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Effect of UV–Vis processing on enzymatic activity and the physicochemical properties of peach juices from different varieties



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ABSTRACT

This work evaluated the effect of ultraviolet and visible light (UV–Vis) processing on the enzymatic activity (polyphenol oxidase and peroxidase) and physicochemical characteristics (pH, $^{\circ}$ Brix, acidity, formol index, phenolics, ascorbic acid, sugars and colour) of peach juices from three different varieties (Baby Gold, Calanda and Planet Top). Irradiation was performed for 120 min, at 25 and 45 $^{\circ}$ C, using a multi-wavelength emission lamp which provided a radiation power of $4.49 \cdot 10^{-2}$ W·cm⁻² at the liquid surface. After the treatments, no significant changes were observed in the pH value, acidity, vitamin C content, sugar content, or lightness. The phenolic content was reduced for the Planet Top juices, while the formol index decreased for the Calanda variety. a^* and b^* CIELab parameters decreased with processing time, contrary to that reported for thermal treatments. In most cases, UV–Vis irradiation was effective at inactivating polyphenol oxidase (PPO) and peroxidase (POD), although sometimes an initial activation was observed. The inactivation stage was described by a first-order model. At 45 $^{\circ}$ C, PPO was almost totally inactivated for the three juices studied, while reductions in POD of up to 60% were registered. Further experiments are suggested to improve the effectiveness of this technology for inactivating enzymes in peach juices.

1. Introduction

Peach juice is a popular product for consumers, being among the five most popular juice flavours in the European Union (AIJN, 2016). For commercialization, it needs to be submitted to a process to inactivate microorganisms and enzymes. Currently, heat processing is the most commonly used means for this purpose, thereby extending its shelf life. However, this process may have adverse effects on the sensory and nutritional qualities of the product (Ibarz, Garza, & Pagán, 1999; Ling, Tang, Kong, Mitcham, & Wang, 2015).

Ultraviolet irradiation in liquid foods is an alternative to thermal treatment. This method is being studied and developed to obtain a better sensory quality in the final product and including microbial safety (Ibarz, Garvín, & Falguera, 2015). Compared with thermal pasteurization, UV-treated juice may have the added benefit of having a more fresh-like quality in addition to being a simpler process with lower operating costs.

Among the non-thermal technologies studied for fruit juice processing, ultraviolet irradiation has shown a high effectivity at inhibiting enzymes (Jiménez-Sánchez, Lozano-Sánchez, Segura-Carretero, & Fernández-Gutiérrez, 2017). This is important since enzymatic reactions can lead to loss of quality during storage, polyphenol oxidase

(PPO) and peroxidase (POD) being the main enzymes that need to be inactivated. According to Manzocco, Quarta, and Dri (2009), ultraviolet (UV) and visible (Vis) light may promote photo-oxidation processes that modify the native structure of the protein, leading to losses in its functional activity due to side-chain oxidation, backbone fragmentation and/or formation of cross-links and aggregates.

The rate of the enzymatic photo-inactivation will depend on the nature and dose of radiation. For instance, a 100-min treatment with UV from a single emission lamp resulted on a reduction of 70% of the initial activity of PPO (Manzocco et al., 2009). Using a multi-wavelength emission lamp, PPO could be completely and irreversibly inactivated in different apple juices, also after 100 min of processing (Falguera, Pagán, & Ibarz, 2011). In fact, multi-wavelength lamps, that emit both ultraviolet and visible radiations, are widely used since they are more powerful than lamps that generate UV.

A considerable number of studies have shown successful results for inactivating PPO and POD by UV–Vis irradiations in model solutions and real fruit juices. For instance, after 15 min of processing, the initial activities of PPO and POD were reduced to < 5% in a coconut water model (Augusto, Ibarz, Garvín, & Ibarz, 2015). In several pear juices, PPO was almost inhibited with a 120-min treatment and POD was null with exposure times up to 60 min (Falguera, Garvín, Garza, Pagán, &

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Ibarz, 2014). Another approach proposed to optimize UV pasteurization consists of performing this treatment at temperatures between 45 and 60 °C (Aguilar, Ibarz, Garvín, & Ibarz, 2016; Gayán, Serrano, Monfort, Álvarez, & Condón, 2013; Sampedro & Fan, 2014).

The effect of UV–Vis irradiation on peach juices has been assayed in terms of colour. However, the impact of this process on the enzymatic activities and other quality parameters has not been investigated. Accordingly, this work aims to analyse the effect of UV–Vis treatments on the enzymatic activity of PPO and POD as well as the physical and chemical parameters in peach juices from three different varieties. In order to elucidate the effect of the temperature, irradiation was performed at 25 and 45 °C. Temperatures above 45 °C were not considered because of the well-known negative effects of the thermal treatment.

2. Materials and methods

2.1. Juice preparation

Peaches from the Baby Gold, Calanda and Planet Top varieties were obtained from local producers in the region of Lleida (Catalonia, Spain). The fruit was washed, each cut into four, pitted, and squeezed with a household juicer. The resulting juice was clarified by centrifugation in an Avanti J-26XP centrifuge (Beckman Coulter, USA) at 13,000 rpm (26,000 g) for 25 min. The temperature was maintained at 4 °C to minimize enzymatic and chemical changes. After centrifuging, the supernatant was recovered and the pellet was discarded. Then, fractions of 800 mL of juice were stored in the freezer at $-20\,^{\circ}\text{C}$.

2.2. UV-Vis irradiation

The UV–Vis irradiation was carried out in a dark chamber containing the reactor and the lamp. The reactor was a $12.5 \times 10.5 \times 10$ cm methacrylate tank which was filled with 800 mL of juice. As is well known, when a fruit juice is irradiated, all the radiation is absorbed in the top few millimetres. In order to avoid this becoming a handicap, the solution was stirred using a magnetic stirrer. This way, all the molecules or microorganisms in the solution can be reached by the radiation. In order to control the working temperature, a refrigeration system consisting of a metallic coil fed with either hot or cold water was coupled to the tank. The temperature was continuously monitored with a R100 thermometer (XS Instruments, Italy) immersed in the juice. The lamp used was a Philips HPM-12 (Philips, Eindhoven, Netherlands) with a nominal power of 460 W that emitted in a range between 250 and 740 nm (Fig. 1). It was centred 23.9 cm above the surface of the liquid in the reactor.

The juice sample was unfrozen at $8\,^{\circ}$ C for $24\,h$ before irradiation started. After that, it was heated until the working temperature was reached. The lamp was turned on $10\,m$ in before placing the juice in the

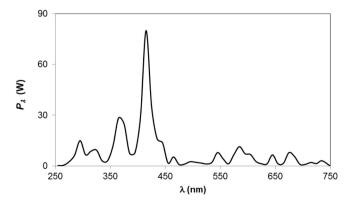


Fig. 1. Emission spectrum of the Philips HPM -12 lamp. P_{λ} is the power of emission at λ wavelength. Data provided by the lamp manufacturer.

chamber. Then, irradiation was carried out for 2 h. Each variety of juice was treated at 25 (UV25) and 45 °C (UV45). The fresh juice without irradiation was considered as the control. Both treatments and determinations were carried out in duplicate. Aliquots to be analysed were taken every 20 min. All the samples were filtered with paper (pore size 20 µm) before analysis.

2.3 Radiation dose

The real power emitted by the lamp was evaluated by actinometry. This consists of monitoring a well-known photo-chemical reaction. In this case, the photo-decomposition of oxalic acid in the presence of uranyl cation was used, as described in a previous work (Aguilar et al., 2016). The incident spectral radiant power on the liquid surface of the reactor P(0) was calculated with the method in Garvín, Ibarz, and Ibarz (2015). In this method, the geometry of a plane photo-reactor, a linear lamp and a linear spherical emission model are considered. The radiation dose applied to the juices was obtained with the following equation:

$$D_r = P(0) \cdot t \tag{1}$$

where D_r is the radiation dose expressed in J·cm⁻², P(0) is the radiant power that insides on the liquid surface of the reactor and t is the exposure time.

2.4. Physical and chemical analysis

The pH was measured with a Crison micropH 2000 pH meter (Crison Instruments, S. A., Alella Spain). Tritratable acidity was determined by potentiometric titration, following the method of the International Federation of Fruit Juice (IFU, 1996a). The soluble solid content was determined using an Atago RX-1000 Digital Refractometer (Atago Co. Ltd., Japan). The total phenolic content was assayed using the spectrophotometric method and the Folin-Ciocalteu reagent, as described by Shaghaghi, Manzoori, and Jouyban (2008). The formol index was evaluated to estimate the amino acid content (IFU, 1986). This is determined by measuring the acidity of the compounds formed in the reaction of formaldehyde and the amino acids in the juice up to a pH of 8.1, using a potentiometric titration. Vitamin C was evaluated by titrating with 2,6-dichloroindophenol, according to the official method AOAC (2007).

The sugar content (sucrose, glucose and fructose) was determined using a 1260 Infinity HPLC chromatograph equipped with an RID detector (Agilent Technologies, Germany) and a Hi-Plex Ca $300 \times 7.7 \text{mm} \times 8 \, \mu \text{m}$ column (Agilent Technologies, GB) at $80\,^{\circ}\text{C}$, with a mobile phase of $0.5\,\text{mL·min}^{-1}$ of ultrapure water (IFU, 1996b). The juice colour was monitored using a Chroma Meter CR-400 tristimulus colorimeter (Konica Minolta Sensing, Inc., Japan) in the CIELab colour space.

2.5. Enzymatic activity

The enzymatic activity of PPO was determined for each sample by measuring the increase in absorbance at 420 nm when the 4-methyl-catechol substrate reacted with the PPO enzyme in the juice (Yerlitürk, Arslan, Sinan, Gencer, & Özensoy, 2008). In each assay, 1.0 mL of the juice sample and 2.5 mL of 4-methylcatechol (Sigma-Aldrich Co, India) 0.01 M in buffer solution at pH 4.0 (citric acid and Na₂HPO₄, Panreac, Spain) were mixed in a 1-cm quartz cuvette. The absorbance at 420 nm was measured every 5 s for 3 min using a Helios Omega spectro-photometer (Thermo Fisher Scientific Inc., USA).

The enzymatic activity of POD was determined for each sample by measuring the increase in the absorbance at 420 nm when the substrate pyrogallol reacted with the POD enzyme present in the juice (Kwak et al., 1995). In each assay, $160 \, \mu L$ of the juice sample, $2.25 \, mL$ of buffer solution at pH 6.0 (citric acid and Na_2HPO_4 , Panreac, Spain) and

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