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Effect of chitosan coating combined with postharvest calcium treatment on strawberry (*Fragaria* × *ananassa*) quality during refrigerated storage

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Abstract

Strawberries ($Fragaria \times ananassa$ Duch.) were coated with either 1% or 1.5% chitosan (CS) or chitosan combined with calcium gluconate (CaGlu). Following treatment, strawberries were stored at 10 °C and 70 \pm 5% RH for one week. The effectiveness of the treatments in extending fruit shelf-life was evaluated by determining fungal decay, respiration rate, quality attributes and overall visual appearance. No sign of fungal decay was observed during the storage period for fruit coated with 1.5% CS (with or without the addition of CaGlu) or 1% CS + 0.5% CaGlu. By contrast, 12.5% of the strawberries coated with 1% CS lacking calcium salt were infected after five days of storage. The chitosan coating reduced respiration activity, thus delaying ripening and the progress of fruit decay due to senescence. Chitosan coatings delayed changes in weight loss, firmness and external colour compared to untreated samples. Strawberries coated with 1.5% chitosan exhibited less weight loss and reduced darkening than did those treated with 1% chitosan, independently of the presence or absence of CaGlu. However, addition of calcium to the 1% chitosan solution increased the firmness of the fruit. Coated samples had greater visual acceptability than had untreated fruits. The addition of calcium gluconate to the chitosan coating formulation increased the nutritional value by incrementing the calcium content of the fruit.

Keywords: Strawberry; Chitosan concentration; Calcium gluconate; Shelf-life; Refrigerated storage

1. Introduction

Strawberry (*Fragaria* × *ananassa*) is a highly perishable non-climacteric fruit. It must be harvested at full maturity to achieve maximum quality in terms of visual appearance (freshness, colour and absence of decay or physiological disorders), texture (firmness, juiciness and crispness), flavour and nutritional value (vitamins, minerals, dietary fibre and phytonutrients). Grey mold, caused by *Botrytis cinerea* Pers. Fr., is the most economically significant postharvest pathogen of strawberry fruits. Strawberry spoilage after

harvest, can also occur by mechanical injury and desiccation.

Low storage temperatures and modified atmospheres with elevated CO₂ levels are common tools for avoiding, at least partially, mold growth and senescence, and extending fruit shelf-life (Manning, 1996). However, prolonged exposure of berries to high CO₂ concentrations can cause off-flavour development (Ke, Zhou, & Kader, 1994).

The use of synthetic chemical fungicides has been the main method for reducing postharvest disease. However, consumer concern over pesticide residues on foods, along with pathogen resistance to many currently used pesticides, has increased the need to find alternative methods for decay control. Recently, biologically active natural products have started to become an effective alternative to synthetic fungicides (Tripathi & Dubey, 2004).

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The preservation of fresh produce can also be achieved by the application of edible coatings. Several mechanisms are involved in extending the shelf-life of fruits and vegetables by film coatings. These include decreasing moisture loss and controlled gas (CO₂/O₂) exchange, resulting in reducing respiration rate. Edible films can also prevent mechanical injury produced during post-harvest handling and processing. Whey protein, calcium caseinate, gluten and polysaccharides, such as cactus mucilage or starch, have also been shown to exert beneficial effects on strawberry fruit quality when applied as coatings (Del-Valle, Hernández-Muñoz, Guarda, & Galotto, 2005; García, Martino, & Zaritzky, 1998; Tanada-Palmu & Grosso, 2005; Vachon, D'ApPrano, Lacroix, & Letendre, 2003).

Chitosan (poly β -(1,4)*N*-acetyl-D-glucosamine) polymer is industrially produced by chemical deacetylation of the chitin found in arthropod exoskeletons. This biopolymer can also be obtained directly from the cell wall of some plant-pathogenic fungi. Chitosan and its derivatives have been shown to inhibit the growth of a wide range of fungi and trigger defensive mechanisms in plants and fruits against infections caused by several pathogens. Chitosan possesses excellent film-forming properties and can be applied as an edible surface coating to fruits and vegetables. Chitosan coatings have been reported to limit fungal decay and delay the ripening of several commodities, including strawberry (El Ghaouth, Arul, Ponnampalam, & Boulet, 1991a; Han, Zhao, Leonard, & Traber, 2004; Ribeiro, Vicente, Teixeira, & Miranda, 2007). Preharvest chitosan sprays have been noted to be effective in controlling postharvest fungal infection in strawberries (Reddy, Belkacemi, Corcuff, Castaigne, & Arul, 2000).

Calcium ions perform multiple roles in plant cell physiology. They are important intracellular messengers, mediating responses to hormones, biotic and abiotic stress signals and a variety of developmental processes (Reddy & Reddy, 2004). They also play an essential role in the structural maintenance of membranes and cell walls. Calcium ions cross-link free carboxyl groups on adjacent polygalacturonate chains present in the middle lamella of the plant cell wall contributing to cell-cell adhesion and cohesion. Preharvest and postharvest treatments with calcium salts have been effective in controlling several physiological disorders, reducing the incidence of fungal pathogens and maintaining fruit firmness (Bakshi, Fa, Gs, & Ta, 2005). Foliar applications of calcium chloride have been reported to delay ripening and retard fungal growth on strawberries (Wojcik & Lewandowski, 2003). Postharvest treatments with calcium salts include dipping, vacuum and pressure infiltration which can be combined with other treatments. Calcium dips alone, or in combination with heat treatments or modified atmosphere, have resulted in improved strawberry shelf-life (Garcia, Herrera, & Morilla, 1996).

The aim of the present work is to study the effect of chitosan coatings combined with calcium gluconate on strawberry (*Fragaria* × *ananassa* cv. Camarosa) quality attributes during refrigerated storage. Strawberries were

treated with 1% or 1.5% chitosan acetate solution, with or without the addition of calcium gluconate. Assessment of the treatments is based on their effects on fungal decay, respiration rate, quality attributes, and the visual appearance of strawberries stored for six days at 10 °C.

2. Materials and methods

2.1. Fruit material

Strawberry fruit (*Fragaria* × *ananassa* Duch. cv. Camarosa) were purchased from a local market. Fruits were harvested and shipped from Palos de la Frontera (Huelva, Spain) in a refrigerated truck on the previous day. Fruits were selected, based on uniformity of size, the absence of physical damage and fungal infection, and >75% of the surface showing red colour.

2.2. Edible coating formulations

Acetic acid, calcium gluconate and high molecular weight chitosan were purchased from Sigma Chemical Co (St. Louis, MO, USA). Coating solutions were prepared by dissolving 1% or 1.5% chitosan in 0.5% acetic acid solution. Chitosan coatings containing calcium gluconate were prepared by dissolving calcium salt at 0.5% or 0.75% in water prior to the incorporation of the acetic acid; chitosan was subsequently added to 1% or 1.5%, respectively.

2.3. Coating application

Strawberries were randomly distributed into five groups. Four groups were assigned to one of four treatments whilst the fifth group provided the untreated control. The treatments consisted in immersing fruits for 5 min in: (a) 1% chitosan acetate; (b) 1.5% chitosan acetate; (c) 1% chitosan +0.5% calcium gluconate; and (d) 1.5% chitosan +0.75% calcium gluconate solution. Fruits were allowed to dry for 2 h at 20 °C and were subsequently stored at 10 °C and $70\pm5\%$ RH.

2.4. Fungal decay

Fungal decay was visually inspected daily during the storage period. Strawberry fruits showing surface mycelial development were considered decayed. Results were expressed as the percentage of fruits infected.

2.5. Respiration rate

Respiration rate was determined by using the static method. Ten berries were placed in hermetically sealed 750 ml glass jars and kept at 10 °C. After 1 h of enclosure, a 100 µl sample was withdrawn from the headspace and analyzed for CO₂ using a gas chromatograph (Hewlett-Packard 5890 series II GC, Agilent Technology, Barcelona, Spain) equipped with a thermal conductivity detector (TCD) and

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