Novel Edible Coating Based on Aloe vera Gel To Maintain Table Grape Quality and Safety

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A novel edible coating based on Aloe vera gel obtained according to SP Patent Filed 200302937 has been used as a means of preservation to maintain the quality and safety of cv. Crimson Seedless table grapes during cold storage and subsequent shelf life. Table grapes have a crucial economic value as a dessert fruit, but once harvested show a reduction of shelf life due to a rapid loss of quality. Uncoated clusters showed a rapid deterioration with an estimated shelf life period of 7 days at 1 °C plus 4 days at 20 °C, based on the fast weight loss, color changes, accelerated softening and ripening, rachis browning, and high incidence of berry decay. On the contrary, those clusters treated with A. vera gel significantly delayed the above parameters related to postharvest quality losses, and storability could be extended up to 35 days at 1 °C. Interestingly, this edible coating was able to reduce the initial microbial counts for both mesophilic aerobic and yeast and molds, which significantly increased in uncoated berries over storage. Moreover, the sensory analyses revealed beneficial effects in terms of delaying rachis browning and dehydration and maintenance of the visual aspect of the berry without any detrimental effect on taste, aroma, or flavors. To the authors’ knowledge, this is the first time A. vera gel has been used as an edible coating in fruits, which would be an innovative and interesting means for commercial application and an alternative to the use of postharvest chemical treatments.

KEYWORDS: Aloe vera; ripening; firmness; color; postharvest; sensory analyses; mesophilic aerobics; yeast and molds

INTRODUCTION

Aloe vera is a tropical and subtropical plant that has been used for centuries for its medicinal and therapeutic properties (1). The two major liquid sources of A. vera are a yellow latex (exudate) and a clear gel (mucilage), which proceeds from the large leaf parenchymatic cells (2). The predominant medical uses of the orally ingested gel juice are against ulcers, gastrointestinal, kidney, and cardiovascular problems and also to reduce the cholesterol and triglyceride levels in blood. Moreover, other properties such as antiinflammatory and antibiotic activities and activities against some diseases (diabetes, cancer, allergy, AIDS) have been reported (1, 3). However, the main use of A. vera gel is in the cosmetic industry, including treatment of burns and scars and in wound healing (4). There are some reports on the antifungal activity of A. vera gel against several pathogenic fungi including Botrytis cinerea (5–7).

Recently, there has been increasing interest for the use of A. vera gel in the food industry as a functional ingredient food in drinks, beverages, and ice creams (8). Nevertheless, most of the so-called Aloe products may contain very small amounts of the active compounds, because the processing techniques used to obtain A. vera gel affect the product quality and the amount of bioactive compounds in the final product. In this sense, a procedure of extraction and processing has been developed to ensure the biological integrity, the sensorial stability, and the quality of the final product for the food industry application (9). In Spain, our group has filed a patent for the use of A. vera gel as an edible coating for postharvest treatment on fruits and vegetables (10).

Table grapes show severe problems during postharvest storage and retailing. The losses of quality are based on weight loss, color changes, accelerated softening and rachis brownint, and high incidence of berry decay (11, 12), which lead to a reduction of shelf life. The most important disease in grapes with severe economical repercussions is gray mold caused by B. cinerea. To solve this problem, chemical fungicides have been used, the most common being SO2 (13). However, the use of a combination of pesticides, the development of fungicide-resistant strains, and the public’s concern for human health and environmental pollution have stimulated the search for new strategies as alternative tools for controlling postharvest decay. These technologies include controlled-atmosphere storage with high
CO₂ (15–25%), hypobaric treatments, heated ethanol, and biocontrol agents (12, 14–17), although occurrence of injuries existed (rachis browning and off-flavors). Finally, the use of modified atmosphere packaging (MAP) has been shown to maintain berry quality (18) and to reduce decay alone or in combination with a SO₂-commercial generator (19).

Edible coatings are traditionally used to improve food appearance and conservation due to their environmentally friendly nature, because they are obtained from both animal and vegetable agricultural products (20). Generally, coatings can be divided into proteins, lipids, and polysaccharides, alone or in combination. They act as barriers to moisture and oxygen during processing, handling, and storage and do not solely retard food deterioration but also enhance its safety due to their natural biocide activity or the incorporation of antimicrobial compounds (21). Other advantages of the use of edible coatings could be the reduction of packaging waste, because they are considered to be biodegradable, and the development of new products. From early evidence of cucumbers coated with wax (22), different compounds have been used as edible coatings to prevent weight loss mainly, including Sepmerfresh (23), milk proteins (24), celluloses (25), lipids (26), starch (27), zein (28), alginate (29), and mucilage (30). In addition, edible coatings based on chitin, chitosan, and their derivatives have shown antimicrobial properties (31, 32). In the revised literature, there is little evidence on the use of edible coatings in table grapes. Thus, methyl cellulose added with antimicrobial substances extended the shelf life and reduced spoilage microorganisms (33), whereas chitosan controlled the gray mold in artificially inoculated berries (34).

The objectives of this work were to analyze the effect of A. vera coating on table grape quality attributes and its role in microbial spoilage during 35 days of cold storage and subsequent shelf life (SL). As far as we know from the literature, this is the first time A. vera gel has been used as an edible coating in fruits, which would be an innovative and interesting means for commercial postharvest application.

MATERIALS AND METHODS

Plant Material and Experimental Design. Table grapes (Vitis vinifera L. cv. Crimson Seedless) were harvested from a commercial farm in Jumilla (Murcia, Spain). At the laboratory, clusters were selected to obtain homogeneous batches based on color, size, absence of injuries, and healthy greenish rachises. Clusters were cut to obtain 120 samples ranging from 150 to 170 g. Half of them were treated with A. vera L. gel (pharmaceutical quality, 100% purity) manufactured by Roig Farma S.A. (Tarrasa, Barcelona, Spain). Treatment was performed by immersion during 5 min with a solution of A. vera diluted 1:3 with distilled water according to our developed patent (10). The other half were immersed in distilled water and served as control. Following treatment, all clusters were air-dried during 30 min before storage at 1 °C and 95% relative humidity (RH) in permanent darkness for 35 days. Ten samples for both treated and control clusters were taken after 7, 14, 21, 28, and 35 days; half of them were immediately analyzed (cold storage), and the remainder were transferred to a chamber under controlled conditions at 20 °C and 90% RH and analyzed after 4 days, to simulate market operations (shelf life, SL) for the analytical determinations.

The visualization of both clusters and grapes after 21 days of cold storage plus 4 days at 20 °C showed that control clusters were unmarketable, with a high incidence of decay and accelerated quality deterioration. For this reason, the predicted sampling schedule of shelf life was stopped at this moment. However, the visual aspect of treated clusters was considered to be acceptable after 35 days at 1 °C plus 4 days at 20 °C.

Respiration Rate and Ethylene. CO₂ and ethylene production was measured by placing each cluster in a 0.5 L glass jar hermetically sealed with a rubber stopper for 1 h. One milliliter of the holder atmosphere was withdrawn with a gas syringe, and the ethylene was quantified using a Hewlett-Packard model 5890A gas chromatograph (Wilmington, DE) equipped with a flame ionization detector and a 3 m stainless steel column with an inner diameter of 3.5 mm containing activated alumina of 80/100 mesh. The column temperature was 90 °C, and injector and detector temperatures were 150 °C. Results were the mean ± SE of four determinations for each cluster and expressed as nanoliter per gram per hour (n = 5). For respiration rate determination, another sample of 1 mL of the same atmosphere was withdrawn and CO₂ quantified using a Shimadzu 14A gas chromatograph (Kyoto, Japan), with a thermal conductivity detector and a molecular sieve 5A column, 80–100 mesh (Carbosieve SI, Supelco Inc., Bellefonte, PA), of 2 m length and 3 mm i.d. Oven and injector temperatures were 50 and 110 °C, respectively. Helium was used as carrier gas at a flow rate of 50 mL min⁻¹. Results were the mean ± SE of four determinations for each cluster and expressed as milligrams of CO₂ per kilogram per hour (n = 5).

Weight Loss. Weights of individual clusters were recorded on the day of harvesting and after the different sampling dates. Cumulative weight losses were expressed as the mean ± SE of percentage loss of original weight (n = 5).

Color. Color was determined using the Hunter Lab System and a Minolta colorimeter model CR200 (Minolta Camera Co., Osaka, Japan). Following the recording of individual L*, a*, and b* parameters, color was expressed as chroma index, and results were the mean ± SE of determinations made on 10 berries for each cluster along the equatorial axis (n = 50).

Firmness. Flesh firmness was determined using a TA-XT2i texture analyzer (Stable Microsystems, Godalming, U.K.) interfaced to a personal computer. For each berry, 1 cm² of the skin was removed and penetration force measurement was individually recorded using a 2 mm diameter probe. Penetration rate was 20 mm min⁻¹ for 10 mm after contacting the flesh, and results were the mean ± SE of determinations made in 10 berries for each cluster (n = 50) and expressed in newtons.

Total Soluble Solids Concentration (TSS) and Total Acidity (TA). TSS was determined in triplicate from the juice obtained from 10 berries for each cluster with a digital refractometer Atago PR-101 (Atago Co. Ltd., Tokyo, Japan) at 20 °C, and results were expressed as the mean ± SE of °Brix (n = 5). The pH of the juice was recorded, and then TA was determined in triplicate from the above juice by potentiometric titration with 0.1 N NaOH up to pH 8.1, using 1 mL of diluted juice in 25 mL of distilled H₂O; results were the mean ± SE expressed as grams of tartaric acid equivalent per 100 g of fresh weight (n = 5).

Sensory Evaluation. Sensory analyses to compare the quality of treated and control table grapes were carried out by 10 trained adults, aged 25–40 years (5 females and 5 males). The panel was trained in a pretest in which berries with extremely low or high attributes (crunchiness, juiciness, sweetness, sourness, and quality) were evaluated (18). A laboratory of sensory analyses with an individual booth for each panelist was used. For rachis, symptoms of dehydration and browning for primary and secondary branches were evaluated (12) on a ranked scale of 1 to 5, where 1 = absence of these symptoms, 2 = slight occurrence, 3 = moderate, 4 = severe, and 5 = extremely severe browning and dehydration. For each treatment and sampling date, the five branches were evaluated by each judge (n = 50). In a second test, each judge evaluated five berries for each cluster for the following characteristics: visual aspect (general aspect), firmness, sweetness, juiciness (amount of free fluid released from the berry during chewing), sourness, and crunchiness (amount of noise generated when the berry was chewed at a fast rate with the back teeth), on a scale of 1–5 (ranked), where 1 = very low, 2 = low, 3 = medium, 4 = high, and 5 = very high. Finally, the occurrence of off-flavors was also tested. For this purpose, judges were screened for their capacity in perceiving off-flavor using a triangle test according to the method of O’Mahony (35). Briefly, each panelist was instructed to cleanse his/her mouth with distilled water, chew the berry, and evaluate the sample using the binary response of yes or no. The sensorial analyses were made after 21 days of cold storage plus 4 days at 20 °C.

Microbiological Analysis. Samples of 10 g from each cluster were obtained under sterile conditions (laminar fume cupboard, gloves, and
RESULTS

Respiration and Ethylene Production Rates. The CO₂ production rate in control berries significantly increased over cold storage, whereas maintenance was shown in Aloe-treated grapes (Figure 1). After 35 days of cold storage, respiration rates were 19.03 ± 0.66 and 13.14 ± 0.69 mg kg⁻¹ h⁻¹ for control and treated clusters, respectively. During SL after cold storage, the reduction of the respiration rate was also significant for Aloe-treated berries compared to controls. Ethylene production rate showed a continuous increase as did the storage, with levels of 0.90 ± 0.11 nL g⁻¹ h⁻¹ after 35 days at 1 °C in control bunches, which were significantly reduced by Aloe treatment (0.45 ± 0.03 nL g⁻¹ h⁻¹). The same observation was noticed after 21 days of cold storage plus 4 days at 20 °C, at which the ethylene production rate doubled in control (0.45 ± 0.03 nL g⁻¹ h⁻¹) with respect to Aloe-treated bunches (data not shown).

Parameters Related to Berry Quality. Weight loss increased during both cold storage and SL of the grapes, but it was significantly greater in control than in Aloe-treated berries (Figure 2). At the end of cold storage, control fruits lost 15.51 ± 0.32%, whereas the loss of weight in Aloe-treated berries was 8.13 ± 0.59%. The same differences were observed after the subsequent SL, at which the percentage of weight loss was double in control compared with that in Aloe-treated fruits after 21 days at 1 °C plus 4 days at 20 °C.

With respect to firmness, table grapes softened during cold storage, but to a greater extent in control than in Aloe-treated berries (Figure 3). At the end of cold storage, control berries showed flesh firmness levels of 2.08 ± 0.13 N; in Aloe-treated berries these levels were significantly higher (2.96 ± 0.15 N). During SL after cold storage the softening process still continued, the loss of firmness being more greatly accelerated in control than in Aloe-treated berries, with significant differences from the first period of SL, immediately after harvest and kept for 4 days at 20 °C.

TSS concentration significantly increased during cold storage in control grapes, from levels at harvest of 20.59 ± 0.10 to 24.97 ± 0.20 °Brix after 35 days at 1 °C (Figure 4). This increase in TSS was more pronounced when berries were kept at 20 °C and was significantly delayed in Aloe-treated fruits. Similarly, the loss of TA was affected by treatment (Figure 5). Thus, grapes treated with Aloe exhibited a significantly lower reduction in acidity compared with controls, the differences being greater when clusters were transferred at 20 °C. After 21 days at 1 °C plus SL, TA levels were 0.33 ± 0.01 and 0.45 ± 0.01 g of tartaric acid equiv 100 g⁻¹ for control and Aloe-treated fruits, respectively.

Color (chroma index) resulted in a sharp increase from the initial value (9.39 ± 0.54) within the first week of cold storage in control berries, showing a continuous increase until the end of the storage period, reaching levels of 18.40 ± 0.38 (Figure 6). On the contrary, color changes were significantly delayed...
by Aloe treatments, the magnitude of the reduction being similar during cold storage or after SL.

**Sensorial Quality.** Panelists evaluated the visual aspect of the rachis and gave the highest scores to those rachises of control clusters, which became significantly different from day 7 of cold storage compared to treated clusters and especially during the subsequent periods of SL (Figure 7). These results indicated severe symptoms of dehydration and browning in control rachises after 7 days at 1 °C plus SL (scores > 3) and slight—moderate effects for those clusters treated with A. vera gel after 28 days of cold storage.

The highest scores after 21 days of cold storage plus 4 days at 20 °C (Figure 8) for berry aspect, firmness, crunchiness, juiciness, and sourness were given to the Aloe-treated berries compared with controls. An inverse tendency was found when sweetness was tasted, because control berries had significantly higher scores. Judges perceived the development of “off-flavors” in Crimson Seedless grapes at 21 days of storage plus 4 days at 20 °C, but the percentage was influenced by treatment. Thus, for control berries, 80% of the panelists found bad aroma and...
“off-flavors”, whereas for those treated with Aloe, none of the panelists found the occurrence of “off-flavors”.

Microbiological Population. At harvest, table grape had 4.6 and 3.5 log CFU g⁻¹ for total mesophilic aerobic and yeast and mold counts, respectively. Following 21 days of cold storage plus 4 days at 20 °C, the microbial populations of Aloe-treated grapes were significantly reduced, the reduction being slightly more effective for yeast and mold counts (1.5 log CFU g⁻¹) than for mesophilic aerobics (2.6 log CFU g⁻¹). On the contrary, significant increases (4.2 log CFU g⁻¹) in yeast and mold populations were observed for control berries (Figure 9). For total mesophilic aerobic, only slight rises were obtained (4.9 log CFU g⁻¹). Similar results were found after 35 days of storage at 1 °C, at which 4.8 and 4.9 log CFU g⁻¹ were counted in control fruits for yeast and mold and mesophilic aerobics, respectively, and were significantly reduced in Aloe-coated grapes (1.9 and 1.4 log CFU g⁻¹, respectively).

DISCUSSION

Table grapes as other many fruits undergo numerous physicochemical, biochemical, and microbiological changes during storage, inducing and accelerating the ripening process and reduction of their SL. These changes are accompanied by economical postharvest repercussions due to weight losses and occurrence of decay caused by fungal plant pathogens. On the other hand, there are growing concerns of consumers regarding the safety of the food products and avoiding the use of chemicals as a means of preservation, but, unfortunately, increasing incidence of foodborne illnesses from pathogenic microorganisms is occurring as availability of fresh foods is increasing (37), resulting in a major public health impact around the world. In this sense, continuing efforts in the processing, preservation, distribution, and marketing are being made worldwide to supply fresh fruits with high quality and safety to consumers. In this work, as an alternative to the use of postharvest chemical treatments, we have studied the application of a novel edible coating based on A. vera gel in preserving table grape quality and safety.

The shelf life of table grapes was affected by respiration rate and weight loss of both berry and rachis, because as grapes lose weight they are more susceptible to fungal decay (12). Control table grapes showed increases in respiration rate during storage and lost 12–14% of their initial weight during cold storage, similar values to those found in other cultivars (18). However, loss of weight in Aloe-treated clusters was significantly reduced (below 5%), which is considered to be acceptable for retailing purposes, and coincided with the reduction in respiration rate. As other edible coatings, A. vera gel prevented moisture loss and controlled respiratory exchange. In general, this positive effect of edible coatings is based on their hygroscopic properties, which enables formation of a water barrier between the fruit and the environment, and thus avoiding its external transference (38). To enhance water barrier efficacy, many formulations are composite coatings of several groups, the most frequently used being polysaccharide–lipid. Thus, increasing lipid content of coating formulations significantly reduced weight loss of mandarins (39). However, A. vera gel, the composition of which is mainly polysaccharides (2), was highly effective as a moisture barrier without the lipid incorporation. The reduction of CO₂ production seems to be a general effect in coated fruits, as has been observed in avocado (40), cut apples (41), and sweet cherry (42).

Texture is an important attribute demanded by consumers and most of the time is responsible for fruit acceptability. The rate and extension of firmness loss during storage are the main factors determining fruit quality and postharvest SL. In fact, Aloe treatment significantly reduced the firmness losses during cold storage and subsequent SL, whereas losses of >50% were detected in control grapes after 21 days of cold storage plus 4 days at 20 °C. The explanation for this firmness maintenance could be related to the lower weight losses, as has been reported in strawberry (27, 30), apple (29), and sweet cherry (23) treated with different edible coatings such as yam starch, cactus mucilage, alginate, gelatin, and Semperfresh. In addition, some effect of A. vera gel on the reduction of β-galactosidase, polygalacturonase, and pectinmethyl esterase activities, the main cell wall degrading enzymes responsible for table grape softening, could not be discarded (43). However, failures in firmness retention have been noted in strawberry and raspberry coated with chitosan, which were attributed to extra liquid residues on the fruit surface and incomplete drying process (44).

Other important parameters determining the quality of table grapes are the TSS and TA, which affect consumer acceptance (45). The coating with A. vera led to a lower increase in TSS and greater TA retention, which indicated that control fruits presented a more pronounced maturation development than coated berries, similarly to that found in starch-coated strawberry (27), and could be related to the higher respiration rate found in uncoated fruits. In addition, the Aloe coating could produce a modification of the internal atmosphere, showing similar effects as MAP (18).

Skin color of table grapes showed lower increases in Aloe-treated than in control berries. Table grapes, such as Crimson Seedless, are rich in anthocyanin compounds, which account for their red color. The ripening process of berries has been correlated to the anthocyanin content, peonidin 3-glucoside being the major anthocyanin in Crimson Seedless (46). At the end of cold storage, and especially at the end of SL, control fruits exhibited a redder and darker color than Aloe-treated ones, showing the aspect of overripe fruit, which is considered to be detrimental to color quality.

The beneficial effect of A. vera coating was also clear in delaying rachis dehydration and browning, because these clusters received lower scores than controls from a trained panel. These symptoms first appeared on pedicels followed by lateral branches and finally on central axis, as has been reported for cv. Flame Seedless table grapes due to increased polyphenol oxidase activity (11). Thus, some effects of the A. vera gel on the activity of this enzyme could not be discarded. Thereafter, the sensorial analyses of the berries rendered higher scores for
all analyzed parameters, with the exception of sweetness. These results are in agreement with the lower weight loss found in Aloe-treated berries and the higher TSS observed in control grapes, which could be due to sugar concentration as a result of water loss by dehydration. Moreover, the A. vera coating imparted an attractive natural-looking sheen to table grapes, which was correlated to lower changes in both skin color and dehydration. It is interesting to point out that none of the judges could discern any bad odor or “off-flavor” attributed to the Aloe treatment.

Edible coatings, apart from acting as a gas barrier, may serve to improve food safety by inhibition or delay of the growth of microorganisms, giving a further step to the concept of active packaging (47). Thus, the use of an edible coating, such as chitosan and yam starch, was a viable alternative for controlling microbial growth in minimally processed carrots (48), strawberries (27), and raspberries (44), as well as in table grapes inoculated with B. cinerea (34). However, Semperfresh resulted in a slight increase in fungal spoilage in coated cherries (49). In this work, A. vera gel was effective in reducing microorganism proliferation in table grape, the effect being higher for yeast and molds than for mesophilic aerobics. Total viable counts were \(<2\) log CFU g\(^{-1}\) for yeast and molds and \(<3\) log CFU g\(^{-1}\) for mesophilic aerobics during cold storage or after 21 days at 1 °C plus SL, which meet the recommended microbiological criteria for nonheated fruit desserts (50), whereas counts of \(\approx5\) log CFU g\(^{-1}\) were observed for control berries. The lower counts for yeast and molds were related to the lower ethylene production in coated clusters, which could indicate that the accumulated ethylene in control bunches proceeds from the fungal metabolism rather than from berry fruit. In fact, it has been reported that B. cinerea produced greater amounts of ethylene as did the concentration of conidia inoculated in vitro (51).

The antifungal activity of A. vera pulp has been documented, including several postharvest fruit pathogens, such as Penicillium digitatum, Penicillium expansum, B. cinerea, and Alternaria alternate (5, 7). The antifungal activity of A. vera was based on the suppression of germination and the inhibition of mycelial growth and could be attributed to the presence of more than one active compound with antifungal activity (6), although the specific action mechanism is still unknown. In addition, A. vera gel has been proven to reduce the growth of 17 bacteria (3), the amount needed for inhibition being lower for Gram-positive than for Gram-negative (52). Although the compound(s) responsible for the antibacterial activity was (were) not elucidated, some individual components found in A. vera gel, such as saponins, acemannan, and anthraquinone derivatives, are known to have antibiotic activity.

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