 2 M NaOH and 8 h reaction time. The hydrolysates were filtered, neutralised with HCl, concentrated to ~20% (w/v), desalted, dried and ground to yield a white water-soluble powder. The products were mainly composed of polysaccharides. The polysaccharide content was calculated at 97.8%, and the yield was 83.6% (w/w).

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