



Quality and biochemical changes of navel orange fruits during storage as affected by cinnamaldehyde -chitosan coating



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ABSTRACT

Quality and biochemical changes of navel orange (*Citrus sinensis* L., Osbeck) fruits in response to cinnamaldehyde-chitosan coating were studied during 120 day of storage at 10 ± 1 °C and 80–90% RH. The results showed that the coating significantly reduced the decay rate and weight loss of the navel orange fruits, delay the decrease of the content of total soluble solids (TSS), titratable acidity (TA) and vitamin C (Vc), effectively inhibited the content of MDA. Furthermore, the coatings maintaining enhanced the activity of SOD, CAT, POD and PPO, delay the senescence of fruits. The high performance liquid chromatography analysis showed that coating treatment inhibited the decrease of total sugar content of fruits and slowed down the decline of total organic acids by slowing down the degradation of sucrose, fructose and citric acid content. Compared with chitosan coating, cinnamaldehyde- chitosan coating could significantly reduce the decay rate and had no adverse effects on fruit quality. Meanwhile, it could improve CAT, SOD and POD activity, induce the activity of PPO to increase, and improve the disease resistance of navel orange fruits. This study suggests that cinnamaldehyde-chitosan coating can extend the storage time and maintain quality of citrus fruit.

1. Introduction

Fresh navel orange (*Citrus sinensis* L., Osbeck) is one of the consumers most favorite fruit for its fleshy crisp, slag, sweet flavor and other characteristics (Zeng et al., 2012). However, navel orange fruits can be harmed by the damage caused by postharvest diseases during storage and transport (Kouassiab and Jijakli, 2012; Zeng et al., 2010). Traditionally, synthetic fungicides are primarily used to control postharvest diseases of fruits, which can easily lead to residues of fruits and endanger consumers' health. In order to find new citrus fruit preservatives that are economical and safe, antibacterial agents such as plant extracts and safe compounds have been introduced as a novel and safe way to extend the post-harvest storage period of citrus (Talibi et al., 2014). Chitosan and carboxymethyl cellulose sodium as a substitute for chemical preservatives were applied to the Postharvest preservation of Citrus (Arnon et al., 2014). Adding carbonate and potassium sorbate in wax is a simple and effective measure to reduce the occurrence of citrus fruit disease (Youssef et al., 2012). The crude extracts of clove bud, *Impatiens balsamina* L. Stems and *Ficus hirta* fruit could inhibit the growth of pathogenic bacteria, reduce fruit decay, improve the quality of Citrus fruits during the storage (Chen et al., 2017; Chen et al., 2015;

Shao et al., 2015).

Guizhi is a dry twig of *Cinnamomum cassia* Presl. As a commonly used Chinese herbal medicine, it has the functions of dispersing cold and relieving pain, irregular menstruation and stopping throbbing (Liang et al., 2008; Zhou et al., 2007). Many studies have identified that the essential oil of cinnamon branch have strong antimicrobial activities (Bullerman et al., 1977; Murmu and Mishra, 2018), so they were used to control the decay and prolong the fresh-keeping period and storage period of fruits. In recent years, researchers found that the essential oil of *Cinnamomum cassia* has strong inhibitory effect on the growth of *Penicillium italicum* and *Penicillium digitatum* (Wan et al., 2017). The chemical constituents of the essential oil of *Cinnamomum cinnamomi* were analyzed by HPLC, and the main components were cinnamaldehyde, cinnamic acid, cinnamic alcohol and coumarin (He et al., 2005). Cinnamaldehyde, the main bacteriostasis component of cinnamon branch essential oil, could effectively enhance the protective effect of citrus fruit *in vivo* and induce the disease resistance of fruit (Duan et al., 2018; Tunc et al., 2007; Wu et al., 2017).

In order to develop cinnamaldehyde in the practical application of fresh citrus fruit, the Newhall navel orange as the research object, studied the effects of cinnamaldehyde - chitosan coating on storage

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quality of citrus fruit.

2. Materials and methods

2.1. Plant materials and experimental procedure

Fruits of navel orange (*C. sinensis* L. Osbeck cv. Newhall) were harvested at the commercially mature stage from Jun-ping Orchard in Nankang city, Jiangxi province, China. The fruits were uniformed in terms of shape, size, color and free of any damage or visual defects. After harvest, the fruits were immediately delivered to the laboratory at Jiangxi Agriculture University (Nanchang, China).

2.2. Preparation of coating

The chitosan was dissolved in 1% acetic acid solution Tween-80 and sodium chloride as co-solvents, stirring at room temperature to completely dissolve the final chitosan coating (CH) concentration of chitosan 1.5% (w/v). Cinnamic aldehyde was added to the chitosan coating to prepare Cinnamaldehyde-Chitosan coating (CI-CH) with cinnamaldehyde concentrations of 1.5% (v / v).

2.3. Fruit treatments

All navel orange fruit pre-cooling sweat 3 d, washed with tap water and air-dried. Fruit of each treatment were soaked either into water (control), 1.5% CH or CI-CH for 30 s. Following air drying, each treatment was composed of three replicates of 300 fruits each. The fruits were packed in perforated, low-density polyethylene bags (d = 0.04 mm) and stored at $10 \pm 1^\circ\text{C}$ and 90–95% (RH) in a fruit refrigerator. Samples were taken initially and at 15-day intervals during storage for quality parameters and other analysis.

2.4. Weight loss determination and decay rate

To determine decay rate and weight loss, samples of 100 fruits per replicate were taken separately during storage. The total fruit weight loss was calculated on initial weight basis and expressed in percentage. Decay rate was expressed by the percentage of fruits indicating fungal infection.

2.5. Fruit quality evaluations of fruit pulp

Quality parameters analysis was performed every 15 days during the storage. A homogeneous sample was prepared from 10 fruit per replicate for measuring Total soluble solids (TSS), Titratable acidity (TA) and vitamin C (Vc). TSS concentration was measured in fruit pulp juice with a RA-250WE digital brix-meter (KYOTO, Tokyo, Japan) TA expressed as percentage (%) of citric acid which was determined by the titration using a standard solution of sodium hydroxide (0.1 M). Vc was assayed by the titration using a solution of 2, 6-dichlorophenolindophenol and the value was expressed in mg ascorbic acid per 100 mL of pulp juice.

2.6. Soluble sugar and organic acid of fruit pulp

Extraction and determination of soluble sugar and organic acid were assayed according to (Chen et al., 2012). Four grams of frozen flesh sample was ground to a powder in liquid nitrogen, add in 5.0 ml of ethanol (80%) solution and water bathed at 35°C for 20 min. The extract was centrifuged at $10,000 \times g$ for 10 min. The residue was extracted twice and the supernatant was collected in volumetric flask to final 25 ml with 80% ethanol. 1 ml extract solution was dried by a vacuum centrifuge concentrator (Eppendorf Company, Germany), then added 1 ml distilled water to dissolve filtered with $\Phi 0.22 \mu\text{m}$, $\Phi 13 \text{ mm}$ water syringe filter (Shanghai Xingya Purification Material Factory, China).

The filtered solution was used for soluble sugar and organic acids analysis.

Toluble sugar and organic acids were measured using high performance liquid chromatography (HPLC, SHIMADZU LC-20 A, Japan). The liquid chromatograph equipped with a degasser, quaternary pump, 20 μL volume injection autosampler, a diode array detector and a differential detector.

The chromatographic conditions for determination of soluble sugar: The chromatographic column was ODS C18 column (4.6 mm \times 250 mm, Waters, USA), column temperature was 30°C ; mobile phase was acetonitrile and water (75: 25); flow rate was 1 mL /min; injection volume was 20 μL ; detection by detector of nitrogen differential detector.

The chromatographic conditions for determination of organic acids: The chromatographic column was ODS C18 column (4.6 mm \times 250 mm, Waters, USA), mobile phase was a solvent system of 50 mM (NH₄)₂HPO₄ phosphate buffer (pH = 2.7 adjusted with phosphoric acid) at a flow rate of 0.5 mL/min. Organic acids were detected at a wavelength of 210 nm. Lab Solutions Software (Shimadzu, Japan) was used to run the HPLC and process the results. Triplicate flesh samples were analyzed.

2.7. Malondialdehyde content of fruit peel

The content of malondialdehyde (MDA) was determined according to (Sofa et al., 2004) and was carried out by thiobarbituric acid colorimetric method.

2.8. Enzymes measurements of fruit peel

Activities of Catalase (CAT, EC 1.1.1.6), Peroxidase (POD, EC 1.11.1.7), Superoxide Dismutase (SOD, EC.1.15.1.1) and Polyphenol oxidase (PPO, EC.1.10.3.2) were assayed according to (Hirai and Ueno, 1977) as described by (Zeng et al., 2012).

2.9. Statistical analysis

The experiment was designed in a fully randomized way. All statistical analysis was carried out using SPSS 11 and Excel (2010). The statistical significance was applied at the level $P < 0.05$.

3. Results

3.1. Effect of different treatments on the decay rate and weight loss of navel orange fruit during storage

The decay rate of navel orange fruit increased continuously during storage (Fig. 1A). The fruits of control and CH were rotted at 30 d after storage, while CI-CH was at 60 d after storage. Compared with the control, the coating treatment could significantly reduce the decay rate, and the rate of CI-CH was significantly less than the 1.5% CH. At 120 d after storage, the decay rates of control, 1.5% CH and CI-CH were 13.67%, 10.33% and 6.33%, and the three were significantly different ($P < 0.05$).

With the prolongation of storage time, the weight loss rate of fruit increased gradually (Fig. 1B). Two coating treatments could reduce the weight loss of fruit, and the weight losses of 1.5% CH and CI-CH fruit were significantly lower than that of control ($P < 0.05$). At 120 d after storage, the weight loss of the fruit treated with 1.5% CH and CI-CH was 0.87% and 1.05%, only 46.52% and 56.15% of the control weight loss.

3.2. Effect of different treatments on the contents of TSS, TA and Vc in navel orange fruit during storage

The content of TSS accumulated in the fruit of navel orange

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