

Understanding texture changes of high pressure processed fresh carrots: A microstructural and biochemical approach

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Abstract

The effects of high pressure processing on textural changes of fresh carrots were studied integrating microstructural and biochemical responses. For all conditions studied, a significant loss in hardness and increase in deformability during cutting was observed after 2 min of processing. Hardness losses of 5, 25 and 50% were found respectively for treatments at 100, 200 and 300 MPa (all at initial temperature of 20 °C). At higher pressure levels no further increase in texture losses occurred. There was limited evidence of recovery of hardness at 300 and 550 MPa.

Analysis of microscopy images provided insight into the mechanisms of textural changes, which included cell deformation related factors such as shape factor and elongation. Linear correlations between tissue hardness and the extent of cell wall breakage during cutting were observed. Textural changes of fresh carrot tissue due to high pressure processing were mainly associated with turgidity loss, a direct result of the applied hydraulic pressure. Changes in biochemical parameters (pectin methylesterase and degree of methylation) were limited and did not highly contribute to texture change at the studied conditions.

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1. Introduction

High pressure processing (HPP) of vegetables has been described as a means of maintaining desirable textural characteristics due to limited tissue damage, cell wall separation and reduced biochemical changes compared to traditional treatments (Islam & Igura, 2003). However, other studies have shown that high pressure can alter phys-

ico-chemical properties of vegetable matrices by inducing changes in their structure (Basak & Ramaswamy, 1998; Butz et al., 2002; Préstamo & Arroyo, 1998). These textural changes are characterized by an initial texture loss, also called instantaneous pressure softening (IPS), followed by a gradual change during pressure hold. The gradual change was described as texture recovery, which in one study reached 100% at low pressures for long processing times (100 MPa/30 min) (Basak & Ramaswamy, 1998).

Microstructure changes are the main contributing factor to textural properties including cellular turgor and cell wall integrity (De Belie, 2002). During thermal or pressure processing, tissue firmness may be lost due to cell wall

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breakdown and loss of turgidity (Dörnenburg & Knorr, 1998; Sila, Smout, Vu, & Hendrickx, 2004). Studies done at the ultrastructural level using cryo-scanning electron microscopy have shown an increase in cell permeability, water movement and soaked appearance in high pressure processed (400 MPa, 30 min, 5 °C) cauliflower, while spinach leaves presented parenchyma disruption and cell collapse (Préstamo & Arroyo, 1998).

Biochemical changes also play an important role in texture changes. Studies done on carrot tissue have indicated that degradation of pectin only occurred in cooked and not in pressurized carrots. This led to a loss in firmness during cooking, while pressurized carrot did not present significant firmness loss; however, an increase in rupture strain was found above 200 MPa (Kato, Teramoto, & Fuchigami, 1997). The main contributors to cooking textural loss are a sequence of semipermeable membrane degradation, cell wall separation and final collapse of the pectin network (which is responsible for cell adhesion) (Lillford, 2000). Added to this, it is important to remember that cell disruption not only influences texture but also a number of biochemical reactions (Aguilera, 2005). Enzymatic degradation shortens demethylated pectin chains resulting in drastic tissue softening. A two-stage sequence explains this tissue softening: a partial demethylation of pectins and associated methanol production as a result of pectin methylsterase activity, followed by depolymerization of the lower degree of methylated pectins and galacturonic acid by polygalacturonase (Vu et al., 2004). Overall, there is already very valuable literature on textural changes due to heat but there is less understanding of textural changes after high pressure processing of vegetables as such. Therefore, the aim of this study was to characterize the textural changes of pressure-treated carrots under ambient temperature conditions (where cooking effects on the cell wall chemistry do not occur) by combining and quantifying mechanical, microstructural and biochemical responses of the tissue with the purpose of understanding the mechanisms occurring for potential use as pre-processing or chilled products.

2. Materials and methods

2.1. Plant material

Carrots (*Daucus carota* L., var. Laguna) were bought at a local Leuven distributor (Belgium) and stored at 4 °C (for a maximum period of 5 days) until further use. Cylinders (12 mm diameter and 10 mm height) of cortex material were obtained by using a stainless steel cork borer (10 samples/treatment).

2.2. High pressure treatment

Carrot cylinders were vacuum packed in a double film polyethylene bag, then processed at pressure levels from 100 MPa to 550 MPa for 2, 10 or 30 min. The high pressure

vessel (Engineered Pressure Systems Intl., Temse, Belgium) of 590 mL volume was temperature controlled using a heli-coidal copper tube jacket in thermal contact with the outer wall of the vessel and connected to a heating/cooling unit (Cryostat TCPS, Kul Circulatiekoeler, Serie P6, Belgium). The pressure transmitting medium was a mixture of propylene and glycol (60% Dowcal N, The Dow Chemical Co., Horgen, Switzerland).

Temperature data were captured by using thermocouples fixed to the lid of the vessel. The maximum temperature reached during processing was 39 °C at 600 MPa, 30 min. Sensors were placed to monitor the temperature of the liquid, middle and the inner border of the vessel together with the pressure profile at 4 s intervals. In addition pressure data were captured (Fig. 1). Compression and decompression rates were 10 MPa/1.6 s and 10 MPa/s, respectively. Data were captured by a SCXI system using LabVIEW (National Instruments, Zaventem, Belgium).

2.3. Water content and water loss

To analyze the moisture content of a sample, carrot cylinders were placed in a 105 °C oven until constant weight was reached (ca. 5 h); each test was carried out in triplicate. To create samples with controlled water loss, carrot samples were placed in a glass desiccator that had been previously equilibrated to a relative humidity of $75.47\% \pm 1.11\%$ (Greenspan, 1977) at ambient temperature using a saturated salt solution (NaCl). The sample mass was monitored as a function of the incubation time and samples were removed at varying extents of moisture loss to a maximum of approximately 14% (based on initial mass).

2.4. Calcium soaking pretreatment

Carrot cylinders were packed in a double film polyethylene bag with a calcium chloride solution (0.5% w/v CaCl_2). A carrot:brine ratio of 12 g:12 mL was used. After packing, the samples were high pressure processed for 30 min and, after pressure release, left at room temperature for another 30 min, resulting in total contact time of 1 h before texture measurements were carried out.

2.5. Pectinmethylesterase (PME) activity assay

Processed and raw samples were frozen with liquid N_2 and stored at -80 °C until required for further analysis. The PME extraction was done by homogenizing 2.5 g of the frozen sample in a pestle followed by the addition of 0.2 M Tris to 1 M NaCl buffer (pH 8.0) in the proportion of 1 (sample weight, kg):1.3 (volume of buffer, L) at 4 °C. Residual PME activity was measured by titration of free carboxyl groups released from a pectin solution (apple pectin, 70–75% degree esterification at 22.5 °C, from Fluka Chemical, Switzerland) using a pH-stat titrator (Metrohm, Switzerland) and 0.01 M NaOH.

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