Effect of thermal and high pressure processing on antioxidant activity and instrumental colour of tomato and carrot purées

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ABSTRACT

Total antioxidant activity, levels of bio-active compound groups and instrumental colour of tomato and carrot purée subjected to high pressure treatment (400–600 MPa/15 min/20 °C) and thermal treatments (70 °C/2 min) were measured. Antioxidant activity in tomato and carrot purée was significantly higher (p < 0.05) than in untreated or thermally processed samples. High pressure treatments at 600 MPa retained more than 90% of ascorbic acid as compared to thermal processing in tomato purées. Heat treatments caused a rapid decrease in ascorbic acid (p < 0.05). Phenolic contents were in general un-affected by thermal or high pressure treatments. Colour parameters were significantly affected (p < 0.05) by thermal and high pressure processing. Principal component analysis (PCA) revealed that the first two components represented 97% and 92% of the total variability in instrumental colour parameters with respect to processing for tomatoes and carrots respectively.

Industrial relevance: This research paper provides scientific evidence of the potential benefits of high pressure processing in comparison to thermal treatments in retaining important bioactive compounds. Antioxidant activity (ARP), ascorbic acid, and carotenoids after exposure to high pressure treatments (400–600 MPa) were well retained. Our results also show that redness and colour intensity of purées were better preserved by high pressure processing than conventional thermal treatment. It would appear from a nutritional prospective, high pressure processing is an excellent food processing technology which has the potential to retain compounds with health properties in foods. Therefore high pressure processed foods could be sold at a premium over their thermally processed counterparts as they will have retained their fresh-like properties.

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1. Introduction

Consumers are demanding high quality and convenient products with natural flavour and taste, and greatly appreciate the fresh appearance of minimally processed food (Oey, Van der Plancken, Van Loey, & Hendrickx, 2008). In order to extend the shelf life of these products they are usually processed thermally using methods such as hot water immersion, however these treatments can cause a reduction in antioxidant capacity (Dewanto, Wu, Adom, & Liu, 2002). Tomato and carrot purée are a particularly good source of carotenoids/phenols which have been shown to have protective effects against cardiovascular disease, diabetes and stroke (Heinonen, Meyer, & Frank, 1998; Scalbert & Williams, 2000). High hydrostatic pressure processing uses water as a medium to transmit pressures from 300 to 700 MPa to foods resulting in a reduction in microbial loads and thus extending shelf life. This can be achieved without heating and therefore the method could be useful for preserving the antioxidant capacity of the foods (Cheftel, 1992; Farr, 2003; Mertens & Knorr, 1992). High-pressure technology has been adapted to the specific requirements of the food industry and a range of pressure-treated products, including fruit and vegetable juices, avocado sauce, stewed packed ham, cooked rice and marinated chicken meat has already been introduced in the European and American market.

High pressure treatment can also have advantages over thermal approaches with regard to the eating quality of the finished product (Deliza, Rosenthal, Abadio, Silva, & Castillo, 2005). This is because small molecules such as volatile flavour compounds and pigments connected with the sensory, eating quality of foods are unaffected by high pressure processing (Cheftel, 1995). While there is a growing (although still limited) body of evidence to suggest that high pressure treatments can inactivate heat resistant spore-forming microorganisms (see for example, Ahn, Balasubramaniam, & Yousef, 2006; Meyer, Cooper, Knorr, & Lelieveld, 2000, 2001; Opstal, Bagamboula, Vannuyse, Wuytack, & Michiels, 2004; Wilson & Baker, 1997), there is very little published data showing what quality improvements, if any, can be achieved relative to conventional thermal processing. HP processing could preserve nutritional value (Oey et al., 2008) and the delicate sensory properties of fruits and vegetables due to its limited effect on the covalent bonds of low molecular-mass compounds.

While many authors have assessed the effect of high pressure processing on the nutritional properties of foods such as antioxidant capacity, few authors have attempted to link their studies with quality...
measurements such as instrumental colour analyses. In this light a principal objective of the present study was to assess the effect of high pressure processing for retaining the antioxidant capacity while also monitoring instrumental colour parameters which can be linked to the visual quality of the purées an important parameter for consumer acceptance.

2. Materials and methods

2.1. Chemicals

2,2-Diphenyl-1-picrylhydrazyl (DPPH), pyrogallol, Folin–Ciocalteu reagent (2 N), sodium carbonate, gallic acid and l-ascorbic acid were obtained from Sigma Aldrich (Dublin, Ireland). Hexane, acetone, metaphosphoric acid and methanol (HPLC grade) were purchased from BDH England (Poole, BH15, ITC).

2.2. Preparation of vegetable purées

Tomatoes (Lycopersicon esculentum cv, Domestica) and carrots (Daucus carota L cv, (L. Monocoty Book, Berlin, France). To minimise oxidation of the purée during processing, it was vacuum mixed (Stephan mixer, Stephan U Söhne GmbH & Co., Hameln, Germany) at 500 rpm at 1 °C. Samples were vacuum packed using Vac-star S220 vacuum sealer (Vicquip Ltd., Dublin, Ireland) and stored at ~21 °C until required for thermal and non-thermal processing. The procedures of freezing, thawing (overnight) and freeze drying were applied to both unprocessed and processed purées.

2.3. High pressure and thermal processing treatments

After thawing overnight at 4 °C the vacuum packed samples (250 g) were placed in a high pressure vessel (100 mm internal diameter × 254 mm internal height, Pressure Engineered System, Belgium) filled with a mixture of water and rust inhibitor (Dowcal N, 60% w/v in distilled water) and subjected to pressures of 400, 500 or 600 MPa for 15 min at an initial ambient temperature of 20 °C. Time taken to reach the target pressure was approximately (8–12 s) and depressurisation took 10 s. Temperature in the sample chamber was monitored during processing. Temperature rises of 16, 19 and 22 °C were recorded during processing at 400, 500 and 600 MPa respectively.

Vacuum packed unprocessed purées (250 g) were boiled in water for 20–25 s at which time they had achieved a core temperature of 70 °C. They were held at this temperature until they had reached a time–temperature (T0, ≥2 min) equivalent to a six log reduction in numbers of vegetative cells of the target pathogen (L. monocytogenes), (FSAI, 2006) as monitored using E-lab time–temperature recorder (Ellab, Ltd., Norfolk, UK). Sample core temperature profile was recorded during the process, using an Ellab E-Val TM TM9608 data module (Ellab [UK] Ltd., Norfolk, England) connected to a laptop. A standard Ellab SSA-12080-G700-TS temperature probe was inserted through an Ellab GKM-13009-C202 packing gland (20 mm) into a vacuum bag. After high pressure and thermal treatments, samples were removed and freeze dried (Model No. A6 13, Frozen in Time Ltd., York, UK) at a temperature and pressure of ~50 °C, 0.03 mbar respectively for two days and tested for antioxidant indices and instrumental colour on the same day. Prior to any experiment, all Ellab unit probes were calibrated against JOFRA (ATC-155B) calibration unit at temperatures of 70 and 80 °C and all results associated with the calibration did not exceed ±0.1 °C.

2.4. Measurement of total antioxidant capacity and phenolic content

Methanolic extracts were prepared by adding 25 ml of HPLC grade methanol added to 1.25 g of freeze dried powder and homogenising for 1 min at 24,000 rpm using an Ultra-Turrax T-25 Tissue homogenizer (Janke & Kunkel, IKA®-Labor technik, Saufen, Germany). The samples were then vortexed with a V400 Multitude Vortexer (Alpha laboratories, North York, Canada) for 20 min at 1050 rpm and centrifuged for 15 min at 3500 rpm (MSE Mistral 3000i, Sanyo Gallenkamp, Leicestershire, UK). 10 ml of the sample was filtered through PVDF Acrodisc syringe filters (pore size 0.45 μm, Sigma, Ireland), and stored at ~20 °C for subsequent analysis. Total antioxidant capacity was measured using the DPPH assay as described by Goupy, Hugues, Boivin and Amiel (1999). Briefly 500 μl of diluted sample and 500 μl of the DPPH (0.238 mg/ml) working solution were added to a micro-centrifuge tube. After vortexing, the tubes were left in the dark for 30 min at room temperature after which the absorbance was measured against methanol at 515 nm using a spectrophotometer (UV-1700 Pharma Spec, Shimadzu, Milton Keynes). Antioxidant activities were expressed as the IC50 i.e., the concentration of antioxidant required to cause 50% reduction in the original concentration of DPPH. For ease of interpretation antiradical powers were also calculated and defined as the inverse of the IC50 value. Finally the antioxidant capacity of the extracts was compared to that of a synthetic antioxidant (Trolox) and expressed as Trolox equivalent antioxidant capacity values (TEAC).

Total phenols (TP) in purée were determined using the Folin–Ciocalteu reagent according to the method of Singleton and Rossi (1965). Briefly 100 μl of methanolic extract, 100 μl of MeOH, 100 μl Folin–Ciocalteu reagent (FC) and 700 μl of Na2CO3 were added to 1 ml microcentrifuge tubes and the samples were vortexed briefly. The tubes were then left in the dark for 20 min at room temperature. Following this, the samples were centrifuged (Eppendorf, Centrifuge 5417R., Germany) at 13,000 rpm for 3 min. The absorbance of the sample was read at 735 nm using aqueous Gallic acid (10–400 mg/l) as a standard. Results were expressed as mg of Gallic acid equivalent per 100 g of dry weight of sample.

2.5. Ascorbic acid analysis

Extraction of ascorbic acid was carried out using 6% metaphosphoric acid and 1.25 g of freeze dried powder as described for antioxidant extractions above. Ascorbic acid determination was carried out by reverse phase high performance liquid chromatography (HPLC) according to the method of Lee and Coates (1999) with slight modifications. A 10 μl of aliquot was injected into the chromatographic column. The chromatographic system (Shimadzu–Model no SPD–M10A VP, Mason Technology, Dublin 8, Ireland) consisted of a pump, a vacuum degasser, a Diode-Array Detector and it was controlled through EZ Start 7.3 software (Shimadzu) at 40 °C. A hypersil ODS column (15 cm × 4.6 cm, 5 μm, Supelco., US) fitted with hypersil ODS guard column (Gemini C18 [4 mm L×3.0 mm ID], Phenomenex., UK) was utilised with a mobile phase (isocratic) of 25 mM monobasic potassium phosphate adjusted to pH 3 at a flow rate of 1 ml/min at 40 °C. The detector was set at 245 nm. For quantification external calibration curves for ascorbic acid in metaphosphoric acid (6%) were prepared at concentrations from 25 μg/ml to 500 μg/ml. The total run time was 4.0 min.

2.6. Total carotenoid content

Total carotenoids were determined using the modified method described by Koca, Burdurulu and Karadeniz (2007). Extractions were carried out using 25 ml of hexane:acetone (7:3) and 0.5 g of freeze dried sample using conditions described for antioxidant extractions. The residue was re-extracted until it became colourless. The filtrates were combined in a separatory funnel and washed with 50 ml distilled water. The water phase was discarded and Na2SO4 (10 g) was added as desiccant. The hexane phase was transferred to a 50 ml volumetric flask and brought to volume with hexane. The absorbance of this solution was then determined at 450 nm using a UV–Vis spectrophotometer. External calibration with authenticated β-carotene standards solutions (0.5 μg)
ml–10 μg/ml) in hexane:acetone (7:3) was used to quantify carotenoids in the solutions. Carotenoid content was expressed as β-carotene equivalents (βCE) in mg/100 g dry weight of sample.

2.7. Instrumental colour analysis

The colour of the samples was measured using a Hunter-Lab colour meter (Hunter Lab DP-9000 colour difference meter, Hunter Associates Laboratory, Virginia, USA) fitted with a 2.5 cm diameter aperture. The instrument was calibrated using the black and white tiles provided. Colour was expressed in Hunter Lab units L*, a* and b*. Samples of purée were filled into plastic Petri dishes (i.d. 50 mm) taking care to exclude air bubbles and placed under the aperture of the colour meter. Eight replicate measurements were performed and results were averaged. In addition, hue angle and chroma were calculated by the following equations.

Hue angle = tan⁻¹(b*/a*)  
(1)

Chroma = √a*² + b*²  
(2)

2.8. Statistics

Data are presented as means ± standard deviation of 3 observations, unless stated otherwise. Three observations of different samples for one treatment were carried out. Differences were considered significant at p < 0.05. A one way analysis of variance (ANOVA) was performed using the GenStat Release 10.1 (PC/Windows XP).The experimental design consisted of 5 treatments (unprocessed, thermally processed, HPP-400, HPP-500, and HPP-600 MPa). Where significant differences were present, individual treatments were compared using the least significant difference (LSD) test. Principal component analysis using “The Unscrambler” software (Version 9.7) was used to study colour parameters subjected to different treatments. As the dimensions of the variables (colour parameters) were different, the data were standardized by the software.

3. Results

3.1. Effect of thermal (TP) and high pressure processing on antioxidant activity of tomato purée

Anti-radical power and other antioxidant indices of tomato purées subjected to thermal (end-point temperature 70 °C for 2 min) and high pressure treatments (400–600 MPa for 15 min) are presented in Table 1. A slight but non-significant decrease in anti-radical power was noted for methanolic extracts of thermally processed tomato purées as compared to the un-processed purée. Antiradical powers of tomato purées processed at 400–600 MPa were significantly higher than thermally processed samples (p < 0.05). In fact samples processed at 400 and 600 MPa had significantly higher ARPs than un-processed samples (p < 0.05). Phenolic contents reported elsewhere (Odriozola-Serrano, Solvia-Fourney, & Martin-Bells, in press; Podsdešek, Sosnowska, & Anders, 2003) and were largely unaffected by processing with the exception of samples processed at 600 MPa which had significantly higher phenolic contents than un-processed samples (p < 0.05). Levels of ascorbic acid and total carotenoids were also in the range of those reported elsewhere (Sánchez-Moreno, Plaza, De Ancos, & Cano, 2006). However, both these parameters were strongly affected by processing. For example, a significant reduction in ascorbic acid levels was noted for all treatments as compared to un-processed samples. This was particularly noticeable following thermal processing which resulted in a 46% reduction in ascorbic acid levels. However, after processing at 600 MPa over 93.7% of ascorbic acid was retained as compared to un-processed samples. A slight but significant decrease in carotenoid content occurred in samples processed at 400 MPa. At 600 MPa a large significant increase (172%) in carotenoids extracted occurred as compared to un-processed samples.

3.2. Effect of thermal and high pressure processing on antioxidant activity of carrot purée

Anti-radical power and other antioxidant indices of carrot purées subjected to thermal and high pressure treatments (400–600 MPa for 15 min at 21 °C) are presented in Table 2. Anti-radical powers of thermally processed samples were not significantly different to values for un-processed purées. However, ARPs of samples processed at 600 MPa were significantly higher than un-processed samples (p < 0.05). Phenolic contents of all high pressure processed samples were significantly higher than thermally processed samples (p < 0.05). Carotenoid content was particularly affected by processing in the tomato purées with all processed samples having higher carotenoid content than un-processed samples (p < 0.05). In fact at pressures of 600 MPa a 58% increase in carotenoid content was noted. Ascorbic acid levels were undetectable in all samples.

3.3. Effect of thermal immersion and high pressure processing on colour parameters of tomato purée

Instrumental colour parameters of tomato purée samples as affected by different processing methods are shown in Table 3. For tomato

<table>
<thead>
<tr>
<th>Samples</th>
<th>Anti-radical power (g/l)</th>
<th>Total phenols mgGAE/100 g</th>
<th>Total carotenoids mg/100 g</th>
<th>Ascorbic acid mg/100 g</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unprocessed</td>
<td>0.37 ± 0.04</td>
<td>360.56 ± 2.89</td>
<td>37.02 ± 3.07</td>
<td>204.83 ± 4.88</td>
</tr>
<tr>
<td>TP</td>
<td>0.34 ± 0.03</td>
<td>341.13 ± 4.83</td>
<td>33.40 ± 1.55</td>
<td>125.14 ± 5.17</td>
</tr>
<tr>
<td>HPP400 MPa</td>
<td>0.43 ± 0.01</td>
<td>337.36 ± 15.31</td>
<td>28.42 ± 2.65</td>
<td>115.25 ± 5.54</td>
</tr>
<tr>
<td>HPP500 MPa</td>
<td>0.40 ± 0.02</td>
<td>367.50 ± 17.58</td>
<td>30.25 ± 7.17</td>
<td>95.67 ± 3.71</td>
</tr>
<tr>
<td>HPP600 MPa</td>
<td>0.47 ± 0.03</td>
<td>371.73 ± 15.15</td>
<td>100.85 ± 0.11</td>
<td>192.13 ± 4.83</td>
</tr>
<tr>
<td>LSD</td>
<td>0.04</td>
<td>24.35</td>
<td>8.44</td>
<td>9.05</td>
</tr>
</tbody>
</table>

Values are means ± standard deviation, n = 3, expressed on dry weight basis. *Least significant difference (p = 0.05).
purées, colour intensity (chroma) was significantly higher for processed samples than un-processed samples in all cases \((p<0.05)\). This effect was particularly noticeable for samples processed at 400 and 500 MPa. Samples processed at 600 MPa had lower colour intensity values than all treatments. In fact redness as measured using hunter \(a^*\) values was significantly higher for all high pressure processed samples as compared to unprocessed samples \((p<0.05)\). Lightness of all processed tomato purées was lower than fresh samples \((p<0.05)\). This effect was particularly noticeable for water immersion cooked samples. For samples processed at 400 and 500 MPa, lightness values were higher than thermally processed samples \((p<0.05)\).

3.4. Effect of thermal and high pressure processing on colour parameters of carrot purée

Colour intensity for pressure treated carrot purées at 600 MPa was significantly higher as compared to all the treatments \((p<0.05, \text{Table 4})\). However, a significant decrease \((p<0.05)\) was observed for thermally processed purées in comparison to the unprocessed purée. On the other hand, redness of carrot purées was significantly higher for high pressure treatments with samples processed at 500 and 600 MPa showing the maximum effect. Although a slight but significant increase was also found for thermally processed purée \((p<0.05)\). Changes in lightness of carrot purées were quite apparent as indicated in Table 4 where \(L^*\) value of purées were significantly higher for all processed samples as compared to fresh samples \((p<0.05)\). For other colour variables (hue angle) an inconsistent effect of processing was apparent, therefore in order to better explain the effect of processing on instrument colour variables in the purées principal component analysis was applied to the data set. A more detailed discussion of the relationship between colour and processing as interpreted using PCA is included in the Discussion section.

4. Discussion

4.1. Study on carrot purée

While increases in antioxidant content as noted for tomato and carrot purées may appear to be difficult to explain in the present study, this effect has been well documented elsewhere and would appear to be related to an increase in extractability of antioxidant components following high pressure treatment rather than an absolute increase. For example, De Ancos, Gonzalez, and Cano (2000) reported that the carotenoid content of high pressure treated tomato purée (600 MPa/25 °C/10 min) was significantly higher than untreated purées. Sánchez-Moreno et al. (2006) also reported that carotenoid content in high pressure treated (400 MPa/25 °C/15 min) tomato purées was significantly higher than untreated or thermally processed samples. Oey, Loey, and Hendrick (2004) reported that the Trolox equivalent antioxidant capacity of carrot juice increased by HP treatment (100–800 MPa/from 30 up to 65 °C/max. 90 min). Anti-radical powers for 400, 500 or 600 MPa were 0.02, 0.03, 0.04 (g/l) \(^{-1}\). With 600 MPa showing an increase of 33.3\% \((p<0.05)\) as compared to unprocessed carrot purées. It has been suggested that food processing such as cooking or grinding might improve lycopene the major carotenoid present in tomatoes bioavailability by breaking down cell walls (Gartner, Stahl, & Sies, 1997). In addition, Nguyen and Schwartz (1999) suggested that homogenization and heat treatment disrupt cell membranes and protein–carotenoid complex, making carotenoids more accessible for extraction. It would appear from the results in the present study that high pressure has a greater effect on disrupting cell membranes and protein–carotenoid complexes than thermal treatment. High pressure processing can be used to inactivate microbiological contamination because it results in damage to cell membranes of contaminating micro-organisms. This has been demonstrated in a number of studies (Hartmann, Mathmann, & Delgado, 2006; Moussa, Perrier, & Gervais, 2007). Other authors have reported that high pressure processing induces textural changes in tomatoes due to physical disruption of cell membranes (Tangwongchai, Ledward, & Ames, 2000).

4.2. Study on tomato purée

Thermal processing did not have a deleterious affect on carotenoid level with non-significant changes \((p>0.05)\) taking place for tomato purées. Hsu (2008) also reported non-significant decrease in total carotenoid content of tomato purées. Ascorbic acid levels in the present study were much more susceptible to degradation following processing than carotenoid contents in the case of tomato purées. Other authors have also reported that ascorbic acid levels decrease following processing. Sánchez-Moreno et al. (2006) reported decrease in total ascorbic acid content in tomato purée after high pressure treatment of 400 MPa/25 °C/15 min. However, it must be noted that in the present study high pressure processing was much more effective than thermal processing in retaining ascorbic acid levels with over 93\% of the ascorbic acid being retained in samples processed at 600 MPa compared to a figure of 38.9\% for thermally processed tomato purées. Yen and Lin (1996) reported that the level of retention of ascorbic acid in guava purée followed the following decreasing order: 400 MPa, 15 min – 88–90 °C, 24 s – 600 MPa, 15 min. In the present study, levels of ascorbic acid were the in the order 600 MPa>water immersed purées>400 MPa>500 MPa. Among all the tomato purées, a positive correlation was seen between anti-radical power and carotenoids \((r=0.537, p=0.039)\) which agrees with the studies conducted by Sánchez-Moreno et al. (2006) who found similar correlations. Phenols were poorly correlated with anti-radical power. Since carotenoids are the major pigments present in both and carrots and tomatoes the consistent increases in hue angle may be a reflection of changes in total carotenoid contents as presented in Tables 1 and 2. For example, carotenoid contents of tomato purées were significantly higher than all other samples and this was reflected in the highest hue angle for these samples. However this effect was not consistent and it appears that a more complex mechanism may be required to explain variations in instrumental colour parameters for the purées.

4.3. Relationship between colour parameters and antioxidant activity

The hue angle and the colour intensity calculated from colour parameters \(L^*, a^*, b^*\) of different non-thermal and thermal treatments are illustrated in Tables 3 and 4, whereas antioxidant indices are shown in Tables 1 and 2 for tomato and carrot respectively. In the case of carrot purée, redness value increased significantly \((p<0.05)\) when subjected to high pressure treatments. This increase was reflected in high ARP values as shown in Table 2. In fact ARP levels of carrot purée were positively correlated with Hunter \(a^*\) values \((r=0.65, p<0.05)\). Furthermore, the colour of carrot purée \((a^*)\) is a direct reflection of carotenoid content which also increased significantly \((p<0.05)\) at higher pressure treatments. This was confirmed by a significant correlation \((r=0.58, p<0.05)\) between carotenoid content and Hunter \(a^*\) values. No significant correlation was observed between ARP and hue angle. Similarly, redness of tomato purée increased at pressure treatments as compared to unprocessed samples. Higher redness at pressure

<table>
<thead>
<tr>
<th>Sample</th>
<th>(L^*)</th>
<th>(a^*)</th>
<th>Colour intensity</th>
<th>Hue angle</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unprocessed</td>
<td>31.46±0.014</td>
<td>14.61±0.027</td>
<td>35.38±0.011</td>
<td>50.61±0.08</td>
</tr>
<tr>
<td>TP</td>
<td>31.60±0.028</td>
<td>15.04±0.070</td>
<td>34.70±0.047</td>
<td>48.68±0.09</td>
</tr>
<tr>
<td>HPT400 MPa</td>
<td>31.71±0.005</td>
<td>15.02±0.010</td>
<td>35.09±0.011</td>
<td>50.14±0.02</td>
</tr>
<tr>
<td>HPT500 MPa</td>
<td>31.17±0.007</td>
<td>16.19±0.011</td>
<td>34.04±0.025</td>
<td>47.15±0.20</td>
</tr>
<tr>
<td>HPT600 MPa</td>
<td>32.35±0.014</td>
<td>16.06±0.035</td>
<td>35.50±0.046</td>
<td>48.58±0.05</td>
</tr>
<tr>
<td>LSD</td>
<td>0.12±0.01</td>
<td>0.01±0.00</td>
<td>0.03±0.00</td>
<td>0.05±0.00</td>
</tr>
</tbody>
</table>

Values are means ± standard deviation, \(n=3\).

*Least significant difference \((p=5\%)\).
treatments may be due to better extractability of carotenoids due to disintegration of chromoplast (Fernandez Garcia, Butz, & Tauscher, 2001; Tauscher, 1998). Apparently, Hunter $a^{\ast}$ value was well correlated with carotenoid content ($r=0.62$, $p<0.05$), whereas good correlation was also found for ARP and redness ($r=0.56$, $p<0.05$), indicating that ARP levels in tomato purée significantly influenced redness of purée.

4.4. Principal component analysis studies on colour parameters

Principal component analysis (PCA) was conducted to analyse the relationships between instrumental measurements and processing treatments for tomato and carrot purée samples. The dominant colour of tomato purée and carrot purée is a mix of red and yellow. Thus, Hunter $L^{\ast}$, $a^{\ast}$ and $b^{\ast}$ values or some combination of $a^{\ast}$ and $b^{\ast}$ should be considered as the physical parameters to describe the visual colour. Different combinations of $L^{\ast}$, $a^{\ast}$, $b^{\ast}$ values ($L^{\ast}$,$a^{\ast}$,$b^{\ast}$, hue angle, colour intensity) were selected to represent tomato colour change after thermal treatment and high pressure processing (Ahmed, Shivhare, & Mandeep, 2002; Ahmed, Shivhare, & Raghavan, 2004; Ahmed, Shivhare, & Singh, 2004) and subjected to PCA. The PCA plots provided an overview of the similarities and differences between samples after thermal and high pressure treatments (400–600 MPa). The PCA score plot for tomato purées (PC1 vs PC2) is shown in Fig. 1a. The first principal component explained 64% of the total variance in the data set with the second explaining 33%; the cumulative explained variance for each additional PC is shown graphically in Fig. 1b. Five groups of samples were visible in score plots for PCs 1 and 2 (Fig. 1a) with the replicates for each sample type being clustered tightly together. A number of major features are apparent from this plot. Firstly, the fresh samples are clustered separately from all of the processed samples in the lower left-hand quadrant of the plot indicating a change in values for the measured parameters after processing. Secondly, scores for one of the processing treatments (HPT600 MPa) are located quite some distance away from all of the other sample groups, indicating that the parameter values after this treatment are quite different from the other treatments, even the other high pressure treatments. Thirdly, it is not apparent that there is any simple linear relationship between the pressure applied during the high pressure treatments and the score plot (i.e. parameter value) locations. It is also clear that processing of any kind generally results in a shift to positive values on PC1 for the tomato purée samples; interestingly, this shift is greater in the case of the thermal treatment than for the high pressure treatments.

PC1 for this sample material is essentially defined by the difference between data recorded on fresh and TP; Fig. 1c shows that the parameters which are important in defining this PC are chroma, $a^{\ast}$ and $L^{\ast}$; hue angle plays almost no role. This suggests that, as tomato purée samples are processed, the values for these chroma, $a^{\ast}$ and $L^{\ast}$ parameters increase; this effect is confirmed in Table 3 for chroma and $a^{\ast}$, although it is not the case for the $L^{\ast}$ parameter, suggesting that changes in this parameter are less important in describing the total variance in the data set. Colour intensity of HPT500, HPT400 or thermally treated purées was significantly higher as compared to unprocessed samples. Some researchers have reported that green colour of vegetables for example green beans tends to get more intense after pressure treatment of 500/1 min/ambient temperature (Krebbers, Matser, Koets, Bartels, & Van den Berg, 2002). The major feature described by PC2 reflects differences between the 600 MPa high pressure treatment and the lower pressure treatments and fresh material. In this case, hue angle values act to produce high scores on PC2 in opposition to values for chroma, $a^{\ast}$ and $L^{\ast}$; this effect is borne out by an examination of the value of these parameters in Table 1. However, the magnitude of the eigenvalues for these latter three parameters is much lower than the positive values for the other two (Fig. 2c). PCA analysis revealed that, thermal treatment had a strong effect on $a^{\ast}$ value and chroma, with values increasing significantly ($p<0.05$) as compared to fresh counterparts. Thermal processing inactivates enzyme pectin-methylesterase, which may decrease methoxyl content (degree of esterification) of pectin molecules to make them more sensitive to crosslinking with calcium ions, hence may affect colour of tomato purée giving greater redness compared to fresh tomato purées (Bao & Chang, 1994). However pressure treatment of 500 MPa caused an increase

![Fig. 1. Principal components analysis (PCA) plots of instrumental colour parameters of tomato purées subjected to thermal and non-thermal (high pressure processing 400–600 MPa) treatments. a) PCA scores plot of different treatments, b) cumulative explained variance plot for the PCA of colour parameters, c) eigenvalues along PC1, and d) eigenvalues along PC2.](image-url)
For carrot purées, the sum of principal components 1 and 2 (PC1 and PC2) accounted for 90% of variations among all the different treatments. PC1 explained the majority of the variations, comprising 52%, while 38% was due to PC2. The explained variance for each PC is shown in Fig. 2b. A similar trend was monitored in the case of carrot purée, where high pressure treatment at 600 MPa caused a significant increase in colour intensity as compared to fresh or thermally processed samples. This is evident in the red colour (a* value) of HP-treated tomato juice compared to conventional hotbreak, attributed to compacting and homogenising effects of the HP treatment. Colour shift in high pressure processed fruit, vegetables and their products can be attributed to textural changes; hence can affect some colour parameters.

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5. Conclusions

High pressure processed purées had significantly higher antioxidant capacities when compared to thermally treated samples (p < 0.05). This was reflected in better retention of carotenoids and ascorbic acid in high pressure treated samples compared to thermally processed samples in tomato and carrot purée samples (p < 0.05). Hence high pressure processing at moderate temperatures can maintain nutritional quality of purées and could be an alternative to thermal processing in producing products of high nutritive value. PCA analysis of the data group of tomato purée and carrot purée was divided into five categories based on colour variations among themselves. All colour parameters were separated based on different type of processing along PC1 and PC2 and more than 90% of the variation was explained. This provides a helpful tool for understanding the effect of processing on colour variation of tomato and carrot purée in a broader spectrum.

Acknowledgements

This project is funded under the Food Research Measure (FIRM) by the Irish Agriculture and Food and Fisheries Development Authority.

References
