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SAFETY AND QUALITY ASSURANCE OF TOMATO AND LETTUCE USING *ALOE VERA* EDIBLE COATING

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Keywords : edible coating, *Aloe vera*, tomato, lettuce

Abstract

Nowadays, urban city inhabitants tend to fulfil their health needs by consuming commercially available ready-to-eat salads, which are normally sold in local supermarkets. The challenge faced by the local supplier is the short shelf-life of the fresh or minimally processed vegetables. This study assessed the application of *Aloe vera* gel as edible coating for tomato and lettuce. The effectiveness of two coating solutions made from fresh *A. vera* gel and spray-dried *A. vera* powder were compared. Evaluations were performed for organoleptic quality, physicochemical characteristics and microbiological assays. Organoleptic test showed that the application of coating was overall acceptable. Noteworthy was the finding at day 15, demonstrating higher freshness score for coated tomato samples compared to uncoated sample. The fresh gel coating application could maintain the texture firmness, as well as reduce the weight loss. The microbiological assays revealed that gel solution made of fresh *Aloe vera* was proven to inhibit the growth of microorganism. Nevertheless, this finding has negative correlation with pH and total soluble solids result. In conclusion, coating solution prepared from fresh *Aloe vera* gel was confirmed to be more effective in assuring the safety and quality of fresh vegetables compared to the gel prepared from spray-dried powder. This opens the possibility of application of the naturally abundant *Aloe vera* as modern edible coating in preparation of ready-to-eat salad that can be conducted not only by major industries, but more importantly by the small and middle local home industry.

1. Introduction

Fruits and vegetables are valuable sources of vitamins, minerals and fibres that are important for human nutrition. However, fruits and vegetables in the form of ready-to-eat (RTE) salad are the kinds that deteriorate easily. Thus, effective yet minimal-invasive preservation methods are required to prevent food spoilage. Food spoilage is not only found in the form of visual appearance, smell or taste of a food product that makes it unacceptable to the consumer. But more importantly, from a health standpoint, food spoiled by microorganism is unsafe to be consumed. *Salmonella sp.* and *Escherichia coli* are types of pathogenic bacteria that have a big influence to the human health, since they are the most common bacteria causing food-borne diseases in developing countries (Del-Portillo, 2000). Food poisoning from *Salmonella sp.* and *Escherichia coli* O157:H7 is related to the consumption of fresh fruits and vegetables, such as mung bean sprouts, tomatoes, watermelons and salads (Lukasik *et al.*, 2001). Therefore, the necessity to find a preservation method for mechanically vulnerable fruits and vegetables is increasing, even more significantly with the growing demand of RTE salad due to arising awareness of people to consume healthy food.

Several food preservation methods are available to reduce food spoilage. These methods vary from thermal processing, drying, freezing, irradiation, high pressure method, as well as addition of salts, antimicrobial agents or other chemical preservatives. However, these practices can not be applied to leaf salad and other mechanically vulnerable fruits due to their undesirable effects resulting from the techniques and the public's concern for human health (Rojas-Graü *et al.*, 2009). Nowadays, the use of modified atmosphere packaging (MAP) stands out among other methods in the effort for preserving freshness and safety of fruits and vegetables that are prone to mechanical damage (Oms-Oliu *et al.*, 2008, Chien *et al.*, 2007). The use of edible coatings with antimicrobial properties or with incorporation of antimicrobial compounds is another alternative to enhance the safety of fresh produce. Edible coatings can lessen the detrimental effects concomitant with minimal processing. They act as a good barrier for the exchange of moisture and oxygen, hence, reducing moisture loss, improving the fruit appearance and even functioning as antimicrobial and antifungal agents (Cha and Chinnan, 2004). Several types of edible coatings have been used for extending shelf-life of fresh commodities. Rojas-Graü *et al*. (2007) reported the efficacy of alginate and gellan edible coatings with the antimicrobial effect of plant essential oils, such as lemongrass, oregano oil and vanillin, to prolong shelf-life of fresh-cut apples. Raybaudi-Massilia e*t al.* (2008) also studied the ability of an alginate-based coating carrying malic acid and essential oils, i.e. cinnamon, palmarosa and lemongrass to improve the shelf-life and safety of fresh-cut melon. Their results showed that incorporation of 0.3% palmarosa oil into the coating is promising, since it could maintain the fruit quality parameters, inhibit the growth of the native microbiota and reduce the population of inoculated *Salmonella enteritidis*. The effect of incorporation of 0.4% of sorbic acid into hydroxypropylmethylcellulose (HPMC) coatings on tomatoes resulted in a significant reduction of *Salmonella* Montevideo (Franssen & Krochta, 2003).

Currently, there is an increasing interest in the use of *Aloe vera* gel in the food industry, beings applied as a source of functional foods in drinks, yoghurts, and ice creams (Kumar-Sampath, K.P. *et al*., 2010). *Aloe vera* gel as antimicrobial coating for fruits and vegetables was proposed by some authors, because of their proven antifungal and antimicrobial activities. The *Aloe vera* gel-based edible coatings was proven to have hygroscopic properties, thus, preventing moisture loss, reducing texture decay and controlling respiratory rate, while reducing microbial proliferation in fresh fruits and vegetables (Jasso de Rodríguez *et al.*, 2005; Valverde *et al.*, 2005; Martínez-Romero et al. 2006).

The objectives of this study were to evaluate the effect of application of Aloe vera as edible coating on tomato and lettuce with regards to their functional properties during storage, as well as its role in controlling microbial spoilage.

2. Materials and methods

Materials

Fresh leaves of *A. vera L.* were purchased from a local market in Bogor, Indonesia. *A. vera* spray-dried powder is obtained from PT. Aloe vera Indonesia. Tomatoes (*Lycopersicon esculentum Mill.*) and lettuce (*Lactuca sativa L.*) were procured from local markets in Bogor and Serpong, Indonesia. They were selected on the basis of size, colour and absence of external injuries.

Methods

Preparation of Coating Solutions

Fresh *A. vera* gel (FAV) was prepared starting with washing and soaking the leaves in chlorine solution 200 ppm for 30 min. After rinsing with boiled water, the gel matrix was separated from the outer cortex of leaves and this colorless *hydroparenchyma* was rinsed again with distilled water to remove the undesired yellow sap. Subsequently, the gel was ground in a blender. The juice was filtered to separate it from the dregs, and then pasteurized at 75°C for 15 minutes. Afterwards, carboxymethylcellulose/CMC (FVH9A, Hercules, Indonesia) and glycerine (Bina Karya Prima, Indonesia) was added to the solution. For tomato coating, 1% w/w CMC and 0.5% glycerine were added. As for lettuce, a lower amount of CMC (0.5% w/w) and glycerine (0.25% w/w) were used to avoid over-gelling of the coating solution. To prepare *A. vera* powder coating (PAV) solution 5% w/w *A. vera* powder were mixed with distilled water, heated at 75°C for 15 minutes, added with the same amount of CMC and glycerine, then cooled to room temperature before application.

Application of Edible Coating Solutions

Tomatoes and lettuces were washed with distilled water and dried before coating. They were dipped completely into the coating solutions at room temperature for 5 minutes, allowed to drain and let dry for about 30 minutes at room temperature. Then they were weighed, for storage put in a plastic packaging and stored at 5 ± 1°C. As uncoated samples (UCS) for control, tomatoes and lettuces were only washed with water and stored under same conditions as those for coated samples. The quality parameters of tomatoes were analyzed at 5 d interval until day 20. Whereas, those of lettuces were measured at 3 d interval until day 9, since a longer storage time caused lettuce samples to be considered unsafe for consumption due to spoilage.

Weight Loss

Water loss was calculated by the following equation: % weight loss = (A-B)/A x 100%, where, A is the initial weight of sample (day 0) and B is the weight after the storage period.

Texture Firmness

Flesh firmness of tomato was determined using Rheometer CR300 (Sun Scientific Co. Ltd., Japan) by pressing a plunger needle with a diameter of 2.5 mm into the tomato on 3 opposing surface. Penetration rate was 60 mm min^{-1} with a maximum load of 2 kg. Texture firmness value was expressed in mm s^{-1}. This measurement was only applied to tomato samples.

pH and Total Soluble Solids (TSS)

All samples were homogenized and the resultant pulp was filtered. The pH was determined using a digital pH meter (Schott, Germany). TSS was measured in triplicate for both control and coated samples by using a handheld refractometer (Atago Co. Ltd., Tokyo, Japan) at room temperature and expressed as the mean ± SD of % Brix.

Total Bacterial Plate Count

Total Plate Count (TPC) was carried out using plate count agar (Oxoid, UK) as the media. The surfaces of the samples were swabbed with sterile cotton swabs, both horizontal and vertical diameter of tomato surface, as well as the axes and diagonals of lettuce surface. Serial dilutions with the same dilutent were performed. Samples were prepared in triplicate and all plates were incubated for 24 h at 36°C.

Sensory Analysis

Sensory analysis was performed by 15 untrained panelists, aged 20-40 years. For tomatoes, the parameters of interest were overall acceptance and freshness. For lettuce samples, the sensory qualities analyzed were appearance, taste, crunchiness, freshness and overall acceptance. The panelists were asked to give a score based on Hedonic scale to the attributes in question. The score is defined on a ranked scale of 1 to 9, where 1 = dislike extremely, 2 = dislike very much, 3 = dislike moderately, 4 = dislike slightly, 5 = neither like or dislike, 6 = like slightly, 7 = like moderately, 8 = like very much, 9 = like extremely.

Statistical Analysis

The data obtained from the experiments were expressed as mean ± SD. They were subjected to single factor analysis of variance (ANOVA) and further analyzed with t-test when the significance of the difference at $p < 0.05$.

3.Results and discussion

3.1. Weight Loss and Texture Firmness

A. vera gel coating was proven effective in delaying weight loss. After 20 days of storage, control sample lost in total 2.88% while the loss of weight in samples treated with fresh *Aloe* gel were the lowest followed by *Aloe* powder (Table 1). The percentage was more than double in control than in fresh *Aloe*-treated fruit after 20 days at 5°C. This result was in accordance to the sensory evaluation result for tomato freshness. The less the weight loss value, the more bound water kept inside the cell structures, making the more rigid, thus, enhancing the juiciness of the tomatoes. Valverde *et al*. (2005) found similar result that *Aloe* coating was effective in lessening the weight loss of table grapes. The mechanism for this positive effect is based on their hygroscopic properties, which enables formation of a barrier to water diffusion between fruit and environment, thus avoiding its external transference (Morillon *et al*., 2002).

With respect to firmness, the measurement used a plunger needle. The deeper the penetration of the plunger needle, the softer the tomato texture, thus the lower the firmness value. The lowest score interprets the softest tomato texture. At day 0, all samples had quite similar firmness value to one another. All tomatoes softened during cold storage, but to a greater extent in control than in coated and *Aloe* powder-coated tomatoes (Figure 1). Fresh *Aloe*-gel coated samples displayed the highest firmness result after 20 days of storage. ANOVA analysis showed that the average firmness of FAV samples were significantly different from PAV and UCS samples. Even though chilling injury was observed for some samples, it was clear that fresh *A. vera* gel was the best coating to maintain the texture consistency of tomato samples, since the gel could protect the outer layer of tomato from possible friction that might damage the outer surface. Furthermore, the fresh gel might also decelerate textural deformation probably caused by microbial spoilage due to its antimicrobial characteristics.

3.2. pH and Total Soluble Solids

The pH values of all tomato samples were indistinguishable (Table 2). pH ranged from 3.93 ± 0.11 to 4.23 ± 0.03 and pH changes from day 0 to 20 were very minimal. This indicates that all samples were generally similar in term of acidity level. This also showed that the application of gel coating did not influence the pH. Thus, pH did not contribute to antibacterial effect of *A. vera* gel.

Total soluble solids for coated and uncoated samples showed no significant difference at any storage time (Table 3). This result is in conformity with the study from Muchtadi and Sugiyono (1989) that showed during the growth and maturity process, tomatoes do not or barely have any increase in their sugar content level. Hence, it can be concluded that in this study the TSS had negative correlation to antibacterial characteristic of *Aloe* edible coating found in the microbial analysis.

3.3. Microbial population

For tomato samples, TPC was performed after the storage time reached 5 and 20 days, whereas for lettuces, it was carried out on day 3 and 6. This was caused by the shorter lifespan of lettuce compared to that of tomato. Spoilage of tomato was not visible at day 20, but observable at day 25. Therefore, day 20 was chosen as the limiting time for tomato sample. As for lettuce, spoilage was detected at day 9, thus TPC was done for day 3 and 6. Total viable counts in tomatoes on day 20 increased significantly for PAV and UCS samples. Whereas, in FAV samples only a slight increase was recorded. It was obvious that samples coated with fresh *Aloe* gel had the least microbial count compared to control as well as those treated with *Aloe* powder. The bacterial count on PAV samples was quite similar to the UCS samples. This signified that the gel made of fresh *A. vera* worked better in retarding microbial growth in both tomato and lettuce compared to the gel made of *A. vera* spray-dried powder. Similar result for tomato and lettuce samples was observed. The reduction of the growth of 17 bacteria by *A. vera* gel has been proven (Reynolds and Dweck, 1999), being more effective against gram positive than gram negative microorganisms (Ferro *et al*., 2003). Some individual components found in *A. vera* gel, such as saponins, acemannan and anthraquinones derivatives, are known to have antibiotic activity, and thus, could be responsible for its antibacterial activity.

3.4. Sensory analysis

In this study, 15 panellists examined the freshness and the overall acceptance of tomato and lettuce samples. These panellists were untrained and unbiased, so the result should demonstrate the acceptance level in normal consumers. For tomatoes, the organoleptic test was only performed until day 20, since the samples were not considered as safe for consumption, because spoilage was detected on day 25. The sensorial analyses of tomatoes rendered generally higher scores for freshness and overall acceptance after 20-day storage (Table 4). These results are in agreement with the lower weight loss observed in *Aloe*-coated tomatoes. Moreover, the *A. vera* coating effected an attractive shiny-looking sheen to tomato skins, which was correlated to lower changes in dehydration. Interestingly, none of panellists could detect any "off-flavor" attributed to the *Aloe* treatment. During storage, the FAV samples could maintain their sensory qualities, whereas the PAV and UCS samples generally underwent decrease in acceptance level over the storage period. Furthermore, it was also recorded that the two samples were not significantly different from each other, as perceived by the panellists.

Appearance, taste, freshness and overall acceptance, the major sensory attributes of lettuce samples, were scored by panel members (Table 5). There were more parameters observed by using sensory analysis for lettuce than for tomato. This is due to the fact that lettuce is a kind of leafy vegetables, making it uneasy to undergo any kind of mechanical test, thus, organoleptic test is the alternative answer. The test was performed in 3 days interval until day 9. The test was halted on day 9, since the samples afterwards showed apparent spoilage already. During storage time, the score of the parameters of lettuce samples tended to decrease. The decline of scores indicated the deterioration of the lettuce quality, which influenced all parameters tested in organoleptic test. For appearance parameter, it could be observed that FAV samples generally were the less preferred one compared to PAV and UCS samples. The *A. vera* gel were still in good intact with lettuce surface, leaving the surface looking thich and the leaves became folded to one another. Due to the thickness of the coating, the leaves of FAV and PAV samples were likely to stick with each other after coating was applied, hence causing some leaves to fold as well. With time the folded side showed darkening in color due to pressure from the thick coating. During processing, many parts of the leaves were also torn due to the weight of the gel itself, thus, worsening the appearance. In taste, FAV scored the least compared to PAV and UCS. Panellists reported the presence of bitterness for the FAV samples and 'strange taste" for PAV samples. The same preference was also observed for freshness attribute. This might be due to the thickness of the coating on lettuce surface, which caused glueyness to the leaves. For overall acceptance, panellist members persistently chose the uncoated samples as their preference. ANOVA analysis showed that UCS samples were notably distinguishable from the other samples. The scores for FAV and PAV samples on day 3 and 6 were more or less comparable. However, the panellists significantly disliked the FAV samples after 9 days of storage. After coating, fresh *A. vera* gel displayed a more consistent intact with the surface of lettuce samples. As a consequence, the thickness of the coating was perceptible, affecting the panellists' appreciation to the samples. Therefore, it could be concluded that *A. vera* gel coating is not suitable for leafy vegetables, such as lettuce.

4.Conclusions

From sensory analysis of tomatoes, both FAV and PAV samples were comparable to the control. At day 15, the uncoated sample was significantly disliked, since it showed signs of deterioration. However, the *Aloe* gel could lessen this perishing signs, resulting in higher overall acceptance value for the treated tomatoes. Nevertheless, it appeared that these treatments were not suitable for lettuce. The panel test resulted in notable difference among the samples, with untreated samples as the most preferred one. Application of *Aloe* coating could retard the weight loss and better maintaining the texture firmness of tomato, where FAV coating performed better than PAV. FAV coating was proven as well to inhibit the growth of microorganism, while PAV coating did not give the effect correspondingly. The pH and TSS measurements showed that all samples were indistinguishable, hence, these parameters were negatively correlated with the observed antibacterial effect of *A. vera* coating. In conclusion, edible coating using *A. vera* gel was proven to be beneficial to preserve the safety and quality of fresh fruit. Further improvement for gel formulation in order to be suitably applied for leafy vegetables should be investigated in the future.

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Table 1 Percentage of weight loss during cold storage of control and *Aloe vera*-coated tomatoes.

Day	FAV	PAV	UCS
5	0.30%	0.23%	1.14%
10	0.16%	0.23%	0.70%
15	0.46%	0.07%	0.45%
20	0.37%	0.39%	0.59%
Total	1.29%	1.55%	2.88%

Table 2 pH values for tomato samples.

Day	FAV	PAV	UCS
0	4.07 ± 0.06	4.01 ± 0.02	4.06 ± 0.06
5	3.93 ± 0.11	3.97 ± 0.06	4.06 ± 0.09
10	4.07 ± 0.06	4.04 ± 0.08	4.14 ± 0.05
15	3.93 ± 0.11	4.23 ± 0.03	3.99 ± 0.26
20	4.08 ± 0.03	4.18 ± 0.10	4.04 ± 0.04

Table 3 Total soluble solids of tomato samples.

Day	FAV (%Brix)	PAV (%Brix)	UCS (%Brix)
0	3.767 ± 0.225	3.733 ± 0.314	3.750 ± 0.394
5	3.783 ± 0.417	3.800 ± 0.358	3.800 ± 0.460
10	3.833 ± 0.234	3.850 ± 0.243	3.933 ± 0.306
15	3.800 ± 0.237	3.850 ± 0.259	3.817 ± 0.256
20	3.917 ± 0.293	3.850 ± 0.259	3.783 ± 0.256

Table 4 The average of organoleptic scores of tomato samples.

Storage time (days)	Type of Coating	Freshness	Overall Acceptance
5	FAV	6.8	6.8
	PAV	6.8	6.9
	UCS	6.9	6.9
10	FAV	6.2	6.5
	PAV	5.9	6.6
	UCS	6.3	7.1
15	FAV	7.0	6.6
	PAV	5.9	6.6
	UCS	5.5	5.5
20	FAV	6.1	6.7
	PAV	5.4	6.3
	UCS	5.8	6.1

Table 5 The average of organoleptic scores of lettuce samples.

Storage time (days)	Type of Coating	Parameters			
		Appearance	Taste	Freshness	Overall Acceptance
3	FAV	5.2	4.4	4.7	5.9
	PAV	5.6	6.3	5.3	6.1
	UCS	6.9	6.6	6.9	7.3
6	FAV	4.3	3.2	3.2	5.1
	PAV	5.1	5.4	4.8	4.9
	UCS	6.6	6.1	6.4	6.8
9	FAV	1.8	2.4	2.4	2.8
	PAV	4.3	4.2	3.9	4.6
	UCS	5.1	5.2	5.1	5.6

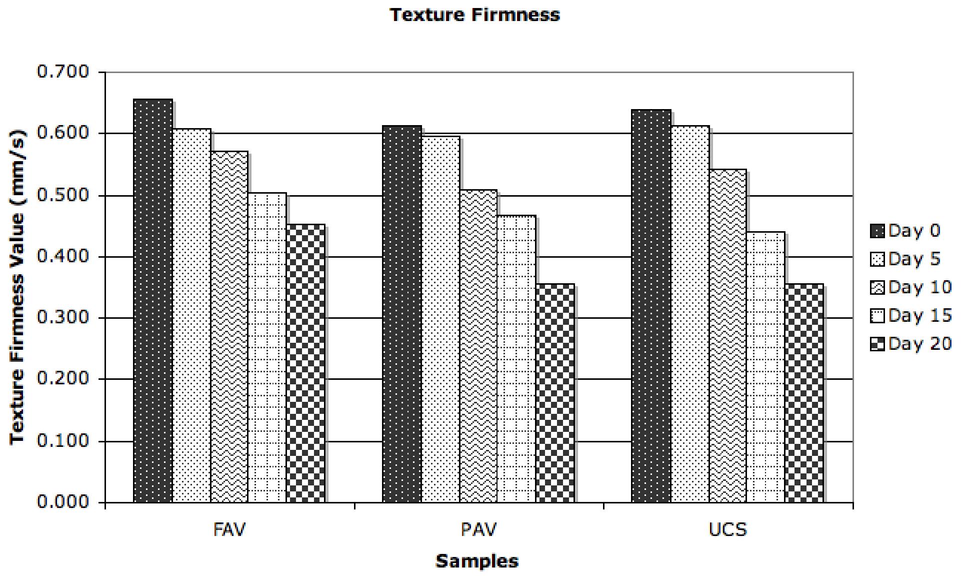

Figure 1 Texture firmness score of tomato samples.

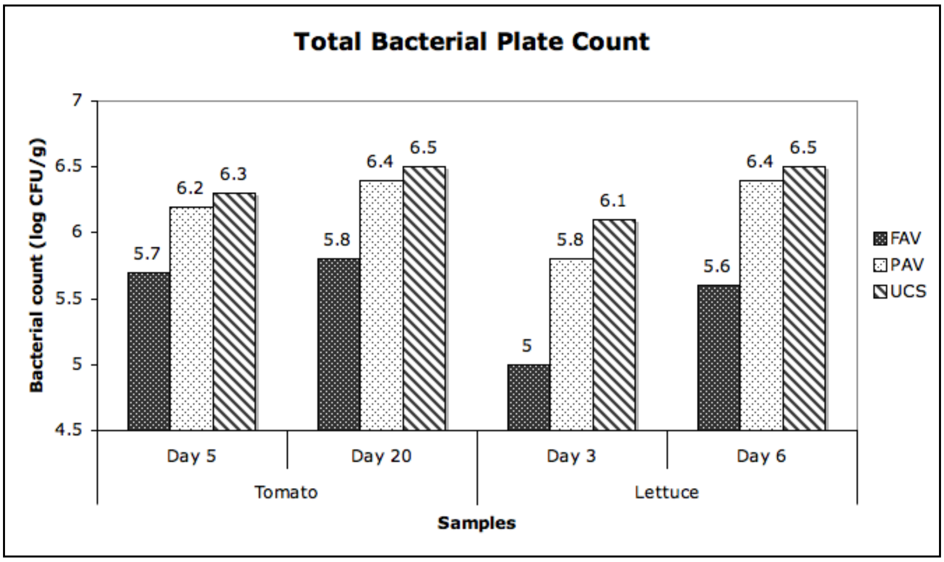

Figure 2 Total bacterial plate count for tomato and lettuce samples.