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Physicochemical properties of apple juice during sequential steps of the industrial processing and functional properties of pectin fractions from the generated pomace

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ABSTRACT

The impact of processing at industrial scale on properties of apple juice and pectin fractions extracted from the generated pomace is scarcely known. In this study, apple juice was collected at selected steps of the industrial production process and evaluated for physical and chemical properties. The generated pomace was recovered and subjected to extraction of pectins according to their solubility. They were evaluated for physicochemical and functional properties. The transmittance for color and clearness of juice increased during clarification step but decreased during concentration. The sugar content was not altered by any step. The content of individual acids showed some changes during the pasteurization. The levels of total and individual phenols tended to increase during the production process. Low levels of 5-HMF (4.0 mg/L) were detected only in the concentrated juice while patulin was absent. The pomace contained only chelator and alkali soluble pectin. In comparison to commercial apple pectin, extracted pectins were of low degree of esterification (67% vs 22.8–44.6%), GalA (654.1 vs 393.5–436.6 mg/g) and Gal (228.6 vs 71.3–115.0 mg/g) content but rich in Ara (27.5 vs 103.4–166.3 mg/g) and of high molecular weight (644.5 vs 1559.6–2360.6 kDa). Their viscosity was lower than that of commercial apple pectin (*k* values of 0.03–0.05 vs 0.14 Pa·s^n, respectively). They showed lower thermal stability than commercial apple pectin. The production steps involving high temperature or enzymes effected the properties of apple juice and extracted pectins.

1. Introduction

Clarified apple juice is one of the most popular processed foods in the world. Several chemical components (i.e. sugars, acids, phenolic compounds, pigments, safety-related compounds, etc.) are involved in juice quality; however, the levels of these compounds are highly influenced by the production process (Kadakal & Nas, 2003; Suárez-Jacobo et al., 2011). The apple pulp and juice are exposed to exogenous enzymes, adsorbent materials, high temperatures, and high osmotic pressure during the production process (Eisele & Drake, 2005). These conditions favor the release, degradation, transformation, and generation of compounds involved in juice quality (Markowski, Baron, Le Quéré, & Płocharski, 2015; Onsekizoglu, Bahceci, & Acar, 2010). The production process determines the color and turbidity as well as the levels of phenols, sugars, acids, the mycotoxin patulin and 5-hydroxymethylfurfural (5-HMF) of clarified juice, among other compounds (Gökmen, Artık, Acar, Kahraman, & Poyrazoğlu, 2001; He, Ji, & Li, 2007; Markowski et al., 2015; Onsekizoglu et al., 2010; Welke, Hoeltz, Dottori, & Noll, 2009). However, these processing effects have mainly been determined under laboratory or pilot plant conditions (Gökmen et al., 2001; Kadakal & Nas, 2003; Onsekizoglu et al.,

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2010; Spanos, Wrolstad, & Heatherbell, 1990; Suárez-Jacobo et al., 2011). The effect of apple juice processing at industrial scale has been scarcely studied and limited to a few processing steps and juice attributes (Welke et al., 2009). Further information in this regard is required to be used as a basis for the improvement of the quality of clarified apple juice.

The pomace is the main waste product generated by the apple juice industry. It represents 25% of fruit weight and its disposal involves costs and environmental pollution (O'Shea et al., 2015). However, it also is an important source of commercial pectin, which is a heteropolysaccharide widely used in many industrial sectors for its functional properties (gelling, stabilizing, emulsifying and thickening agent) (Wang, Chen, & Lü, 2014). The functionality of this type of apple pectin depends of its physicochemical characteristics, which are highly influenced by apple variety and ripening stage as well as the using of enzymes during juice production (Canteri-Schemin, Fertonani, Waszczynskyj, & Wosiacki, 2005; Fischer, Arrigoni, & Amadò, 1994). The physicochemical properties of total pectin from apple pomace have extensively been studied (Kammerer, Kammerer, Valet, & Carle, 2014; Kosmala et al., 2010; Min, Bae, Lee, Yoo, & Lee, 2010; O'Shea et al., 2015; Wang et al., 2014). However, the physicochemical and functional properties of pectin fractions from apple pomace have received scarce attention (Kosmala et al., 2010). The objective of this work was to determinate the impact of selected steps of the production process at industrial scale in apple juice quality as well as to evaluate the physicochemical and functional properties of different pectin fractions obtained from the generated pomace.

2. Materials and methods

2.1. Apple juice and pomace

The samples of juice and pomace from 'Golden Delicious' apples were obtained under industrial scale conditions. Triplicate samples of juice (1 L) and pomace (3 Kg) were weekly collected during four weeks. Juice was collected at the pressing, pasteurization, clarification, and concentration step while pomace only at pressing step. The first step consisted in pressing apple cubes (2 mm) previously treated with pectin lyase for 60 min at 25 °C. The pomace was collected at this step and stored at −70 °C until pectin extraction. The juice was pasteurized at 90 °C for 60 s and then quickly cooled (50 °C). The clarification step involved the treatment with a mixture of polygalacturonases, pectinase and glucoamylase for 100 min at 55 °C followed by ultrafiltration (UF) with tubular membranes of molecular weight cut-off (MWCO) of 100 kDa. The concentration consisted of two heating steps, one at 55 °C (23 min) and other at 70 °C (60 min). The average production flow was 3000 L/h. The juice samples were immediately stored at −70 °C until analysis. The juice samples were analyzed for total soluble solids content (TSS %) to determine the dehydration level during the production process and then adjusted to 11.2 °Brix before analysis of the physicochemical attributes. Several pectins were sequentially obtained from the apple pomace according to their solubility and evaluated for physicochemical and functional properties.

2.2. Miscellaneous evaluations in juice

The content of TSS (%) was determined in three subsamples (0.5 mL) of each juice using an automatic digital refractometer RX 5000 (ATAGO, Tokyo, Japan). The titratable acidity was determined in tree subsamples (5 mL) of each juice according to Eisele and Drake (2005). The total phenolic content was determined three times in each juice by the Folin Ciocalteu method at 750 nm, according to Spanos et al. (1990), and expressed as mg of chlorogenic acid equivalent per L of juice. Three subsamples (8 mL) of each juice were evaluated for color and clearness using a Thermo Spectronic 20D + spectrophotometer (Thermo Scientific, Wisconsin, USA) at 440 and 625 nm (transmittance), respectively. These measurements were performed according to Kadakal and Nas (2003).

2.3. Individual sugars and organic acids

The evaluation of sugars and organic acids was performed according to Ornelas-Paz et al. (2017), with slight modifications. For sugars, three subsamples of each juice were centrifuged (24652 *g*/20 min/4 °C) and then diluted ten times with water. The pH of diluted juice was adjusted at 7 with 10% NaOH, filtered and manually injected (20 μL) into a HPLC system (Varian Inc., CA, USA), which was equipped with a refractive index detector (Star Model 9040). The sugars were separated in a SUGAR SC 1821 (8.0 × 300 mm) (SHODEX, Tokyo, Japan) ion exchange column at 70 °C. The mobile phase was HPLC grade water at a flow rate of 0.6 mL/min. For organic acids, three subsamples of each juice were filtered and directly injected (20 μL) into the HPLC system described above but fitted to a UV-Vis detector (Model 9050). Acids were separated in an Aminex HPX-87H (7.78 × 300 mm) ion exchange column (Bio-Rad Laboratories., CA, USA). The separation was performed at 60 °C using 5 mM H_2SO_4 as mobile phase at a flow rate of 0.4 mL/min. The organic acids were monitored at $\lambda = 210$ nm. The identification and quantification of sugars and acids were performed using standard compounds.

2.4. Analysis of phenolic compounds

The extraction and chromatographic separation of phenolic compounds were based in the methodology described by Ornelas-Paz et al. (2017). Three subsamples of each juice were centrifuged (24652 *g*/20 min/4 °C), filtered and manually injected (20 μL) into an Agilent 1200 series HPLC system (Agilent Inc., CA, USA) equipped with a diode array detector. The separation of phenolic compounds was performed in a ZORBAX XDB-C18 column (4.6 × 150 mm) (Agilent Inc., CA, USA) at 30 °C. The phenolic compounds were monitored at $\lambda = 280$, 320, 350 and 520 nm. The mobile phase consisted of 2% acetic acid (A) and acetonitrile (B), according to the following gradient: 100% A at 0 min, 93% A at 12 min, 89% A at 20 min, 86% A at 35 min, 84% A at 36 min, 82% A at 41 min, 76% A at 48 min, 70% A at 54 min, and 65% A at 59 min. The flow rate was 1 mL/min. Reference compounds were used for identification and quantification purposes.

2.5. Analysis of patulin and 5-HMF

Three subsamples (10 mL) of each juice were individually placed in a separatory funnel, vigorously mixed with ethyl acetate (20 mL), and kept in repose until separation of phases. The organic phase was recovered while the aqueous phase was extracted two times more with ethyl acetate (20 mL each time), recovering the organic phase after each washing. The organic phases were pooled and washed two times with 1.5% Na_2CO_3 (10 mL). The organic phase was recovered and evaporated at reduced pressure at 40 °C. The residue was dissolved in 1 mL of water at pH 4 (adjusted with acetic acid), filtered, and manually injected (100 μL) in the Agilent 1200 series HPLC system described above. The separation of patulin and 5-HMF was performed according to Gökmen and Acar (1999), using a ZORBAX XDB-C18 (4.6 × 150 mm) (Agilent Inc., CA, USA) column at 30 °C. The patulin and 5-HMF were monitored at 276 and 284 nm, respectively. The mobile phase consisted of water (pH 4, A) and acetonitrile (B) according to the following gradient: 99% A at 0 min, 95% A at 10 min, 90% A at 15 min, 50% A at 20 min, 100% B