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Effects of carboxymethyl cellulose and chitosan bilayer edible coating on postharvest quality of citrus fruit

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ABSTRACT

There is increasing public interest in development of edible natural biodegradable coatings to replace the currently used commercial synthetic waxes for maintaining postharvest quality of citrus fruit. We tested the efficacy of a newly developed polysaccharide-based edible bilayer coating comprising carboxymethyl cellulose (CMC) and chitosan in preserving postharvest quality of various citrus fruit, including 'Or' and 'Mor' mandarins, 'Navel' oranges, and 'Star Ruby' grapefruit after simulated storage and marketing. In all citrus species, it was found that the CMC/chitosan bilayer coating was equally effective as the commercial polyethylene wax in enhancing fruit gloss. Furthermore, the CMC/chitosan bilayer coating slightly increased fruit firmness, especially of oranges and grapefruit, but was mostly not effective in preventing post-storage weight loss. Both the CMC/chitosan bilayer coating and the commercial wax had no significant effects on juice total soluble solids and acidity levels, and had similar effects on gas permeability, as indicated by only slight increases in internal CO_2 levels and in juice ethanol accumulation after storage. Sensory analyses revealed that neither the CMC/chitosan bilayer coating nor the commercial wax coating had any deleterious effects on flavor preference of 'Navel' orange and 'Star Ruby' grapefruit. However, application of the commercial wax, and moreover the CMC/chitosan bilayer coating, resulted in a gradual decrease in flavor acceptability of 'Or' and 'Mor' mandarins because of increased perception of off-flavors. Overall, we showed that the CMC/chitosan bilayer edible coating sufficiently enhanced fruit gloss, but was not effective in preventing postharvest weight loss. Furthermore, flavor quality was slightly impaired in mandarins but not in oranges and grapefruit.

1. Introduction

Citrus fruit are coated with commercial waxes on the packing line, in order to enhance gloss and reduce water loss and shrinkage (Petracek et al., 1998). However, wax coatings may also impair fruit quality, as they may restrict gas exchange through the peel, thus resulting in development of anaerobic conditions in the internal atmosphere of the fruit and accumulation of ethanol, and sensation of off-flavors (Baldwin et al., 1995; Hagenmaier and Shaw, 1992; Hagenmaier, 2000; Porat et al., 2005).

Recently, because of rising public concern regarding human health issues and environmental protection, there has been increased interest in development of natural biodegradable edible coatings for maintaining postharvest quality of fruit and vegetables. These would replace the currently used commercial synthetic waxes, composed mainly of oxidized polyethylene (Debeaufort et al., 1998; Embuscado and Huber, 2009; Valencia-Chamorro et al., 2010; Dhall, 2013). In general, natural edible coatings are composed of polysaccharides, proteins or lipids, or of various composites of these (Valencia-Chamorro et al., 2010; Dhall, 2013).

So far, most evaluations of edible coatings for citrus fruit focused on hydroxypropyl methylcellulose (HPMC)/beeswax/shellac composites (Navarro-Tarazaga et al., 2007, 2008; Valencia-Chamorro et al., 2011; Contreras-Oliva et al., 2011, 2012). However, although these composite coatings retained fruit quality during postharvest storage, they require the use of powerful organic solvents, such as ammonia, to dissolve the shellac, which itself may also restrict gas exchange and enhance development of anaerobic conditions and off-flavors (Navarro-Tarazaga et al., 2007; Contreras-Oliva et al., 2011, 2012). Moreover, we found that coatings containing beeswax appeared opaque. Several other studies have examined the efficacy of chitosan-based edible coatings in preserving postharvest quality of citrus fruit (Chien and Chou, 2006; Chien et al., 2007; Contreras-Oliva et al., 2012), and in this respect it is worth notice that chitosan exhibits proven antimicrobial and antifungal properties, therefore, its application as a coating material assists in prevention

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of microbial spoilage, and it might replace the use of synthetic fungicides (Devlieghere et al., 2004; Dutta et al., 2009; Elsabee and Abdou, 2013). Other types of edible coatings tested on citrus have been based on sucrose and locust bean gum (Rojas-Argudo et al., 2009; Tao et al., 2012).

In systematic preliminary studies in our laboratory, we evaluated the efficacy of various polysaccharide-based edible coatings, including cellulose derivatives and chitosan, which are relatively inexpensive and easy to dissolve, and we found that carboxymethyl cellulose (CMC) coatings provided a uniform and stable matrix and maintained high structural integrity during storage but, on the other hand, imparted little gloss. In contrast, a chitosan coating provided high gloss but as a stand-alone single-layer film was unstable and tended to peel off. Therefore, in the present study, we evaluated the efficacy of a CMC/chitosan bilayer edible coating, formed by first applying CMC and then chitosan, because this bilayer coating provided a uniform and stable matrix and imparted high gloss. The effects of this edible coating on postharvest quality were tested with various citrus fruit: 'Or' and 'Mor' mandarins, 'Navel' oranges, and 'Star Ruby' grapefruit. We chose to apply these compounds as separate layers, because the CMC/chitosan composite formulation proved non-homogenous whereas, in contrast, application of chitosan upon the CMC layer resulted in a clear, homogenous and stable coating. Furthermore, the proposed CMC/chitosan bilayer coating has the advantage of combining the beneficial properties of each individual ingredient into a superior coating formulation (Hagenmaier and Baker, 1995).

2. Materials and methods

2.1. Plant material and storage conditions

'Or' and 'Mor' mandarins (*Citrus reticulata* Blanco), 'Navel' oranges (*Citrus sinensis* [L]. Osbeck), and 'Star Ruby' grapefruit (*Citrus paradisi* Macf.) were harvested from commercial orchards at the peak of the harvesting season of each variety. On the day of harvest the fruit were transferred to the Department of Postharvest Science at the Volcani Center, where they were sorted against external defects, cleaned by rinsing with tap water under a set of brush rollers, and dried by passing through a hot-air tunnel at 37 °C for ~1 min. After application of the various coatings, as described in Section 2.2, the mandarins and oranges were stored for 4 weeks at 5 °C, and the grapefruit for a similar period at 10 °C, all under simulated commercial postharvest handling and marketing conditions. Fruit quality was evaluated after an additional 5 days under shelf-life conditions at 20 °C. The relative humidity was ~90–95% during cold storage and ~80–85% during shelf-life. Each treatment comprised four cartons, each containing 30 fruit – a total of 120 fruit per treatment.

2.2. Preparation and application of edible coatings and commercial wax

The CMC/chitosan bilayer coating was applied in two steps: first with the CMC formulation, and then by applying chitosan solution. The fruit were then completely dried by passing through a hot-air tunnel at 37 °C. The CMC sodium salt (BDH Chemicals Ltd., Dorset, UK) was dissolved in sterilized double-distilled water (DDW) by stirring at 80 °C to obtain a final concentration of 1.5% (w/v). Medium-molecular-weight chitosan (200–800 cP) (Sigma–Aldrich, St Louis, MO, USA) was dissolved in sterile DDW, acidified with 0.7% (v/v) acetic acid, to obtain a final concentration 1.0% (w/v). The 1.5% CMC and 1.0% chitosan solutions were applied manually with paint brushes. Some of the fruit were left untreated as controls, or coated with commercial polyethylene-based waxes commonly used for citrus fruit in Israel: grapefruit and oranges were coated with 'Zivdar' wax formulation and mandarins with 'Tag' wax formulation (Safepack Products Ltd., Kfar Saba, Israel).

2.3. Determination of fruit gloss, firmness and weight loss

Fruit visual appearance was evaluated subjectively by 5 people. Fruit gloss was evaluated on a 0–10 scale in which 0 = no gloss and 10 = very glossy.

Fruit firmness was evaluated by measuring the force required to compress the fruit to 95% of its initial width using an Inspekt 5 dynamic firmness analyzer (Hegewald and Peschke, Nossen, Germany); the presented results are means ± S.E. of measurements obtained with 10 different fruit per treatment.

Fruit weight loss was evaluated by weighing the same fruit before and after storage, and the data are means of 15 measurements ± S.E. and the results are presented as percentages of weight losses.

2.4. Internal atmosphere analysis

One-milliliter samples of the fruit internal atmospheres were withdrawn through a syringe inserted through the fruit blossom ends. The CO_2 concentrations in the gas samples were determined with a Gow-Mac Series 580 gas chromatograph (Gow-Mac, Canton, Massachusetts, USA) fitted with a Alltech Chromosorb 80/100 (1/8 in. × 1.2 m) column, followed by a molecular sieve 5A 45/60 (1/8 in. × 1.2 m), with helium used as a carrier gas. The injector, oven and detector temperatures were 35, 115 and 150 °C, respectively. Each measurement was applied to gas samples from nine different fruit.

2.5. Determination of juice TSS and acid contents

Total soluble solids (TSS) content in the juice was determined with a Model PAL-1 digital refractometer (Atago, Tokyo, Japan), and acidity percentages were measured by titration to pH 8.3 with 0.1 M NaOH in a Model CH-9101 automatic titrator (Metrohm, Herisau, Switzerland). Each measurement comprised five replications, each using juice collected from three fruits, i.e., a total of 15 fruit per measurement.

2.6. Determination of juice ethanol concentrations

Ethanol concentrations in the juice were determined according to Davis and Chace (1969). Ten-milliliter aliquots of juice, extracted from three different fruit, were incubated in 50 mL Erlenmeyer flasks at 30 °C for 30 min. In parallel, 50 mL Erlenmeyer flasks containing 10 mL of solutions containing ethanol at 100 $\mu L L^{-1}$ were incubated at the same temperature and used as internal standards for quantity evaluations. After incubation 2 mL gas samples were withdrawn from the Erlenmeyer headspaces into syringes, and their ethanol levels were determined with a Varian 3300 gas chromatograph (Hewlett–Packard, Bloomington, IL, USA) equipped with a flame ionization detector and a 1/8 in. × 1.2 m Supelco Co 80/100 column, with helium used as a carrier gas. The injector, oven and detector temperatures were 80, 150 and 200 °C, respectively. The presented results are means ± S.E. of four replicate samples.

2.7. Sensory evaluations

Fruit sensory quality was evaluated by a trained panel according to Tietel et al. (2011a,b). In all cases, fruit were hand-peeled, and separated segments were cut into halves and placed in glass cups, identified by randomly assigned three-digit codes; each treatment comprised a pooled mixture of six to eight cut segments from six