Effects of conventional and ultrasound blanching on enzyme inactivation and carbohydrate content of carrots

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Abstract

There is a growing interest in the use of ultrasound (US) as an alternative to conventional processes. Although US have previously been applied as a pretreatment of fruits and vegetables, no investigation has been done on the usefulness of US for carrot blanching, paying special attention to its effect on enzyme inactivation and leaching losses. In the present paper, the influence of US (in bath and with probe) on peroxidase (POD) and pectinmethylesterase (PME) inactivation and on the loss of total soluble solids and carbohydrates by leaching has been evaluated. Results of this preliminary study have also been compared with those obtained after conventional (hot water and steam) blanching of carrots. The highest enzyme inactivation was obtained with the conventional treatments performed at high temperatures and with the US-probe treatments with heat generation. Carrots blanched by US-probe for 10 min at a temperature up to 60ºC, showed similar characteristics than those conventionally treated at 60ºC for 40 min. Although the efficiency of US was limited for total inactivation of POD and PME activity, this treatment resulted to be advantageous in terms of time for blanching at mild temperatures. US-probe treatments could also be considered as an advantageous alternative to low temperature-long time conventional treatments for those applications in which partial inactivation of PME is required for better preservation of carrot structure.

Keywords: Carrot, blanching, ultrasound, peroxidase, pectinmethylesterase, carbohydrates.

Introduction

Carrot (*Daucus carota* L.) is considered one of the most important vegetables due to its pleasant flavour, nutritive value and great health benefits related to its antioxidant, anticancer, antianemic, healing and sedative properties [1, 2]. Carrot is constituted by, approximately, 90% of water and 5% of carbohydrates; vitamins and minerals, among other constituents, are also present at lower concentrations [3]. Although carrots are widely consumed as fresh vegetables, due to their perishable nature, they are also subjected to different processes such as freezing, canning or dehydration to extend their shelf life for distribution and storage. Prior to these processes, carrots are usually blanched in hot water or steam for air removal, stabilization of colour, hydrolysis and solubilisation of protopectin and inactivation of microorganisms and enzymes [4-6].

Enzymes such as peroxidase (EC 1.11.1.7, POD) and pectinmethylesterase (EC 3.1.1.11, PME) are of considerable importance since they can be involved in different degenerative modifications of vegetables [7]. Particularly, POD catalyses a great number of oxidation-reduction reactions and it is considered among the most heat-stable enzymes in plants. POD is widely used as an index of blanching since if this enzyme is inactivated, it is quite unlikely that other enzymes are active. Therefore, it has been accepted as a general rule in the food industry that if there is no activity of peroxidase, no activity of other heat-resistant enzymes such as catalase, should be detected. However, complete inactivation of peroxidase has been shown not to be necessary for quality preservation in frozen vegetables [8]. In relation to PME, this enzyme has an important role in textural changes of unblanched vegetables since it catalyses the de-esterification of pectin to pectic acid which facilitates the link of calcium and magnesium, increasing the firmness of the cellular wall [9]. In some cases, a certain residual PME is preferred since, after drying, the texture of rehydrated product can be improved [10, 11]; this is possible by blanching at low temperature and long-time (LTLT). Despite the beneficial effects of blanching depend on the degree of thermal treatment applied, the quality and bioactivity of the final product can be negatively affected due to the destruction of nutrients relatively unstable to heat, the loss of water-soluble components by leaching and the changes in texture with this sample pretreatment [12, 13].

On the other hand, as a result of the increased consumer's awareness of the relationship between diet and health, the food industry is greatly interested in the search for mild processing technologies which give rise to final products with improved characteristics as compared to those obtained by conventional thermal treatments, being high-intensity ultrasound (US) one of the emerging processes

whose applications in the food industry have been recently reviewed [14]. In this respect, there are some studies on the use of ultrasound as a pre-treatment before conventional drying and as a medium to assist osmotic dehydration of vegetable and fruits [15-19]. Most of these works have been carried out in ultrasonic baths at mild temperatures or have been mainly focused on the kinetic of moisture loss during drying; US showing a noticeable reduction in the overall drying time together with a variable loss of total sugars. In the case of carrots, hardly any research has been carried out on the potential of US as an alternative to conventional blanching with hot water or steam. Rawson et al. [19] reported higher retention of carotenoids in hot air and freeze dried carrots previously subjected to US than in samples blanched with hot water at 80°C for 3 min. However, to the best of our knowledge, no previous work has been done on the effect of ultrasound on important enzymes related to carrot blanching. Therefore, this paper has been devoted: (i) to study the influence of US pre-treatments, with probe and in bath, on the inactivation of POD and PME, and (ii) to determine the changes in total soluble solids and major and minor carbohydrates of US-processed carrots. US pretreatment results have been compared with those obtained in conventional heat blanching processes (steam and hot water 60-95 °C).

Materials and methods

Sample preparation

A big batch of fresh carrots (*Daucus carota* L. var. Nantesa) was purchased from a local market in Madrid (Spain) and was stored at 4 °C for less than a week until processing. Carrots were properly washed in tap water to remove external impurities. Then, samples were cut in slices of 24 mm in diameter and 4 mm thickness and as minced carrots (1-2 mm).

Processing

Table 1 summarizes all blanching processes (conventional and by ultrasound) carried out.

Ultrasound treatments

For US treatments, samples of 40 g were added to the 250-mL Erlenmeyer flasks filled with 200 mL of distilled water. Two sets of experiments were carried out: (i) in bath and (ii) with an ultrasonic probe.

(i) Erlenmeyers containing the carrot samples were placed in a temperature-controlled ultrasound bath (30-70 ± 1 ºC) (SONICA SWEEP SYSTEM EP 2200, SOLTEC, Italy), operating at 45 kHz, and carrot samples were US-treated at 40 and 60 °C for 30 and 60 min (USB 40-30; USB 40-60; USB 60-30; USB 60-60). The soak water was preheated at the selected temperature.

(ii) In the case of the assays with probe, Erlenmeyers with carrot samples were sonicated in an ultrasonic system (450 Digital Sonifier, Branson Ultrasonics Coorporation, Danbury, CT, USA). This sonicator is equipped with a temperature sensor (error ± 0.1 ºC) and a tip of 13 mm diameter directly attached to a disruptor horn (20 kHz, 400 W full power) and immersed 2 cm in depth with respect to the liquid surface (**Figure 1**). Experiments were carried out at low temperature (≤ 35 °C) for 15 and 60 min (USP 35-15, USP 35-60) by immersing the samples in an ice-water bath. Additional assays were done with generation of heat: temperatures up to 60 and 70 °C being achieved after 10 min (USP 60-10) and 15 min (USP 70-15) of sonication, respectively. In this case, the ice-water bath was removed.

The ultrasound density, calculated according to Jambrak et al. [16], was 0.04 and 0.26 Wcm^{-3}, respectively, for bath and probe experiments.

Conventional blanching treatments

Using the same carrot - distilled water ratio as above mentioned, carrot samples were subjected to blanching with boiling water for 1 min (CB-1), with water at 95 °C for 5 min (C95-5) and at 60 °C for 40 min (C60-40) using a magnetic stirrer (200 rpm) with temperature control (IKA RCT Basic Labortechnik, Staufen, Germany). For CS-2 treatments (steam blanching), an autoclave (CERTOCLAV CV-EL GS, Austria) was used.

All assays (ultrasound and conventional) were performed in duplicate. After treatments, samples were cooled in an ice-water bath and conveniently drained and dried with absorbent paper to remove the excess of distilled water.

Sample characterization

The dry matter (DM) content of carrots was gravimetrically determined by drying the samples in a conventional oven at 102 ºC until constant weight, according to the AOAC method (950.01, 1990) [20]. The same method was used to determine the leaching loss during blanching. The percentage of leached solids was referred with respect to the initial weight of raw carrot (%).

The pH of blanching water was determined using a pH meter (Mettler-Toledo GMBH, Schwenzenbach, Switzerland).

Enzymatic determinations

Determination of peroxidase (POD) activity

The POD activity was determined as described by Shivhare et al. [2] with slight modifications. Blanched carrots (2 g) were crushed in a domestic chopper (BRAUN, Germany) and, after addition of 5 mL of phosphate buffer solution (pH 6.5; 0.1 M), samples were homogenized for 30 s at 18000 rpm and 4 ºC using an Ultra-Turrax T-25 homogenizer (IKA Labortechnik, Janke & Kunkel, Saufen, Germany). The slurries were subsequently filtered through a medium-grade paper filter (Whatman no. 40) and the filtrates were centrifuged at 5000 ×*g* (Eppendorf, F-45-12-11, Hamburg, Germany) for 20 min. The POD substrate solution was daily prepared by mixing phosphate buffer solution (pH 6.5; 0.1 M), guaiacol (0.1% v/v) and hydrogen peroxide (0.1% v/v). The supernatants (60 µL) were added to 870 µL of enzymatic substrate solution. Residual POD activity was measured at 470 nm and 25 ºC in a spectrophotometer (Power Wave XS Microplate, BIO-TEK) using the KC Junior Data Reduction software. The enzyme activity was determined from the slopes of linear progress curves generated on the recorder, and the slopes of raw samples were considered as indicatives of 100% of residual activity. The lower the value of the slopes calculated for blanched samples, the higher inactivation of POD in these samples. All determinations were carried out in duplicate.

Determination of pectinmethylesterase (PME) activity

The PME activity was determined in blanched carrots as described by Lemmens et al. [11]. Tris(hydroxymethyl-aminomethane) hydrochloride buffer (0.2 M; pH 8) containing 1 M NaCl was added

to carrots (ratio buffer:carrots, 1.3-1). The samples were stirred for 2 h at 750 rpm and 22 ºC using a Thermomixer (Eppendorf, Germany). The supernatants were recovered after filtration (Whatman no. 40) and then used to measure the residual PME activity by a titrimetric method (pH 7 and 22 ºC). The enzymatic substrate (0.35% apple pectin solution, containing 0.125 M NaCl) was demethoxylated by the residual enzyme, and the released carboxyl groups were titrated with 0.01 M NaOH. The residual PME activity was expressed as percentage respect to the raw sample, which was considered with 100% activity. All extracts were made and titrated in duplicate.

Carbohydrate determination by GC

Carrot samples were freeze-dried and grinded to powders with a laboratory mill (Janke and Kunkel IKA A-10, Labortechnik, Staufen, Germany) and soluble sugars were extracted according to the method reported by Soria et al. [21] with slight modifications. Grinded carrots (30 mg) were weighted in a polyethylene tube and extracted with 2 mL of Milli-Q water under stirring at room temperature for 20 min. Then, 8 mL of absolute ethanol were added followed by 0.2 mL of an ethanolic solution 10 mg mL^{-1} of phenyl-β-D-glucoside (Sigma Chemical Co., St. Louis, MO, USA) used as internal standard. After stirring for 10 min, samples were centrifuged at 10 °C and 9600 ×*g* for 10 min and the supernatant was collected. The precipitate was subjected to a second extraction with 10 mL of 80% ethanol under the same conditions to obtain recovery values close to 100%. Finally, 2 mL of supernatant was evaporated under vacuum at 40 °C. The extracts were prepared in duplicate.

The analysis was performed by GC as described by Soria et al. [21] with a gas chromatograph (Agilent Technologies 7890A) equipped with a flame ionization detector (FID) and using nitrogen as carrier gas at a flow rate of 1 mL min^{-1}. The trimethylsilyl oxime (TMSO) derivatives, prepared as described by Montilla et al. [22], were separated using a HP-5MS capillary column (5% phenyl methylsilicone, 30 m x 0.25 mm i.d. x 0.25 μm film thickness; J&W Scientific, Folsom, CA, USA). The oven temperature was held at 200 ºC for 11 min, then increased to 270 ºC at a heating rate of 15 °C min^{-1} and to 300 ºC at 3 ºC min^{-1} and finally raised to 315 ºC at 15 ºC min^{-1}, remaining at this temperature for 3 min. Injector and detector temperatures were 280 ºC and 315 °C, respectively. Injection was carried out in split mode (1:40).

Data acquisition and integration was done using Agilent ChemStation Rev. B.03.01 software (Wilmington, DE, USA). Identification of TMSO derivatives of carbohydrates was carried out by comparing the experimental retention indices with those of standards.

Quantitative data (mg g^{-1} DM) were calculated from FID peak areas. Standard solutions of fructose, glucose, sucrose, *scyllo*- and *myo*-inositol (all of them from Sigma Chemical Co.) over the expected concentration range in carrot extracts were prepared to calculate the response factor relative to the internal standard.

Soluble sugar content of blanching water (1 mL) was analysed using the same method, after addition of an ethanolic solution 0.5 mg mL^{-1} of phenyl-β-D-glucoside (0.4 mL) as internal standard. Samples were prepared in duplicate.

Statistical analyses

Data were subjected to one-way analysis of variance (Fisher's least significant difference (LSD) procedure) by applying the Statgraphic 4.0 software (Statistical Graphics Corp., Rockville, MD, USA) for Windows. The significance of differences was defined as $P < 0.05$.

Results and discussion

Effects of US and conventional blanching on enzyme inactivation

Table 2 lists the results corresponding to the enzymatic (POD and PME) activity of carrot samples subjected to the different blanching treatments under study. Considering POD activity, high temperature short time conventional blanching treatments (CB-1 and C95-5) gave rise to the total inactivation of this enzyme, in agreement with Kidmose and Martens [23] and with Shivhare et al. [2] that inactivated POD after 7 and 4 min at 80 and 90 °C, respectively. These authors also indicated that inactivation time of catalase and POD during steam blanching was consistently higher than in hot water. Similarly, in the present paper, some residual POD activity was detected in steam blanched carrots.

A certain effect of sample geometry was detected in samples subjected to mild conventional blanching treatments (CS-2, C60-40), with the highest inactivation of POD in minced as compared to

sliced carrots (Table 2). The highest residual activity (40.9%) was observed for sliced carrots blanched at 60 °C for 40 min. Lemmens et al. [11] reported residual POD activities of 70% after blanching treatments carried out under the same conditions, but with samples of 10 mm thickness.

In general, in the US blanching study, the reduction of POD activity was more evident for assays carried out with probe as compared to those with US bath, probably due to the higher acoustic density in the former experiments (0.26 Wcm^{-3} *vs.* 0.04 Wcm^{-3}). No inactivation of POD was detected in carrot samples US treated in bath at 40 °C (USB 40-30 and USB 40-60), while a significant inactivation of POD was observed at 60 °C, being this effect particularly noticeable after 60 min treatment of carrot slices. This could be due to the fact that, in minced carrots, the formation of sample aggregates, confirmed by visual inspection, might avoid the transfer of thermal and acoustic energy and, therefore, give rise to less cavitation phenomenon.

In carrot slices blanched in the ultrasonic bath, a higher inactivation of POD for treatment USB 60-30 and USB 60-60 (25.5 and 11.9% of residual activity, respectively) can be observed, as compared with the results obtained for the conventional blanching C60-40 (40.9%), indicating the usefulness of the combined effect of temperature and ultrasound for enzyme inactivation.

A noticeable reduction of POD activity with time was observed during US treatments with probe at temperatures lower than 35 °C; values of residual activity close to 60% being reached after 60 min, irrespective of carrot geometry. However, to obtain higher inactivation (17.4 and 6.7% residual POD activity in sliced and minced carrots, respectively), the application of US with heat generation was necessary; pretreatment USP 70-15 providing the highest enzyme inactivation. In addition, similar results of POD inactivation were obtained for carrots processed by either US (USP 60-10) or by conventional mild temperature treatments (C60-40).

Although it is difficult to exactly determine the effect of sample geometry on enzyme inactivation, the larger specific area would be the main factor to explain the higher inactivation of minced carrots after treatments carried out at high temperature (conventional and US with probe at 60 and 70°C). On the contrary, this factor seems not to be as significant in US blanching treatments carried out in bath, probably due to the previously mentioned formation of aggregates taking place in minced carrots.

With respect to US probe experiments, the combined effect of ultrasonic waves and heat treatment on enzyme inactivation appears to be more effective than US on its own. De Gennaro et al. [24], in a kinetic study carried out in solution on the inactivation of peroxidase type VI from horseradish,

found a considerable reduction in the *D* value when US were applied at 80 °C. According to Cruz et al. [25], who studied the peroxidase inactivation kinetics in watercress by thermosonication, the reduction of specific activity could be related to the conformation changes in the tertiary structure of the enzyme, and in the three-dimensional structure of the active site affecting the enzyme-substrate interaction.

Total inactivation of PME (Table 2) was achieved after conventional treatments CS-2, CB-1 and C95-5, whereas heating at 60 °C for 40 min (C60-40) preserved approximately 60% of the enzymatic activity. Similarly, Lemmens et al. [11] found 80% of PME residual activity at 60 °C and total enzyme inactivation at 90 °C during the blanching of carrots by microwave, ohmic and conventional heating. Comparing PME results with those of POD shown above, the lower stability of PME at high temperatures was confirmed [26-28]. However, in the case of LTLT treatments (C60-40), the presence of two isoenzymes of PME (bound and free) with different susceptibility to heat, could explain its higher residual activity as compared to POD [27].

During the US bath assays, no inactivation of PME was detected in USB 40-30 treated carrot samples and 60 min of treatment or higher temperature (60 °C) were needed to achieve a significant reduction of the activity of this enzyme. The application of the experimental setting of Figure 1 (with and without heat generation) did not produce either an important deactivation of PME. Thus, after US treatments, the values of enzymatic residual activity were always within the range 50-80%, and no conclusions derived from the sample geometry and/or processing temperature could be obtained. An additional advantage of US probe is to obtain a higher POD inactivation that with US bath while remain a high activity of PME that can contribute to the textural stability of samples.

Variable results have been reported on the inactivation of PME in tomato juice [29-31]. In all these cases, the application of US resulted in the reduction of PME activity dependent on the media in which the enzyme was suspended and on the ultrasound processing conditions. In addition, previous papers have also shown surprising results during the inactivation of PME by thermal treatment. Thus, in potato, Abu-Ghannam and Crowley [32] found 60% of residual activity after treatments at 65-90°C for 5 min and 0% at 80°C for 10 min, whereas in samples treated at 65°C for 15 min a 85% of residual activity was detected, probably due to some reactivation effect.

All these results underline the difficulty to identify the mechanism responsible for enzyme deactivation during sonication. Inactivation of enzymes by US is mainly attributed to a mixture of mechanical and chemical effects of cavitation, which are the formation, growth and implosion of bubbles

caused by US [29]. The sonochemically generated radicals can oxidise the residues of amino acids such as tryptophan, tyrosine, hystidine and cysteine that are involved in the catalytic activity and stability of several enzymes. Free radicals have been reported to participate in the ultrasonically-induced inactivation of horseradish peroxidase and catalase, among other enzymes [31]. Moreover, ultrasound efficacy is dependent upon numerous extrinsic and intrinsic operating parameters [33].

Effects of US and conventional blanching on total soluble solids and carbohydrates

Fructose, glucose and sucrose were the major carbohydrates in all the blanched samples analysed, regardless of the blanching treatment applied. Minor carbohydrates such as *scyllo*-inositol, *myo*-inositol and sedoheptulose were also present in all the samples under study.

Tables 3 and **4** list, respectively, the loss of total soluble solids and of low-molecular-weight carbohydrates due to leaching during the blanching of carrots by conventional and US treatments. As expected, the losses of total soluble solids were higher in minced over sliced carrots since the surface:volume ratio is 2-fold higher in the former. For both types of geometry, blanching treatment CS-2 provided the lowest loss of total soluble solids and carbohydrates in carrot samples. With the exception of CS-2 and CB-1 samples, all carrots presented a slight decrease in the pH values of the blanching water (results not shown). This could probably be due to the fact that, under these conditions, a higher amount of organic acids could be transferred to water by carrot leaching [34].

With respect to major low-molecular-weight carbohydrates, glucose and fructose were the main lost carbohydrates, followed by sucrose, probably due to the higher diffusivity and solubility of monosaccharides as compared to sucrose [35]. Machewad et al. [36] reported total soluble sugar losses of 62.5% in the conventional blanching of carrots carried out in boiling water for 5 min, whereas Nyman et al. [37] found 24 and 38% losses of soluble solids and carbohydrates, respectively, in carrots blanched in boiling water for 7 min. All these differences might be attributed, among other factors, to the different sample geometry and water/sample ratio used in the reported studies.

Minor carbohydrates were lost in variable amounts depending on the carbohydrate and the assayed treatment. The most striking result was the high leaching loss of sedoheptulose for any of the blanching treatments evaluated with values in the range 18-66%, higher than those obtained for *scyllo*-inositol (0-54%) and *myo*-inositol (0-57%).

Regarding samples processed by US pretreatments, the main losses were detected when samples were treated with generation of heat for longer times. For US bath and US probe blanching treatments carried out at low temperatures (USB 40-30 and USP 35-15), very low losses of total soluble solids (3-7%) and carbohydrates (3-10%) were found. In general, higher sugar losses were observed by other authors for papayas (13.8%), banana (21.3%), pineapples (23.2%) and Malay apples (17%) after 30 min treatment at 30 °C in an ultrasonic bath of 45 kHz [15, 38-40]. These differences could be due to the different susceptibility of vegetable substrates to the effects of US.

In the assays with US probe, taking into account only the effect of US (USP 35-15 and USP 35-60), the total soluble losses were low even after 60 min (< 6.5% in slices). However, higher losses were observed after treatments carried out at a final temperature of 60 or 70 °C, with values close to 37% in the latter. Finally, USP 60-10 gave rise to similar losses of total soluble solids and carbohydrates than conventional blanching at mild temperature (C60-40); particularly for minced carrot samples.

Conclusions

This work presents preliminary results on the efficiency of different conventional and US treatments for blanching of carrots. Although further research on additional indicators would be necessary to draw definite conclusions, it seems that US for blanching purposes is more convenient with probe and heat generation. According to the obtained results, among the US treatments of carrot samples assayed, those carried out at temperatures up to 70 °C gave rise to the highest enzymatic deactivation (90 and 50% POD and PME inactivation, respectively), with losses of total soluble solids ~ 37% and up to almost 50% of total carbohydrates. Moreover, US blanching with probe at temperatures up to 60 °C for 10 min presented similar values of enzyme inactivation and similar losses by leaching than the conventional treatment at 60 °C 40 min. Therefore, the application of US for carrot blanching, under these conditions, could constitute an adequate treatment with similar effects to LTLT conventional blanching but with a noticeable reduction of time. These treatments could also be considered as an advantageous for those applications in which partial inactivation of PME is required for better preservation of carrot structure. The results obtained in this work may contribute to broaden the application of US as an effective procedure for blanching of vegetables, particularly under mild conditions.

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Figure Captions

Fig. 1 Experimental set-up for US treatments with probe. [1]Depth of the probe in the sample (2 cm)

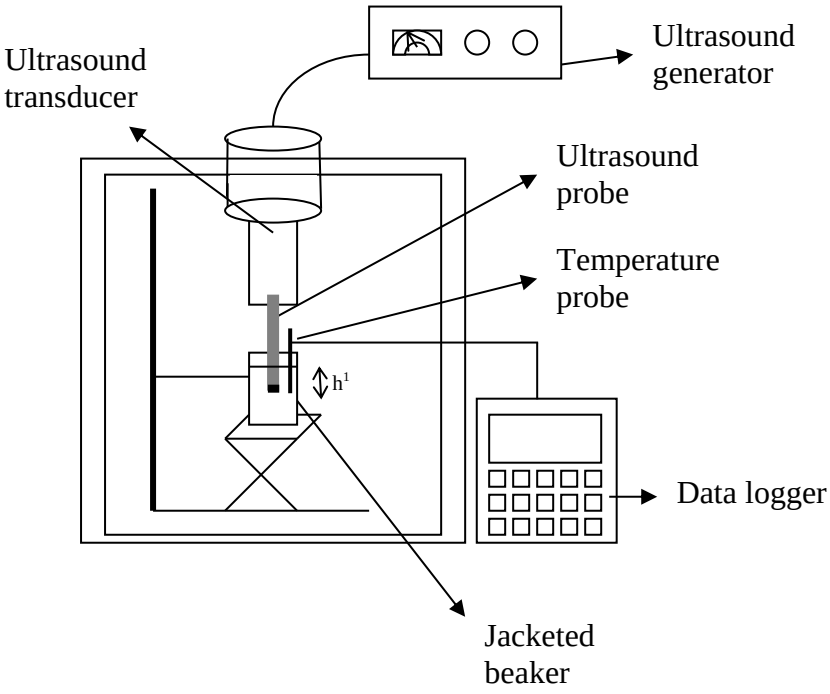

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Table 1 Processing conditions used during the blanching of carrot samples by conventional and ultrasound (in bath and with probe) treatments

Blanching	Samples	Temperature (ºC)	Time (min)	US density (Wcm^{-3})[1]
Conventional	CS-2	Steam	2	-
	CB-1	98	1	-
	C95-5	95	5	-
	C60-40	60	40	-
US (in bath)	USB 40-30	40	30	0.04
	USB 40-60		60	
	USB 60-30	60	30	
	USB 60-60		60	
US (with probe)	USP 35-15	≤35	15	0.26
	USP 35-60		60	
	USP 60-10	≤ 60	10	
	USP 70-15	≤70	15	

[1] Determined according to Jambrak et al. (2007)

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478**Table 2** POD and PME residual activity (%) in minced and sliced carrot samples after the 479different conventional and ultrasound blanching treatments. Mean of two replicates ± standard 480deviation.

	POD (%)		***PME (%)***	
Samples	***Minced***	***Sliced***	***Minced***	***Sliced***
Raw	100.0 ± 0.0 d[1]	100.0 ± 0.0 d	100.0 ± 0.0 a	100.0 ± 0.0 a
CS-2	6.8 ± 1.6 a	15.4 ± 0.7 a	0.1 ± 0.1 b	0.2 ± 0.1 b
CB-1	1.0 ± 0.0 b	1.0 ± 0.0 b	0.1 ± 0.1 b	0.1 ± 0.1 b
C95-5	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b	0.0 ± 0.0 b
C60-40	12.4 ± 3.2 c	40.9 ± 6.4 c	62.9 ± 0.7 cd	56.8 ± 5.3 c
USB 40-30	100.0 ± 0.1 d	100.0 ± 0.2 d	100.0 ± 0.0 a	100.0 ± 0.0 a
USB 40-60	100.0 ± 0.3 d	100.0 ± 0.1 d	79.9 ± 0.8 g	73.9 ± 8.0 f
USB 60-30	63.4 ± 6.9 e	25.5 ± 2.3 e	69.4 ± 4.8 f	52.4 ± 4.2 e
USB 60-60	63.4 ± 4.0 e	11.9 ± 0.5 a	68.4 ± 3.3 df	67.3 ± 1.6 d
USP 35-15	78.5 ± 5.7 f	71.4 ± 2.3 f	62.7 ± 1.2 c	61.8 ± 4.2 c
USP 35-60	58.3 ± 3.0 g	60.3 ± 8.2 g	49.0 ± 3.0 e	54.6 ± 4.3 e
USP 60-10	10.4 ± 0.1 ac	41.7 ± 8.4 c	69.1 ± 5.9 f	56.7 ± 8.0 c
USP 70-15	6.7 ± 1.4 a	17.4 ± 2.6 a	78.4 ± 1.8 g	53.5 ± 2.1 c

481[1]Samples with the same lower-case letter (a-g) within the same column showed no statistically 482significant differences for their mean values at the 95.0% confidence level.

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Table 3 Loss of total soluble solids by leaching determined in the blanching water of carrot samples submitted to different conventional and ultrasound treatments. Mean of two replicates ± standard deviation

	Leaching loss (%)	
Samples	*Minced*	*Sliced*
CS-2	0.7 ± 0.3 a[1]	0.6 ± 0.3 a
CB-1	11.9 ± 0.2 b	9.6 ± 0.9 b
C95-5	31.4 ± 0.2 c	19.2 ± 0.9 c
C60-40	26.5 ± 3.9 d	15.2 ± 0.6 d
USB 40-30	7.1 ± 0.2 e	3.2 ± 1.0 ae
USB 40-60	36.6 ± 4.6 f	15.0 ± 0.4 d
USB 60-30	48.5 ± 0.1 g	24.2 ± 1.0 f
USB 60-60	52.4 ± 0.1 g	37.7 ± 5.8 g
USP 35-15	6.2 ± 2.1 e	3.1 ± 0.6 ae
USP 35-60	13.5 ± 0.3 b	6.3 ± 0.5 be
USP 60-10	26.4 ± 0.1 d	19.1 ± 0.2 c
USP 70-15	37.4 ± 1.1 f	35.6 ± 0.0 g

[1]Samples with the same lower-case letter (a-g) within the same column showed no statistically significant differences for their mean values at the 95.0% confidence level

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Table 4 Loss (%) of major and minor carbohydrates in carrot samples blanched under different conventional and ultrasound treatments. Mean of two replicates ± standard deviation

Sample	Fructose		Glucose		Sucrose		*Scyllo*-inositol		*Myo*-inositol		Sedoheptulose		Total carbohydrates	
	Minced	Sliced	Minced	Sliced	Minced	Sliced	Minced	Sliced	Minced	Sliced	Minced	Sliced	Minced	Sliced
CS-2	0.4±0.2 a	1.3±0.0 a	0.3±0.1 a	0.1±0.0 a	0.3±0.1 a	0.3±0.0 a	0.3±0.0 a	0.0±0.0 a	0.4±0.1 a	2.9±0.0 a	18.4±0.3 a	18.3±0.0 a	1.3±0.4 a	1.2±0.3 a
CB-1	23.5±1.4 b	9.6±0.1 bc	22.6±0.9 b	10.7±0.4 bc	14.3±1.7 b	10.5±0.6 b	17.2±2.2 bc	14.2±1.4 bcd	18.8±0.8 b	19.4±0.7 b	34.7±0.9 b	30.5±0.9 b	18.0±1.5 b	11.6±0.5 b
C95-5	48.0±1.3 c	28.2±0.4 d	48.0±1.8 c	27.7±0.9 d	40.2±6.3 c	20.6±1.6 c	32.7±3.5 d	27.9±1.2 e	42.6±5.5 cd	29.9±0.6 c	51.6±0.2 c	41.5±0.9 c	43.1±4.6 c	23.9±0.7 c
C60-40	44.6±3.7 cd	30.4±5.3 d	45.0±2.2 c	28.9±5.8 d	31.0±4.7 d	22.5±5.3 c	39.5±4.1 de	28.8±7.1 e	39.0±4.1 cd	31.8±8.2 c	47.8±6.3 c	43.4±5.4	36.1±4.2 d	25.8±4.5 c
USB 40-30	19.5±2.7 be	3.6±1.0 ab	12.5±0.9 de	2.5±1.5 a	5.3±0.9 ae	2.9±1.2 a	20.7±6.4 c	3.2±0.6 a	13.5±5.3 be	4.7±1.3 a	33.3±6.2 b	21.5±1.2 a	10.0±1.4 e	4.0±1.2 a
USB 40-60	35.1±6.0 f	21.7±3.6 e	29.7±6.5 f	20.6±4.3 e	28.5±6.8 d	10.7±0.5 b	47.6±5.2 ef	14.6±6.0 cd	45.8±2.7 d	19.1±7.9 b	46.2±5.5 c	37.1±6.0 bc	30.7±6.5 d	15.3±1.9 b
USB 60-30	58.0±3.3 g	46.5±4.1 f	58.1±3.4 g	46.2±4.8 f	47.6±3.9 f	37.6±5.0 d	53.7±3.9 f	39.2±8.3 f	57.3±7.0 f	33.7±1.7 cd	63.8±2.9 d	48.9±1.8 de	51.6±3.8 fg	40.8±2.0 d
USB 60-60	56.5±3.2 g	53.9±6.8 g	57.9±2.6 g	56.0±7.1 g	56.4±1.3 g	40.8±5.3 d	52.1±4.7 f	44.1±6.7 f	55.8±0.7 f	54.4±6.3 e	65.9±5.3 d	64.2±7.0 f	57.1±0.3 g	46.4±5.9 e
USP 35-15	8.7±1.3 h	4.5±2.1 ab	8.7±1.8 d	4.6±1.7 ab	7.2±2.5 abe	1.6±1.1 a	8.6±1.1 ab	3.4±0.1 a	10.4±0.1 e	3.5±0.3 a	24.7±2.1 ae	20.0±0.7 a	8.6±1.9 e	3.5±0.2 a
USP 35-60	18.1±3.0 be	5.1±1.3 ab	16.9±2.6 be	4.4±1.4 ab	9.2±0.1 be	4.8±1.9 ae	14.7±0.7 bc	5.1±1.2 abc	16.4±0.7 be	6.5±1.8 a	32.2±2.6 be	22.8±2.1 d	13.0±1.0 be	5.8±1.8 a
USP 60-10	40.9±5.6 df	13.4±0.3 c	43.0±4.9 c	13.8±0.2 ce	25.6±3.5 d	8.4±0.3 be	31.1±8.1 d	23.8±2.1 de	35.4±3.0 c	16.9±0.3 b	50.7±2.3 c	31.6±0.1 b	31.9±0.5 d	11.3±0.3 b
USP 70-15	56.9±0.1 g	49.6±1.1 fg	56.3±0.2 g	47.5±0.7 f	45.1±0.1 cf	25.1±0.6 c	42.3±1.6 e	45.5±9.2 f	54.6±1.2 f	41.7±3.4 d	64.7±0.2 d	54.0±4.5 e	49.6±0.1 f	33.8±0.7 f

Samples with the same lower-case letter (a-h) within the same column showed no statistically significant differences for their mean values at the 95.0% confidence level

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