Food Chemistry 142 (2014) 19–26

Contents lists available at SciVerse ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem

Effect of ozone pre-conditioning on quality and antioxidant capacity of papaya fruit during ambient storage

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ARTICLE INFO

Article history:
Received 26 March 2013
Received in revised form 5 July 2013
Accepted 8 July 2013
Available online 17 July 2013

Keywords:
Physico-chemical
β-Carotene content
Firmness
Ascorbic acid content
Sensory attributes

ABSTRACT

The objective of this study was to compare the physico-chemical characteristics and antioxidant activity of ozone-treated papaya fruit and untreated fruit. Freshly harvested papaya fruit were exposed continuously to ozone fumigation (0, 1.5, 2.5, 3.5 and 5 ppm) for 96 h prior to ambient storage at 25 ± 3 °C and 70 ± 5% relative humidity (RH) for up to 14 days. The fruit exposed to 2.5 ppm ozone had higher levels of total soluble solids (25.0%), ascorbic acid content (12.4%), β-carotene content (19.6%), lycopene content (52.1%), and antioxidant activity (30.9%), and also reduced weight loss (11.5%) at day 10 compared to the control. The sensory attributes of papaya treated with 2.5 ppm ozone was superior in sweetness and overall acceptability. These results support the application of ozone as a non-thermal and safe food preservation technique for papaya which can benefit both the producers and consumers.

1. Introduction

Epidemiological studies indicate that high fruit and vegetables consumption contribute to the prevention of chronic diseases, including cardiovascular disease and certain types of cancer (Arai et al., 2000). Papaya is a tropical fruit with important antioxidant properties and is in high demand in international markets. High amounts of antioxidants, both of hydrosoluble and lipid-soluble varieties, including vitamins C, E and A in papaya have numerous health benefits, ranging from reduced risks of developing cardiovascular diseases, macular degeneration to protection against colon and prostate cancer.

According to FAO statistic data, about 11.2 million metric tonnes of papaya produced in 2010 from 433,500 ha in the various countries. In 2010, Malaysia produced papaya for domestic and export market with about 46,000 metric tonnes (FAOSTAT, 2012). After Mexico, which supplies 40% of the world market, Malaysia is the second largest exporter of papaya, supplying 25% of the market. Brazil, the third largest exporter of papaya, gives greater priority to the hygienic aspect of the fruit, right from the moment it is harvested at the orchards until the stage where it reaches the packaging facilities. It is noted that agronomic conditions, variability among cultivars, divergence among seasons, stage of ripeness and postharvest manipulation, all could be possible factors explaining the differences of antioxidant capacity and quality of fruit (Kevers et al., 2007).

Ozone (O_3), or triatomic oxygen, is naturally produced from oxygen as a result of lightning or UV light interaction (Kim, Yousef, & Dave, 1999). The possibility of using ozone as an alternative sanitizing technology has the potential to replace regular sanitizers such as chlorine. Ozone at concentrations of 0.15–5.0 ppm was shown to inhibit the growth of spoilage bacteria as well as yeasts (Jay, Loessner, & Golden, 2005). Ozone treatments have been effective in extending the shelf-life of oranges, raspberries, grapes, pears and apples (Skog & Chu, 2001). Ozone may induce oxidative stress in fresh fruit, which promotes various physiological responses, including synthesis of antioxidants, polyamines, ethylene, phenolic compounds, and other secondary metabolites (Forney, 2003).

Information available on changes of individual phenols and carotenoids in papaya during ozone treatment is limited. The objective of this study was to investigate the effects of exposure to ozone on the physical quality parameters (weight loss, fruit firmness, peel colour), chemical composition (total phenolic content, ascorbic acid content, titratable acidity, soluble solids concentration, β-carotene, lycopene content) and antioxidant activity of papaya fruit.

2. Materials and methods

2.1. Plant material

Mature green papaya cv 'Sekaki' of colour index 2 (green with trace of yellow) were obtained from a local fruit wholesaler at

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http://dx.doi.org/10.1016/j.foodchem.2013.07.039

Pasar Borong Selangor, Malaysia on the day of harvest. Fruit of uniform size (800–1200 g), shape, maturity and free from any indication of mechanical injury, insect damage/infestation or pathogenic infection were selected for the experiment. Fruit were washed with clean distilled water and air-dried at ambient temperature (25–28 °C) before 96 h of ozone exposure. Fruit were evaluated following 0, 2, 4, 6, 8, 10, 12 and 14 days of ambient storage (25 ± 3 °C, 70 ± 5% RH).

2.2. Ozone fumigation

The fumigation system, comprised of chambers (112.0 cm length × 47.0 cm width × 43.0 cm height) constructed from 5 mm thick polycarbonate equipped with four 12 V fans positioned directly below the sample platform to ensure a well-mixed atmosphere, was located in the postharvest laboratory, School of Biosciences, The University of Nottingham Malaysia Campus. Ozone was introduced to each chamber by an ozone generator (Model MedKlinn Professional Series, MedKlinn International Sdn. Bhd., Malaysia) and the ozone concentration was controlled manually using a sensor (Eco-Sensor, Model OEM-2, MedKlinn International Sdn. Bhd., Malaysia). The ozone concentration in the chamber was recorded via an ozone analyser (Model IN-2000 L2-LC, IN USA Incorporated, USA). The chambers were maintained at 25 ± 3 °C and 70 ± 5% RH, and monitored with the aid of temperature/humidity sensors (Model U14-001, HOBO LCD Data Logger, Onset Computer Corporation, USA). A replicate of 10 fruits were placed in a single layer of the chamber. Each chamber was prepared as a replicate. There were four replications for each ozone treatment. The fruit was fumigated with ozone concentrations of 1.5, 2.5, 3.5 or 5 ppm for 96 h. Fumigation period of 96 h was chosen because it had shown the best suppression of papaya anthracnose disease in earlier study done by Ong, Kazi, Forney, and Ali (2012).

2.3. Physical quality parameters

Three fruits in each replication were marked and weighed (using a digital balance, EK-600H, Japan) before storage for weight loss determination. The fruit were then weighed at 2-day intervals over the storage period. The same three fruit in each replication were weighed repeatedly throughout the storage period. The results were expressed as percentage weight loss relative to the initial weight.

Fruit firmness was assessed using the Instron Universal Testing Machine (Model 5540, USA). Three fruits in each replication were penetrated using the 50 mm diameter probe, at a speed of 50 mm min^{-1} on three points in the equatorial region of the whole piece of fruit. The compression force measured at the maximum peak of the recorded force on the chart was expressed in Newtons (N).

Fruit peel colour on three points of the whole piece of fruit was determined using the Minolta CR-300 Chroma Meter (Minolta Corp., Japan). The colour determination made on papaya peels was expressed as values of lightness (L^*), chroma (C^*) and hue angle (h°). The values of L^*, forms the vertical axis with values ranging from 0 = black to 100 = white, a^* (red–green axis) and b^* (yellow–blue axis) which represent coordinates in the colour chart indirectly reflecting C^* and h°. The C^* which refers to the vividness of colour was computed from values of a^* and b^*, i.e. $C^* = (a^{*2} + b^{*2})^{1/2}$, which represents the hypotenuse of a right angled triangle with values ranging from 0 = least intense, to 60 = most intense. The h° was referred to as colour, and was the angle of tangent$^{-1} b^*/a^*$, where 0° = red purple, 90° = yellow, 180° = bluish-green and 270° = blue.

2.4. Chemical quality parameters

During storage, the soluble solids concentration (SSC), titratable acidity and ascorbic acid of the fruit pulp were analysed. Fruit pulp samples from each replication (10 g) were homogenised using a kitchen blender (Model HR2094, Philip, Malaysia) with 40 ml of distilled water, and filtered through muslin cloth. The fruit pulp was sampled from the equatorial region of the whole piece of fruit and was a pooled sample from each replicate. A drop of the filtrate was then used to determine the SSC (°Brix) using a Palette Digital Refractometer (Model: PR-32α) from Atago Co, Ltd (Japan), which was calibrated with distilled water prior to taking readings. The readings were multiplied by the dilution factor to obtain the original SSC (%) of the papaya pulp.

The titratable acidity (TA) was analysed using the titration method described by Ranganna (1999). Ten grams of pulp tissue were homogenised with 40 ml of distilled water using a kitchen blender (Model HR2094, Philip, Malaysia) for 2 min. The mixture was then filtered through muslin cloth. Five millilitres of filtrate with one to two drops of 0.1% phenolphthalein as a pH indicator was titrated against 0.1 N sodium hydroxide (NaOH) to a pink endpoint (at least for 10 s). Titratable acidity was expressed as percentage of citric acid equivalents.

Ascorbic acid was determined by using the 2,6 dichlorophenol-indophenol dye titration method (A.O.A.C., 1990). Samples were prepared with 10 g of fruit pulp tissue from three fruit, homogenised in 90 ml of 3.0% metaphosphoric acid (HPO_3) solution and the filtrate transferred into a 100 ml volumetric flask. The volume was made up to 100 ml using 3.0% HPO_3. An aliquot of the sample (10 ml) was taken and titrated against 2,6 dichlorophenol-indophenol dye until a pink colour persisted for 15 s. The ascorbic acid content was expressed as mg per 100 g of fresh fruit.

2.5. β-Carotene and lycopene content

Carotenoids were determined by the method described by George, Tourniaire, Gautier, Groupy, and Rock (2011). Ground papaya pulp tissues (10 g) from each replication (four replicates) were mixed with 50 ml acetone for 20 min in the dark and the mixture was filtered through carded cotton. Carotenoids from the remaining material were subsequently extracted in the same way twice by mixing with 30 ml acetone for 5 min and combining the filtrates in a separating funnel. To the combined filtrates 75 ml petroleum ether was added and the organic phase was washed three times with 50 ml water. Remaining water was removed with anhydrous sodium sulphate, and the volume was made up to 100 ml with petroleum ether. The optical density of the layer was measured with a spectrophotometer (Biochrom, Libra S12, UK) at 450 and 503 nm using petroleum ether as a blank. The concentrations of lycopene and β-Carotene were determined spectrophotometrically using the following equations (Lime, Griffiths, O'Connor, Heinzelman, & McCall, 1957):

$$[C]_{\beta\text{-carotene}} = 4.624 \times A_{450} - 3.091 \times A_{503}$$

$$[C]_{\text{lycopene}} = 3.956 \times A_{450} - 0.806 \times A_{503}$$

where $[C]$ is the concentration of carotenoid expressed in mg100 g^{-1} Fresh weight (FW), and A_{450} and A_{503} represent the absorbance at 450 nm and 503 nm, respectively.

2.6. Phenolic content and antioxidant activity

The fruit extract was prepared using the modified method of Chew et al. (2011). Papaya pulp tissues (2 g) from each replication (four replicates) were homogenised in a glass tube with 10 ml of