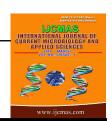
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Original Research Article

Application of Biodegradable *Aloe vera* gel to control post harvest decay and longer the shelf life of Grapes

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ABSTRACT

Keywords

Grapes; Aloe vera; shelf life; control post harvest decay The search for biodegradable, safe, healthy and environmental friendly treatments for increasing storage period of fruits has lead to use of edible, biodegradable films or coatings. The aim of this work was to analyze effects of biodegradable *Aloe vera* gel (0, 1, 5 and 10% w/v) coating on green grape berries, stored at 15 °C for 40 days in air tight container. Treated berries (5 % and 10 %) showed minimum weight loss, lesser browning, shattering, cracking and reduced bacterial and fungal count, which significantly increased in uncoated berries over storage and flavor related factors such as total soluble solids (TSS), titratable acidity (TA) levels were observed maximum in treated grape berries (5 % and 10 %) and sensory analyses of treated berries revealed beneficial effects in terms of delaying rachis browning and maintenance of the visual aspect of the berry without any detrimental effect on taste, aroma, or flavors. This work evaluates the use of *Aloe vera* as biopreservative, which is an economical and eco-friendly.

Introduction

Grapes are one of the important fruit crop in the world, with production rates of approx. 50 million tons per year (Schieber et al., 2001). Grapes can be decay during pre harvest and post harvest and post harvest decay of grapes can be due to physical, physiological, or pathological factors. Rachis dehydration is a main decay factor (physical deterioration) of grapes during the pre or postharvest (Crisosto et al. 1994, Davood et al., 2013). Skin browning of grapes is another main physiological problem associated with

mature table grape cultivar (Vial et al. 2005)

Major cause for grape spoilage is fungal infection, which decreases the production of fruit (Thanaboripat, 2008). Fungicides spray prevent decay of grapes but because of health hazard effects with application of fungicides, have became restrictive, there is a necessity of natural substitutes, such as the application of essential oils of plants which shows higher effectiveness in preventing decay of table grapes (Valverde et al., 2005; Martinez-Romero et al.,

2007). Aloe vera has medicinal properties, is a tropical and subtropical plant that has been used from ancient time (Eshun et al., 2004). The gel of Aloe vera leaves is the colorless mucilaginous, obtained from the parenchymatous cells. Application of aloe vera gel in the food industry is increasing day by day as resource of drinks, beverages and ice creams (Eshun and He, 2004).

Aloe vera is a stem less and very shortstemmed succulent plant belongs to family Liliaceae (Surjushe A, 2008 and Ni et al., 2004). The medical uses of the gel juice (orally) are against skin diseases, constipation, radiation injury gastrointestinal, kidney and cardiovascular problems (Leila Akbari et al., 2013), reduce the cholesterol and triglyceride levels in blood. Recently other important property of Aloe vera has been reported such as anti-inflammatory and antibiotic activities against some diseases like diabetics, cancer, allergy and AIDS (Eshun et al., 2004; Reyhlds et al., 1999; Arowora et al., 2013). Aloe vera gel is also used in the cosmetic industry, including treatment of burns and scars and in wound healing (Aburjai et al., 2004). The antifungal activity of aloe vera gel has observed against several pathogenic fungi including Botrytis cinerea, main causative agent to decay grape fruit (Jasso de Rodriguez et al., 2005; saks and Barkai-Golan, 1995).

Edible coatings have been used since ancient time to protect perishable food stuffs from deterioration by retarding dehydration, suppressing respiration, improving textural quality to retain volatile flavor compounds, and reducing microbial growth (Debeaufort et al. 1998; Ozdemir et al.2010). Due to consumer demand for food without chemical

preservatives has resulted in application of natural antimicrobials preservatives and antimicrobial films and fungicide application can be reduced (Elmer and Reglinski, 2006). To avoid fruit spoilage it is essential to preserve fruits and it has been estimated that around 25% to 80% of harvested fresh fruits are wasted due to spoilage (Quezada *et al.*, 2003).

There are natural preservatives which are used as edible surface coatings for vegetables and fruits such as waxes but these coatings commonly contain ingredients such as polyethylene, carnauba and candelilla (Hagenmaier and Baker, 1995; Debeaufort et al., 1998; Alleyne and Hagenmaier, 2000). Amarante et al. (2001); Jeong et al., (2003) have studied wax coating as fruits preservatives and increase the shelf life, slows down ripening, retards water loss, reduces decay and enhances visual quality. The Aloe gel is made up of water, amino acids, vitamins, lipids, sterols, tannins, and enzymes (Shelton, 1991) and contains phenol, saponin, anthraquinones components, have anti-bacterial, antiviral and antifungal properties. Aloe vera has shown antibacterial property against gram positive and gram negative pathogens (Adetunji, 2008). Aspergillus, Fusarium and Penicillium are fungal species which are responsible for oxidation and spoilage of food (A. Babaei et al., 2013). Aloe vera can be applied as edible coatings for fruits as its biological activities prevent loss of moisture, firmness, control respiration rate and maturation development, delay oxidative browning, reduce and microorganism proliferation. (Valverde et al., 2005; Matinez Romero et al., 2005; Ahmed et al., 2009).

In analysis of the fact that increasing consumer demand for natural

preservatives and there are no recent report on the utilization of *Aloe vera* in increasing the shelf-life of grapes, this study was conducted to prevent decay and increase shelf period of grapes with the application of aloe vera gel, which is an economic and biodegradable natural preservative.

Materials and Methods

Fruit

Fresh healthy Indian Grapes 'Thompson', 'Sharad', 'Seedless' green color grapes were purchased from local market of Gwalior (M.P.).

Surface Preparation of Grape

The primary purpose of surface preparation was to remove all contaminants that would hinder proper coating adhesion and to render a sound, clean substrate, suitable for firm bonding.

Preparation of edible coatings (Preparation of *Aloe vera* gel)

Matured leaves of Aloe vera plant was harvested and washed with a mild chlorine solution of 25%. Aloe vera gel matrix was then separated from the outer cortex of leave and this colorless hydroparenchyma was grounded in a blender and resulting mixture was filtered to remove the fibers, and fresh Aloe vera gel was obtained. The gel matrix was pasteurized at 70°C for 45min. The gel was cooled immediately for stabilization and ascorbic acid (1.9 -2.0g L-1), citric acid (4.5 - 4.6gL-1) was added to maintain the pH at 4. The viscosity of the stabilized Aloe vera gel and stored in brown Amber bottle to prevent oxidation of the gel (He et al., 2005 and Adetunji et al 2012).

Application of *Aloe vera*

The experiment completely was randomized design. Two hundred (200) wholesome grapes single berries were randomly selected into four main groups, each three group of 50 (fifty grapes) was coated with 1.0%, 5.0%, 10.0% Aloe vera, while another group of 50 grapes berries were not coated (control, 0.0%). Aloe vera gel dilutions were prepared with distilled water as 0% (control), 1%, 5% and 10% then berries were dipped for 2-3 sec. Berries were dried, placed in air tight plastic container and stored at refrigerated temperature $(15^{\circ}C,$ 96-98%RH). Physicochemical parameters were carried out weekly in triplicates.

Grape quality assessment

Bunches of each treatment were examined by evaluation of berry and rachis appearance, incidence of cracked and shattered berries, decay, browning, acceptability, and flavor by a panel of 5 judges (Xu et al. 2007). The visual characteristics, including berry and rachis appearance, were scored in daylight. Fruit flavor was evaluated under red light in a taste room in order to avoid the interference of visual judgment.

So the quality of the grapes was assessed at the end of storage by evaluation of berry, incidence of cracked, brown, and shattered berries, berry color, taste, and weight loss.

Physicochemical Analysis

pH and titratable acidity

Whole fruit were passed through an electric juicer (Inalsa, India) and filtered through cheesecloth for the measurement of pH and titratable acidity (TA). pH was

measured by digital pH meter (WTW 526, Germany). TA was determined by titration of 5 ml juice diluted with 25 ml distilled water to pH 8.2 with 0.1 N NaOH and expressed as percentage.

Total phenolic content

(TPC) Total phenolic content was determined using the Folin-Ciocalteu method as described by Singleton et al. (1999),with minor modifications. Polyphenol extraction was carried out with 10 ml acidic methanol added to 1 gm of fine powder of specimen, kept at 4 °C, with the mixture then filtered through ordinary filter paper. Next, 150 µl of this extract was diluted with 350 µl of distilled water, and then 2.5 ml of Folin-Ciocalteu reagent and 2 ml of 7.5% sodium carbonate were added to the mixture. This reaction solution was shaken in a shaker and kept in the dark for 2 h. The absorbance of the samples was measured at 765 nm with a UV spectrophotometer. Gallic acid was used as a standard for obtaining the calibration curve.

Data were expressed as milligram of gallic acid equivalent (mg GAE) per gram of fruit fresh weight.

Microbial limit test

Daily observations were taken and on 7th, 21st, 28th and 40th day microbial count was estimated by the microbial limit test (MLT) along with pathogen estimation. With the Microbial Limit Test (MLT) analysis of total number of viable aerobic count with in limit was estimated. For this purpose 10 gm of sample was dissolved in 90 ml of buffer peptone water and 1 ml of each pretreated dilution sample solution to four sterile petri plates two plates for Bacteria and remaining two for Fungi

count, then sterile 15 ml SCDA (soyabean casein digest agar) at not more than 45°C poured in to two plates labeled for bacterial count and 15 ml of sterile liquefied SDA (sabraud dextrose agar) at not more than 45°C poured in to two plates labeled for fungal count. Then the plates were incubated at 30°C to 35°C for 5 days for bacterial count and 20°C to 25°C for 5 days for fungal count respectively. The number of cfu per gm of the sample was calculated as examined. The number of colonies should be in limit as: For Bacterial Count = NMT 1000 cfu per ml or gm. For Fungal Count = NMT 100 cfu per ml or gm.

Results and Discussion

The results indicated that Aloe vera gel treatment may be used as biopreservative to grapes for retarding quality losses. In table 1: The maximum total soluble solid (TSS) for coated grapes was 12.3 with 10.0 % application while that of uncoated oranges was found to be 11.9 at the end of There was no significant difference in TSS values with coated grapes. The pH of the grape juice was found to be gradually increasing during the course of storage as shown in table. The final value of pH for uncoated oranges was 4.86 and for coated grape was 4.90 + 0.01. There was no significant difference between the two treatments (5.0% and 10.0%). It was found that coated grapes had higher value at the end of storage period; this was due to the semipermeability created by aloe vera coatings on the surface of the fruit, which might have modified the internal atmosphere i.e. endogenous O2 and CO2 concentrations in the fruit, thus retarding ripening (Lowings and Cutts, 1982; Bai et al., 1988).

Results show the effects of Aloe vera coatings on titratable acidity (TTA) during

Quality Parameter	Methods of evaluation and units					
Berry appearance	Visual index of grape:					
(Figure - 2)	1 = excellent, $2 = $ good, $3 = $ slightly dull and shattered, $4 = <50%$					
	brownish and shattered berries, $5 = >50\%$ brownish and shattered					
	berries					
Flavor (Table -1)	Flavor acceptability using a 5-point scale:					
	1 = excellent, 2 = good, 3 = acceptable, 4 = poor, 5 = unacceptable					
Over all acceptability	Acceptability was evaluated on a scale of 9 to 1					
(Table -1)	where 9 = excellent, no defects; 7 = very good, minor defects; 5 =					
	fair, moderate defects; 3 = poor, major defects; 1 = unusable					

Table.1 Shows Changes in physio-chemical and overall acceptability of *Aloe vera* coated grapes during storage (Kittur et al., 2001) and Patricia et al., 2005)

Sr. No	Aloe vera coating (%)	Storage (days)	TSS	TTA (%)	pН	Flavor	Overall acceptability
1	0.0% 1.0% 5.0% 10.0%	0	12.0 12.0 12.0 12.1	0.30 0.31 0.30 0.31	4.87 4.88 488 4.86	1 1 1 1	9 9 9
2	0.0% 1.0% 5.0% 10.0%	40	11.9 12.0 12.2 12.3	0.25 0.29 0.30 0.30	4.86 4.90 4.91 4.91	4 2 1 1	1 7 9

Figure.1 Changes in total phenolic content of table grapes (Green) treated with *aloe vera* during storage at 15°C

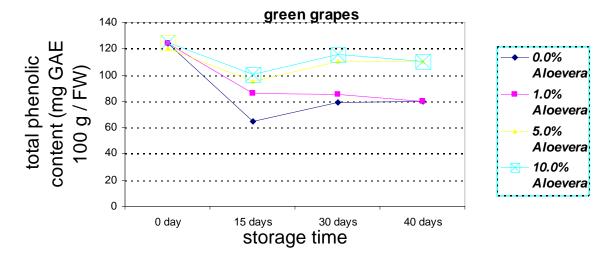


Figure.2 Visual index for Berry appearance on 0 day, 15th day and 40th day

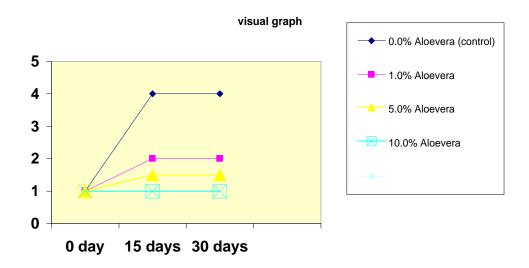


Figure.3 In the experiments disease severity assessment was performed according to the following empirical scale: 0 = healthy berry; 1 = one very small lesion (beginning of infection); 2 = one lesion, 3 = several lesions or 25% of the berry infected; 4 = 26-50% of the berry surface infected, sporulation present; 5 = more than 50% of the berry surface infected, sporulation present. Less disease severity assessment were shown with treatment of 5.0% and 10.0% *Aloe vera*.

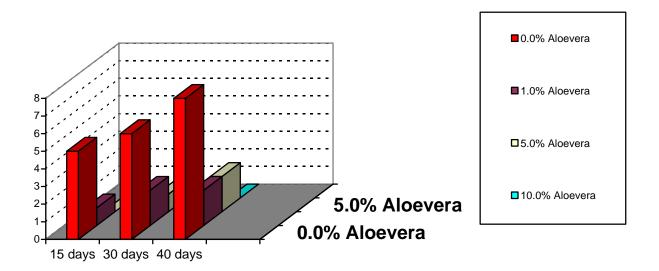


Figure.3.1 Number of infected berries recorded on the 40th day and graph indicates less infected berries with 5.0% and 10.0% *aloe vera* coating so more effectiveness was recorded with these

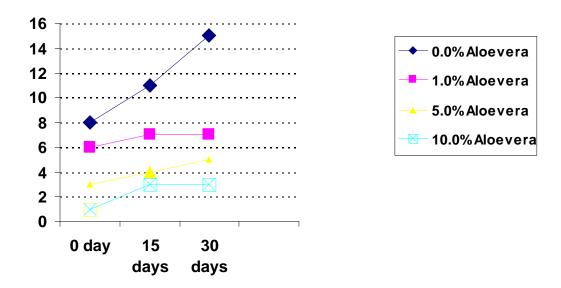
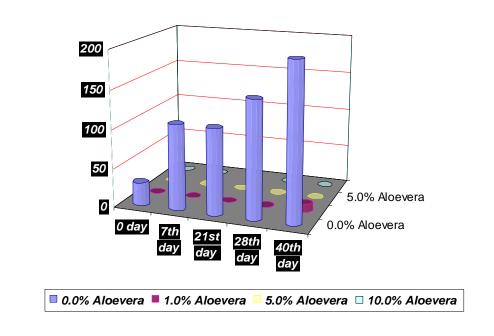


Figure.3.2 Shows microbial count (fungal growth in cfu/ml) on SCA media plates by microbial limit test, it has been observed bacterial counts (cfu/ml)



storage. TTA is directly related to the concentration of organic acids present in the fruits. No significant difference was found in titratable acidity of coated and uncoated grapes. The final value of TTA of uncoated grapes at the end of storage was found to be 0.30-0.05%, while that of coated grapes was 0.30± 0.01%. The decreasing acidity at the end of storage might be due to the metabolic changes in fruits resulting from the use of organic respiratory process; acids in observation was in agreement with the findings of Echeverria and Valich (1989).

Visual index: Visual index of grapes: 1 = excellent, 2 = good, 3 = slightly dull and shattered, 4 = <50% brownish and hattered berries, 5 = >50% brownish and shattered berries. Graph indicates untreated grapes shows highest index line indicating by blue color.

Figure shows the effects of coating on firmness also. There was a decreasing trend in firmness in both coated and uncoated oranges during the course of storage. However, the control (untreated fruits) exhibited higher loss in firmness than the coated samples; this was due to the effects of waxing which delayed softening in coated samples. This is in agreement with the observation by Hagenmaeir and Baker (1995) who found that coating with wax reduces shriveling of oranges than the control. The retention of firmness in coated fruits was due to reduction in degradation of insoluble protopectins to more soluble pectic acid and pectin. It was found that during fruit ripening, depolymerization or shortening of chain length of pectin substances occurs with an increase in pectineesterase and polygalacturonase activities (Yaman and Bayoindirli, 2002). Hence low oxygen and high carbon-dioxide concentrations reduce

the activities of these enzymes and allows retention of the firmness during storage (Salunkhe et al., 1991, Yaman and Bayoindirli, 2002; Patricia et al., 2005).

Figure 3. In the experiments disease severity assessment was performed according to the following empirical scale: 0 = healthy berry; 1 = one very smalllesion (beginning of infection); 2 = onelesion, 3 = several lesions or 25% of the berry infected; 4 = 26-50% of the berry surface infected, sporulation present; 5 = more than 50% of the berry surface infected, sporulation present, 6 = 100% of the berry surface infected, sporulation present. Less disease severity assessment were shown with treatment of 5.0% and 10.0% Aloe vera.

Figure 3.2. Shows microbial count (fungal growth in cfu/ml) on SCA media plates by microbial limit test, it has been observed bacterial counts (cfu/ml) were with in limit with treated samples, where as untreated samples showed bacterial counts as more than the limit of 100 cfu /ml. The shelf life of grapes is affected by respiration rate and weight loss, as grapes lose weight, more susceptible to fungal decay. Control (untreated grapes) showed higher fungal growth than A. vera-treated grapes. It has been reported that the antifungal activity of A. vera is based on the suppression of germination and inhibition of mycelial growth (Ali et al., 1999).

In conclusion, to our knowledge this is the first time that *A. vera* gel, applied as edible coating in grapes, has beneficial effects in retarding the ripening process. This treatment was effective as a physical barrier and thus reduced the weight loss and lowered the decay rate during postharvest storage. The results of this paper show that *A. vera* gel could be

applied as a postharvest treatment to inhibit microbial spoilage and reduce decay incidence during postharvest storage of grapes. In addition, the Aloe treatments led to grapes with better freshness without browning symptoms, and lower decay incidence after 40 days of cold storage. Bioactive molecules present in *Aloe vera*, such as aloe-emodin and aleonin have shown antifungal activity against *Aspergillus*, *Cladosporium* and *Fusarium*.

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