

## PAPER

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2025, 3, 239Pasteurization of mandarin juice by ohmic heating  
and evaluation of its shelf-life under refrigerated  
and ambient conditionsSomnath Basak,<sup>ID</sup> Piyush Thakur and Snehasis Chakraborty<sup>ID</sup>\*

Mandarin juice was treated inside a bath ohmic heater at 15 different combinations of voltages (120, 160, and 200 V) and treatment times (30, 60, 90, 120, and 150 s) targeting 85 °C. The come-up time to reach 85 °C for 120, 160, and 200 V were 240, 180, and 100 s, respectively. The system performance ranged between 88% and 98%. Ohmic heating rates increased with the applied voltage reaching 0.545 °C s<sup>-1</sup> at 200 V. Ohmic heating at 200 V/30 s and 160 V/90 s produced a microbially safe mandarin juice (5 log reduction in natural microflora); whereas 200 V/30 s, 160 V/90 s, and 120 V/150 s treatments led to an enzymatically stable (>99% spoilage enzyme inactivation) mandarin juice. Under the pasteurized condition of 200 V/30 s, there was only a 10% and 8.4% loss in vitamin C and total phenolics in the juice. Elevated antioxidants were observed after ohmic heating. Lightness and total color change ( $\Delta E^*$ ) of the juice increased with ohmic heating. The maximum browning of 66.74 was observed in the juice treated at 120 V/150 s. The juice treated at 200 V/30 s showed a shelf life of 42 and 15 days under refrigerated and ambient conditions, respectively.

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## Sustainability spotlight

To achieve adequate microbial lethality and enzymatic stability in food products, the conventional pasteurization methods require long holding times at high temperatures. This detrimentally results in a loss of bioactive compounds and nutritional appeal of the product. Ohmic heating helps in attaining the desired temperature in a smaller duration, leading to lower process time. The advantages of uniform heating, high energy conversion efficiencies, no surface fouling, and less maintenance cost due to no moving parts advocate for ohmic heating as a sustainable technology. This also aligns well with the United Nation's Sustainable Development Goal 12 of "Responsible Consumption and Production"; wherein ohmic heating can sustainably help in the improvement of the shelf-life of fresh produce and prevent food wastage.

## 1 Introduction

Citrus juices are an important source of bioactive compounds and micronutrients; and their consumption has been correlated with a reduced incidence of several chronic diseases.<sup>1</sup> The citrus fruits can be classified into four major categories, namely mandarins, sweet oranges, grapefruit & pomelo group, and the common acid group members such as the acid lime and the sweet lime.<sup>2</sup> The common mandarin fruit (*Citrus reticulata* Blanco) is rich in nutrients and bioactive compounds, possessing nutritional value.<sup>3</sup> The popularity of mandarin fruit due to its "simple-to-peel" characteristic has propelled an increase in mandarin consumption in comparison to the "hard-to-peel" grapefruit and pomelo.<sup>4</sup> Mandarin is reported to exhibit antioxidant, anti-diabetic, anti-inflammatory, and antihyperlipidemic activities.<sup>5</sup>

Although the acidity of citrus juices acts as a barrier to the growth of foodborne pathogens, outbreaks of *Salmonella*, and *Escherichia coli* O157:H7 in fruit juices have been reported quite often.<sup>6</sup> In such a scenario, the pasteurization of fruit juices is extremely necessary to prevent the emergence of fruit juice-associated outbreaks.<sup>7</sup> Some of the pasteurization conditions employed for fruit juices include 65 °C|30 min, 77 °C|1 min, and 88 °C|15 s.<sup>8</sup> The conventional thermal processing used for pasteurization has a very high come-up time (in the range of 15–30 min), drastically reducing the nutritional quality and sensory acceptability of juices.<sup>9</sup> Therefore, novel thermal methods are being employed to preserve the nutritional quality and also achieve microbial and enzymatic lethality. Ohmic heating is being projected as a great alternative to conventional thermal pasteurization.<sup>10</sup>

Ohmic heating, or Joule effect heating, is a process in which heat is generated internally within a food material when an electric current is passed through it. The internal heat generation in the food material is due to the resistance offered by food materials to an electric current; i.e., food material acts as

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a resistor in the circuit. Ohmic heating does not depend on the heat transfer between solid-liquid interfaces or solid-solid interfaces; therefore, it is a form of internal energy generation technology, not thermal transfer technology.<sup>11</sup> The advantages of ohmic heating include uniform heating, high energy conversion efficiencies, no surface fouling, target temperature being achieved very quickly, and less maintenance cost due to no moving parts,<sup>12</sup> increasing demand in the food industry. Due to uniform or rapid heating, nutritional or structural properties of food materials are minimally affected by ohmic heating.<sup>13</sup> Apart from the electric field, a concentric magnetic field is also created near the current carrying electrodes. A Lorentz force is experienced by the charged particles, wherein the particle attains a velocity which is perpendicular to the direction of the force and the magnetic field.<sup>14</sup> A higher rate of temperature rise is expected as the charged particles in the juice will experience several forces from different directions at a particular point, resulting in greater agitation of the particles. Ohmic heating has several potential applications such as extraction,<sup>15</sup> fermentation,<sup>16</sup> pasteurization,<sup>17</sup> drying,<sup>18</sup> juice expression,<sup>19</sup> and frying.<sup>20</sup> Ohmic heating is best suited for the aseptic processing of viscous fluids or particulates.<sup>21</sup> However, no study has been undertaken to recommend a pasteurization condition for mandarin juice. Therefore, this study aims to study the effect of various ohmic heating conditions on the various quality attributes of mandarin juice.

In this study, the ohmic heating treatment of mandarin juice was undertaken at different voltage and treatment durations. The physicochemical properties, along with the microbial and enzymatic quality of juice, were examined to understand the impact of process variables. Furthermore, a microbially safe and enzymatically stable sample after ohmic heating was selected. Finally, the properties of the juice were also studied during storage under refrigerated and ambient conditions to determine the shelf-life of the ohmic heating-pasteurized and equivalent thermally treated juice.

## 2 Materials and methods

### 2.1 Materials

Fresh, ripe and mature mandarins (*Citrus reticulata*) were purchased from the local market in Matunga, Mumbai, India. The fruits (~10 cm diameter) were free from any physical damage and infestation. The fruits were washed with 200 ppm sodium hypochlorite solution, peeled, deseeded, and extracted using a juice extractor (JL02, Anjali Kitchenware, India). The average juice yield was around 55%, excluding the peel, seeds, and pulp. The juice was filtered using a press filter (Microfilt India, Mumbai, India) with a 100 µm mesh. All the other chemicals were of analytical grade and procured from HiMedia Pvt. Ltd, Mumbai, India. The pH, total soluble solids (TSS), and Brix to acid ratio of juice were  $3.2 \pm 0.2$ ,  $10.82 \pm 0.37$  °Bx, and  $17.60 \pm 1.3$ , respectively. The viscosity of strained juice was  $496.64 \pm 0.02$  mPa s.

### 2.2 Pasteurization in a batch ohmic heating unit

The heating section had a capacity of 6 L and consisted of a cuboidal shape cell (620 mm × 100 mm × 100 mm). Six

stainless steel (SS-304) electrodes ( $\Phi$  2 mm) with a silver coating were placed 1.25 cm apart to facilitate temperature uniformity. A J type thermocouple (Pencil type, Thermonic India) was placed between the electrodes (Fig. 1). The thermocouple is attached to a temperature controller (SELEC TC523 PID, Mumbai, India). The voltage regulator (Variacco Electricals, Mumbai, India) is connected to regulate the voltage between electrodes. The variable power supply was obtained using a voltage regulator (0–250 V) from a domestic supply (220 V and 50 Hz). Electrodes were attached to a voltage regulator, and a thermocouple is connected to the temperature controller. The voltage was varied at three levels, *i.e.*, 120, 160, and 200 V and the time was varied on five levels, *i.e.*, 30, 60, 90, 120, and 150 s, making the cumulative number of treatments  $3 \times 5 = 15$ . The treatment temperature was maintained constant at 85 °C. The come-up time for 2 L of juice to reach 85 °C for 120, 160 and 200 V was 240, 180, and 100 s, respectively. The ohmic heating rate of the mandarin juice was calculated by plotting come-up time with respect to temperature.

To understand the performance of the ohmic heating system, the system performance coefficient (SPC) was calculated as the ratio of the sensible heat absorbed by the juice ( $\text{Heat}_{\text{abs}}$ ) and the energy delivered to the juice system ( $\text{Energy}_{\text{del}}$ ), as given in eqn (1).<sup>9</sup> The specific heat of the juice was assumed to be constant in the studied temperature range. The electrical energy given to a system and heat required to reach the final temperature ( $T_f$ ) are calculated using the voltage ( $V$ ), current ( $I$ ), and initial temperature ( $T_i$ ). The SPC during the ohmic heating was calculated from the mass of the juice ( $m$ ) with a specific heat ( $C_p$ ) that was processed in  $t$  seconds.

$$\text{SPC} = \frac{\text{Heat}_{\text{abs}}}{\text{Energy}_{\text{del}}} = \frac{mC_p(T_{\text{final}} - T_{\text{initial}})}{\sum VIt} \quad (1)$$

For high-moisture foods, the  $C_p$  of the juice above the freezing point can be calculated using the Seibel empirical formula, wherein  $X_m$  refers to the moisture content of the juice.<sup>22</sup>

$$C_p = 3.35X_m + 0.837 \quad (2)$$

The current ( $I$ ) during the treatment of mandarin juice at 120, 160, and 200 V was recorded between 35 and 60 °C to determine the electrical conductivity ( $\sigma$ ).<sup>23</sup>

$$\sigma = \frac{L}{A} \times \frac{I}{V} \quad (3)$$

The  $L/A$  represents the cell constant of a continuous ohmic heater where  $L$  is the length of the electrode through which current is generated in the sample and  $A$  is the cross-section area normal to the current flow.  $V$  represents the voltage and  $I$  represents the current. The cell constant of the ohmic heater was  $1.59 \text{ cm}^{-1}$  when filled with up to 2 L of juice. The methodology for calculating electrical conductivity for a multiple-electrode assembly has been adopted from Bhattacharjee and Chakraborty.<sup>9</sup> The setup for determining electrical conductivity



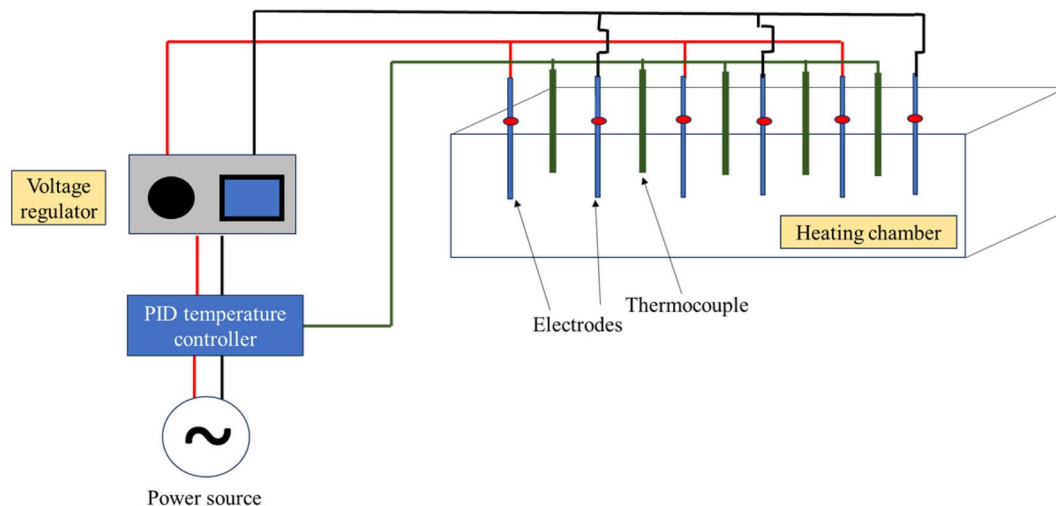


Fig. 1 The schematic of the ohmic heating chamber used for the pasteurization of mandarin juice.

includes two multimeters functioning as voltage and current indicators. The multimeters were calibrated before each set of current and voltage readings to ensure accuracy. In this configuration, two adjacent electrodes were connected to one terminal of the voltage transformer, while the other two electrodes were connected to the opposite terminal. A known current was applied between the two outer electrodes, and the resulting voltage drop was measured across the inner electrode pair.

### 2.3 pH, total soluble solids and titratable acidity

The pH of the juice was estimated by using a bench-top pH meter (Labserve, Fisher Scientific). A handheld refractometer (Erma 0–32 ° Brix, 182 Japan) was used to determine the TSS. The titratable acidity (TA) was determined by titrating the juice against a standard NaOH solution. The TA was expressed in % (wt/v) citric acid (anhydrous).

### 2.4 Color

CIE  $L^*$  (lightness),  $a^*$  (green to red) and  $b^*$  (blue to yellow) values of the juice were recorded using a HunterLab colorimeter (LabScan XE, Hunter Associates Laboratory, USA) in reflection mode (D65/10°). The browning index (BI) and total color difference ( $\Delta E^*$ ) were calculated using the following equations:<sup>24</sup>

$$\Delta E^* = \sqrt{(L^* - L_0^*)^2 + (a^* - a_0^*)^2 + (b^* - b_0^*)^2} \quad (4)$$

$$BI = \frac{31}{0.172} \left[ \frac{a^* + 1.75 \times L^*}{5.645 \times L^* + a^* - 3.012 \times b^*} \right] \quad (5)$$

### 2.5 Ascorbic acid, total phenolics, and antioxidant activity

The ascorbic acid (AA), total phenolic content (TPC), and antioxidant capacity (AOX) of the juice were determined using the spectrophotometric methods described by Chakraborty *et al.*<sup>25</sup>

The ascorbic acid (AA) content was expressed as mg AA/100 mL of juice. TPC was estimated using Folin–Ciocalteu's phenol reagent method and expressed in mg gallic acid equivalent (GAE)/100 mL of juice. The AOX activity was reported in mg GAE/100 mL of the juice.

### 2.6 Enzyme activity

The assays of peroxidase (POD), polyphenol oxidase (PPO), and pectin methyl esterase (PME) were conducted as per the protocol suggested by Chakraborty *et al.*<sup>26</sup> and Sahoo & Chakraborty.<sup>27</sup> The protein concentration in the crude extract was estimated using the Bradford method, which used bovine serum albumin as the reference protein. The rate of change in product formation per unit mass of the protein was considered to be enzyme activity. The residual activity was calculated, considering the untreated sample's activity to be 100%.

### 2.7 Microbial enumeration

The indigenous microflora of the untreated and the enzymatically stable (complete inactivation, *i.e.*, >99% inactivation) juices were enumerated using the serial dilution-pour plate method.<sup>28</sup> While the aerobic mesophiles (AM) were incubated on the plate count agar (HIMEDIA® M091) for 24 h at 37 °C, the yeasts and molds (YM) were incubated on yeast and mold agar (HIMEDIA® M424) at 30 °C for 3–5 days. The limit of detection was 10 cfu mL<sup>-1</sup>.

### 2.8 Shelf-life estimation

The best ohmic heating condition was selected as the minimum intensity required to achieve 5 log cycle reduction in the natural microflora population along with >99% enzyme inactivation. The best treatment (200 V/30 s) was chosen for further shelf-life experiments. Soon after the ohmic heating treatment, the pasteurized juice was transferred to sterile coextruded ethylene vinyl alcohol (EVOH) copolymer pouches (thickness 90 microns) with a water vapor permeability of 0.047 g mm<sup>2</sup> m<sup>-2</sup> h<sup>-1</sup> kPa<sup>-1</sup>.



The AM and YM counts of the juice were tracked on every 7th day till 42 days of refrigerated ( $4 \pm 1$  °C) and ambient storage ( $27 \pm 1$  °C) at 85% R. H. The PME activity, color, and TPC were also monitored.

A control untreated sample and equivalent thermally treated sample (90 °C for 60 s of holding) were also checked for their shelf-lives for comparison. The thermal treatment condition was selected for its ability to achieve greater than  $5 \log_{10}$  cfu mL<sup>-1</sup> reduction in YMC and AMC, along with undetectable enzyme activity. While an open-tank treatment was performed for the batch ohmic heating, in-pouch thermal treatment was performed as the reference treatment. The in-pouch treatment was performed to prevent any possibility of post-processing contamination. The processing temperature was maintained at 90 °C in a thermostatic water bath (EIC23, Expo-Hi-Tech, Mumbai, India). 500 mL of the juice was packed in an EVOH pouch and placed in the center of the water bath to facilitate heating of the juice from 27 to 90 °C. A dummy pouch containing the juice and J-type pencil thermocouple (Thermonic India) was also placed in the water bath to estimate the come-up time. Throughout the heating process, it was ensured that the juice inside the pouch is completely dipped in the water. The come-up time for the thermal treatment (90 °C) was  $12.1 \pm 0.8$  min. The thermally pasteurized and the ohmic heated samples have been referred to as ETT and OHT, respectively. The samples stored under refrigerated conditions have been referred to as ETTR and OHTR, while the pasteurized juices stored under ambient conditions have been referred to as ETTA and OHTA throughout the manuscript.

## 2.9 Statistical analysis

All the tests and analyses were conducted in triplicate. One-way analysis of variance (ANOVA) at a significance level of 0.05 was performed using Tukey's HSD test using SPSS software (IBM SPSS Statistics, version 16.0, USA).

# 3 Results and discussion

## 3.1 Heating rate, electrical conductivity of mandarin juice and SPC of ohmic heating

The potential difference across the electrodes significantly affects the heating rate of mandarin juice during ohmic treatment. Heating rates of the juice at three different voltages are shown in Fig. 2. The ohmic heating rates were  $0.234$  °C s<sup>-1</sup>,  $0.333$  °C s<sup>-1</sup>,  $0.545$  °C s<sup>-1</sup>, at 120, 160, and 200 V respectively (Fig. 2(A)). The time required to reach 60 °C at 200 V was 1.6 and 2.2 times lower than that for 160 and 120 V. The heating rate is affected by the electrical conductivity of juice and the electric field strength during the ohmic heating treatment. Ohm's law states that the current flowing through the two points between a conductor is directly proportional to the potential difference across those points. Thus, when higher voltage gradients were applied, the samples showed a linear trend of temperature rise from 30 to 60 °C. At higher voltages, the augmentation of the movement of water molecules and ions is greater, leading to higher heating rates. This significantly reduces the process

time. However, bubble formation was observed above 95 °C, which reduces the heating rate significantly. Darvishi *et al.*<sup>29</sup> reported a similar bubble formation in pomegranate juice during ohmic heating. The formation of a bubble is due to electrochemical reactions, and fruit juices are generally acidic, leading to hydrogen gas formation by electrochemical reactions.<sup>30</sup>

The range of electrical conductivity of mandarin juice was between 1.2 and 2 S m<sup>-1</sup>. The electrical conductivity of mandarin juice showed a linear relationship with the temperature at three different voltages (Fig. 2(B)). The increasing movement of ions and the decreasing viscosity of the juice at higher temperatures and voltages result in greater electrical conductivities.<sup>31,32</sup> Jha *et al.*<sup>33</sup> and Yildiz *et al.*<sup>34</sup> reported that electrical conductivity increases with temperature due to a reduction in drag force for the movement of ions. The electrical conductivity of juice also depends on the solid content of juice; at low solid content, the drag force for the movement of ions is less, which provides high electrical conductivity.<sup>30</sup> The current intensity and electrical conductivity are expected to increase with temperature, probably due to a decrease in the viscosity of the juice and frictional drag force between the ions.<sup>35</sup> Karakavuk *et al.*<sup>36</sup> observed that the electrical conductivity of the grape juice ranged between 0.19 and 0.85 S m<sup>-1</sup>, when exposed to a voltage gradient of 13 to 17 V cm<sup>-1</sup>. Norouzi *et al.*<sup>35</sup> also observed electrical conductivity in the range of 0.70 to 2.52 S m<sup>-1</sup> when sour cherry juice was pasteurized by ohmic heating at voltage levels of 30 to 50 V.

The SPC of the system was 98% when treatment was conducted at 120 V, and as the voltage increased, the SPC value started to decrease. At 200 and 160 V, efficiency of a system falls to 88% and 90%, respectively. These values of SPC indicate that at a higher voltage gradient, the energy provided to the system is not thoroughly utilized for heating the juice. Heat loss (2%) was significantly less at 120 V, indicating efficient conversion of electrical energy to heat energy. This is in line with the findings of Icier *et al.*,<sup>23,37</sup> wherein SPC increases from 47 to 92%, with a decrease in voltage gradient. The low energy losses at low voltage gradient can be attributed to the small heat transfer area of the cell and thereby lower convection losses. Higher voltages induce chemical, physical, and electrochemical changes in the juice, which result in greater energy losses.<sup>38</sup>

## 3.2 pH, total soluble solids and titratable acidity

Initially, the pH of untreated mandarin juice was 3.31, which largely remained unaffected after the ohmic heating treatment. Similarly, the total soluble solids of the untreated sample were  $10.82 \pm 0.37$  °Brix, which remained constant after the ohmic heating treatments. In other words, ohmic heating did not induce any major hydrolysis of complex polysaccharides into monosaccharides and disaccharides. The titratable acidity of the juice decreased from 0.58% to 0.41 and 0.48% at a treatment voltage of 200 V and 160 V for 150 s, respectively (Table 1). There is some literature reporting a similar trend. For instance, Darvishi *et al.*<sup>29</sup> observed a slight decrease in acidity after ohmic heating due to the hydrolysis of juice and corrosion of



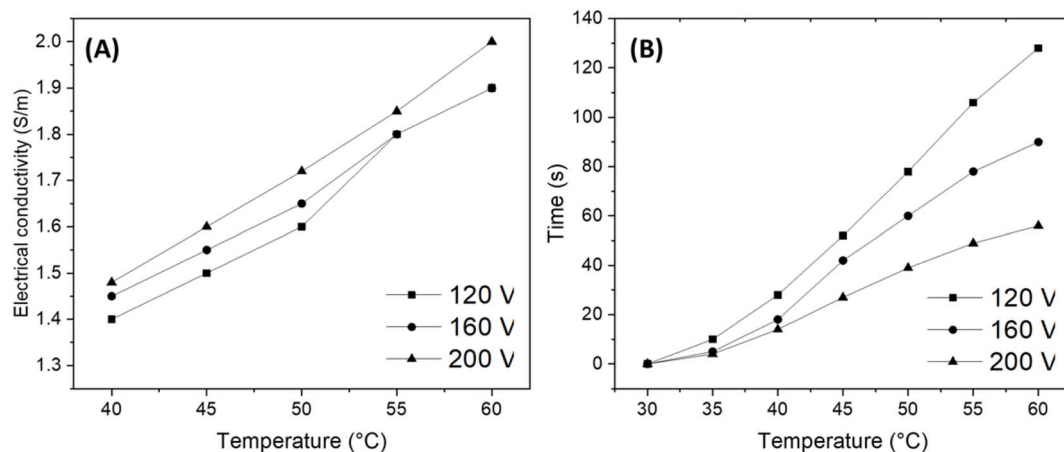


Fig. 2 The (A) electrical conductivity and (B) heating rate of the mandarin juice subjected to ohmic heating at 120, 160, and 200 V. The heating rate for 120, 160, and 200 V was 0.234, 0.333, and 0.545 °C s<sup>-1</sup>, respectively.

Table 1 The pH, total soluble solids (TSS) and titratable acidity of the mandarin juice subjected to different ohmic heating treatments at varying voltages and durations of treatment<sup>a</sup>

Voltage (V)	Time (s)	pH	TSS (°Bx)	Titratable acidity (% (wt/v) citric acid)
Untreated		3.32 ± 0.02 <sup>a</sup>	10.82 ± 0.37 <sup>a</sup>	0.58 ± 0.04 <sup>a</sup>
120	30	3.34 ± 0.01 <sup>a</sup>	10.76 ± 0.28 <sup>a</sup>	0.56 ± 0.03 <sup>a</sup>
	60	3.33 ± 0.01 <sup>a</sup>	10.47 ± 0.38 <sup>a</sup>	0.56 ± 0.04 <sup>a</sup>
	90	3.33 ± 0.01 <sup>a</sup>	10.81 ± 0.49 <sup>a</sup>	0.58 ± 0.04 <sup>a</sup>
	120	3.33 ± 0.01 <sup>a</sup>	10.78 ± 0.39 <sup>a</sup>	0.58 ± 0.05 <sup>a</sup>
	150	3.34 ± 0.01 <sup>a</sup>	10.72 ± 0.29 <sup>a</sup>	0.52 ± 0.05 <sup>a</sup>
160	30	3.36 ± 0.01 <sup>a</sup>	10.69 ± 0.34 <sup>a</sup>	0.56 ± 0.04 <sup>a</sup>
	60	3.37 ± 0.01 <sup>a</sup>	10.72 ± 0.44 <sup>a</sup>	0.55 ± 0.05 <sup>a</sup>
	90	3.37 ± 0.01 <sup>a</sup>	10.66 ± 0.35 <sup>a</sup>	0.52 ± 0.04 <sup>ab</sup>
	120	3.37 ± 0.01 <sup>a</sup>	10.45 ± 0.43 <sup>a</sup>	0.51 ± 0.04 <sup>ab</sup>
	150	3.37 ± 0.01 <sup>a</sup>	10.56 ± 0.51 <sup>a</sup>	0.48 ± 0.03 <sup>b</sup>
200	30	3.35 ± 0.01 <sup>a</sup>	10.45 ± 0.54 <sup>a</sup>	0.57 ± 0.04 <sup>a</sup>
	60	3.35 ± 0.01 <sup>a</sup>	10.44 ± 0.22 <sup>a</sup>	0.56 ± 0.05 <sup>ab</sup>
	90	3.35 ± 0.01 <sup>a</sup>	10.63 ± 0.11 <sup>a</sup>	0.53 ± 0.04 <sup>ab</sup>
	120	3.35 ± 0.01 <sup>a</sup>	10.52 ± 0.23 <sup>a</sup>	0.49 ± 0.05 <sup>bc</sup>
	150	3.34 ± 0.01 <sup>a</sup>	10.45 ± 0.44 <sup>a</sup>	0.41 ± 0.04 <sup>c</sup>

<sup>a</sup> Different superscript letters within one column indicate significant ( $p \leq 0.05$ ) differences among means determined by Tukey's test.

electrodes. The reason behind this is the conversion or degradation of organic acids such as citric and ascorbic acid. Besides, ascorbic acid can participate in non-enzymatic browning reactions, which gets aggravated at higher voltage and long treatment durations.<sup>39</sup>

### 3.3 Ascorbic acid, total phenolics and antioxidant activity

The ascorbic acid content of fresh mandarin juice was 48 mg/100 mL. The AA degradation was in the range of 10 to 53.8% for the tested voltage–time combinations. As the voltage increases, the come-up time decreases, leading to a shorter cumulative treatment time. The minimum ascorbic acid degradation was observed for a treatment duration of 30 s at 200 V. A reduced come-up time correlated well with greater ascorbic acid retention. The highest loss of 53.8% was observed at 120 V for a treatment duration of 150 s. This can be attributed

to the highest come-up time at a lower voltage. The ascorbic acid (AA) retention of guava, sapota, and papaya was tested by Athmaselvi *et al.*<sup>40</sup> at different voltage gradients (10 and 20 V cm<sup>-1</sup>), temperatures (70, 80, 90 and 100 °C) and electrode combinations (stainless steel and titanium electrodes). A higher voltage resulted in greater ascorbic acid degradation due to electrochemical reactions and electrode corrosion. This result is contrary to our results. This can be attributed to the lower come-up time at higher voltages and the type of electrode (silver coated SS 304) which reduces the corrosion and electrochemical reactions during treatment. However ascorbic acid degradation is more sensitive to the time of treatment compared to an electric field. Assiry *et al.*<sup>38</sup> reported that ascorbic acid degradation of orange juice within a temperature range of 65–90 °C was unaffected by the electric field strength. Interestingly, Doan *et al.*<sup>41</sup> observed an increase in AA content in pomelo juice by





5.9% for a treatment at  $30 \text{ V cm}^{-1}$ . The ohmic heating treatment breaks down the bonds that are associated with AA and the pomelo juice matrix, thereby leading to an increase in AA content.

A similar trend was also observed for the phenolics and antioxidant capacity (Fig. 3(B) and 4(A)). The total phenolic content of untreated juice was  $53.3 \text{ mg GAE}/100 \text{ mL}$ . The least and greatest reduction in phenolics was 8.4 and 27.1%, under treatment conditions of  $200 \text{ V}$  for  $30 \text{ s}$  and  $120 \text{ V}$  for  $150 \text{ s}$ , respectively (Fig. 3). According to Brochier *et al.*,<sup>42</sup> the TPC degradation of sugarcane juice was 11–18% between  $50$  and  $80^\circ \text{C}$  for 6–12 min at a fixed voltage. Brochier *et al.*<sup>42</sup> reported that the degradation of phenolics in sugarcane juice was 14% at  $3.9 \text{ V cm}^{-1}$  and 10% at  $20.5 \text{ V cm}^{-1}$ . The antioxidant capacity (AOC) of untreated mandarin juice was  $21.2 \text{ mg GAE}/100 \text{ mL}$ . Interestingly, the antioxidant activity of the treated juices was

significantly greater than that of the untreated juice. It increased with the treatment voltage. The maximum antioxidant activity ( $26.3 \text{ mg GAE}/100 \text{ mL}$ ) was observed at  $200 \text{ V}/30 \text{ s}$  ( $85^\circ \text{C}$ ) at the same point where the least AA and TPC degradation was observed. While a lower come-up time at higher voltages is expected to better preserve the AOC, the greater electro-permeabilization at higher voltages can facilitate greater extraction of antioxidants while maintaining the integrity of the product.<sup>43</sup>

### 3.4 Color

The visual and sensory acceptability of juices is highly dependent on the color of the juice. The  $L^*$  value of untreated mandarin juice was 16.13 which increased with an increase in treatment voltage, implying an increase in the brightness of the

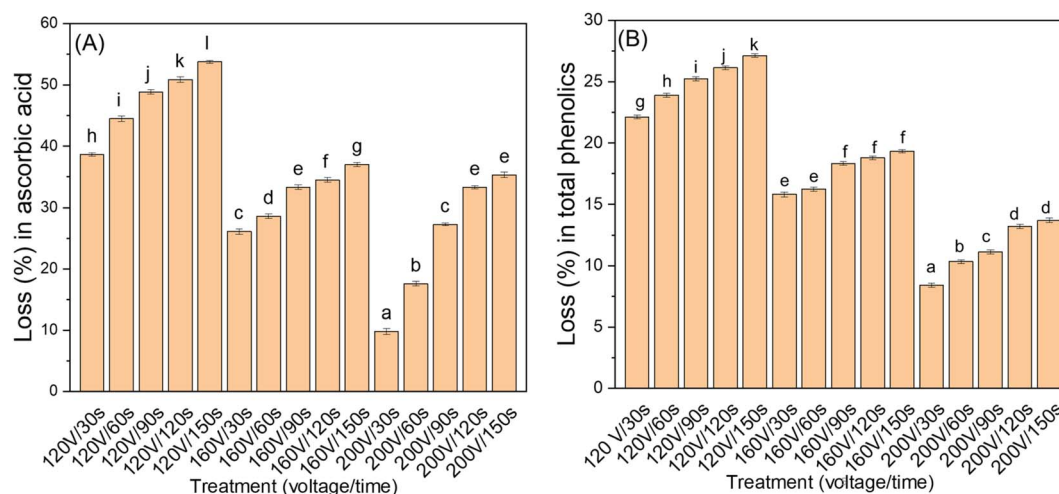


Fig. 3 The loss of (A) ascorbic acid and (B) total phenolics in the mandarin juice, when subjected to ohmic heating at 15 various combinations of voltage and treatment time. The dissimilar alphabets on the top of the bar denote that the mean values are statistically different at  $p < 0.05$ .

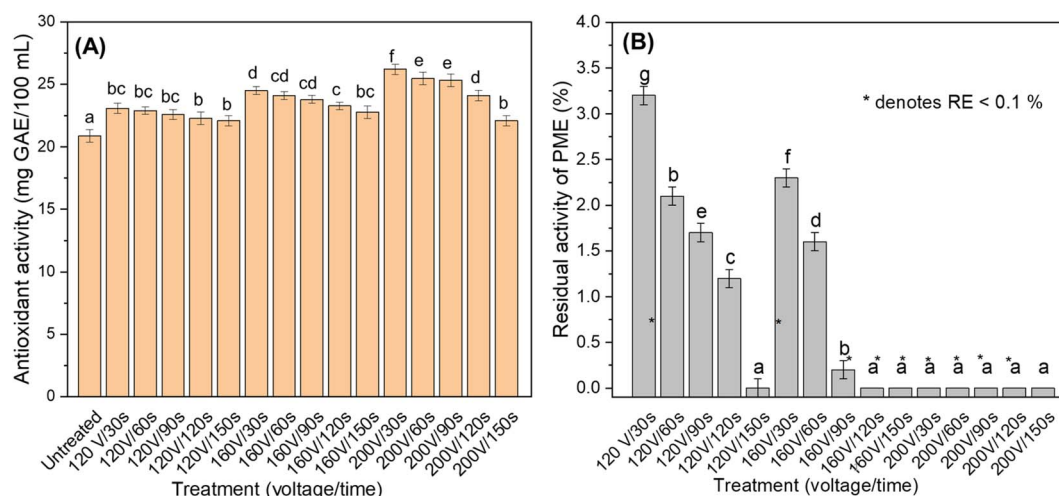


Fig. 4 The (A) antioxidant activity of the mandarin juice, when subjected to ohmic heating at 15 various combinations of voltage and treatment time. (B) The residual PME activity of the juice treated by ohmic heating at several voltage–time combinations. The dissimilar alphabets on the top of the bar denote that the mean values are statistically different at  $p < 0.05$ . RE, residual PME activity.



juice. Similarly, the  $a^*$  and  $b^*$  values significantly increased after the ohmic heating treatment. Notably, the  $b^*$  values for ohmic-treated samples were in the range of 3.89 to 4.88, which is substantially greater than the  $b^*$  value of the untreated mandarin juice (2.69). This is indicative of the increase in the yellowness of juice with voltage. The lower degradation of carotenoids at higher voltage due to a lower come-up time can be attributed to this trend.<sup>17</sup> It is worth noting that  $L^*$  significantly increased after the ohmic heating treatment compared to the untreated sample. This increase was so significant that it was a major reason for a drastic increase in  $\Delta E^*$ . For instance, the maximum  $\Delta E^*$  was significantly higher for 200 V-treated juice; however, the BI values of the juice treated at 120 and 160 V were similar. The increased  $L^*$  or lightness of the juice can probably be due to the better extraction of carotenoids and phenolics at higher voltages. The higher  $b^*$  values also support the trend, indicating a greater yellowness in the juice. Therefore, the higher  $\Delta E^*$  should not be mistaken for greater browning of the juice.

The browning index of ohmic-treated mandarin juice was in the range of 62.65 to 66.74 (Table 2). The maximum browning was observed in the juice treated at 120 V for 150 s, which can be attributed to the highest come-up time as compared to the other voltage treatments. Non-enzymatic browning results in the loss of nutrients, formation of brown pigments, off-flavour and loss of colour. Makroo *et al.*<sup>44</sup> observed a similar phenomenon for ohmic heating of watermelon juice. Although darkening of the juice was observed after the treatment of the juice, the  $L^*$  decreased to a greater extent for thermal processing than ohmic heating. The authors attributed this trend to the greater degree of PPO inactivation in ohmic heating. Ascorbic acid degradation is considered one of the main reasons for browning in citrus juices. Leizeron and Shimoni<sup>17</sup> reported the same trend in ohmic-treated orange juice. The change in color ( $\Delta E^*$ ) of juice increases as the treatment time increases. The maximum change

was seen in the sample treated at 150 s and minimum in a sample treated at 30 s at any given voltage. The sample treated at 200 V shows less color change compared to the sample treated at 120 V (Table 2). For instance, the total color change of guava juice during ohmic heating decreases with an increase in voltage gradient.<sup>45</sup> The reason is due to less exposure time at a higher voltage. The color change may occur due to the heat-induced browning of juice in the presence of metal ions, oxygen, and carbonyl groups that act as a precursor in enzymatic browning.<sup>17</sup> Saberian *et al.*<sup>46</sup> did not report any change in browning in aloe vera gel after the ohmic heating. Sabancı and İcier<sup>47</sup> ascribed the color change and browning in sour cherry juice to the degradation of monomeric anthocyanins after the ohmic heating.

### 3.5 Enzymatic activity

PPO and POD result in enzymatic browning of juices, leading to a loss of nutritional content, sensory appeal and economic value of the product. Furthermore, enzymes like PME result in a loss of turbidity and viscosity, leading to phase separation in juices.<sup>48</sup> The PPO and POD enzymes were more susceptible than PME in mandarin juice and were completely inactivated after ohmic heating treatments of 120 V for 60 s, 160 V for 30 s and 200 V for 30 s, respectively. PPO and POD were inactivated by 92.4 and 95.7%, respectively, after ohmic heating at 120 V for 30 s. PME was also completely inactivated after ohmic heating at 160 V for 90 s and 120 V for 150 s (Fig. 4(B)). For a specific voltage, the enzyme activity decreased with increasing duration of the treatment. Brochier *et al.*<sup>42</sup> also established that PPO is more labile than POD in sugarcane juice during ohmic heating. Demirdöven & Baysal<sup>49</sup> reported 96% reduction in PME activity in orange juice after a treatment of 42 V cm<sup>-1</sup> at 69 °C. Similarly, Saxena *et al.*<sup>50</sup> noticed a 97.8% reduction in PPO activity after a treatment of 32 V cm<sup>-1</sup> at 90 °C for 5 min. The mechanism of enzyme inactivation by ohmic heating is a combination of thermal and nonthermal mechanisms. Thermal denaturation

**Table 2** The color parameters ( $L^*$ ,  $a^*$  and  $b^*$ ) of the mandarin juice subjected to ohmic heating treatments of varying voltages and durations of treatment<sup>a</sup>

Voltage (V)	Time (s)	$L^*$	$a^*$	$b^*$	$\Delta E^*$	BI
Untreated		16.13 ± 0.12 <sup>a</sup>	0.93 ± 0.11 <sup>a</sup>	2.69 ± 0.12 <sup>a</sup>	—	62.65 ± 0.12 <sup>a</sup>
120	30	17.98 ± 0.11 <sup>d</sup>	1.87 ± 0.21 <sup>cd</sup>	3.89 ± 0.05 <sup>b</sup>	2.40 ± 0.11 <sup>b</sup>	65.55 ± 0.10 <sup>b</sup>
	60	17.89 ± 0.06 <sup>d</sup>	1.99 ± 0.13 <sup>d</sup>	3.78 ± 0.12 <sup>b</sup>	2.33 ± 0.12 <sup>b</sup>	65.52 ± 0.13 <sup>b</sup>
	90	17.81 ± 0.10 <sup>d</sup>	2.02 ± 0.06 <sup>d</sup>	3.99 ± 0.08 <sup>b</sup>	2.39 ± 0.09 <sup>b</sup>	66.07 ± 0.10 <sup>c</sup>
	120	17.53 ± 0.06 <sup>c</sup>	1.85 ± 0.13 <sup>cd</sup>	3.94 ± 0.09 <sup>b</sup>	2.10 ± 0.08 <sup>a</sup>	65.92 ± 0.09 <sup>c</sup>
	150	16.98 ± 0.12 <sup>b</sup>	2.11 ± 0.09 <sup>d</sup>	3.99 ± 0.10 <sup>b</sup>	1.95 ± 0.10 <sup>a</sup>	66.74 ± 0.09 <sup>c</sup>
160	30	18.87 ± 0.11 <sup>f</sup>	1.64 ± 0.11 <sup>c</sup>	4.12 ± 0.12 <sup>b</sup>	3.17 ± 0.11 <sup>d</sup>	65.25 ± 0.10 <sup>b</sup>
	60	18.33 ± 0.13 <sup>e</sup>	1.89 ± 0.12 <sup>cd</sup>	4.01 ± 0.11 <sup>b</sup>	2.74 ± 0.12 <sup>c</sup>	65.63 ± 0.10 <sup>b</sup>
	90	18.42 ± 0.12 <sup>c</sup>	1.78 ± 0.10 <sup>cd</sup>	4.21 ± 0.09 <sup>b</sup>	2.88 ± 0.10 <sup>c</sup>	65.86 ± 0.08 <sup>b</sup>
	120	18.21 ± 0.15 <sup>de</sup>	1.75 ± 0.09 <sup>cd</sup>	4.16 ± 0.10 <sup>b</sup>	2.68 ± 0.14 <sup>c</sup>	65.85 ± 0.13 <sup>bc</sup>
	150	17.98 ± 0.09 <sup>d</sup>	1.83 ± 0.11 <sup>cd</sup>	4.23 ± 0.13 <sup>b</sup>	2.58 ± 0.11 <sup>c</sup>	66.24 ± 0.12 <sup>d</sup>
200	30	21.33 ± 0.11 <sup>h</sup>	1.24 ± 0.21 <sup>b</sup>	4.76 ± 0.21 <sup>c</sup>	5.61 ± 0.17 <sup>f</sup>	64.78 ± 0.16 <sup>b</sup>
	60	20.76 ± 0.09 <sup>g</sup>	1.68 ± 0.18 <sup>c</sup>	4.98 ± 0.12 <sup>c</sup>	5.22 ± 0.13 <sup>ef</sup>	65.95 ± 0.14 <sup>c</sup>
	90	20.88 ± 0.10 <sup>g</sup>	1.59 ± 0.12 <sup>c</sup>	4.99 ± 0.13 <sup>c</sup>	5.32 ± 0.12 <sup>f</sup>	65.81 ± 0.13 <sup>c</sup>
	120	20.55 ± 0.12 <sup>g</sup>	1.62 ± 0.09 <sup>c</sup>	4.86 ± 0.09 <sup>c</sup>	4.98 ± 0.10 <sup>e</sup>	65.78 ± 0.11 <sup>c</sup>
	150	20.42 ± 0.22 <sup>g</sup>	1.51 ± 0.12 <sup>c</sup>	4.88 ± 0.11 <sup>c</sup>	4.85 ± 0.16 <sup>e</sup>	65.76 ± 0.15 <sup>c</sup>

<sup>a</sup> Different superscript letters within one column indicate significant ( $p \leq 0.05$ ) differences among means determined by Tukey's test. The  $\Delta E^*$  and BI refer to the total color difference and browning indices, respectively.



of enzymes occurs when heat disrupts the enzyme's secondary and tertiary structures by breaking hydrogen bonds, ionic bonds, and hydrophobic interactions. Apart from the heat-induced denaturation of enzymes, the presence of an electric field can modify the molecular spacing and promote inter-chain reactions in enzymes.<sup>44</sup> The non-thermal effect of ohmic heating is due to the translation and rotation of the enzyme molecules in response to the applied external electric field. This can be majorly attributed to the charge and dipole moment in the enzyme molecule. A greater electric field gradient results in greater collision between the molecules, that can have a detrimental effect on enzyme activity.<sup>42</sup> The electrostatic forces generated by the electrodes during ohmic heating result in conformational alterations in the tertiary structure.<sup>51</sup> The induction of molecular movement due to the dissipation of energy by friction resulting from the oscillating electric field reduces the enzyme activity.<sup>43</sup> The susceptibility of the enzymes to denaturation by ohmic heating is increased due to the metallic prosthetic groups interacting with electric fields generated during the treatment.<sup>52</sup>

### 3.6 Microbial quality

It has been reported that spoilage enzymes exhibit greater resistance to thermal treatment compared to microbes in fruit juices.<sup>48</sup> Our goal is to identify a treatment condition that achieves both microbial safety (5 log reduction) and enzymatic stability (99% inactivation of the target enzyme). Therefore, only samples showing 99% enzyme inactivation after treatment were further analyzed for microbial enumeration. It is evident that the remaining treatment conditions, while potentially meeting microbial safety, do not satisfy both criteria of microbial safety and enzymatic stability. The enzymatically stable samples of mandarin juice (*i.e.*, 200 V for 30 s, 160 V for 90 s, and 120 V for 150 s) were taken for microbial testing. The initial AM and YM counts in the untreated juice sample were 6.9 and 6.2 log<sub>10</sub> cfu mL<sup>-1</sup>, respectively. No aerobic mesophiles were detected in the juice samples treated at 200 V for 30 s and 160 V for 90 s. A 5.5 log reduction in AM population was observed in the 120 V/150 s treated juice. The YM count was undetected in all the enzymatically stable samples. The reduction of AM and YM population was greater than 5 log<sub>10</sub> cycle, indicating adequate pasteurization. The application of ohmic heating treatment reduced all microbiological counts, with values which are below the quantification levels. The microbial inactivation induced by ohmic heating is a combination of thermal and nonthermal effects. While the thermal effects of ohmic heating result in protein denaturation, nonthermal effects such as electroporation result in cell lysis and death. Yoon *et al.*<sup>53</sup> revealed that the exuded intracellular material was greater in ohmic heating than in the conventional heating treatment. This suggests that nonthermal effects of ohmic heating play a significant role in microbial inactivation. The external electric field induces a transmembrane potential in the phospholipidic membrane.<sup>54</sup> The increase in permeability thereof would result in irreversible leakage of proteins, nucleic acids and coenzymes.

### 3.7 Shelf-life analysis

**3.7.1 Microbial and enzymatic quality.** The ohmic heating condition of 200 V/30 s was selected, as it was the minimum intensity required to achieve 5 log cycle reduction in the natural microflora population along with >99% enzyme inactivation. Furthermore, the greatest retention in ascorbic acid (90.2%) and phenolics (91.6%) was also observed under the treatment condition of 200 V/30 s.

The AM and YM are the main microbes responsible for the spoilage of fruit juices under refrigerated conditions.<sup>55</sup> The AM and YM counts of untreated juice stored at 4 °C crossed the value of 7 log<sub>10</sub> cfu mL<sup>-1</sup> on the 7th day of storage. The AM and YM counts of the untreated juice sample stored at 27 °C exhibited the same trend wherein the counts increased to 7.6 log<sub>10</sub> cfu mL<sup>-1</sup> and 7.4 log<sub>10</sub> cfu mL<sup>-1</sup>; on the 7th day of ambient storage. The ETTR and OHTR exhibited undetectable AM and YM counts throughout the storage period of 42 days. The microbial counts in the sample stored at 27 °C started increasing rapidly from the 14th day of the storage period. On the 27th day of ambient storage, the respective AM and YM counts were 6.7 and 6.3 log<sub>10</sub> cfu mL<sup>-1</sup> for the ETT sample, while the respective AM and YM counts in the juice pasteurized by ohmic heating were 6.1 log<sub>10</sub> cfu mL<sup>-1</sup> and 6.4 log<sub>10</sub> cfu mL<sup>-1</sup> on the 27th day (Fig. 5). The sample stored at 4 °C exhibited no growth in microbial counts compared to the sample stored at 27 °C, in which microbial count rapidly increased from the 27th day of storage. As per the microbiological standards for fruit products set by the Food Safety and Standards Authority of India (FSSAI), the permissible AM and YM counts in pasteurized juices are 4 and 3 log<sub>10</sub> cfu mL<sup>-1</sup>, respectively.<sup>56</sup> While the standard set for unpasteurized juices mandates a maximum permissible limit of 7 log<sub>10</sub> cfu mL<sup>-1</sup> of natural microflora. For the ETTR and OHTR samples, the microbial counts were well below the prescribed limit for 42 days. For the juices stored at 27 °C, the AM and YM counts crossed the value of 4 and 3 log<sub>10</sub> cfu mL<sup>-1</sup> after 15 days. Therefore, it can be summarized that ohmic heating treatment increased the shelf life of juice stored under refrigerated and ambient conditions to 42 and 15 days, respectively. Alcántara-Zavala *et al.*<sup>57</sup> reported that ohmic heating at 65 °C for 5 min preserved pulque, a probiotic beverage until 22 days with greater sensory acceptability. The sudden spurt in growth under ambient conditions indicates a sublethal effect on the microorganisms, which regenerate after cell damage. Saxena *et al.*<sup>50</sup> observed that the untreated sugarcane juice converted to a viscous gel on the 6th day of refrigerated and ambient storage. Similar to our study, the authors also did not observe any significant increase in AM and YM at the end of 10 days of refrigerated storage. The juice at room temperature had a rapid growth of YM, thereby limiting the shelf-life to less than 10 days of storage. Debbarma *et al.*<sup>58</sup> also achieved a shelf-life of 10 days of refrigerated storage of carrot juice treated by ohmic heating treatment at an electric field intensity of 17 V cm<sup>-1</sup> for a processing time of 40 s.

Enzyme activity is a crucial factor for determination of cloud stability and sensory acceptability of juice. The residual PME activity in untreated juice stored at 4 and 27 °C exhibited a decrease in activity. The ETTR and OHTR maintained low





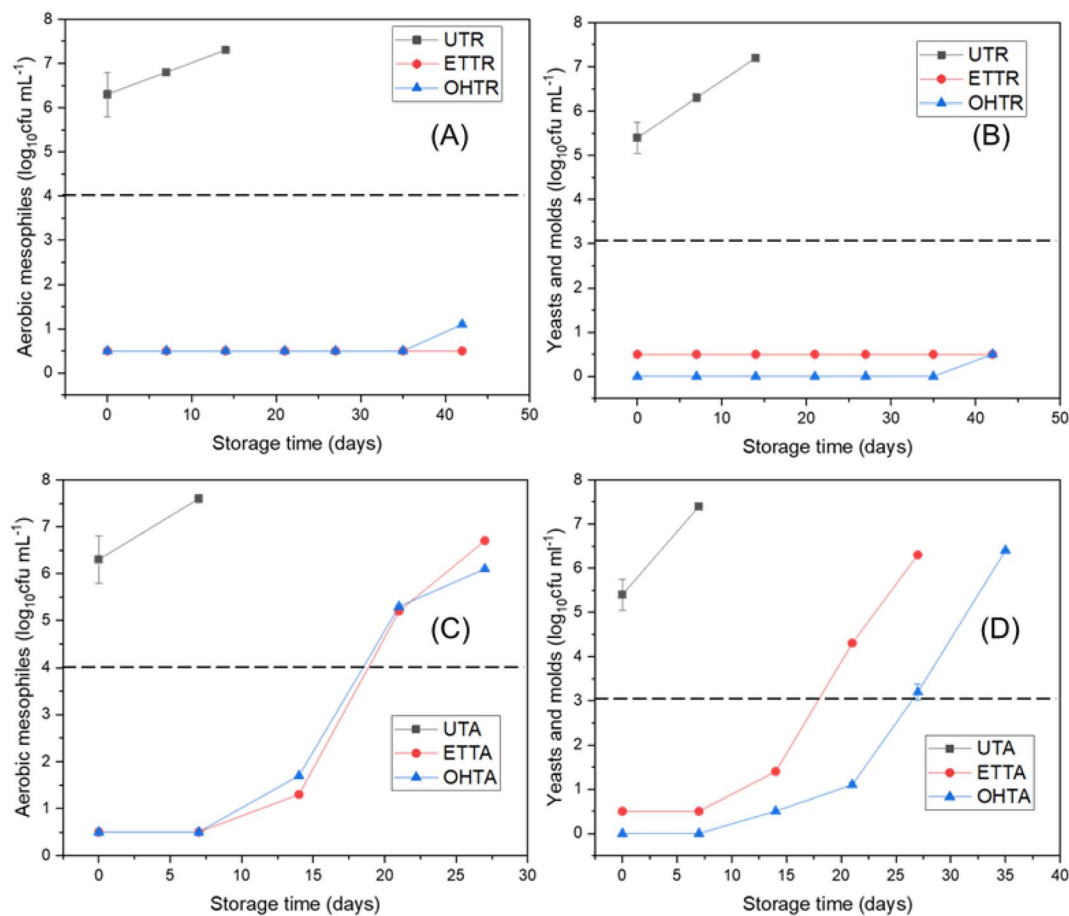


Fig. 5 The change in (A) aerobic mesophile and (B) yeast and mold count of the juice of the untreated, ohmic heating-pasteurized and thermally treated sample under refrigerated storage conditions. The change in (C) aerobic mesophile and (D) yeast and mold count of the juice of the untreated, ohmic heating-pasteurized and thermally treated sample under ambient storage conditions. UTR and UTA refer to the untreated juice stored under refrigerated and ambient conditions, ETTR and ETTA refer to the thermally treated juice stored under refrigerated and ambient conditions, and OHTR and OHTA refer to the ohmic heating-pasteurized juice stored under refrigerated and ambient conditions, respectively.

enzyme activities (<0.5%) throughout the refrigerated storage period of 42 days (Fig. 6). However, under ambient conditions, the ETTA and OHTA exhibited an increase in enzyme activities up to 10%. This indicates that the ohmic heating treatment successfully reduced the enzyme activity soon after the treatment and achieved a low enzyme activity throughout the storage period. Saxena *et al.*<sup>50</sup> reported a similar trend in PPO activity in ohmic heated sugarcane juice during refrigerated storage. This was also true in the case of PPO and POD in carrot juice after ohmic heating followed by storage.<sup>43</sup> A slight increase in PME activity after 2-week storage of a pasteurized orange-carrot juice blend was reported by Rivas *et al.*<sup>59</sup> In this study, the slight increase in PME activity might be due to the activation of the reversible latent form at ambient temperature.

**3.7.2 Color.** Initially, the  $L^*$ ,  $a^*$ , and  $b^*$  of the untreated sample were 16.13, 0.93, and 2.69, respectively. During storage,  $L^*$  and  $b^*$  values showed a decreasing trend and  $a^*$  values showed an increasing trend for both samples stored at 4 and 27 °C (Tables 3 and 4). This can be attributed to oxidative degradation and browning reactions, which often occur during juice storage, especially as lightness ( $L^*$ ) decreases and redness

( $a^*$ ) increases due to pigment changes and Maillard reactions. The  $\Delta E^*$  value of all the samples stored at 4 and 27 °C shows a decreasing order for both ETT and OHT samples. The  $\Delta E^*$  was in the range of 2.03 to 5.60 for the OHTR throughout the storage period. Similarly, the range of  $\Delta E^*$  was from 1.2 to 3.5 for the ETTR. The ETT samples exhibited a comparatively lower change in  $\Delta E^*$  in comparison to OHT samples throughout the storage period under refrigerated and ambient conditions. However, a relatively higher BI was observed in ETTR than in OHTR. The increase in the BI with the storage time can be ascribed to the diffusion of oxygen through the packaging material, which can degrade the phenolics and result in non-enzymatic browning. The presence of oxygen accelerates the degradation of phenolic compounds and promotes browning reactions, leading to the observed color changes. As already discussed, the initially high  $\Delta E^*$  of the treated juices at the start of the storage study was due to the increased  $L^*$  values, which decreased significantly during storage. Alcántara-Zavala *et al.*<sup>57</sup> also observed a decrease in  $L^*$  values of pulque juice after ohmic heating and thermal treatment for 22 days of refrigerated storage. The conventional pasteurization technique had a lower lightness value (26.79) as



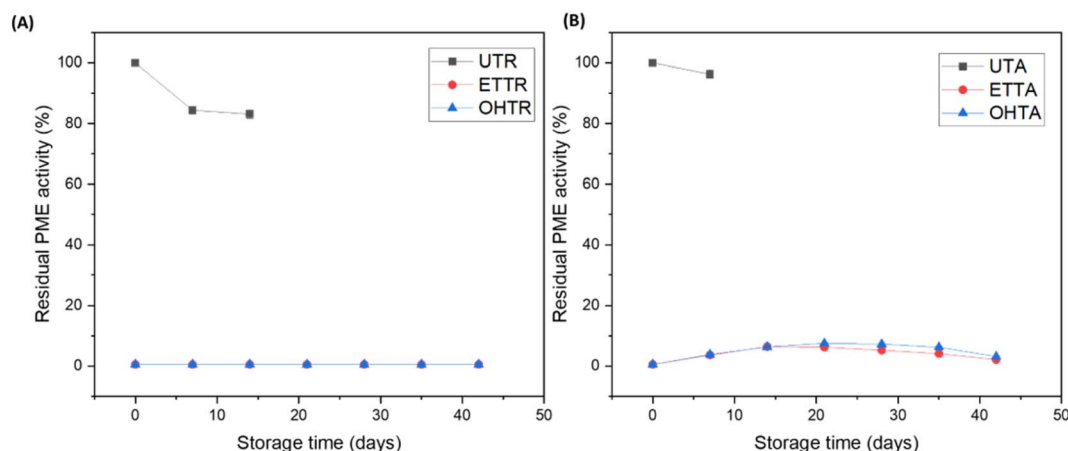


Fig. 6 The residual PME activity of the untreated, ohmic heating-pasteurized and thermally pasteurized sample under (A) refrigerated and (B) ambient conditions. UTR and UTA refer to the untreated juice stored under refrigerated and ambient conditions, ETTR and ETTA refer to the thermally treated juice stored under refrigerated and ambient conditions, and OHTR and OHTA refer to the ohmic heating-pasteurized juice stored under refrigerated and ambient conditions, respectively.

Table 3 The color parameters ( $L^*$ ,  $a^*$  and  $b^*$ ) of the mandarin juice pasteurized by ohmic heating (OHT) and equivalent thermal treatment (ETT) until 42 days of refrigerated storage<sup>a</sup>

		$L^*$	$a^*$	$b^*$	$\Delta E^*$	BI
UTR	0	$16.13 \pm 0.12^b$	$0.93 \pm 0.05^a$	$2.69 \pm 0.10^b$		$62.65 \pm 0.09^a$
	7	$14.13 \pm 0.11^a$	$1.05 \pm 0.11^b$	$2.28 \pm 0.12^a$	$2.05 \pm 0.11^c$	$62.83 \pm 0.09^a$
OHTR	0	$21.33 \pm 0.12^i$	$1.24 \pm 0.09^b$	$4.76 \pm 0.22^f$	$5.60 \pm 0.15^h$	$64.78 \pm 0.13^b$
	7	$20.81 \pm 0.11^h$	$1.26 \pm 0.08^b$	$4.21 \pm 0.14^e$	$4.93 \pm 0.16^g$	$64.03 \pm 0.11^b$
	14	$19.12 \pm 0.13^g$	$1.31 \pm 0.11^{bc}$	$4.19 \pm 0.18^{de}$	$3.37 \pm 0.18^f$	$64.86 \pm 0.15^b$
	21	$19.01 \pm 0.21^g$	$1.43 \pm 0.14^c$	$4.07 \pm 0.17^{de}$	$3.23 \pm 0.21^f$	$64.82 \pm 0.18^b$
	28	$18.71 \pm 0.19^f$	$1.56 \pm 0.11^c$	$3.91 \pm 0.21^{de}$	$2.92 \pm 0.15^e$	$64.80 \pm 0.16^b$
	35	$18.22 \pm 0.17^e$	$1.61 \pm 0.13^{cd}$	$3.86 \pm 0.13^{de}$	$2.49 \pm 0.15^d$	$65.03 \pm 0.09^b$
	42	$17.75 \pm 0.15^d$	$1.67 \pm 0.11^{cd}$	$3.67 \pm 0.15^{de}$	$2.03 \pm 0.12^c$	$64.96 \pm 0.11^b$
ETTR	0	$19.25 \pm 0.13^g$	$1.28 \pm 0.09^b$	$4.13 \pm 0.11^{de}$	$3.45 \pm 0.10^f$	$64.63 \pm 0.09^b$
	7	$18.91 \pm 0.22^f$	$1.35 \pm 0.21^{bc}$	$4.07 \pm 0.09^{de}$	$3.13 \pm 0.16^f$	$64.77 \pm 0.13^b$
	14	$18.65 \pm 0.23^f$	$1.47 \pm 0.12^c$	$4.01 \pm 0.13^{de}$	$2.89 \pm 0.16^c$	$64.93 \pm 0.14^b$
	21	$17.73 \pm 0.19^d$	$1.67 \pm 0.23^{cd}$	$3.91 \pm 0.16^{de}$	$2.14 \pm 0.15^c$	$65.49 \pm 0.13^b$
	28	$17.11 \pm 0.15^c$	$1.72 \pm 0.11^d$	$3.73 \pm 0.15^d$	$1.63 \pm 0.12^b$	$65.54 \pm 0.11^b$
	35	$16.65 \pm 0.32^b$	$1.85 \pm 0.22^d$	$3.61 \pm 0.22^{cd}$	$1.40 \pm 0.23^a$	$65.73 \pm 0.16^b$
	42	$16.19 \pm 0.29^b$	$1.87 \pm 0.17^d$	$3.49 \pm 0.18^c$	$1.24 \pm 0.08^a$	$65.78 \pm 0.12^b$

<sup>a</sup> Different superscript letters within one column indicate significant ( $p \leq 0.05$ ) differences among means determined by Tukey's test. The  $\Delta E^*$  and BI refer to the total color difference and browning indices, respectively. UTR: untreated juice stored under refrigerated conditions; OHTR: ohmic heating-pasteurized juice stored under refrigerated conditions; ETTR: thermally pasteurized juice stored under refrigerated conditions.

compared to the ohmic heating treatment (65 °C for 7 min) (28.95). Since both ohmic heated (OHT) and thermally treated (ETT) samples achieved complete enzyme inactivation, the better color retention in OHT samples during storage is likely due to the greater retention of phenolic compounds and ascorbic acid, which act as natural antioxidants.<sup>60,61</sup> These compounds help prevent oxidative browning over time. In contrast, the ETT samples, with lower initial phenolic content and longer exposure to heat (due to the extended come-up time of 12.1 min), may have experienced greater degradation of these protective compounds, leading to increased browning and a higher BI during storage.

**3.7.3 Total phenolic content.** Apart from microbial and enzymatic stability, retention of a high phenolic content at the

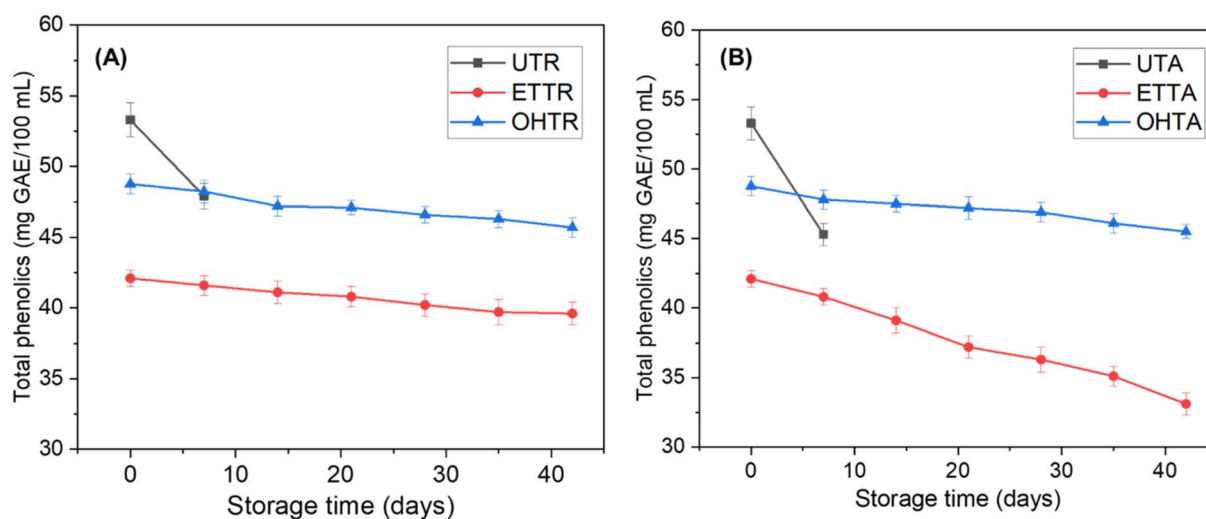
end of the respective shelf-lives is necessary for the preservation of the nutritional content of juices. The total phenolics content of untreated juice was 53.3 mg GAE/100 mL which was reduced to 47.2 and 45.3 GAE/100 mL for samples stored at 4 °C and 27 °C, respectively on the 8th day of storage. The initial phenolic content of OHT and ETT was 48.76 and 42.1 mg GAE/100 mL, respectively (Fig. 7). This further signifies that phenolics were preserved to a greater extent during the process of ohmic heating than in thermal treatment, probably due to the lower come-up time in the former. The degradation of phenolics in the samples stored at 4 and 27 °C showed the same trend; however, the samples stored at 27 °C exhibited a more significant degradation. The degradation of phenolics is enhanced at



**Table 4** The color parameters ( $L^*$ ,  $a^*$  and  $b^*$ ) of the mandarin juice pasteurized by ohmic heating (OHT) and equivalent thermal treatment (ETT) until 42 days of ambient storage<sup>a</sup>

		$L^*$	$a^*$	$b^*$	$\Delta E^*$	BI
UTA	0	$16.13 \pm 0.11^c$	$0.93 \pm 0.08^a$	$2.69 \pm 0.03^b$		$62.65 \pm 0.10^a$
	7	$13.12 \pm 0.15^a$	$1.07 \pm 0.09^b$	$2.01 \pm 0.15^a$	$3.09 \pm 0.13^d$	$62.70 \pm 0.12^a$
OHTA	0	$21.33 \pm 0.23^h$	$1.24 \pm 0.03^c$	$4.76 \pm 0.07^e$	$5.61 \pm 0.12^g$	$64.78 \pm 0.20^c$
	7	$20.65 \pm 0.22^g$	$1.29 \pm 0.08^c$	$4.17 \pm 0.13^d$	$4.77 \pm 0.16^f$	$64.06 \pm 0.19^b$
	14	$19.42 \pm 0.23^f$	$1.35 \pm 0.03^c$	$3.94 \pm 0.22^d$	$3.54 \pm 0.23^e$	$64.26 \pm 0.18^b$
	21	$18.46 \pm 0.24^e$	$1.46 \pm 0.02^d$	$3.81 \pm 0.05^d$	$2.64 \pm 0.18^c$	$64.61 \pm 0.18^b$
	28	$17.31 \pm 0.26^d$	$1.57 \pm 0.10^d$	$3.73 \pm 0.19^d$	$1.70 \pm 0.17^b$	$65.22 \pm 0.17^c$
	35	$15.12 \pm 0.11^b$	$1.69 \pm 0.05^{de}$	$3.42 \pm 0.12^c$	$1.46 \pm 0.11^{ab}$	$66.11 \pm 0.11^d$
	42	$14.89 \pm 0.13^b$	$1.77 \pm 0.04^e$	$3.31 \pm 0.19^c$	$1.62 \pm 0.16^b$	$66.12 \pm 0.10^d$
ETTA	0	$19.25 \pm 0.21^f$	$1.28 \pm 0.09^c$	$4.13 \pm 0.17^d$	$3.45 \pm 0.19^e$	$64.63 \pm 0.16^c$
	7	$18.85 \pm 0.20^e$	$1.37 \pm 0.12^{cd}$	$4.01 \pm 0.13^d$	$3.06 \pm 0.08^d$	$64.71 \pm 0.15^c$
	14	$18.32 \pm 0.20^e$	$1.51 \pm 0.13^d$	$3.84 \pm 0.04^d$	$2.54 \pm 0.17^c$	$64.81 \pm 0.14^c$
	21	$17.65 \pm 0.19^d$	$1.69 \pm 0.09^{de}$	$3.62 \pm 0.22^c$	$1.94 \pm 0.16^b$	$64.94 \pm 0.19^c$
	28	$17.01 \pm 0.18^d$	$1.75 \pm 0.04^e$	$3.41 \pm 0.13^c$	$1.40 \pm 0.16^{ab}$	$64.92 \pm 0.13^c$
	35	$16.52 \pm 0.13^c$	$1.87 \pm 0.03^f$	$3.28 \pm 0.15^c$	$1.18 \pm 0.12^a$	$65.08 \pm 0.13^c$
	42	$15.65 \pm 0.21^{bc}$	$1.91 \pm 0.09^f$	$3.19 \pm 0.18^c$	$1.20 \pm 0.20^a$	$65.48 \pm 0.19^c$

<sup>a</sup> Different superscript letters within one column indicate significant ( $p \leq 0.05$ ) differences among means determined by Tukey's test. The  $\Delta E^*$  and BI refer to the total color difference and browning indices, respectively. UTA: untreated juice stored under ambient conditions; OHTA: ohmic heating-pasteurized juice stored under ambient conditions; ETTA: thermally pasteurized juice stored under ambient conditions.



**Fig. 7** The change in total phenolics of the untreated, ohmic heating-pasteurized and thermally pasteurized juice stored under (A) refrigerated and (B) ambient conditions. UTR and UTA refer to the untreated juice stored under refrigerated and ambient conditions, ETTR and ETTA refer to the thermally treated juice stored under refrigerated and ambient conditions, and OHTR and OHTA refer to the ohmic heating-pasteurized juice stored under refrigerated and ambient conditions, respectively.

higher temperatures due to the enhancement in the rate of the degradation reactions.<sup>62</sup>

Furthermore, a lower enzyme activity at lower temperatures can also be one of the possible reasons for lower degradation at 4 °C than at 27 °C. To summarize, the ohmic heating-treated sample had 12.5% greater phenolics than thermally treated juice at the end of their respective shelf-lives at 4 °C.

## 4 Conclusion

The ohmic heating rates increased with applied voltage reaching  $0.545 \text{ } ^\circ\text{C s}^{-1}$  at 200 V. The system performance

decreased with applied voltage; however, 88% efficiency was obtained in every case. The ohmic heating at 200 V/30 s produced a microbially safe (5 log reduction in natural microflora) and enzymatically stable (>99% inactivation of PPO, POD, and PME) mandarin juice. Under the pasteurized condition of 200 V/30 s, 90% vitamin C and 91% phenolics were retained in the juice. The juice treated at 200 V/30 s showed a shelf life of 42 and 15 days under refrigerated and ambient conditions, respectively. Further study should explore a continuous ohmic heating system for the juice and its industrial scale-up.



## Data availability

Data will be made available on request.

## Conflicts of interest

The authors declare no conflict of interest.

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