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Effect of Ultrahigh-Temperature Continuous Ohmic Heating Treatment on Fresh Orange Juice

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The scope of this study is the effect of ohmic heating thermal treatment on liquid fruit juice made of oranges. Effects of ohmic heating on the quality of orange juice were examined and compared to those of heat pasteurization at 90 °C for 50 s. Orange juice was treated at temperatures of 90, 120, and 150 °C for 1.13, 0.85, and 0.68 s in an ohmic heating system. Microbial counts showed complete inactivation of bacteria, yeast, and mold during ohmic and conventional treatments. The ohmic heating treatment reduced pectin esterase activity by 98%. The reduction in vitamin C was 15%. Ohmicheated orange juice maintained higher amounts of the five representative flavor compounds than did heat-pasteurized juice. Sensory evaluation tests showed no difference between fresh and ohmicheated orange juice. Thus, high-temperature ohmic-heating treatment can be effectively used to pasteurize fresh orange juice with minimal sensory deterioration.

KEYWORDS: Ohmic heating; electroheating; orange juice; pasteurization; flavor; vitamin C

INTRODUCTION

Ohmic heating of food products involves the passage of alternating current though them, thus generating internal heat as a result of electrical resistance (1). This technology provides rapid and uniform heating, resulting in less thermal damage to the product (2). In addition, the absence of a hot surface in ohmic heating reduces fouling problems and thermal damage to the product. Therefore, a high-quality product with minimal structural, nutritional, or organoleptic changes can be manufactured in a short operating time (3). The key to the successful implementation of an ohmic process is the rate of heat generation, the electrical conductivity of food material, and the way the food flows through the heater. Changes in electrical conductivities of vegetable samples and meat were studied and shown to be affected by a number of factors, for example, field strength, soluble solids, melting of fats, and cell structure changes (4-6). Palaniappan et al. (7) also determined the electrical conductivity of orange and tomato juices using a static device. They concluded that electrical conductivities of tomato and orange juice increase linearly with temperature and decrease with solids content. In addition, they determined that electrical conductivity tended to increase as particle size decreased, but a general conclusion cannot be reached without accounting for particle shape and orientations.

Although the technology of ohmic heating appears to be promising and highly effective, there is little information concerning the effects of this technique on specific food products compared to conventional pasteurization. Therefore, orange juice was selected as a model system to investigate the effects of ohmic-heating technology on liquid food products. The quality parameters concerning orange juice, such as inactivation of microorganisms and enzymes, heat-sensitive compounds, and physical characteristics, play an important role in the industry also. Lima et al. (8) studied the degradation of vitamin C in orange juice subjected to electrical and conventional heating. Experiments were performed using a static ohmic-heating apparatus, and matching heating histories were applied for both conventional and ohmic heating. A statistical analysis showed that the electrical field had no significant effect on ascorbic acid degradation. A continuous alternating current electric field was applied to orange juice containing *Bacillus subtilis* spores to examine the inactivation effect of electrical equipment (9). Electrical treatment at 121 °C under pressurized conditions was found to reduce the viable *B. subtilis* spores in orange juice.

The objectives of our study were to investigate the effects of ohmic heating on inactivation of microorganisms and pectin esterase (PE) in orange juice and to compare the quality of ohmic-heated orange juice including vitamin C, flavor compounds, browning, and color to that of conventionally pasteurized juice.

MATERIALS AND METHODS

Preparation and Processing of Orange Juice. Shamuti oranges (Somitz Ltd., Ramat-Tzvi, Israel) were processed in a citrus juice extractor 291 (FMC Co., Lakeland, FL) to produce freshly squeezed juice. Orange juice was filtered using a sieve for the separation of large particles (1 mm diameter holes). The fresh orange juice was thermally processed in a 50 kW pilot scale Electroheating system (Raztek, Sunnyvale, CA; **Figure 1**). The system consists of two feeding tanks: one for a salt solution and the other for the untreated product. The untreated product continuously enters the system via a mono pump

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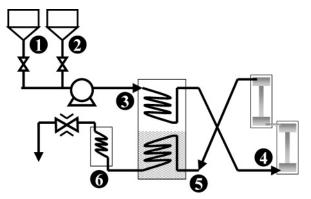


Figure 1. Scheme of the electroheating system, including salt solution tank (1), product tank (2), preheating (3), electroheating (4), rapid cooling (5), and secondary cooling (6).

 Table 1. Thermal Treatment Parameters at All Conditions: F Values (Minutes)

flow rate	time at holding	temperature		
(L/min)	tube (s)	90 °C	120 °C	150 °C
3	1.13	$1.89 imes 10^{-5}$	1.89×10 ⁻²	18.9
4	0.85	$1.41 imes 10^{-5}$	$1.41 imes 10^{-2}$	14.1
5	0.68	$1.13 imes 10^{-5}$	1.13×10^{-2}	11.3

(A.P.V. Baker, Peterborouth, U.K.). Initially, the product is pumped to the first part of the system, the rapid cooler, where the product is preheated (10, 11). The rapid cooler consists of a tank containing two sets of coiled tubes: an upper coil tube for the untreated product flow and a lower coil for the heated product flow. The upper part of the tank is saturated with steam, and the lower part is filled with water, and the whole system is under vacuum to maximize the heat transfer. The hot product passes through the tube, the water is quickly boiled, and the cold product passes through the tube above the boiled liquid and is heated by the steam condensation. Following preheating, the product enters the electroheater at a temperature that is the exact mean between its temperature at the tank (point 3, Figure 1) and the treatment temperature (point 5, Figure 1) (12-15). The electroheating unit consists of two pairs of adjacent graphite electrodes, with a 20 cm gap between each pair of electrodes. The product flows along the axis between the electrodes. The system utilizes an alternating current at a frequency of 50 Hz and at maximum voltage of 8 kV. The system controller automatically determines the necessary current and voltage using a feedback from a thermocouple at the exit from the heating chamber to heat the product to the predetermined treatment temperature. Following electroheating, the product enters a 120 cm holding tube, in which the thermal treatment takes place. This tube connects the exit of the electroheater to the entry of the rapid cooler (point 5, Figure 1). Then the product is cooled rapidly in the lower part of the rapid cooler. Finally, the product is cooled to room temperature in a tubular heat exchanger.

The installation was pressurized with a pressure valve to provide a backpressure of ~12 atm and prevent boiling of the superheated product. A presterilization step of the electroheating system was carried out by circulating a sodium chloride solution with the same electrical conductivity of orange juice ($\sigma = 0.36$ S/m at 25 °C) at 120 °C for over 20 min. After processing, orange juice samples were collected aseptically into sterilized jars for analysis and stored on ice.

A matrix of three set point temperatures and three flow rates was tested. Orange juice was treated by the ohmic-heating system at set point temperatures of 90, 120, and 150 °C and in flow rates of 3, 4, and 5 L/min. **Table 1** summarizes the corresponding *F* values (minutes) of every combination of temperature and flow rate in the thermal treatments. The *F* values were calculated with 120 °C as the reference temperature, on the basis of z = 10 °C, which was taken as a representative for the *z* value of PE ranging from 6.5 to 13 °C in the temperature range of 80–90 °C (*16, 17*). Conventional pasteurization

of fresh orange juice was conducted at 90 °C for 50 s using a plate heat exchanger (A.P.V. Baker) with a 1 L holding tube. The corresponding *F* value is 8.33×10^{-3} min.

Microbial Counts. Microbial counts followed Israeli standard 885, "Microbiological test methods for foodstuffs: general laboratory rules", which follows ISO 7218-1996, "Microbiology of food and animal feeding stuffs—general rules for microbiological examinations" methods. Inactivation of microorganisms by thermal treatments was determined by total plate counts of the heated samples using orange serum agar (OSA) and yeast and mold counts using oxytetacycline glucose yeast extract agar (OGYE) with a selective supplement. OSA, OGYE, and OGYE selective supplement were purchased from Oxoid (Hampshire, U.K.). Samples of 1 mL of fresh and treated orange juice were diluted with 0.1% peptone water (Bactro, Sparks, MD) to 10⁻¹ dilution. From each dilution, two samples were plated. Plates were incubated at 30 °C for 48 h.

Pectin Esterase Activity. Determination of residual PE activity after thermal treatments was based on the formation of galacturonic acid and determined titrimetrically as described by Rouse and Atkin (*18*). Specifically, a 3 mL aliquot of orange juice sample was added to 50 mL of substrate solution containing 1.0% citrus pectin (Fluka, Buchs, Germany) and 0.2 M NaCl (Bio-lab, Jerusalem, Israel). During hydrolysis at 30 °C, the pH was maintained at 7.5 by the addition of 0.1 N NaOH (Antibioticos, Ronado, MI) using an automatic pH-stat (Titrino 718, Metrohm, Herisau, Switzerland). The consumption of 0.1 N NaOH was recorded during a 10 min reaction period. PE activity unit (PEU) and the relative PE activity (%) were calculated according to the following equations:

$$PEU = \frac{(mL \text{ of } NaOH) \times (0.1 \text{ N } NaOH)}{(3 \text{ mL of sample}) \times (10 \text{ min})} \times 10^{4}$$

relative PE activity (%) =

 $\frac{\text{PEU of thermally treated orange juice}}{\text{PEU of fresh orange juice}} \times 100$

Vitamin C Concentration. Determination of the ascorbic acid concentration in orange juice was performed according to the method of Yeom et al. (19). The analysis was conducted using a reverse-phase high-performance liquid chromatograph (RP-HPLC), HP 1100, equipped with a diode array detector at 254 nm and controlled by ChemStation software package (Hewlett-Packard, Wilmington, DE). HPLC analysis was carried out on a reverse-phase C_{18} column (25 cm \times 4.6 mm SupelcoSil column, Supelco Inc., Bellefonte, PA). Samples were eluted at a flow rate of 1 mL/min with 10% methanol in citrate solution (pH 2.9) as a mobile phase. The mobile phase was filtered using a 0.45µm membrane filter (Millipore, Bedford, MA). The orange juice samples were centrifuged at 12500g for 10 min in a 4214 ACL microcentrifuge (ACL International, Milano, Italy) to remove pulp and coarse cloud particles. Twenty microliters of the supernatant was injected manually to the column. The elution time of ascorbic acid and dehydroascorbic acid was 3.4 min. A standard calibration curve of L-ascorbic acid (Aldrich Chemical, Co., Milwaukee, WI) and dehydroascorbic acid in concentrations ranging from 10 to 80 mg/100 mL was used to quantify vitamin C. Because the curves were identical for both ascorbic acid and dehydroascorbic acid, we used the measured value as total vitamin C.

Flavor Compound Analysis. Measurements of flavor compounds in orange juice were performed by headspace solid-phase microextraction gas chromatography (SPME-GC) following the method of Jia et al. (20). A 1.0-mL aliquot of fresh and thermally treated orange juice was transferred into a sealed 7-mL vial. A SPME fiber with 65 μ m polydimethylsiloxane—divinylbenzene (Supelco, Inc.) coating was manually inserted into the headspace of the vial containing orange juice for adsorption of the flavor compounds. The vial was incubated at 60 °C for 20 min. The SPME fiber was then injected into the GC injection port at 220 °C and kept for 2 min. The separation of the flavor compounds was accomplished by a Hewlett-Packard 6890 GC equipped with a capillary column (Innowax, 30 m × 0.25 mm i.d., 0.25 μ m, Agilent Technologies) and a flame ionization detector. The temperature was programmed from 60 to 180 °C at rate of 5 °C/min and held for a final 2 min. The GC chromatograph peak area was calculated using ChemStation software package (Hewlett-Packard). The identification of flavor compounds in orange juice was performed by comparing the retention time with that of five standard compounds: α -pinene, myrcene, limonene, octanal, and decanal (Sigma Chemical Co.). The standard calibration curve of each flavor compound was obtained by plotting the GC peak area against a known concentration in deodorized orange juice.

Browning Measurement. Browning was determined on the basis of the method of Meydav et al. (21) and Yeom et al. (19). Specifically, orange juice was centrifuged at 12500g for 10 min in a 4214 ALC microcentrifuge (ACL International). Supernatant was collected and clarified utilizing a 0.45- μ m filter (Millipore). The browning index was determined as the absorbance at 420 nm in spectrophotometer (Ultraspec 2100, Biochrom, Cambridge, U.K.,) at room temperature.

Color Determination. Orange juice color was measured using a chroma meter (Minolta, CR-300, Tokyo, Japan). A sample of 7 mL was placed in a cell and measured for *L*, *a*, and *b* values. An increasing *L* value represents an increase in lightness (L = 0, dark; L = 100, light). An increase in the value of *a* indicates an increase in redness (-a = green, +a = red).

Sensory Evaluation. Sensory tests were conducted to evaluate the sensory attributes of ohmic-heated orange juice. A triangle test was performed to compare fresh, pasteurized, and ohmic-heated orange juices. It should be noted that, for this experiment, the pasteurized and ohmic-heated orange juices were thermally treated at the same *F* value of 8.33×10^{-3} min to create a basis for comparison. In this test, each panelist received three coded samples, of which two were identical and one was different. The panelist was asked to identify the different sample. Each set of three samples was repeated 25 times. Statistical level of confidence was 5% (22).

°Brix Measurement. °Brix indicates the soluble solids content and was measured by a table refractometer, AR-4 (Kruss, Hamburg, Germany).

Data and Statistical Analysis. All experiments were performed in duplicates, and the results are expressed as the average. Statistical analysis was performed for the determination of significant differences in the processing treatments. Statistical analysis was conducted with JMP 4.0.4, statistical discovery software (SAS Institute Inc.), and the data analysis tool pack of the Microsoft Excel software.

RESULTS AND DISCUSSION

Effects of Ohmic Heating on Microorganisms. Citrus juices are characterized by high acidity conditions, which lead to the growth of yeast and mold in addition to a few types of low acid tolerant bacteria (23). To avoid microbial spoilage, it is necessary to cause inactivation by applying heat. Both ohmic and conventional thermal treatments reduced microbial counts by at least 2-3 orders of magnitude compared to their number in fresh orange juice, which was $\sim 10^{2.5}$ colony-forming units (CFU)/mL. Although there was no detectable difference between ohmic and conventional heating concerning inactivation of microorganisms, a true comparison of this aspect of the techniques should be done by inoculation of samples with a reference microorganism. The results indicate that the significant parameter in the inactivation of microorganisms is the thermal effect, regardless of the kind of thermal treatment. These results are supported by the findings of Palaniappan et al. (24). In their work, suspensions of yeast cells (zygo Saccharomyces bailii) and cells of Escherichia coli were subjected to conventional and ohmic heating. It was found that at thermally lethal conditions, any lethal effect caused by electricity was insignificant when compared to that produced by heat. A more recent study was conducted on Bacillus subtilis spores treated by conventional or ohmic heating under identical temperatures histories (25). This study showed that spores heated at 92.3 °C had significantly smaller D values when heated using an ohmic rather than a conventional method. It was concluded that spore

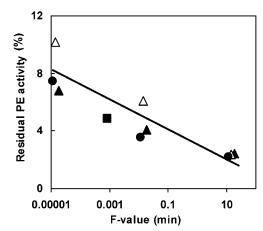


Figure 2. Effects of thermal treatments on pectin esterase activity, compared to its activity in fresh orange juice. Thermal treatments are expressed as *F* value (minutes). Type of thermal treatment: ohmic heating [3 (\triangle), 4 (\triangle), and 5 (\bigcirc) L/min]; conventional pasteurization (\blacksquare).

inactivation during ohmic heating was primarily due to the thermal effect. However, there was an additional killing effect caused by the electric current. In our study we attributed the inactivation of microorganisms to heat only.

Effects of Electrical Heating on Pectin Esterase Activity. PE causes cloud loss in orange juice by de-esterification of pectin; thus, thermal treatment is applied to inactivate the enzyme. The design for thermal pasteurization of orange juice is based on the thermal destruction characteristics of pectin esterase, which is more thermally stable than many vegetative microorganisms. The z value of PE ranges from 6.5 to 13 °C in the temperature range of 80-90 °C (16, 17). The measured PE activity after thermal treatments is presented in Figure 2. PE activity is presented as percentage of PE activity in fresh orange juice. Generally, as the impact of thermal treatments during ohmic heating increases by applying higher temperatures or times, the residual PE activity decreases (p < 0.05). During ohmic heating, PE activity showed a reduction of 90-98% compared to its activity in fresh orange juice. Under conventional pasteurization conditions, the residual PE activity was reduced to 5%. Thermal treatments were preformed as a combination of various temperatures and flow rates. Therefore, effects of ohmic and conventional treatments were investigated either by temperature, flow rate, or the interaction between these factors. For all ohmic-heating treatments, no significant difference was found between different flow rates or the interaction between temperature and flow rate (p > 0.05), but only for temperature (p < 0.001). A regression line was drawn for ohmicheating results, and the result obtained for conventional pasteurization was found to be within the 95% confidence interval of the ohmic-heating regression line. This indicates that there was no significant effect to the type of thermal treatment on PE inactivation.

Influence of Ohmic Heating on Vitamin C Concentration. Vitamin C is subjected to degradation due to nonenzymatic reactions. Among other factors, these reactions are accelerated because of exposure to elevated temperatures during thermal process. Vitamin C concentration decreased in 7–25% compared to fresh orange juice (Figure 3). As can be drawn from the results, despite the increase in *F* value applied during ohmicheating treatments, the reduction of vitamin C maintained at ~15% (p > 0.05). The reason for this interesting result is the very short residence time during ohmic heating (<2 s), which allows retention of heat-sensitive compounds. At all ohmicheating treatments, no significant effect was found for flow rates

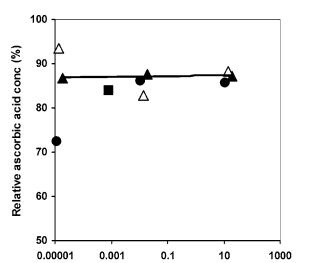


Figure 3. Influence of ohmic and conventional pasteurization treatments on ascorbic acid concentration compared to fresh orange juice. Thermal treatments are expressed as *F* value (minutes). Type of thermal treatment: ohmic heating [3 (\blacktriangle), 4 (\triangle), and 5 (\bigcirc) L/min]; conventional pasteurization (\blacksquare).

F-value (min)

or the interaction between temperature and flow rate (p > 0.05). A decrease of 16% in the concentration of ascorbic acid was observed after conventional pasteurization compared to fresh orange juice. This result is similar to the ohmic-heating results, because it was found within the confidence interval of 95% of the ohmic-heating regression line. Lima et al. (8) examined ascorbic acid degradation in pasteurized orange juice during conventional and ohmic heating. They performed matching time—temperature histories in both conventional and ohmic-heating batch treatments. As in this study, they also found that the type of heating had no significant effect on vitamin C degradation. They measured a decrease of 21-23% in ascorbic acid during thermal treatments at 90 °C for 30 min.

Effects of Electrical Heating on Browning in Orange Juice. Nonenzymatic browning may result in the formation of offflavor, a decrease in nutrient content, a loss of color, and, above all, the appearance of brown pigments Ascorbic acid degradation is considered to be a major chemical reaction responsible for browning in citrus juices. The results of browning index in treated orange juice are presented in Figure 4. Browning measurements of both ohmic and conventional juices were higher than the result obtained for fresh orange juice, which was 0.14. Browning levels in ohmic-heated orange juices showed significant increase with the increase in F values (p <0.0001). The browning levels measured after ohmic heating ranged from 0.25 to 0.43 depending on temperature. The browning index of conventionally pasteurized orange juice was 0.27, as was expected for ohmic-heated orange juice processed at an equivalent F value. The increase in absorbance indicates initiation of browning reactions due to exposure to high temperatures during thermal treatments. It should be noted that these levels of browning are not visible to the consumer (26). In addition, the effects of ohmic and conventional heating on lightness (L) are shown in Figure 5. The lightness of ohmicheated juices showed a significant decrease with the increase in F values (p < 0.05). As expected, ohmic-treated orange juice browns during processing compared to fresh orange juice. The L value of fresh orange juice was 99.3. This observation indicated that the browning reactions were initiated by thermal treatment. The decrease in lightness of ohmic-heated juices correlates with the increase in their browning levels ($r^2 = 0.73$,

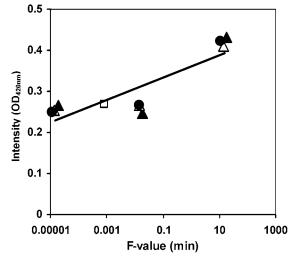


Figure 4. Effects of ohmic and conventional heating treatments on browning index. Thermal treatments are expressed as *F* value (minutes). Type of thermal treatment: ohmic heating [3 (\blacktriangle), 4 (\triangle), and 5 (\bigoplus) L/min]; conventional pasteurization (\blacksquare).

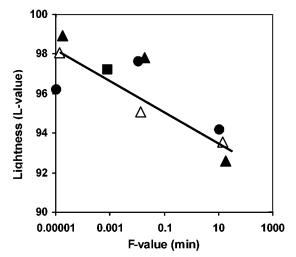


Figure 5. Changes in lightness in thermally treated orange juice. Thermal treatments are expressed as *F* value (minutes). Type of thermal treatment: ohmic heating [3 (\blacktriangle), 4 (\triangle), and 5 (\bigcirc) L/min]; conventional pasteurization (\blacksquare).

p < 0.001). The *L* value of conventional pasteurized orange juice was 97.2, which was also lower than the *L* value of fresh juice

Effects of Ohmic and Conventional Heating on Flavor Compounds. Flavor compounds of orange juice are 0.02% of the total weight: 75-98% of the flavor compounds are hydrocarbons, 0.6-1.7% aldehydes, 1% esters, 1% ketones, and 1-5% alcohols (27). Thermal treatment can negatively affect the flavor of the juice. Irreversible damage to the citrus juice flavor results from chemical reactions accelerated during the heating process. In this study we focused on five major flavor components: limonene, pinene, myrcene, octanal, and decanal (20). The results of flavor components concentration in processed orange juice are presented in Figure 6. All of the flavor compounds show the same concentration profile. Higher concentrations were obtained for ohmic-heated orange juice samples than for conventionally pasteurized orange juice. Statistical significance was found for limonene, myrcene, pinene, and decanal (p < 0.05), but not for octanal (p > 0.05). Flavor components concentrations in pasteurized juice were outside the 95% confidence interval of the ohmic-heating regression

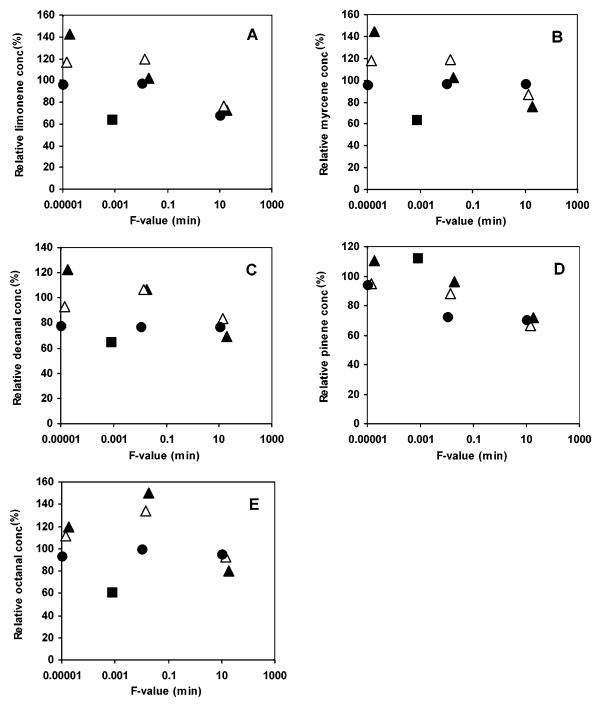


Figure 6. Influence of electrical and conventional heating treatments on the concentration of flavor compounds: limonene (**A**), myrcene (**B**), decanal (**C**), pinene (**D**), octanal (**E**). Thermal treatments are expressed as *F* value (minutes). Type of thermal treatment: ohmic heating [3 (\blacktriangle), 4 (\triangle), and 5 (\bigcirc) L/min]; conventional pasteurization (\blacksquare).

line. Furthermore, the relative concentrations during 90 and 120 °C ohmic-heating treatments were >100% for all flavor compounds. After ohmic-heating treatment at 150 °C, flavor compounds concentrations were <100%, but still higher than pasteurized ones (**Figure 6**). The flavor compounds in fresh orange juice are considered as 100%. Thus, the results indicate retention and even release of flavor compounds in ohmic-heated orange juice. This interesting observation can be explained by two corresponding phenomena. On the one hand, thermal treatment may cause a release of bonded components from the medium. On the other hand, due to the short residence time during ohmic heating, the released flavor compounds are not quickly degraded as during conventional pasteurization. There-

fore, higher flavor compounds concentrations were obtained after ohmic heating. These findings are supported by the study conducted by Yeom et al. (19), investigating the effects of pulsed electric fields (PEF) on the quality of orange juice. They also encountered the phenomenon that the same flavor compounds concentration in orange juice after PEF were higher than those obtained after conventional pasteurization. They concluded that the lower initial heat load during PEF treatment might cause fewer chemical reactions, resulting in more retention of flavor compounds.

Sensory Evaluation of Fresh and Thermally Treated Orange Juice. Sensorial tests for food products are conducted, among other purposes, to provide for preliminary analysis to

 Table 2. Sensory Evaluation Results of Fresh, Conventionally

 Pasteurized, and Ohmic-Heated Orange Juice^a

no. of correct answers	treatment		
14*	fresh vs pasteurized OJ		
9	fresh vs ohmic heated OJ		
16*	pasteurized vs ohmic heated OJ		

^a Triangle tests were preformed, n = 25, $\alpha = 0.05$; *, statistically significant.

measure the flavor of food and its acceptance or to measure flavor changes due to change in processing (22). It is important to mention that ohmic and conventional pasteurization treatments were applied at the same F value of 8.3×10^{-3} min to create a basis for comparison. The sensory evaluation results are summarized in Table 2. Results for sensory evaluation of fresh and thermally treated orange juice indicate that a panelist could distinguish between fresh and pasteurized samples and between pasteurized and ohmic-heated orange juice (p < 0.05). It should be noted that tasters could not differentiate between fresh and ohmic-heated orange juice (p > 0.05). As shown previously, fresh and ohmic-heated orange juices exhibited similar levels of flavor compounds concentrations. When fresh and ohmicheated samples are compared, the panelist actually compares between taste, odor, and color. Given no visible change in color, tasters had to determine only according to flavor. Thus, due to the similar flavor profiles of fresh and ohmic-heated orange juice, panelists could not distinguish between them, as the two juices resembled each other.

To conclude, the results of the present study suggest that continuous ohmic-heating thermal treatment can be used to effectively pasteurize freshly squeezed orange juice. This technique is efficient in the required microbial and enzymatic inactivation, as well as in keeping the good quality of the juice. This property of the treatment results in the high retention of the sensorial attributes of the fresh juice, as supported by chemical analyses of major flavor compounds as well as sensory evaluation. The most interesting aspect of the ohmic-heating treatment is the lack of overheating due to heat transfer aspects. Further studies are being conducted to explore the effect of this treatment on the long-term stability and shelf life of such products.

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