



# Effect of citric acid combined with UV-C on the quality of fresh-cut apples



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## ABSTRACT

The aim of this study was to evaluate the effect of 0.5% citric acid (CA), UV-C alone, and their combination (CA + UV) on native microflora survival, phenolic composition, browning, firmness and weight loss of fresh-cut 'Fuji' apples during refrigerated storage. Results showed that CA, UV alone and UV + CA had no significant effect on the firmness and total phenolic content but increased the polyphenol oxidase activity of fresh-cut apples. CA alone aggravated the browning and increased the weight loss of fresh-cut apples, while UV or UV + CA treatment significantly decreased the occurrence of browning reactions and reduced the weight loss as compared to control. The bacterial count of fresh-cut apples were reduced by 1.5, 2.1 and 2.6 log CFU/g after treatment with CA, UV and CA + UV, respectively. All these treatment retarded the microbial growth during storage and CA + UV treatment maintained the lowest growth rate among all treatment samples. Furthermore, the content of chlorogenic acid, (–) epicatechin, (+) catechin and caffeic acid in CA + UV treated sample was higher than the control after 15 d storage. Overall, these results suggest that CA + UV might be applied as an effective and safe preservative for fresh-cut apples.

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## 1. Introduction

Consumption of fresh-cut fruits and vegetables has become increasingly popular over the past decade due to greater interest in healthy and nutritious diets and changes in consumer lifestyles (Sipahi et al., 2013). However, processing operations such as peeling, cutting, and slicing lead to tissue damage, thereby causing adverse reactions in the produce. The various detrimental changes include browning, off flavor, loss of texture, and higher microbial load (Azarakhsh et al., 2014). Given that fresh-cut products are ready to eat and not subjected to further microbial killing steps, high quality ingredients, good manufacturing hygienic production and correct storage are very important to ensure product safety (Little et al., 2007). Increased research efforts are now being focused on various means to impart the desired decontamination effect without impairing the fresh-like attributes of the produce.

Currently, the use of chlorine as a produce disinfectant is probably the most widely adopted method in the produce industry and it would not be possible to sell fresh-cut salads and vegetables at markets without chlorine (Gil et al., 2009). However, the use of chlorine and chlorine-based derivatives for washing is becoming

increasingly challenged. Considerations have included public health concerns with chlorine and its by-products and an increasing awareness of the negative environmental impact of chlorine (Parish et al., 2003). Citric acid is generally recognized as safe (GRAS) for use as a food ingredient and is often used to control the microbial contamination of fresh-cut fruits and vegetables (Park et al., 2011; Rahman et al., 2011). It is one of the organic acids that are naturally present in fruit and it does not adversely affect taste and flavor of plant commodities. Jiang et al. (2004) and Queiroz et al. (2011) reported that the application of citric acid can prevent browning and maintained the quality of fresh-cut samples.

Shortwave ultraviolet radiation (UV-C) is an alternative and inexpensive method to reduce the number of microorganisms on the surface of fresh-cut fruits and vegetables. UV-C treatments (0.5–20 kJ/m<sup>2</sup>) decrease microbial growth by inducing the formation of pyrimidine dimers that alter the DNA helix and block microbial cell replication (Nakajima et al., 2004). It does not leave residues and has been claimed as forming only non-toxic by-products. The United States Food and Drug Administration has approved UV-C light as a disinfectant technology for surface treatment of food (Manzocco et al., 2011a). Although it has been claimed that UV-C treatment does not produce undesirable byproducts that could change taste, odor and color, some authors as reviewed by Shama and Alderson (2005) have reported skin discoloration in tomatoes, browning of calyxes in strawberries and

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increasing susceptibility to brown rot in peaches. The influence of UV-C on the quality (color, texture, taste and aroma) of fresh-cut apples during storage are also quite diverse depending on the cultivar of apples and the dosages UV applied (Manzocco et al., 2011a; Gómez et al., 2010).

Fresh-cut apples, which contain antioxidants and other nutrient components, have recently emerged as popular snacks in food service establishments, school lunch programs, and for family consumption (Guan and Fan, 2010). They generally have a short shelf life because of enzymatic browning, tissue softening, and microbial growth (Tappi et al., 2014). Therefore, the present study was conducted to evaluate the effect of CA and UV alone and in combination on microbial growth, total phenolics, phenolic composition, browning and firmness of fresh-cut “fuji” apples during storage at 5 °C.

## 2. Materials and methods

### 2.1. Sample preparation and treatments

Apples (Fuji) were purchased from a local wholesale distributor at commercial maturity. Apples were selected for uniform size and appearance. These fruit were rinsed gently with tap water by hand and dried naturally. The apples were then peeled, cored and cut into 1 cm thick cubes (average weight at  $1 \pm 0.3$  g) with a sharp stainless steel knife. The knife and cutting board were washed with deionized water and rinsed with 1000 µL/L sodium hypochlorite solution prior to use.

Fresh-cut apples were divided into four groups for different treatment: (a) dipped in 0.5% citric acid (CA) solutions for 5 min and air-dried; (b) each side of fresh-cut apples exposed to UV-C lamp (maximal emission at 253.7 nm, TUV-15W G13 T8 55 V, Philips, Holland) for 5 min; (c) dipped in CA and then exposed to UV-C; (d) untreated samples were used as control. After treatment, fresh-cut apples were placed in plastic foam tray (15 × 21 × 2.5 cm size) and wrapped with PE cling film and stored at  $5 \pm 2$  °C for 15 d.

### 2.2. Microbiological analysis

Microbiological analysis was carried out according to the method of Wu and Chen (2013) with slight modifications. Ten grams of samples were combined with 50 mL of 0.1% peptone and homogenized in a stomacher (BagMixer-400W, Interscience, France) at high speed for 1 min. The samples were serially diluted with 0.1% peptone water and surface plated (0.1 mL or 1 mL) in triplicate on plate count agar (PCA, Qingdao Hope Bio-Technology Co., Ltd., Qingdao, China) to detect total bacteria. 3 M<sup>TM</sup> Petrifilm<sup>TM</sup> EC (AOAC Official Method 991.14 and 998.08) to detect *Escherichia coli*/coliform, and 3 M<sup>TM</sup> Tecra<sup>TM</sup> Salmonella Visual Immunoassay (AOAC Official Method 998.09) for *Salmonella* spp. The plates for total bacteria were incubated at 37 °C for 48 h, and plates for *E. coli* and coliform were incubated at 35 °C for 24 h and 48 h, respectively. All microbiological analysis was carried out in triplicate and the results were expressed as log<sub>10</sub> colony forming units per gram (log<sub>10</sub> CFU/g). For *Salmonella*, the samples were primary enriched in Buffered Peptone Water for 24 h at 35 °C followed by the secondary enrichment in Rappaport Vassiliadis Broth for 20 h at 42 °C. For post-enrichment, samples were enriched in M-Broth for 8 h at 36 °C. The results were interpreted comparing wells of the samples to the well of positive control.

### 2.3. Total phenolics and phenolic composition

#### 2.3.1. Samples extraction

Fresh-cut apples were homogenised in a Waring blender and a 10 g sample was added to 40 mL of 80% ethanol. After 1 h of

extraction with magnetic stirring at 900 rpm at room temperature, the extract was centrifuged (Allegra X-30R, Beckman Coulter, USA) at  $10,000 \times g$  for 10 min at 4 °C. Supernatant was collected and residues were re-extracted twice. All three supernatants were combined for subsequent analyses.

#### 2.3.2. Total phenolics

Total phenolics content was determined according to the Folin–Ciocalteu method (Suresh et al., 2013). Extract (100 µL) was mixed with 3.9 mL of water, followed by addition of 250 µL of Folin–Ciocalteu reagent and 750 µL of sodium carbonate solution. The mixture was allowed to react in a vortex mixer and then incubated for 2 h at room temperature ( $22 \pm 2$  °C) in dark. Absorbance of the mixture was measured at 765 nm by spectrophotometer (Hitachi U-2800 Spectrophotometer, Japan). Total phenolics content was determined from a calibration curve prepared with standard gallic acid solution and was expressed as mass of gallic acid equivalents per fresh weight mass of fruit, mg/kg.

#### 2.3.3. Phenolic composition

Phenolic composition was measured on an Agilent Technologies (Waldbronn, Germany) 1100 Series HPLC system equipped with a photodiode array detector (DAD) according to Hagen et al. (2006) with some modifications. Ten microlitres of filtered samples were separated on a Hypersil BDS C18 column (250 mm × 4.6 mm, 5 µm) (Thermo, Bellefonte, PA, USA) eluted with a mobile phase consisted of 2% (v/v) acetic acid in water (solvent A) and 2% (v/v) acetic acid in water and acetonitrile (50:50, v/v, solvent B) at a flow rate of 16.67 µL/s. The phenolic compounds were eluted by a linear gradient from 10 to 55% B in 50 min and monitored at 254 nm, 280 nm and 320 nm. Standard curves for each polyphenol standard were prepared with five different concentration levels and triplicate injections at each level. The concentration of the phenolic compounds was expressed as the mass of phenolics per fresh weight mass of fruit, mg/kg.

### 2.4. Measurement of color

The color of fresh-cut apples was measured with a Minolta Chroma Meter Model CR-300 (Minolta, Tokyo, Japan), on using the CIELAB color parameters,  $L^*$  (lightness),  $a^*$  (green chromaticity), and  $b^*$  (yellow chromaticity). The results were expressed as a mean value from three replications of the 10 measured samples. Whiteness index (WI) and browning index (BI) were calculated as follows (Palou et al., 1999):

$$WI = L^* - 3b^*$$

$$BI = \frac{[100(x - 0.31)]}{0.172}$$

$$\text{where } x = (a^* + 1.75 L^*) / (5.645 L^* + a^* - 3.012 b^*)$$

### 2.5. Measurement of weight loss

Weight loss during storage of treated and untreated fresh-cut apples was recorded using a balance (Precisa 180 A, Switzerland) with a precision of  $\pm 0.0001$  g. Measurements were replicated 10 times. Results were expressed as percentage change in weight with respect to fresh sample without treatment or storage. The weight loss can be expressed in the following formula as:

$$\text{Weight loss(\%)} = \frac{(m_t - m_c)}{m_c} \times 100$$

where  $m_c$  is the initial weight of fresh-cut apples and  $m_t$  is the weight of sample at time  $t$ .

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