Food Chemistry 198 (2016) 113–118

Contents lists available at ScienceDirect

Food Chemistry

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

Optimization of pectin extraction from banana peels with citric acid by using response surface methodology

Túlio Ítalo S. Oliveira [1], Morsyleide F. Rosa [2], Fabio Lima Cavalcante [1], Paulo Henrique F. Pereira, Graham K. Moates, Nikolaus Wellner, Selma E. Mazzetto [1], Keith W. Waldron, Henriette M.C. Azeredo [2,*]

Institute of Food Research, Norwich Research Park, Colney, Norwich NR4 7UA, UK

ARTICLE INFO

Article history:
Received 18 February 2015
Received in revised form 18 July 2015
Accepted 20 August 2015
Available online 21 August 2015

Keywords:
Polysaccharides
Biopolymers
Response surface methodology
Banana
Pectin

ABSTRACT

A central composite design was used to determine effects of pH (2.0–4.5), extraction temperature (70–90 °C) and time (120–240 min) on the yield, degree of methoxylation (DM) and galacturonic acid content (GA) of pectins extracted from banana peels with citric acid. Changes in composition during the main steps of pectin extraction were followed by Fourier transform infrared (FTIR) spectroscopy. FTIR was also used to determine DM and GA of pectins. Harsh temperature and pH conditions enhanced the extraction yield, but decreased DM. GA presented a maximum value at 83 °C, 190 min, and pH 2.7. The yield of galacturonic acid (YGA), which took into account both the extraction yield and the pectin purity, was improved by higher temperature and lower pH values. The optimum extraction conditions, defined as those resulting in a maximum YGA while keeping DM at a minimum of 51%, were: 87 °C, 160 min, pH 2.0.

1. Introduction

Bananas (*Musa acuminata*, AAA Group) are one of the most important fruit crops, with a global annual production that surpassed 100 million tons in 2011 (The World Banana Forum, 2014). Bananas are mostly consumed raw, and their processed products include banana flour, chips, and puree (which can be used to produce nectars, smoothies, and a variety of other products). Banana peels constitute about 30% of the fruit, and represent an environmental problem because of their large nitrogen and phosphorus contents as well as their high water content, making them highly susceptible to microbial degradation (González-Montelongo, Lobo, & González, 2010). The use of banana peels as a source of high value compounds such as pectin (Happi Emaga, Ronkart, Robert, Wathelet, & Paquot, 2008b), cellulose nanofibers (Tibolla, Pelissari, & Menegalli, 2014), and phenolic compounds (González-Montelongo et al., 2010; Rebello et al., 2014) is interesting not only from an economic point of view, but also from an environmental perspective.

Some previous studies were carried out to investigate effects of process variables on extraction of pectin from banana peels. Qiu et al. (2010) investigated the effects of pH, extraction time, temperature, and salting out time on pectin extraction by using an enzymatic method. Happi Emaga et al. (2008b) evaluated the differences in pH, temperature and time on pectin extraction from banana peels using sulfuric acid. However, no previous report has been found on studying effects of process variables on pectin extraction from banana peels using organic acids. On one hand, strong mineral acids are cheaper and more effective than organic acids; on the other hand, organic acids are more interesting than strong acids from an environmental point of view (Chan & Choo, 2013; Pinheiro et al., 2008). Moreover, because of their lower dissociation constant, organic acids have a lower hydrolyzing capacity than mineral acids, so they will be less likely to cause proton-catalyzed depolymerization of pectins (Kermani et al., 2014). That may be especially important during the mixing step, when the local acid concentrations can fluctuate dramatically. Pectin yields from extraction of apple pomace, cocoa husks, and passion fruit peel with citric acid were found to be similar to those obtained with hydrochloric acid (Canteri-Schemin, Fertonani, Waszczynskyj, & Wosiacki, 2005; Chan & Choo, 2013; Kliemann et al., 2009).

The objective of this study was to evaluate the influence of pH, temperature, and time on pectin extraction from banana peels with citric acid.

* Corresponding author.
E-mail addresses: tulioufc@gmail.com (T.Í.S. Oliveira), morsyleide.rosa@embrapa. br (M.F. Rosa), fabiolima2008@hotmail.com (F.L. Cavalcante), selma@ufc.br (S.E. Mazzetto), henriette.azeredo@embrapa.br (H.M.C. Azeredo).
[1] Permanent address: Federal University of Ceara, Chemistry Department, Campus do Pici, Bloco 940, CP 6021, CEP 60455-760 Fortaleza, CE, Brazil.
[2] Permanent address: Embrapa Tropical Agroindustry, R. Dra. Sara Mesquita, 2270, CEP 60511-110 Fortaleza, CE, Brazil.

http://dx.doi.org/10.1016/j.foodchem.2015.08.080
0308-8146/

2. Materials and methods

2.1. Chemical composition of banana peel powder and changes in FTIR spectra

Banana peels at ripening stage 6 (fully ripe bananas, ready for consumption) were collected from Cavendish bananas purchased in the local market (Norwich, UK). The peels were immersed in sodium metabisulfite solution (1% w/v) for 24 h, oven-dried at 60 °C for 24 h, and milled to 0.5 mm in a Retsch Brinkmann ZM-1 centrifugal grinding mill (Retsch GmbH, Haan, Germany).

The ash, extractive, and Klason lignin contents of the banana peel powder were determined according to methods TAPPI t413 OM-93 (TAPPI, 1993), TAPPI T204 cm-97 (TAPPI, 1997), and TAPPI T222 om-22 (TAPPI, 2000), respectively. Hemicellulose and α-cellulose contents were analyzed as described by TAPPI T203 cm-99 (TAPPI, 2009), and holocellulose, according to Yokoyama, Kadla, and Chang (2002).

Fourier transform infrared (FTIR) spectra were collected from the banana peel powder, the alcohol insoluble residue (AIR), and pectin, in the frequency range of 4000–800 cm^{-1} (128 scans at 2 cm^{-1} resolution) on a Nicolet Magna-IR 860 FTIR spectrometer (Thermo Nicolet, Madison, WI, USA). Samples were placed on a GoldenGate diamond ATR accessory (Specac, Orpington, Kent). The empty crystal was used as reference. The spectra were compared in order to understand the changes that occurred during the pectin extraction process.

2.2. Preparation of the alcohol-insoluble residue (AIR)

The pectin extraction follows a preparation of an alcohol insoluble residue, in order to remove low molecular weight compounds, including traces of free galacturonic acid (Happi Emaga et al., 2008b).

The alcohol insoluble residue (AIR) preparation was based on the method by Waldron and Selvendran (1990), with some modifications. 100 g of milled peels were washed three times with ethanol, to remove any alcohol soluble phenolics (the first time with 600 mL of boiling 70% v/v ethanol solution for 5 min, the second time with 600 mL of boiling absolute ethanol for 5 min, the third time with 600 mL of absolute ethanol at room temperature for 5 min), then washed in 200 mL acetone. Between washings, the material was filtered through a 10 μm nylon mesh. The AIR was air dried at room temperature.

2.3. Pectin extraction

The pectin was extracted from the AIR with citric acid solution (AIR: citric acid solution ratio, 1:20 w/v), according to a central composite design (CCD) with 3 variables: pH of the citric acid solution, temperature and time of extraction (Table 2). The extraction was carried out in a water bath under stirring (150 rpm). After centrifugation (1147 g, 30 min, 10 °C), the supernatant was vacuum filtered, added to the same volume of absolute ethanol, and the pH was adjusted to 3.5 (pH of minimum pectin solubility) with KOH. The mixture was stirred for 30 min, left to precipitate at 4 °C for 2 h, and centrifuged (15 min, 4 °C, 3500 rpm). The pellet was collected, washed with ethanol 70% (v/v), centrifuged again (20 min, 4 °C, 3500 rpm), and dried at room temperature. It was then diluted/dispersed in water (Ystral, 20 min), had its pH adjusted to 7 (with KOH), and was again dried and milled to a fine powder using a basic mill (A10, IKA GmbH, Germany).

2.4. Pectin characterization and statistical analyses

The degree of methoxylation (DM) and the galacturonic acid content (GA) of the banana peel pectin were determined from FTIR spectra (collected as previously described). The DM was determined as described by Manrique and Lajolo (2002), with some modifications. Pectin standards with known DM values of 31% (P-9311), 67% (P-9436), and 89% (P-9561) were obtained from Sigma (Steinheim, Germany), their FTIR spectra were recorded (in triplicates), as well as the spectra of four other samples obtained from blends of the pectin standards (with DM values of 40, 49, 60, and 78). The DM determination was based on the band areas at 1700–1750 cm^{-1} (methyl esterified uronic acids, EUA) and 1600–1630 cm^{-1} (free uronic acids, FUA), calculated by using Origin Pro 9, multiple peak fit function, Gaussian fitting (OriginLab, Northampton, USA). A calibration curve was obtained by plotting DM against the ratio between the EUA peak area over the sum of the EUA and FUA peak areas of the standards (Eq. (1)), and used to determine the DM of the banana peel pectin from all treatments.

$$DM = 126.3R_A + 2.493 \quad (R^2 = 0.974) \tag{1}$$

$$R_A = \frac{A_{EUA}}{A_{EUA} + A_{FUA}} \tag{2}$$

DM: degree of methoxyl esterification; A_{EUA}: area corresponding to the peak at 1700–1750 cm^{-1} (methyl esterified uronic acids); A_{FUA}: area corresponding to the peak at 1600–1630 cm^{-1} (free uronic acids).

The determination of the galacturonic acid content (GA) was modified from the method described by Monsoor, Kalapathy, and Proctor (2001). A set of 10 calibration galacturonic acid standards was prepared by blending polygalacturonic acid (95%, 81325, Fluka) with cellulose (Sigmacell type 20, Sigma) to obtain standards with galacturonic acid contents of 0% (pure cellulose) to 90% (w/w) galacturonic acid. Peak areas were measured as the area above the baseline between 1840 cm^{-1} and 1550 cm^{-1}, which was used to calculate the total carbonyl peak area (Monsoor et al., 2001). A calibration curve was obtained by plotting GA against the total carbonyl peak area (Eq. (3)), and used to determine the GA of the banana peel pectin from all treatments.

$$GA(\%) = 0.7934 \times A_{1550-1840} - 0.1985 \quad (R^2 = 0.951) \tag{3}$$

GA: galacturonic acid content; $A_{1550-1840}$: area above the baseline between 1840 cm^{-1} and 1550 cm^{-1}, corresponding to the total carbonyl peak area.

The yield of galacturonic acid (YGA), defined by Eq. (4), was considered as a good measure of the performance of the extraction process, since it took into account both the extraction yield (EY), but also the purity of the material extracted (GA, galacturonic acid content).

$$YGA(\%) = \frac{EY(\%) \times GA(\%)}{100} \tag{4}$$

Results of the extraction yield, degree of methoxylation, galacturonic acid content and yield of galacturonic acid were analyzed using the software Minitab® 16 (Minitab Inc., State College, PA, USA). The regressions to represent the responses were obtained and evaluated in terms of their determination coefficients (R^2 values) and the significance of their F values.

3. Results and discussion

3.1. Chemical composition of banana peel powder and changes in FTIR spectra

The banana peel powder has less than 40% holocellulose, 48% of it being α-cellulose (Table 1), and the remaining 52% being hemicelluloses and pectin. When compared to the values reported by Oberoi, Vadlani, Saida, Bansal, and Hughes (2011) for composition