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Effect of chitosan/nano-silica coating on the physicochemical characteristics of longan fruit under ambient temperature

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ABSTRACT

A novel chitosan/nano-silica hybrid film was prepared using tetraethoxysilane as precursor by in situ sol–gel process, and characterized by transmission electron microscopy. Its effect on preservation quality of longan fruits (*Dimocarpus longan* Lour. cv Shijia) was investigated under ambient temperature. The present study revealed that the excellent semi-permeable film of chitosan/nano-silica markedly extended shelf life, reduced browning index, retarded weight loss and inhibited the increase of malondialdehyde amount and polyphenoloxidase activity in fresh longan fruit. In addition, the peroxidase activity of longan fruit coated with hybrid film was lower than that in other treatment fruits. Decreases in the contents of total soluble solids, titratable acidity and ascorbic acid were also significantly inhibited by hybrid films. These data indicated that the chitosan/nano-silica coating might provide an attractive alternative to improve preservation quality of fresh longan fruits during extended storage.

1. Introduction

Longan (*Dimocarpus longan* Lour.) belongs to a non-climacteric tropical and subtropical fruit, and is widely cultivated in many countries around the world, especially in China, Thailand and Vietnam (Jiang et al., 2002). Nevertheless, Fresh longan fruit has a very short shelf life, deteriorating within a few days under ambient temperature depending on the cultivar. The major factors reducing longan storage life and marketability are pericarp browning and microbial decay. This short shelf life greatly limits consumption, so a attention has been paid to the storage techniques of fresh longan fruits, including chemical reagent processing, edible coating, modified atmosphere packing and refrigeration (Tian et al., 2002; Jiang and Li, 2001; Thavong et al., 2010;). Recently, the most popular commercial means for prevention of longan fruit decay and browning was SO_2 fumigation and fungicide dips because they were effective and inexpensive (Jiang et al., 2002). However, SO_2 fumigation and fungicide leave sulphite residues and toxic ingredients that may have adverse effects on human health. As food safety awareness gradually increases in the international market, alternative means for decay control without toxic effects in harvest longan fruit are needed.

Application of edible coatings is promising to improve the quality and extend shelf life of fruits and vegetables (Lin et al., 2011). Chitosan, a versatile biopolymer derived from deacetylation of chitin had been widely applied in the fresh-keep field owing to its good biocompatibility, biodegradability, antibacterial activity and capacities to form film (Lin et al., 2011). Due to its unique physicochemical properties, it could help lessen moisture loss and slow respiration by reducing fruit oxygen uptake from the environment (Jiang and Li, 2001). The chitosan coating against a wide variety of microorganisms including fungi, algae and some bacteria had been also reported (Zheng and Zhu, 2003). Based on previous studies, chitosan coating had been successfully applied to prolong storage life and control decay of many fruits (Jiang and Li, 2001; Lin et al. 2011; Abbasi et al., 2009; Meng et al., 2008; Yu et al., 2012; Fisk et al., 2008; Dong et al., 2004). However, some physical properties such as unsound strength, water permeability and gas transmission of chitosan films had narrowed its application (Yeh et al., 2007).

To further improve the physicochemical properties of chitosan coating and prolong the fruit storage life, organic/inorganic hybrid materials are promising systems due to their synergistic properties. These are easily produced through sol–gel chemistry which is a very versatile method allowing incorporation of inorganic components like metal alkoxide namely, Si, Ti, Zr or even bioactive materials to modify final chemical and physical properties of

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materials (Yeh et al., 2007; Lai et al., 2006; Dhanasingh et al., 2011). The narrower distribution of particles and the reduced particle size due to in situ development of the metal oxide network leads to better strength. Incorporation of essential nano-silica into chitosan coating may greatly enhance the coating's antimicrobial properties (Dhanasingh et al. 2011), restrict enzyme immobilization (Lei et al., 2007) and inhibit fruit decay (Yan et al., 2011). By evaporating chitosan, porous silica with a high specific area is formed (Dhanasingh et al. 2011). Therefore, Chitosan/silica hybrid material can provide an alternative way as novel biomaterials for fruit storage application.

To date, there are few published data on the effects of chitosan/nano-silica composite on fruit storage (Yan et al., 2011; Yu et al. 2012), especially in fresh longan fruit. Therefore, the objective of this study attempts to elucidate the potential of chitosan/nano-silica coating on the most suitable extension of the storage shelf life of longan fruit, and further to investigate the fruit quality attributes. Finally, we investigated those together with the effects of chitosan/nano-silica coating on polyphenoloxidase (PPO) activity, peroxidase (POD), weight loss, pericarp browning index, malondialdehyde (MDA), total soluble solids (TSS), titratable acid (TA) and vitamin C (Vc) during longan storage under ambient condition.

2. Materials and methods

2.1. Materials

Fresh and mature longan fruits (*D. longan* Lour. cv Shijia) were harvested from 27th June to 5th July in a commercial orchard located in Zhanjiang, Guangdong province, China, and immediately transported to the Southern Subtropical Crop Research Institute of CATAS (Zhanjiang, China). The fruits were selected for uniformity in size, shape and color without signs of mechanical damage, blemishes, disease and pest damage. The storage temperature was maintained at 25 °C with ca. 65% relative humidity.

High molecular weight chitosan, with a deacetylation degree of 75.6% (CAS Number 9012-76-4, Batch 10305DD, Sigma–Aldrich, USA), and 98% glacial acetic acid are used to prepare the film-forming dispersions. Tetraorthosilicate (TEOS) from Sigma–Aldrich (USA) has 98% purity. All other chemicals belong to analytical grade.

2.2. Preparation of hybrid film

Chitosan/nano-silica hybrid film was prepared according to Yeh et al. (2007) with minor modification. Chitosan (2.0%, w/v) coating solution was dispersed in an aqueous solution of glacial acetic acid (0.5%, v/v). The solution was well stirred with a magnetic stirrer for 48 h at room temperature to form a homogeneous mixture. Required amount of this solution was taken in a 500 ml bottle, and measured amount of TEOS was added to it. And then, the solution was repeatedly stirred with a magnetic stirrer until the mixture solution became clear and coating formation. Stoichiometric amount of an equal mixture of ethanol and water was added to the solution and allowed to stir for 20 h at room temperature. The water to TEOS ratio was 1:4. The pH of the solution was adjusted to 5.6 with 1 M NaOH. The 500 ml final volume solution was vacuumed for 2 h to remove trapped air bubbles during the mixing.

2.3. Coating and storage conditions of longan fruits

Selected fruit samples were distributed randomly into four groups. Each group contained 100 fruits. Four different treatments were used: (1) control; (2) chitosan coating (2%); (3) nano-silica (30%) and (4) chitosan/nano-silica hybrid film. All the treatments were performed in triplicate. The fruits were dipped into the chitosan, nano-silica and chitosan/nano-silica solutions for 4 min. An aqueous solution of glacial acetic acid (0.5%, v/v) was used for the immersion of control samples. They were placed in polyethylene bags (100 fruits/bag) at room temperature with 70–80% relative humidity.

2.4. Browning assessment and weight loss

Pericarp browning was estimated by measuring the extent of total brown area on each fruit surface using the following scale: 1 = no browning; 2 = slight browning; 3 = <25% browning; 4 = 25–50 browning; 5 = >50 browning. The browning grade was calculated using the following formula: $\sum$(browning scale $\times$ percentage of corresponding fruit within each class). Fresh weight loss was monitored every two days during fruit storage at room temperature, and 30 fruits in each replication for each treatment were weighted from the beginning to the end of each storage period.

2.5. Extraction and assay of TSS, TA and Vc

TSS and TA were measured following the procedures as described by Fisk et al. (2008). Pulp (30 g) from 30 fruits was homogenized and then centrifuged at 15,000*g* for 20 min. The supernatant was collected to measure TSS using a digital refractometer (PAL-α, ATAGO, Tokyo, Japan). TA was determined by titration with 0.1 M NaOH to pH 8.2 and expressed as citric acid (mass/mass) on the basis of fresh weight.

The contents of Vc were measured by 2,6-dichlorophenolindophenol titration (Abbasi et al. 2009). Briefly, tissue (50 g) from 30 fruits was immediately homogenized in 50 ml of 0.02 g/ml oxalic acid solution and then centrifuged at 15,000*g* and 4 °C for 15 min. Total of 10 ml supernatant were titrated to a permanent pink color by 0.1% 2,6-dichlorophenolindophenol titration. Vc concentration was calculated according to the titration volume of 2,6-dichlorophenolindophenol and expressed as milligram per 100 g of fresh weight.

2.6. Extraction and assay of MDA content

MDA content was measured according to the reported method of Yang et al. (2010) with a slight modification. Pulp (3 g) from 30 fruits were homogenized with 15 ml of 10% trichloroacetic acid and centrifuged at 15,000*g* for 20 min. One milliliter of supernatant was mixed with 3 ml of 0.5% 2-thiobarbituric acid, heated at 95 °C for 20 min, and then immediately cooled in an ice-water bath. The absorbance was read at 532 nm after centrifugation at 3000*g* for 10 min and the value for non-specific absorbance 600 nm was subtracted. The amount of MDA was estimated as follows: (μM/g FW) = $[6.45\ (OD_{532} - OD_{600}) - 0.56\ OD_{450}] \times 5$ ml/0.25 g.

2.7. Extraction and assay of PPO and POD activities

For measuring PPO activity, pericarp tissues (4 g) from 30 fruits were homogenized with 0.1 M Na_2HPO_4-NaH_2PO_4 buffer (pH 6.8) containing 0.2 mM EDTA and 2% insoluble polyvinylpyrrolidone in a chilled pestle and mortar. The homogenate was centrifuged at 12,000*g* for 20 min at 4 °C and the supernatant was collected for determination of enzyme activity. PPO activity was assayed with catechol as a substrate according to a spectrophotometric procedure. The enzyme solution (0.1 ml) was rapidly added to 2.9 ml of 10 mM catechol prepared in 0.01 M sodium phosphate buffer (pH 6.8). One unit of PPO activity was defined as the amount of enzyme that caused a change of 0.001 in absorbance per minute at 410 nm.