



Extraction optimization and physicochemical properties of pectin from melon peel



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ABSTRACT

In this study, acidic extraction (citric acid) was used for the extraction of pectin from melon peel. A central composite design (CCD) was applied to study the effect of temperature (35–95 °C), time (40–200 min), pH (1–3) and solvent to sample ratio (10–50 v/w) on the yield and degree of esterification (DE). The results showed that the yield and DE ranged from 2.87 to 28.98% and 1.33–29.33%, respectively. Also, the highest extraction yield (29.48 ± 1.7%) was obtained under optimal conditions (pH of 1, temperature of 95 °C and ratio of 10 v/w after 200 min). The protein, ash, carbohydrate and galacturonic acid content of pectin obtained under optimal conditions were about 1.5, 3.5, 10 and 48%, respectively. Rheological experiments determined that melon peel pectin dispersions behaved as a weak gel at concentrations of 1%w/v. The emulsifying activity was 35% and also, the emulsion stability was higher at 4 °C than at 23 °C.

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1. Introduction

Pectin is a complex polysaccharide in the primary cell wall and middle lamella of higher plants. Pectin mainly consists of D-galacturonic acid (GalA) and monosaccharides including L-rhamnose, L-arabinose, D-galactose, and D-xylose [1]. In addition, GalA residue can be partly methyl-esterified [2].

Pectin is applied in food science and nutrition, cosmetics, and the pharmaceutical industry. It is widely used for thickening, gelling, and as an emulsifying agent in jams, soft drink, fish, meat, and milk products [3,4]. Furthermore, pectin has useful application in medicine where it helps in lowering serum cholesterol level, removing heavy metal ions from the body, stabilizing blood pressure, restoring intestinal functions [5], and in weight reduction [6]. This wide variety of applications for pectin accounts for its substantial consumption worldwide, and demand is growing. Annual consumption of pectin worldwide is currently approximately 45,000 tones [7].

Commercial pectin is produced from various different plant sources, the most common of which are apple pomace and citrus peel [3,8,9]. Producers and researchers have identified other sources for pectin production from agricultural by-products such

as cacao pod husks [10,11], peach pomace [12], sunflower heads [13], durian rind [14], banana peel [15], soy hull [16], pomegranate peel [17], and papaya peel [18].

Melon belongs to the family *Cucumis melo* L. and its peel is a potential source of pectin. Iran is a main producer of melon after China and Turkey. Melon is generally cultivated in the arid and semi-arid regions of Iran [19]. Fruit peel waste is sometimes used as animal feed, and this may be problematic in terms of environmental pollution. However there has been some interest in using this by-product to develop commercially valuable ingredients. Accordingly, melon processing industries have been searching for alternative applications and processing options for melon by-products such as peel as a source of many important natural compounds such as pectin, flavonoids, carotenoids, limonene, and polymethoxy flavones [20].

Acidic extraction is the most common procedure used to extract pectin from agro-industrial by-products and the process is normally based on protopectin hydrolyzation at a high temperature [11]. Research has demonstrated that the most important factors affecting yield and chemical and functional properties of the produced pectin were: type of acid, extraction temperature, time, pH and solvent to sample ratio (SSR) and research has shown that these factors have a considerable influence on product outcome [7,21]. Many researchers have investigated and optimized the effects of temperature, processing time, pH and SSR on pectin and polysaccharides extracted from agricultural by products [8,22].

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The main advantage of RSM is reducing the number of experimental trials and allowing evaluation of multiple parameters and their interactions [23]. RSM has been used in other studies to optimize conditions for extraction of pectin from agricultural by-products [25] but no specific information has been published to date on producing pectin from melon peel. The aim of this research was to optimize the effects of operating factors in the acidic extraction process (temperature, extraction time, pH and SSR) on yield of pectin extracted from melon peel using RSM and study of functional properties of pectin extracted from these optimal extraction conditions.

2. Materials and methods

2.1. Preparation and chemical analysis of melon peel

The commercial Iranian melon cultivars (*C. melo* L) used in this study was collected from a local fruit market. The fruit peels were removed from pulp. Afterwards, the peels were washed and dried in an oven at 80 °C for 1 day. Subsequently, dried peels were milled and the obtained powder were sieved (40-mesh size) and packed in dark bags and stored in a dry environment prior to experimental analyses [26].

2.2. Extraction and purification

Initially, the effect of acid type containing tartaric acid, citric acid, hydrochloric acid, acetic acid, lactic acid, nitric acid, phosphoric acid, and sulfuric acid on the pectin yield was investigated. Pectin extraction was performed at 90 °C for 90 min (SSR of 30 v/w), using acidified water at pH of 2.5. Afterwards, the most suitable acid was selected for further studies to adjust the pH for acidic extraction. In the next step, pectin was extracted at different conditions and the effect of temperature (35, 50, 65, 80, and 95 °C), extraction time (40, 80, 120, 160, and 200 min), pH (1.0, 1.5, 2.0, 2.5, and 3.0), and SSR (10:1, 20:1, 30:1, 40:1, and 50:1 v/w) on the extraction yield was investigated (Table 1). Afterwards, the hot extract was filtered through two layers of gauze and the filtrate was centrifuged at 5000g for 30 min to achieve higher purifications of pectin in the solution. Then, the supernatant was precipitated with 96% ethanol (1:2%v/v). After storing the solution for 24 h at 4 °C, the polysaccharides were washed two times with ethanol. Finally, the precipitated pectins were dried under vacuum at 50 °C to a constant weight [27]. The yield of extraction was calculated according to the following formula [24]:

$$\% \text{Yield} = \frac{\text{weight of dried extracted pectin (g)}}{\text{weight of dried powder (g)}} \times 100 \quad (1)$$

2.3. Physicochemical composition

The melon peel powder and the extracted pectin from melon peel were analyzed for crude protein ($N \times 6.25$) and total ash content [28]. Total carbohydrates were determined by the phenol-sulfuric acid photometric method, using D-glucose as reference [29]. The galacturonic acid content was determined by the meta-hydroxydiphenyl method using D-galacturonic acid as external standard [30].

2.4. Determination of the degree of esterification (DE)

The degree of esterification was determined by a titrimetric method described by Silva et al. [31]. Dried pectin (20 mg) was moistened with ethanol and dissolved in 20 ml of deionized water at 40 °C. After complete dissolution of pectin, five drops of phenolphthalein were added to the solution. Then the solution was

titrated with 0.5 M NaOH and the volume of NaOH solution used for color change was recorded as the initial titer (V_1). Subsequently, 10 ml of 0.5 M NaOH was added and then the solution was shaken strongly and was allowed to rest for 15 min. In the next step, 10 ml of 0.5 M HCl was added and the solution was shaken until the pink color was disappeared. The solution was titrated with 0.5 M NaOH for the last step, and the volume consumed was recorded as saponification titer or final titer (V_2). The DE was calculated according to the following formula:

$$DE = \frac{V_2}{V_1 + V_2} \times 100 \quad (2)$$

2.5. FT-IR spectroscopy

FT-IR spectrum of the extracted pectin was obtained at the medium resolution. The spectrum was recorded in the transparent mode from 4000 to 400 cm^{-1} (Spectrum 100, Perkin-Elmer, InK, MA, USA).

2.6. Experimental design

The extraction parameters were optimized using Response Surface Methodology (RSM). A central composite design (CCD) was employed in this regard. Temperature (X_1), extraction time (X_2), pH (X_3) and SSR (X_4) were chosen as independent variables.

The whole design consisted of 27 experimental points carried out in random order, which included 16 factorial points, 3 center points and 8 axial points. Three replicates at the center of the design were used to allow the estimation of a pure error sum of squares (Table 1). It should also be noted that all computation and graphics in this study were conducted using the statistical software Design Expert 7.0 and Excel.

2.7. Emulsifying properties

Emulsifying activity of pectin samples and the stability of prepared emulsions were assessed according to the method described by Dalev and Simeonova [32]. Determination of emulsifying activity is based on the ratio of the emulsified layer volume and the whole volume of the solution. Emulsions were prepared by adding 3 ml of vegetable oil to 3 ml of pectin solution (0.5%w/w), containing 0.02% sodium azide as a bactericide. The mixture was homogenized at 12,000 \times g for 3 min at room temperature in an Ultra-Turrax T-25 homogeniser (Ika T25 Digital Ultra-Turrax, Staufen, Germany).

Samples were centrifuged at 527 \times g for 5 min at 23 °C. After centrifugation, the whole volume of the solution (Wv) and the emulsified layer volume (ELV) were determined. Emulsifying activity (EA) was calculated as:

$$EA(\%) = \frac{ELV}{Wv} \times 100 \quad (3)$$

Similar emulsion samples were prepared to study the emulsion stability (ES) after 1 and 30 days of storage at 4 and 23 °C. The samples were centrifuged at 527 \times g for 5 min at 4 °C and 23 °C. The initial emulsified layer volumes (VEi) were measured. After each storage period, the samples were centrifuged and the remaining emulsified layer volumes were measured (VEr). Emulsion stability was calculated using the following relation:

$$ES(\%) = \frac{VEr}{VEi} \times 100 \quad (4)$$

2.8. Rheological properties

Pectin dispersions were prepared by dissolving pectin with the concentration of 1%w/v in deionized water. Small deformation oscillatory and viscosity measurements were performed using a

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