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#### Short communication

### Maintaining postharvest qualities of three leaf vegetables to enhance their shelf lives by multiple ultraviolet-C treatment



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#### ABSTRACT

It is well reported that ultraviolet-C (UV-C) treatment enhances the storage or shelf life of many fruits and vegetables. However, biochemical changes associated with this treatment are largely unknown in leaf vegetables. The object of the present study is to evaluate the effects of UV-C treatment on the postharvest quality and biochemistry of three kinds of leaf vegetables (i.e. spinach, leek and cabbage). These vegetables were respectively exposed to single and multiple UV-C irradiations at the same energy level of 2.46 kJ/m<sup>2</sup>, and subsequently stored at 4 °C for 5 days. Total soluble protein, vitamin C and chlorophyll a were determined every day. As a result, the multiple UV-C treatment maintained significantly better quality parameters of postharvest leaf vegetables over the experiment, compared to the single treatment (p < 0.05). We therefore summarize that the multiple UV-C treatment can be an effective alternative to maintain the quality and to enhance the shelf life of postharvest leaf vegetables. © 2016 Elsevier Ltd. All rights reserved.

#### 1. Introduction

Decay of postharvest fruits and vegetables is usually one of the most important factors that cause huge economic loss of agriculture. Although a large number of synthetic microbicides have been developed to prevent the rapid deterioration of these agricultural commodities, large postharvest loss (over 50%) still exists in developing counties (Martinez-Romero et al., 2007). Furthermore, most chemical preservatives or microbicides are found to be harmful to the health of customers, especially children (Kregiel, 2015). This fact explains why many food researchers recently focus mainly on the development of safe and nontoxic solutions to enhance the quality of postharvest commodities. For example, treatment of gamma ray (<sup>60</sup>Co or <sup>137</sup>Cs) is now regarded as an effective and healthful sterilization, and has been already used to improve the shelf life and quality of vegetables and fruits (George, Razali, Santhirasegaram, & Somasundram, 2015). However, the widespread application of gamma rays is subject to both the high cost in equipment and the potential risk of its own ion source

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# (D'Hallewin, Cubaiu, Ladu, & Venditti, 2013). Compared to gamma rays, ultraviolet-C (UV-C) is safer to operate and not in need of expensive equipments or high cost investment (Pinheiro, Alegria, Abreu, Goncalves, & Silva, 2015). Therefore, UV-C treatment is widely applied to enhance the quality of vegetables and fruits on shelf or during storage.

It is well known that an appropriate dose of UV-C treatment can greatly improve the quality of vegetables and fruits under the optimal conditions. After treatment of 7 kJ/m<sup>2</sup> UV-C, pimientos maintained at 10 °C for 18 days exhibit a significantly lower level of chilling injury, decay, electrolyte leakage as well as respiratory rate, and finally obtain a better quality compared with the control (George et al., 2015; Guo, Zhu, & Hou, 2015; Pinheiro et al., 2015; Rodoni, Concellon, Chaves, & Vicente, 2012). Furthermore, pimientos treated by10 kJ/m<sup>2</sup> UV-C can be reserved at 0 °C for 21 days, without any changes in color and quality (Wen et al., 2015). Also, the irradiation of 4 kJ/m<sup>2</sup> UV-C can keep the hardness and color of tomato (Pinheiro et al., 2015). UV-C treatment for 5-10 min can significantly reduce the decay of strawberry when stored at 10 °C, and can also prevent the deterioration and brown stain of grape under low illumination intensities (D'Hallewin et al., 2012). Although these studies seem to indicate positive consequences of UV-C treatments on postharvest vegetables, more experimental evidences are needed to better explain the mode of action of UV-C

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treatments in leaf vegetables where fewer reports were referable and accessible.

Leaf vegetables, such as leek, spinach and cabbage, are extremely welcome in our meal and provide our organism with kinds of necessary nutritious components. However, these vegetables are fairly vulnerable to decay and deterioration, meanwhile, their nutritious components are easy to lose during storage or on shelf (Giovenzana, Beghi, Buratti, Civelli, & Guidetti, 2014). Also, there are few reports on the effect of UV-C on postharvest quality of leaf vegetables. In this study, we therefore aimed to evaluate the influences of different UV-C treatments (by single and multiple irradiations) on postharvest qualities of three leaf vegetables (i.e. leek, spinach and cabbage) and to determine changes of the quality parameters, such as soluble protein, vitamin C (Vc), and chlorophyll a content. Finally, we expected to develop an optimal method to improve postharvest qualities of these three leaf vegetables during storage.

#### 2. Materials and methods

#### 2.1. Sample and experiment

Leaf vegetables (leek, spinach and cabbage) which are the most important commercial vegetables in China were obtained in October 2015 from a local vegetable greenhouse near Henan University of Urban Construction, Pingdingshan, China. Defect-free vegetables with uniform size, color and maturity were selected, cleaned using 100  $\mu$ L/L<sup>1</sup> of 5% sodium hypochlorite and subsequently sterile water, normally air-dried, and randomly divided into three sets of 1 kg vegetable each. One vegetable set was used as control, and the other two were separately prepared for single and multiple UV-C treatments. Each treatment was carried out in triplicate. The samples were finally kept at 4 °C using low density polyethylene (LDPE) wraps up to *in vivo* assays.

#### 2.2. UV-C treatment

UV-C treatment was performed using unfiltered germicide emitting lamps (General Electric, 15 W G15T8, Phillips, Netherlands), which were installed inside a metal cabinet  $(120 \times 60 \times 50 \text{ cm})$  and whose most emitted radiation (82%) lies in the UV-C range of 250-280 nm (data not shown) which agrees with the energy level of UV-C irradiation (Pinheiro et al., 2015). Samples of leek, spinach and cabbage were displayed on a stainlesssteel tray without overlapping the samples, and irradiated with the germicide-emitting lamps located 20 cm above and below the surface holding the trays. Each irradiation lasted for 5 min and the trays were rotated 90° every 2.5 min to ensure uniform irradiation. For single UV-C irradiation, samples were lighted only once at the beginning. For multiple irradiations, the samples were treated once every day, and the control was placed under the same conditions without any UV-C irradiation. UV lamps were warmed up for 30 min before UV-C irradiation to ensure reliable results. The intensity of UV-C radiation ( $\lambda = 254$  nm) was monitored using an Optronic Model 340 UV-vis radiometer (Lutron Electronic Co., Ltd., Taiwan). Total radiation energy for 5 min' exposure was 2.46 kJ/m<sup>2</sup>, and UV-C irradiation was carried out in a dark room to minimize the influences of other lights. Ozone generated during the irradiation was removed through a fume hood on the cabinet. After treatment, samples were respectively packaged using the LDPE wraps and stored at 4 °C.

#### 2.3. Sampling

Before and after UV-C irradiation, sampling was performed

every day during the storage period to evaluate the decay or appearance. Analyses of total soluble protein, vitamin C (Vc) and chlorophyll a were carried out on sampled vegetables immediately and after 1, 2, 3, ..., 5 days of UV-C treatment throughout the whole storage. About 50 g vegetables were sampled for assay each time, and all measurements were carried out in triplicate.

#### 2.4. Measurement of quality parameters

Chlorophyll a was measured using acetone repeated freezing and thawing-extraction method (Inskeep & Bloom, 1985), with some modification, described below. For total chlorophyll extraction, 2 g of vegetable leaf was grinded into powder using liquid nitrogen and then transferred into 80% (v/v) acetone in the dark. Afterwards, samples were homogenized using a tissue lyser at 12,000 g for 15 min at 4 °C. The supernatant was finally diluted one time for measurements of OD<sub>645</sub> and OD<sub>663</sub>, and the content of chlorophyll a was calculated by Aron formula (Inskeep & Bloom, 1985). The remaining supernatant was kept at -40 °C for soluble protein and vitamin C (Vc) determinations. Soluble protein content was analyzed strictly according to the Bradford technique (Bradford, 1976) for the quantitation of microgram quantities of protein using chymotrypsinogen A as standard. Vc content was estimated referring to the method of Pinheiro et al. (Pinheiro et al., 2015). Each assay was performed in triplicate.

#### 2.5. Statistical analysis

Analysis of variance and Duncan's multiple range tests with significance at p < 0.05 were performed using the SAS program (SAS Institute, Inc., USA). Data (n = 3) were expressed as the means  $\pm$  standard deviation (Origin version 8.0, Microsoft corp., USA).

#### 3. Results and discussion

#### 3.1. Effect of UV-C treatment on soluble protein

A large number of studies have shown that exposure to low doses of UV-C can be able to delay deterioration of some fruits (D'Hallewin et al., 2013; Guo et al., 2015) and can reduce decay of vegetables such as carrots (Balestrazzi et al., 2010), onions (Rodov, Tietel, Vinokur, Horev, & Eshel, 2010), tomatoes (Liu, Jahangir, & Ying, 2012), and zucchini squash (Erkan, Wang, & Krizek, 2001). Villa-Rojas et al (Villa-Rojas, Lopez-Malo, & Sosa-Morales, 2011) reported both shelf life and quality of postharvest strawberries were maintained by UV-C combined with heat treatments. In this study, low-dose UV-C treatment showed a notable effect on total soluble protein contents of spinach (Fig. 1a), leek (Fig. 1b) and leek (Fig. 1c). Multiple UV-C irradiations could be able to keep soluble protein out of decline in these three vegetables. By contrast, soluble proteins in both the control and single irradiation groups had significantly decreased at different levels (p < 0.05). For spinach, its soluble protein still maintained about 18 mg/g for five days when treated by multiple irradiations, while soluble proteins in both control and single irradiation groups had declined to less than 10 mg/g by day 5. Soluble protein in leek treated by multiple irradiations was about 33 mg/g though there was a little of decrease to 29 on day 1, remaining there before slightly decreasing to 23 mg/g on day 5. However, protein contents of control and single irradiation groups almost kept the same pace to decline, and remained at 10 mg/g and 15 mg/g after day 3, respectively. As for cabbage, soluble protein content in multiple-irradiation group was well kept at 18 mg/g by day 5, while this content in single-irradiation group decreased to 12 mg/g and then remained there by day 5. The

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