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## Effect of ultraviolet irradiation combined with chitosan coating on preservation of jujube under ambient temperature



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Shaoying Zhang\*, Youwei Yu, Chunling Xiao, Xiangdong Wang, Yaofen Lei

College of Engineering, Shanxi Normal University, 1 Gongyuan Street, Linfen City 041004, China

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#### ABSTRACT

The effect of ultraviolet irradiation combined with chitosan coating on fresh jujube under ambient temperature was investigated. In the first period, the jujubes were irradiated for 4, 6, 8, 10 min at 253.7 nm ultraviolet, or coated with 1%, 1.5%, 2% and 2.5% of chitosan, respectively. In the second period, the jujubes were firstly irradiated 6 min, and then coated with 1.5% of chitosan. The result showed that combined treatment could reduce decay incidence of jujubes and restrain increase of respiration rate, weight loss, malonaldehyde content and electrolyte leakage of jujubes. Furthermore, the activities of superoxide dismutase, peroxidase, and catalase were maintained at higher level, and the decrease of ascorbic acid and chlorophyll was restricted. The present data indicated that treating post-harvest jujubes with ultraviolet irradiation combined with chitosan coating was an effective method, and might be considered in commercial application for preservation of postharvest fruit.

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#### 1. Introduction

Jujube, a popular fruit to human health, is abundance in carbohydrate, protein, fat, crude cellulose, mineral and functional compositions such as vitamin C, vitamin B<sub>1</sub>, riboflavin and polyphenols (Li, Fan, Ding, & Ding, 2007). After harvest, the sugar content of fresh jujube is high and becomes nutrient for spoilage bacterial growth, resulting in rot. In addition, the fresh jujube loses water, which leads to shrinkage. Thus, the commodity value of jujube was discounted. To improve the physiological quality and prolong the shelf life of fresh jujube, many methods including lower temperature, controlled atmosphere storage, using preservative, or coating edible film were taken into account (Sun, Liu, Zhu, Zhou, & Wang, 2007; Wu, Wang, Shi, Xue, & Gao, 2010; Zhong & Xia, 2007). Physical method such as irradiation has little effect when the fresh fruit is infected during storage period. Although chemical can control certain diseases effectively, its residue brings potential toxicity on human health. The preserving effect of biological control is mostly unsatisfactory at present. Thus, single physical, chemical or biological methods were limited (Yao & Tian, 2005; Zhao et al., 2009).

Ultraviolet ray is an electromagnetic wave between 100 and 400 nm. The wavelengths between 250 and 260 nm are easily

absorbed by nucleic acid of microorganism. After absorbing ultraviolet, the microorganism is led to death owing to nucleoprotein molecular structure variation and metabolism impediment (Fredericks, Toit, & Krügel, 2011). Applying ultraviolet irradiation to disinfect germ is convenient and simple in operation, and ultraviolet irradiation cannot bring second pollution as chemical do (Serpieri et al., 2000). Besides sterilization, ultraviolet irradiation can adjust biosynthesis in plant. For example, the flavonoid biosynthesis, a secondary metabolism, was facilitated, the increase of malonaldehyde was repressed, and the shelf life was prolonged in Shiitake mushroom after ultraviolet irradiation (Jiang, Lu, Jiang, Pei, & Ying, 2010). Ultraviolet irradiation made the rotten rate decrease by 10% and antioxidant activity increase in blueberry fruit during storage (Penelope, Collins, & Howard, 2008).

Chitosan, a natural alkaline polysaccharide, has become one of the most popular edible film materials in recent years owing to its non-toxicity and superior biocompatibility. It has good property to form film, which can partly hold back the carbon dioxide generated in fruit respiration to release, and adjust oxygen of the air into the tissue of fruit (Majeti & Kumar, 2000). Meanwhile chitosan coating could lessen water transpiration of fruit. The effective  $-NH_3^+$  group of chitosan could respond with lipoid and protein of germ cell membrane, which caused pathogens cell death and repressed harmful miscellaneous germ growth (Aider, 2010; Zhou, Xu, & Ma, 2009). At present, chitosan coating had been successfully used for preservation in grape, litchi and beef, etc. (Beverly, Janes, Prinyawiwatkul, & No, 2008; Jiang, Li, & Jiang, 2005; Meng, Qin, & Tian, 2010).

<sup>\*</sup> Corresponding author. Tel.: +86 357 2092489; fax: +86 357 2051000. *E-mail address:* zsynew@163.com (S. Zhang).

The objective of this work was to investigate the effect of ultraviolet irradiation combined with chitosan coating on jujube during storage under ambient temperature. We tried to explore an integrated strategy to preserve fresh jujube and provide reference for other vegetable and fruit preservation.

#### 2. Materials and methods

#### 2.1. Materials and reagents

The jujubes (*Ziziphus jujuba* Mill. cv. Lizao) were picked at a preclimacteric but physiological mature stage from a local orchard in Yaodu District (Shanxi, China) at noon. After the jujubes were selected for their uniform shape, size, color, and absence of defects, they were quickly transported to laboratory in open cartons.

Chitosan (water-soluble, molecular weight of about 200 kDa and 85% deacetyl degree) was purchased from AK Biotech Co., Ltd. (Shandong, China). Thiobarbituric acid, methionine and nitroblue tetrazolium (biochemical reagent) were purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai China). Alfa Aesar Company (Tianjin, China) supplied other regents (AR).

#### 2.2. Fruits treatments

The jujubes were cleaned with tap water, dried under natural wind. Moreover, the experiment was carried out through two periods. In the first period, some jujubes were wholly dipped into 1%, 1.5%, 2% and 2.5% of chitosan solution for 3 min. Afterward, the jujubes were taken out and put in ventilation to dry. And the others were irradiated at 253.7 nm ultraviolet with quartz lamp (30W, Beijing Haoteguang Ultraviolet Co., Ltd., Beijing, China) over 40 cm height of single layer jujubes for 4, 6, 8, and 10 min. In the second period, the jujubes were firstly irradiated 6 min at 253.7 nm ultraviolet over 40 cm height of single layer jujubes, and then they were wholly dipped into 1.5% of chitosan solution for 3 min. Afterward the jujubes were taken out and put in ventilation to dry.

The untreated jujubes served as control samples. In each treatment, about 100 fruits were used and each treatment was performed in triplicate. All samples were placed in plastic boxes and stored under ambient temperature with 85% of relative humidity. The related parameters of the jujubes were determined periodically every other three days during the storage time from 0 to 15 days. The jujube tissue used in assay was prepared as followed. A little part of each fruit (10 units) was cut and all the parts were together ground at about 4 °C with ice bath. Thus, the homogeneous slurry tissue of 10 jujubes was obtained. The tissue was weighed and further homogenized with certain buffer according to determined parameter.

#### 2.3. Decay incidence, weight loss and respiration rate

Fifty jujubes were observed every four days. If visible decay appeared in the surface of jujube, the jujube was considered rotten. Decay incidence was calculated as followed, decay incidence  $(\%) = (n/m) \times 100$ , where *n* is the rotten fruit number; *m* is the total fruit number during storage and is about 50 in the experiment. Weight loss rate was calculated as followed, weight loss  $(\%) = [(m_0 - m_1)/m_0] \times 100$ , Where  $m_0$  was the initial weight and  $m_1$  was the weight during storage.

Respiration rate was determined as described previously (Zhu & Zhou, 2007). About 500 g jujubes were sealed in a 10 L glass for 2 h. The carbon dioxide concentration was measured using TEL7001 dioxide analyzer (Telaire Co, USA). The respiration rate was expressed as mg  $CO_2$  kg<sup>-1</sup> h<sup>-1</sup>.

2.4. Determination activities of superoxide dismutase, catalase and peroxidase

Superoxide dismutase (SOD) activity was determined using a modified method (Zhao et al., 2009). Briefly, tissue (2.0 g) from ten fruits was homogenized with 10 mL of 50 mmol L<sup>-1</sup> phosphate buffer (pH 7.8), and centrifuged at 10,000g for 15 min at 4 °C with a centrifuge Eppendorf 5417R (Germany). The supernatant was collected as crude enzyme. The reaction mixture containing 0.1 mL extracted enzyme, 50 mmol L<sup>-1</sup> sodium phosphate buffer (pH 7.8), 13 mmol L<sup>-1</sup> methionine, 75  $\mu$ mol L<sup>-1</sup> nitroblue tetrazolium (NBT), 10  $\mu$ mol L<sup>-1</sup> EDTA, and 10  $\mu$ mol L<sup>-1</sup> riboflavin, was illuminated using a fluorescent lamp (60  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup>) for 10 min, and the absorbance at 560 nm was recorded by a spectrophotometer (UV-1100, Shanghai Meipuda Instrument, Shanghai, China). An aliquot of identical solution was placed in dark and served as blank control. One unit of SOD activity was defined as the amount of enzyme per gram that catalyzed 50% decrease of the SOD-inhibitable NBT reduction.

Catalase (CAT) activity was assayed by the method of García, Iribarne, Palma, and Lluch (2007) with slight modifications. Tissue (2.0 g) from ten fruits was homogenized with 10 mL 50 mmol L<sup>-1</sup> phosphate buffer (pH7.0, containing 1% PVP), and centrifuged at 10,000g for 15 min at 4 °C. The supernatant was collected as crude enzyme. A reaction mixture contained 1.5 mL 50 mmol L<sup>-1</sup> phosphate buffer (pH 7.0, containing 1% PVP), 0.5 mL 0.1 mol L<sup>-1</sup> H<sub>2</sub>O<sub>2</sub>, 1.5 mL de-ionized water and 0.2 mL crude enzyme. The absorbance of mixture was measured at 240 nm. One unit of enzyme activity was defined as the amount of enzyme that caused a change of 0.1 in absorbance at 240 nm per minute per gram.

Peroxidase (POD) activity was assayed by the method of Quiroga, Forchetti, Taleisnik, and Tigier (2001) with. The crude enzyme of POD was prepared as SOD enzyme. The assay mixture contained 0.5 mL extracted enzyme, 1.5 mL 20 mmol  $L^{-1}$  sodium phosphate buffer (pH 7.8), 1 mL 0.25% guaiacol (w/v), and 0.02 mL 0.75% H<sub>2</sub>O<sub>2</sub> (v/v). POD activity was measured by an increase in absorbance at 460 nm. One unit of POD activity was defined as a 0.01 increase in absorbance at 460 nm per min per gram.

## 2.5. Determination of malonaldehyde content and electrolyte leakage

Malonaldehyde (MDA) content was determined as described previously by Xing, Wang, Feng, and Tan (2008) with slight modification. Tissue (2.0 g) from ten fruits was homogenized with 0.2 mL 10% thiobarbituric acid (TCA). The extraction solution was filtrated, and then 5 mL 0.5% TCA was added to the solution. Boiled for 10 min and then cooled, the solution was centrifuged at 10,000g for 15 min at 25 °C. The absorbance of supernatant using TCA solution as blank was measured at 532, 600 and 645 nm. The malonaldehyde content was calculated as followed, MDA (µmol 100 g<sup>-1</sup>) = [6.45 × ( $D_{532} - D_{600}) - 0.56 \times D_{450}$ ] × 100.

Electrolyte leakage was determined as described previously (Antunes, Dandlen, Cavaco, & Mgjuel, 2010) with slight modifications. Jujube fruits were sliced into small discs 0.05 cm thick and washed three times with de-ionized water to remove surface-adhered electrolytes. After dried with filter paper, 10 discs were placed in vials containing 40 mL de-ionized water. The water was stirred slowly, and conductivity was measured as  $C_1$  with a Conductivity Meter DDS-307, Shanghai Sophisticated Scientific Instrument (Shanghai, China). The water was boiled for ten minutes and cooled quickly. Afterward, the water was diluted to 40 mL with de-ionized water and the conductivity was measured as  $C_2$ . Electrolyte leakage was calculated through the formula, electrolyte leakage (%) =  $C_1/C_2 \times 100$ .

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