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Suitability of different varieties of peaches for producing minimally processed peaches preserved by high hydrostatic pressure and selection of process parameters

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ABSTRACT

Fresh-cut products represent an easy way to include fruits in everyday meals. The aim of this work was to evaluate the suitability of two cultivars for high pressure processing (HPP), and to establish the principal parameters leading to a better quality preservation of minimally processed peaches. *Prunus persicae* cv. Flavorcrest and cv. Romea were subjected to different HPP-treatments according to a factorial design. The factors were: pressure level (500, 600 and 700 MPa) and holding time (1 and 5 min), applied at room temperature. Several determinations were carried out over the samples: texture parameters, ascorbic acid content, total phenols, and polyphenoloxidase and alcohol dehydrogenase activity. Results showed that only the 700 MPa treatments, for both holding time evaluated, provoked a significant decrease in the hardness of the HPP-product. Romea had lower polyphenoloxidase activity and higher ascorbic acid and total phenols content than Flavorcrest. The application of 600 MPa-5 min to Romea peaches successfully prevented enzymatic browning, with the additional advantage of rendering higher concentrations of ascorbic acid and phenols. This last aspect would be an asset either for the development of high-quality products or, as a pre-treatment, for increasing the yield of polyphenols to be recovered from fruit products waste.

1. Introduction

Over the last years, there has been a significant increase in the offer of minimally processed fruits in retail and food services. The main driver of this tendency is the increasing demand for convenience products suitable for the modern lifestyle, considering that they offer the advantage of saving time and effort. However, it is well-known that these products represent a technological challenge, since processes such as peeling and/or cutting, normally used for the manufacturing of fresh-cut fruits, bring about a faster physiological deterioration, biochemical changes and microbial spoilage, which altogether may result in degradation of the color, texture and flavor. As a consequence, different strategies have been developed to obtain products able to keep, during long-time storage, the freshness and quality of the original commodity. In this regard, one of the most successful approaches assayed so far has been the combination of different preservation technologies, acting synergistically, which is referred to as hurdle technology. Under this framework, high pressure processing (HPP) constitutes a promising non-thermal technology having proved highly successful for the preservation of minimally processed fruits, in combination with other processes (e.g. organic acids dipping, vacuum packing, and refrigerated storage) (Denoya, Vaudagna, & Polenta, 2015).

Since the quality and shelf-life of minimally processed fruit products is affected by several factors, it is important to develop these products under a holistic approach that includes a detailed selection of the type of cultivar, processing conditions, and storage atmosphere and temperature. Cultivar selection is probably the most important topic in these products, since genetic characteristics are closely linked to quality-related aspects such as flesh texture, skin color, and browning potential (Gorny, Hess-Pierce, &

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Kader, 1999; Putnik, BursaćKovačević, Penić, Fegeš, & Dragović-Uzelac, 2016).

The selection of varieties able to render products that adequately responds to the modern market requirements and consumer demands constitutes one of the key aspects in the development of fresh-cut fruits. In spite its relevance, studies focusing this aspect are rather scarce. In the case of peaches, the different varieties can be classified into three main categories: varieties for fresh consumption, varieties for canning, and multipurpose varieties. It was found in fruits such as melon, that the correct selection of the cultivar most suitable for preservation by HPP, had a significant impact over important quality aspects of the final product, such as vitamin C content, and color preservation, after HPP and during refrigerated storage (Wolbang, Fitos, & Treeby, 2008).

In a previous work carried out in our laboratory with peach pieces treated with pressures between 400 and 600 MPa, and holding times between 1 and 9 min, we found that 600 MPa attained the highest level of inactivation for two deterioration-related enzymes: polyphenoloxidase (PPO), the main enzyme that catalyzes browning, and alcohol dehydrogenase, an enzyme related to the induction of fermentation under anaerobic condition. In turn, this level of pressure caused no significant changes on the color parameters, and only a minor variation in the texture (Denoya et al., 2016). The incomplete inactivation attained in that study highlights the importance of assessing in depth whether the application of higher pressures would render higher levels of inactivation of the enzymes associated with deterioration. This could also offer the additional advantage of reducing the holding time, taking into account that the longer this process parameter, the more expensive would be the treatment.

According to the mentioned above, the aim of the present work was to evaluate the suitability of two peach cultivars for this kind of product, and to establish the main parameters (pressure level and holding time) leading to a better preservation of the product in terms of sensorial attributes (such as texture and color) and of health promoting compounds content (such as ascorbic acid and total phenols), while maximizing the inactivation of the deterioration-related enzymes.

2. Materials and methods

2.1. Plant material

Peaches (*Prunus Persica* (L.) Batch) from two different varieties: cv. Flavorcrest (used for fresh consumption and canning) and cv. Romea (only used for canning) were harvested from an experimental orchard in San Pedro, Buenos Aires, Argentina (Latitude 33°41_S, Longitude 59°41_W) and carefully selected according to their uniform size and ground color. Flavorcrest cultivar presented a mean value of 12 °Brix while Romea cultivar presented a mean value of 14 °Brix. The firmness in both varieties was in a range between 20 and 30 N. Peaches were stored in a cold chamber at 0 °C and 90–95% relative humidity for two weeks before processing.

2.2. Sample preparation

Prior to processing, the fruits from each variety were washed in running tap water. Cylinders (15 mm in length, 15 mm in diameter) of parenchyma tissue were cut using a stainless steel cork borer and knife. To obtain homogenous samples, the cylinders were taken from the middle zone of the mesocarp, parallel to the major axis of the fruit. Subsequently, the cylinders were dipped into tap water containing 20 ppm of HClO for 2 min. The cylinders were then drained and dipped into an aqueous solution containing 1 g/100 mL ascorbic acid (ACS, Biopack, Argentina) and 1 g/100 mL citric acid (USP, Anedra, Austria) for 2 min to prevent surface browning and to wash the remaining HClO. The cylinders were drained again, pooled, and vacuum-packed in Cryovac BB2800 bags (O_2 transmission rate: 6–14 cm^3/m^2/24 h at 23 °C, 1 atm, Sealed Air, Argentina) filled randomly with eight units each, using a double chamber vacuum packaging machine (Rapivac, Model Maximax 800, Argentina). For each variety, the samples were subjected to HPP treatments with different pressure levels and holding times, selected according to the experimental design (see below). HPP was performed in a high hydrostatic pressure system with a vessel of 2 L capacity (Stanted Fluid Power Ltd. High Pressure Iso-Lab System Model: FPG9400:922, Stansted, UK) and a maximum working pressure of 900 MPa. A mix of distilled water and propylene glycol (70/30, v/v) was used as the compression fluid. Pressure was increased at 5 MPa s^{-1}. The HPP treatments were carried out at an initial temperature of 21–24 °C and this parameter was increased by adiabatic heating. The maximum temperature of the compression fluid (at the end of compression stage) was 35 °C for 500 MPa, 38 °C for 600 MPa and 40 °C for 700 MPa and upon pressure release reduced to 20 °C.

2.3. Experimental design

A completely randomized factorial design (3 × 2) was applied for the experiments of each variety. The factors were: pressure level (500 MPa, 600 MPa, 700 MPa) and holding time (1 and 5 min). A total of 48 bags for each variety were prepared, eight for each treatment (each combination of pressure level and holding time). Eight cylinders from different bags were analyzed for each treatment, to carry out texture determinations. Three pooled samples were prepared from different cylinders to carry out biochemical determinations (PPO and ADH activity and contents of total phenols and vitamin C).

2.4. Analyses

2.4.1. Texture Profile Analysis (TPA)

Instrumental approaching for texture of fresh-cut peaches was performed by running a Texture Profile Analysis. Eight cylinders per treatment were compressed twice to 75% of their original height (1 s interval) simulating mastication. A Texture Analyzer model TA-XT plus (Stable Micro Systems LTD, Surrey, England) was used at room temperature and the following conditions were set according to the instrument manufacturer's recommendations: 3.0 mm/s pre-test speed, 0.8 mm/s test speed, 3.0 mm/s post-test speed and 25% strain. The trigger force was 0.049 N. A 35-mm diameter cylindrical probe (P/35) was used to assure that the surface area of the peach cylinder was completely covered by the probe. Force–distance–time data were recorded for two cycles.

2.4.2. Enzyme activities

2.4.2.1. Enzyme extraction. Enzymes were extracted according to the method described by Denoya, Nanni, Apóstolo, Vaudagna & Polenta (2016). Briefly, 7 g of peach cylinders were homogenized with 20 mL of 0.1 M phosphate buffer pH 7.3 containing 1 g/100 mL insoluble polyvinylpolypyrrolidone (PVPP, Sigma, USA) as a phenolic scavenger and 0.5 mM phenylmethylsulfonyl fluoride (PMSF, Sigma, Germany) as a protease inhibitor. Then, the homogenate was centrifuged at 10,000 × g for 15 min at 4 °C. The supernatant was used as the enzyme source in the following experiment.

2.4.2.2. PPO activity assay. The PPO activity assay was carried out according to the method described by Denoya, Nanni, Apóstolo, Vaudagna & Polenta (2016). PPO catecholase activity was

followed by measuring the initial rate of increase in absorbance at 400 nm due to the production of 4-*tert*-butyl-*o*-benzoquinone with a spectrophotometer (Gilford/CIBA-Corning Diagnostics, USA). The activity was expressed as units (U) per gram of fruit. One U represents the amount of enzyme necessary to change A_{400nm} in 0.01/min at 30 °C.

2.4.2.3. ADH activity assay. The ADH activity assay was carried out according to the method described by Denoya, Nanni, Apóstolo, Vaudagna & Polenta (2016). ADH activity was followed by measuring the initial rate of increase in absorbance at 340 nm due to NADH (β-Nicotinamide adenine dinucleotide, reduced) accumulation during ethanol oxidation with a spectrophotometer (Gilford/CIBA-Corning Diagnostics, USA). The activity was expressed as units per gram of fruit; one unit represents the amount of NADH produced in μM per min at 30 °C.

2.4.3. Total phenols

The total phenols extraction was carried out according to Pace, Cefola, Renna, & Attolico (2011) with some modifications and the determination was carried out according to Singleton, Orthofer, & Lamuela-Raventós (1999, pp. 152–178). Peach cylinder portions (2 g) were homogenized with 2 mL of aqueous methanol (90%, v/v). Afterwards, samples were vortexed for 2 min, sonicated for 10 min and centrifuged for 10,000 × g for 10 min at 4 °C. A volume of 0.5 mL of the supernatant from each sample was mixed with 2.5 mL of Folin-Ciocalteau reagent in a tube. The mix was vortexed and was allowed to react for 8 min. Then, 4 mL of a saturated Na_2CO_3 (aq.) solution (75 g/L) was added to each tube. After the solution had been standing for 2 h, the absorbance was read at 760 nm against a blank using a UV–Vis spectrophotometer (Lambda Bio 20, Perkin Elmer, USA). The content of total phenols was calculated on the basis of a calibration curve with gallic acid and was expressed as milligrams of gallic acid per 100 g of fresh fruit.

2.4.4. Ascorbic acid

2.4.4.1. Extraction. The ascorbic acid extraction was carried out according to the method described by Valente, Albuquerque, Sanches-Silva, & Costa (2011) with slight modifications. Two grams of fruit from each sample previously frozen at −80 °C were extracted with 20 mL of 10 g/100 mL percloric acid and 1 g/100 mL metaphosphoric acid (aq.) solution by using a Power Gen 1000 Homogenizer (Fisher Scientific, Germany). After that, samples were vortexed, sonicated for 10 min and centrifuged (10,000 × g at 4 °C) for 20 min. The supernatant was separated and filtered (0.2 μm filter pore) to be immediately analyzed.

2.4.4.2. Determination. The determination was carried out by reversed phase HPLC with a Shimadzu 10 A High Performance Liquid Chromatographer equipped with two LC-10Ai pumps and a SPD-10 AVP UV/Vis detector (Shimadzu, Japan). The column utilized was a C_{18} Hypersil GOLD (4.6 mm ID x 250 mm). The mobile phase consisted in a 0.01 M KPO_4H_2 solution (90:10 v/v water: methanol). Its pH was adjusted to 2.5 with phosphoric acid. The flow was 0.5 mL/min, the injection volume was 20 μl and the wavelength selected for the detection was 254 nm. The results were expressed as milligrams of ascorbic acid per 100 g of fresh fruit.

2.5. Statistical analysis

Differences were tested for significance by analysis of variance, which was performed using the General Linear Model procedure from SAS (Version 9.2 2002–2003 SAS Institute Inc. Cary, NC, USA). Duncan's test (significance level of 0.05) was performed for the data presented in tables.

3. Results

3.1. Texture Profile Analysis

Table 1 shows the values of hardness (maximum force attained during the first compression cycle, N) for the different combinations pressure level-holding time. Even though other TPA parameters were also determined, they are not shown since only hardness and chewiness showed significant differences among treatments. Chewiness, in turn, showed a tendency similar to hardness (data not shown), which is mainly due to the fact that it represents a secondary parameter, defined by the multiplication of three primary parameters: hardness, springiness and cohesiveness. By comparing these parameters one to another, the values of hardness were much higher than those of cohesiveness and springiness. In addition, the fact that the last parameters showed no significant differences among treatments evidences that chewiness variations were mainly due to changes in hardness.

As shown in Table 1, hardness was significantly affected by the pressure level, with 700 MPa causing the higher decrease (statistically significant at 5% level), while the holding times had no effect on this parameter.

3.2. PPO activity

In the case of PPO activity (Table 1), results evidenced the existence of pronounced and significant differences ($p < 0.05$) among treatments, which can be attributed to both the effect of variety and pressure exposure (HPP). Interestingly, the differences between varieties could be clearly detected even prior to the application of HPP. Thus, the PPO activity in untreated Flavorcrest peaches was more than two times higher in comparison to Romea (308.1 U/g vs 135.9 U/g, respectively). This fact highlights the relevant implication of an adequate selection of the raw material, which will eventually have an impact in the development of deterioration processes in the final product. No residual PPO activity was detected after the HPP treatment in Romea peaches subjected to either 600 and 700 MPa, while in Flavorcrest, a residual PPO activity could be still measured after the application of all the treatments assayed, with the lowest values (statistically significant, $p < 0.05$) being detected for the highest levels of pressure. In the 500 MPa treatment, the residual PPO activity was much higher in Flavorcrest than in Romea. In turn, the holding time had a significant effect on the enzymatic activity ($p < 0.05$) only for Flavorcrest peaches and for 600 MPa. All in all, the positive effect of HPP and the selection of a suitable variety as a raw material on the prevention of enzymatic browning was clearly evident.

3.3. ADH activity

In the case of ADH, the enzymatic activities in the untreated fruit from Flavorcrest and Romea were not significantly different, (0.159 U/g vs 0.121 U/g, respectively). After HPP application, the results showed that both factors (level of pressure and holding time) had a significant effect ($p < 0.05$) on the residual activity of the enzyme. In particular, for fruit subjected to HPP at 600 MPa, there was a significant effect of holding time, with treatments at 600 MPa for 5 min and at 700 MPa for 1 and 5 min being the most effective for the inactivation of ADH. Interestingly, as shown in Table 1, the inhibition pattern for both varieties was quite similar.

3.4. Ascorbic acid content

Table 2 shows the ascorbic acid content (mg/100 g) for the two varieties of peaches subjected to the different treatments assayed.

Table 1
Results for hardness (N), one of the parameters of Texture Profile Analysis, Polyphenoloxidase (PPO) and alcohol dehydrogenase (ADH) Activity (U/g) of fresh-cut cylinders from *Prunus persica* cv. Flavorcrest and cv. Romea and of the same product subjected to different high pressure processing (HPP) treatments.

Variety	Flavorcrest		Romea	
Level of pressure (MPa)	Holding Time (min)			
	1	5	1	5
Hardness (N)				
Fresh-cut fruit	27 ± 3		23 ± 3	
500	26 ± 3 aA	21 ± 3 aA	20 ± 3 aA	21 ± 4 aA
600	23 ± 2 aA	19 ± 1 aA	18 ± 4 aA	16 ± 3 aA
700	17 ± 3 bA	16 ± 2 bA	12 ± 2 bA	12 ± 2 bA
PPO Activity (U/g)				
Fresh-cut fruit	308 ± 2		136 ± 2	
500	160 ± 4 aA	156 ± 2 aA	49 ± 1 A	34 ± 1 A
600	120 ± 2 bA	104 ±2 bB	ND	ND
700	110 ± 2 cA	112 ± 2 bA	ND	ND
ADH Activity (U/g)				
Fresh-cut fruit	0.159 ± 0.001		0.121 ± 0.001	
500	0.068 ± 0.002 aA	0.050 ± 0.004 aA	0.098 ± 0.003 aA	0.074 ± 0.004 aA
600	0.039 ± 0.004 bA	0.013 ± 0.001 bB	0.034 ± 0.006 bA	0.021 ± 0.001 bB
700	0.010 ± 0.001 cA	0.001 ± 0.001 bA	0.010 ± 0.001 cA	0.012 ± 0.003 bA

For each variety and HPP treatment with different holding time, means for treatments with different level of pressure followed by the same lowercase letter were not significantly different according to Duncan's test (p = 0.05). For each variety and treatment with different level of pressure, means at different holding time followed by the same uppercase letter were not significantly different according to Duncan's test (p = 0.05). Data expressed as means ± Std. Error (Hardness n = 8, Enzymes activity n = 3). ND means = undetected.

In all cases, the ascorbic acid contents of Romea peaches were markedly higher than those of Flavorcrest. This difference was also observed in the fresh-cut peaches, before the application of the HPP treatments (53 mg/100 g vs 41 mg/100 g for Romea and Flavorcrest, respectively). Among the different HPP treatments, Romea peaches subjected to 500 MPa showed the lowest values of ascorbic acid ($p < 0.05$). A similar trend was found in the case of Flavorcrest, although the values among the different pressure levels were not significantly different for 5 min of holding time. Regarding the percentage of loss of ascorbic acid in comparison to the untreated fruit, Flavorcrest peaches treated at 500 MPa presented the highest decrease, attaining approximately 30%. Differently, with the other treatments the peaches lost only minor amounts of this compound (up to 95% of retention). It is highly remarkable the fact that in Romea peaches, no loss of ascorbic acid occurred as a consequence of the treatments (100% of retention compared to the fresh-cut fruit without the application of HPP). What is more, there was an increase of up to 24% in the ascorbic acid concentration in the peaches treated with the highest levels of pressure with respect to the untreated ones. On the other hand, the holding time had no significant effect on this parameter ($p < 0.05$), regardless of the variety and the pressure level.

Table 2
Results for ascorbic acid (mg/100 g) and total phenols (mg GA/100 g) contents of fresh-cut cylinders from *Prunus persica* cv. Flavorcrest and cv. Romea and of the same product subjected to different high pressure processing (HPP) treatments.

Variety	Flavorcrest		Romea	
Level of pressure (MPa)	Holding Time (min)			
	1	5	1	5
Ascorbic Acid (mg/100 g)				
Fresh-cut fruit	41 ± 2		53 ± 1	
500	30 ± 1 b A	29 ± 3 a A	55 ± 1 b A	54 ± 1 b A
600	39 ± 1 a A	35 ± 2 a A	66 ± 1 a A	65 ± 1 a A
700	39 ± 1 a A	37 ± 1 a B	65 ± 2 a A	65 ± 2 a A
Total Phenols (mg GA/100 g)				
Fresh-cut fruit	73 ± 3		80 ± 1	
500	69 ± 4 c A	79 ± 1 c A	107 ± 2 b A	103 ± 1 b A
600	81 ± 2 b B	91 ± 1 b A	120 ± 3 a A	128 ± 2 a A
700	90 ± 3 a A	99 ± 2 a A	130 ± 1 a A	127 ± 1 a A

For each variety and HPP treatment with different holding time, means for treatments with different level of pressure followed by the same lowercase letter were not significantly different according to Duncan's test (p = 0.05). For each variety and treatment with different level of pressure, means at different holding time followed by the same uppercase letter were not significantly different according to Duncan's test (p = 0.05). Data expressed as means ± Std. Error (n = 3).

3.5. Total phenols content

No significant difference was found in the total phenols content of the untreated fresh-cut fruit between the two varieties of peaches (73 mg Gallic Acid equivalents (GA)/100 g vs 80 mg GA/100 g for Flavorcrest and Romea, respectively). Table 2 shows the total phenols content of the two varieties of peaches, subjected to the different treatments assayed. It is important to highlight that the values obtained in the Romea peaches were significantly higher than those obtained in Flavorcrest ones, independently of the treatment. In both varieties, peaches subjected to 500 MPa presented the lowest ($p < 0.05$) values of total phenols among all treatments. In the case of Flavorcrest, peach pieces subjected to 700 MPa had significantly higher content of total phenols ($p < 0.05$) than those treated at 600 MPa. Similarly to the case of ascorbic acid in Romea, the holding time had no significant effect ($p < 0.05$), regardless of the pressure level, while in Flavorcrest, there was an increase ($p < 0.05$) in total phenol content only for the 600 MPa treatments applied for 1 and 5 min.

In terms of percentage, results showed that in the case of Romea, the application of all HPP treatments induced an increase in the concentration of total phenols by comparison to the fruit without the application of HPP, this raise being of about 30% for the treatments of 500 MPa, and between 50 and 60% for higher levels of pressure. In the case of Flavorcrest, the total phenol content remained invariable after the exposure to 500 MPa, while an increase of 11% and 23% was observed for the treatments at 600 MPa for 1 min and at 700 MPa for 1 min, respectively, and 25% and 36% for the treatments at 600 MPa for 5 min and at 700 MPa for 5 min, respectively.

4. Discussion

4.1. Texture profile analysis (TPA)

Maintaining the texture of HPP-treated fruits represents one of the key aspects to be taken into account for the optimization of the

HPP-process. In a previous study with HPP-treated peaches cylinders (Denoya et al., 2016), we found that the level of pressure (in a range between 400 and 600 MPa) had no significant effect in the textural parameters. With this in mind, the range was extended in the present work up to 700 MPa. However, the results showed that this level of pressure provoked a significant decrease ($p < 0.05$) of the hardness, compared to the other treatments. A similar effect was reported by Kingsly, Balasubramaniam, & Rastogi (2009), who observed that 700 MPa caused a significant decrease in pineapple cylinders's hardness when compared to 50, 100 and 300 MPa. These results clearly suggest the inconvenience for this type of product to apply pressure levels higher than 600 MPa, since the textural characteristics of the fresh fruits would be negatively affected. Interestingly, the observed effect was comparable between both varieties assayed, which evidenced the similarity of the mechanical properties in peaches.

4.2. PPO activity

It is well-known that enzymatic browning represents one of the main alterations related to minimally processed fruits. Therefore, PPO activity could give relevant information to define the aptitude of a specific variety for HPP, by predicting the browning potential of the cultivar. In fact, among the parameters measured, this one presented the largest differences between the two varieties. These differences were observed even before the application of HPP, which evidenced that they are inherent to the raw material and are related to the genetic differences between varieties. Such varietal differences were also informed in other fruits such as strawberries, which also showed important differences in the residual PPO activity after HPP application (Terefe et al., 2013).

In the present study, Flavorcrest showed significant differences ($p < 0.05$) in PPO activity among the different holding times after the exposure to 600 MPa, with the combination 600 MPa-5 min being more effective to inactivate PPO than 600 MPa-1 min. In a previous work with peach puree, Khalil, Al-Zubaidy & Abdulaziz (2011) observed that at the same pressure level (600 MPa), an increase in the holding time from 1 to 3 min had a significant effect in the reduction of the browning index (a parameter closely related with PPO activity). This fact makes evident that the selection of the adequate treatment intensity, as defined by the combination of pressure level and holding time, would be a key factor for the effectiveness of a treatment, in an important aspect such as limiting the browning potential of a product.

4.3. ADH activity

Another factor that greatly affects fresh-cut fruits quality is the induction of the fermentative metabolism. This alteration can be even magnified in fruits subjected to HPP, since the product should be vacuum packed in films of low oxygen permeability, before the treatment application. Therefore, the monitoring of marker enzymes related to alteration such as ADH, whose inhibition leads to the prevention of this metabolic change, represents a key aspect to avoid the fermentation in HPP-treated fruits. According to our experience, if this aspect is properly controlled, the vacuum packing technology can be very helpful, by acting as an additional preservation strategy (Denoya, Vaudagna, & Polenta, 2015). In the present study, results were promising, considering the inhibitory effect exerted by the HPP treatments on the ADH activity, which was significantly decreased in both varieties, and positively influenced by the level of pressure and by the holding time. This evidenced the sensitivity of the enzyme to the HPP-treatments assayed. In this regards, the most effective combinations were 600 MPa-5 min and 700 MPa for 1 and 5 min, which rendered the lowest levels of enzyme activity. From the biochemical point of view, it can be hypothesized that the sensitivity to these treatments is related to the multimeric nature of this enzyme, taken into account the important effect that HPP generally has over enzymes conformed by more than one monomer (Dallet & Legoy, 1996).

4.4. Ascorbic acid content

In the present study, results showed that the immersion of peach cylinders in the 1 g/100 mL ascorbic acid solution greatly increases the content of this compound in fresh cut peaches, which normal range in fresh peaches is in between 3 and 12 mg/100 g (Tavarini, Degl'Innocenti, Remorini, Massai, & Guidi, 2008). This fact evidences the positive effect of the dipping step, since in addition to preventing the fruit browning, it also substantially contributes to improving the nutritional quality of the product (Lee & Kader, 2000).

In the case of Romea, the ascorbic acid content was completely maintained, even showing an increase of 24% in the case of peach cylinders treated at 600 and 700 MPa, compared to the concentration prior to the pressurization. Similar levels of increase were reported by Kaushik, Kaur, Rao, and Mishra (2014) in mango puree subjected to 600 MPa for 1 s, which had an ascorbic acid content 29% higher than the untreated fruit. These authors highlight the improvement in the extractability of the compound induced by the HPP, which would be linked to the increase in the permeabilization of the compressed cells, and the concomitant release of the cytosolic content into the extracellular space. The effect of the HPP on the micro- and ultra-structure was also observed in fresh cut peaches in our previous work (Denoya et al., 2016). It is important to emphasize the positive effect of this technology in enhancing the extractability of bioactive compounds from fruits and vegetables, thus rendering a product with functional characteristics.

It should be taken into account that the interaction between factors such as variety and pressure, can greatly affect the organolpeptic and nutritional quality of products such as minimally processed peaches. In this regard, the HPP provoked opposite effects on the ascorbic acid content between the two peach variety included in the study. Thus, the content of this compound was decreased in Flavorcrest nearly 30% in the case of the lowest level of pressure (500 MPa), while for the more intense treatments the decrease was around 5%.

We suggest that the final ascorbic acid content after a treatment exposure is the result of a balance between the degradation of the compound catalyzed by the presence of baro-resistant oxidative enzymes during or after HPP and the increase in the extractability. The differences between varieties could be related to different resistance of the oxidative enzymes to the HPP.

Other studies has previously highlighted the necessity of meticulously assessing the varietal aptitude of the raw material for the development of high-quality HPP-treated products (Oms-Oliu, Odriozola-Serrano, Soliva-Fortuny, Elez-Martínez, & Martín-Belloso, 2012), considering the important effect of different factors on main variables such as the ascorbic acid content.

4.5. Total phenols content

As mentioned above, we observed in a previous study that HPP causes an important compression in the fruit microstructure, in addition to other alterations such as disruptions of the cell walls and membranes, resulting in an increase of the permeability (Denoya et al., 2016). These phenomena can modify the distribution of phenolic compounds and increase the rate of mass transfer from the internal part of the tissue into the interstitial space. Interestingly, from a technological point of view, these effects can enhance

the penetration of solvents into the cells, favoring the extraction of phenols and other soluble compounds (Vega-Gálvez et al., 2014).

The present work evidences that for both varieties assayed (Flavorcrest and Romea), the total phenols content was increased after the application of HPP treatments, with higher difference with regards to the untreated peaches for the higher levels of pressure applied. The percentages of increase were comparable, or even higher, than those reported in previous studies with fruits. Among then, Barba, Esteve, & Frigola (2013) found that the total phenols content of blueberry juice increased by 13 and 27% after the exposure to 200 MPa for 5 and 15 min respectively, and by 24% after the application of a 400 MPa treatment for 15 min. These authors emphasize that, similarly to other simple structures, the phenolic compounds are quite resistant to the HPP treatments. In turn, Kaushik et al. (2014) reported that the exposure of mango puree to pressures between 100 and 600 MPa for 1 s increased the extraction of phenolic compounds by 9 and a 19% respectively. As mentioned above, the increase in the total phenols content can be linked to the higher extractability induced by the HPP in other compounds such as ascorbic acid (Keenan et al., 2010), which can be considered as an important additional advantage of this technology.

5. Conclusions

The present study is a contribution towards the establishment of process parameters and raw material selection for the successful application of HPP in fruits. In this regards, it was proved that pressure levels up to 600 MPa can be applied to minimally processed peaches with no negative effect on the texture, in either of the two varieties studied. On the contrary, the exposure to 700 MPa provoked a significant decrease in the hardness of the product. Marked differences were found between the two varieties in biochemical variables such as PPO activity, ascorbic acid and total phenols contents, which highlights the relevance of the adequate selection of the raw material for the development of a high quality HPP-treated product. In this sense, varieties with lower PPO activity would be more suitable for HPP, since they are expectedly less prone to develop enzymatic browning. This was the case of Romea, for which a 600 MPa treatment was able to totally inhibit the PPO activity, with the additional advantage of rendering a product with high concentrations of ascorbic acid and total phenols, compared to Flavorcrest.

Regarding the effect of the treatment intensity, it was shown that the exposure to higher levels of pressure (600 and 700 MPa) improved the characteristics of the product, leading to a more efficient inactivation of the enzymes, and a better extractability of ascorbic acid and phenolic compounds. However, pressure levels higher than 600 MPa (700 MPa in the present study) caused negative effect on the texture of the final product, by inducing a significant decrease in the hardness, which would prevent the recommendation of this condition.

The holding time with the best performance was 5 min, which caused the highest level of inactivation of the enzymes analyzed, and rendered the highest concentration of phenols, no significant changes in ascorbic acid, and with only minor changes in the textural parameters.

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References

Barba, F. J., Esteve, M. J., & Frigola, A. (2013). Physicochemical and nutritional characteristics of blueberry juice after high pressure processing. *Food Research International, 50*(2), 545–549. http://dx.doi.org/10.1016/j.foodres.2011.02.038.

Dallet, S., & Legoy, M.-D. (1996). Hydrostatic pressure induces conformational and catalytic changes on two alcohol dehydrogenases but no oligomeric dissociation. *Biochimica et Biophysica Acta (BBA) - Protein Structure and Molecular Enzymology, 1294*(1), 15–24. http://dx.doi.org/10.1016/0167-4838(95)00250-2.

Denoya, G. I., Nanni, M. S., Apóstolo, N. M., Vaudagna, S. R., & Polenta, G. A. (2016). Biochemical and microstructural assessment of minimally processed peaches subjected to high-pressure processing: Implications on the freshness condition. *Innovative Food Science & Emerging Technologies, 36*, 212–220.

Denoya, G. I., Polenta, G. A., Apóstolo, N. M., Budde, C. O., Sancho, A. M., & Vaudagna, S. R. (2016). Optimization of high hydrostatic pressure processing for the preservation of minimally processed peach pieces. *Innovative Food Science & Emerging Technologies, 33*, 84–93. http://dx.doi.org/10.1016/j.ifset.2015.11.014.

Denoya, G. I., Vaudagna, S. R., & Polenta, G. (2015). Effect of high pressure processing and vacuum packaging on the preservation of fresh-cut peaches. *LWT - Food Science and Technology, 62*(1, Part 2), 801–806. http://dx.doi.org/10.1016/j.lwt.2014.09.036.

Gorny, J. R., Hess-Pierce, B., & Kader, A. A. (1999). Quality changes in fresh-cut peach and nectarine slices as affected by cultivar, storage atmosphere and chemical treatments. *Journal of Food Science, 64*(3), 429–432. http://dx.doi.org/10.1111/j.1365-2621.1999.tb15057.x.

Kaushik, N., Kaur, B. P., Rao, P. S., & Mishra, H. N. (2014). Effect of high pressure processing on color, biochemical and microbiological characteristics of mango pulp (Mangifera indica cv. Amrapali). *Innovative Food Science & Emerging Technologies, 22*, 40–50. http://dx.doi.org/10.1016/j.ifset.2013.12.011.

Keenan, D. F., Brunton, N. P., Gormley, T. R., Butler, F., Tiwari, B. K., & Patras, A. (2010). Effect of thermal and high hydrostatic pressure processing on antioxidant activity and colour of fruit smoothies. *Innovative Food Science & Emerging Technologies, 11*(4), 551–556. http://dx.doi.org/10.1016/j.ifset.2010.07.003.

Khalil, T. A. K., Al-Zubaidy, M. I., & Abdulaziz, O. F. (2011). Browning reaction kinetics of high hydrostatic pressure-treated peach puree during storage. *International Journal of Food Engineering, 7*(4), 1–12. article 2.

Kingsly, A. R. P., Balasubramaniam, V. M., & Rastogi, N. K. (2009). Effect of high-pressure processing on texture and drying behavior of pineapple. *Journal of Food Process Engineering, 32*(3), 369–381. http://dx.doi.org/10.1111/j.1745-4530.2007.00221.x.

Lee, S. K., & Kader, A. A. (2000). Preharvest and postharvest factors influencing vitamin C content of horticultural crops. *Postharvest Biology and Technology, 20*(3), 207–220. http://dx.doi.org/10.1016/S0925-5214(00)00133-2.

Oms-Oliu, G., Odriozola-Serrano, I., Soliva-Fortuny, R., Elez-Martínez, P., & Martín-Belloso, O. (2012). Stability of health-related compounds in plant foods through the application of non thermal processes. *Trends in Food Science & Technology, 23*(2), 111–123. http://dx.doi.org/10.1016/j.tifs.2011.10.004.

Pace, B., Cefola, M., Renna, F., & Attolico, G. (2011). Relationship between visual appearance and browning as evaluated by image analysis and chemical traits in fresh-cut nectarines. *Postharvest Biology and Technology, 61*(2–3), 178–183. http://dx.doi.org/10.1016/j.postharvbio.2011.03.005.

Putnik, P., Bursać Kovačević, D., Penić, M., Fegeš, M., & Dragović-Uzelac, V. (2016). Influence of cultivar, anti-browning solutions,packaging gasses, and advanced technology on browning in fresh-cut apples during storage. *Journal of Food Process Engineering*. http://dx.doi.org/10.1111/jfpe.12400.

Singleton, V. L., Orthofer, R., & Lamuela-Raventós, R. M. (1999). *[14] Analysis of total phenols and other oxidation substrates and antioxidants by means of folin-ciocalteu reagent Methods in Enzymology* (Vol. 299). Academic Press.

Tavarini, S., Degl'Innocenti, E., Remorini, D., Massai, R., & Guidi, L. (2008). Preliminary characterisation of peach cultivars for their antioxidant capacity. *International journal of food science & technology, 43*(5), 810–815.

Terefe, N. S., Kleintschek, T., Gamage, T., Fanning, K. J., Netzel, G., Versteeg, C., et al. (2013). Comparative effects of thermal and high pressure processing on phenolic phytochemicals in different strawberry cultivars. *Innovative Food Science & Emerging Technologies, 19*, 57–65. http://dx.doi.org/10.1016/j.ifset.2013.05.003.

Valente, A., Albuquerque, T. G., Sanches-Silva, A., & Costa, H. S. (2011). Ascorbic acid content in exotic fruits: A contribution to produce quality data for food composition databases. *Food Research International, 44*(7), 2237–2242. http://dx.doi.org/10.1016/j.foodres.2011.02.012.

Vega-Gálvez, A., López, J., Torres-Ossandón, M. J., Galotto, M. J., Puente-Díaz, L., Quispe-Fuentes, I., et al. (2014). High hydrostatic pressure effect on chemical composition, color, phenolic acids and antioxidant capacity of Cape gooseberry pulp (Physalis peruviana L.). *LWT - Food Science and Technology, 58*(2), 519–526. http://dx.doi.org/10.1016/j.lwt.2014.04.010.

Wolbang, C. M., Fitos, J. L., & Treeby, M. T. (2008). The effect of high pressure processing on nutritional value and quality attributes of Cucumis melo L. *Innovative Food Science & Emerging Technologies, 9*(2), 196–200. http://dx.doi.org/10.1016/j.ifset.2007.08.001.