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Effect of Alginate-Chitosan Edible Film Coating on Vitamin C loss of Fresh Cut Sliced Pineapple (*Ananas Comosus* (L.) *Merr*)

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Abstract. Pineapple as tropical fruit with high content of vitamin C, is a perishable fruit easily loses vitamin C due to air oxidation. Fresh fruit coating is required using environmentally friendly biodegradable materials. A study on the effect of alginate-chitosan film coating on the loss of vitamin C in fresh cut pineapple has been carried out. Pineapple coating was carried out by dipping fresh cut sliced pineapple in alginate-chitosan hydrosol which had been prepared in mass ratio alginate and chitosan of 1:1 at pH 5.28. Vitamin C content of pineapple fruit at storage temperatures of 4, 13 and 27 °C was measured by UV-Vis spectrophotometer at maximum wavelength of 660 nm. The stability test of vitamin C content due to film coating was carried out at various storage period of 1, 5, 24 hours, 3 and 7 days at temperature of 4 °C. Biodegradability test of alginate-chitosan film was carried out through a burried test and the degradation of the functional groups of the film was characterized using FTIR Spectrophotometer. Alginate-chitosan film coating can reduce the loss of vitamin C in fresh cut sliced pineapple at all storage period. The reduction of vitamin C content up to 7 days of storage period was 8.81%. The percentage of decomposition of the alginate-chitosan film reached 83% on the 28th day of the burried test with the loss of protonated amine and carboxylate functional groups. It was suggested that alginate-chitosan film coating could be a useful treatment of maintaining vitamin C content in fresh cut sliced pineapple.

INTRODUCTION

Pineapple is one of Indonesia's favorite fruit, which is produced in almost all part of Indonesia with production reached 2.45 million tons in 2020 [1]. Due to its execellent taste and ascorbic acid (vitamin C) content, pineappleis known as queen of fruits [2]. Vitamin C in pineapple acts as an antioxidant that can reduce damage caused by free radicals that can trigger cancer [3,4]. In General, pineapples are in demand for fresh consumption due to its maintained content of vitamins dan enzyme, besides being processed into canned fruit, syrup, jelly, vinegar [5]. Processing of fresh-cut pineapple involves grading, washing, peeling, slicing, and packaging affects the levels of vitamin C in the pineapple [6]. Proper packaging for fresh pineapple is needed to prevent the reduction of vitamin C levels in pineapples, one of which is using edible film. Edible coating could improve shelf-life of fresh cut fruits, provide good apperance and brightness, hence fruits more attractive to the market [7,8].

Natural polymers such us starch, lipid, polysacharide, and protein are widely used as edible coating for fruits and vegetables [9,10]. Due to its excellent characteristic including film-forming capacity, good mechanical as well as antimicrobial properties, and can form transparent film, chitosan based-edible coating has been choosen [11]. Chitosan coating can extend shelf-life of sweet cherry [12], strawberry [13], banana [14], Ber fruit [15], mango [16] etc. But, chitosan has limitation in acidic condition, hence modification with other polymer is needed.

In previous study [17], chitosan was interacted with alginate to form polyelectrolite complex alginate-chitosan that has better mechanical strength and antibacterial activity than native polymer, chitosan and alginate. Interaction of amina protonated of chitosan with carboxylate groups of alginate in polyelectrolite complex alginate-chitosan produce tight structure. It is expected alginate-chitosane coating may inhibit the loss of vitamin C in fresh fruit. In this study, the fresh cut sliced pineapple was coated with alginate-chitosan film. The effect of film coating to vitamin C loss of fruit in various temperature and period storage was determined. The biodegredability as well as FTIR analysis of alginate-chitosan film was carried out.

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MATERIAL AND METHODS

Material

The reagent used in this study were analytical grade, chitosan (deacetylation > 90%, Mol. wt. 300 KDa-400 KDa), sodium alginate (Mol. wt. 12Kda-40 KDa) were obtained from Sigma-Aldrich (USA). Natrium hydroxide, glacial acetic acid, ascorbic acid, oxalic acid, ammonium molybdate, sulfuric acid were purchased from Merck. Pineapple (Ananas comosus (L.) Merr) was obtained from local bazaar at Mataram-West Nusa Tenggara.

Work Procedures

Preparation of Chitosan Alginate Hydrosol and Coating on Pineapple Fruit

Alginate-chitosan hydrosol was prepared following the method of Hermanto et al [18] by mixing alginate hydrosol and chitosan hydrosol in a ratio (1:1). Alginate hydrosol was made by mixing 1 gram of alginate into 25 mL of distilled water using a magnetic stirrer until homogeneous. Chitosan hydrosol was prepared by dissolving 1 g chitosan in 25 mL in acid condition (using 1 mL of glacial acetic acid). Both hydrosols were left overnight to remove air bubbles. Furthermore, the two hydrosols were mixed by adding alginate hydrosol to chitosan hydrosol in stirring condition. pH of solution was adjusted until 5.28 by adding 10% (w/v) NaOH. The hydrosol coating on pineapple was conducted by modified the method [19] by dipping fresh pineapple cut with a size of 1×1×1 cm into chitosan-alginate hydrosol for 30 s and dried using a fan. The pieces were placed on styrofoam and covered with plastic wrap. All samples were stored at 4, 13 and 27 °C for 6 days. Meanwhile, the stability test was done by stored the samples at 4 °C for 1, 5, 24 hours, 3 and 7 days.

Determination of Vitamin C Levels

Determination of vitamin C levels was carried out using spectrometric method with ammonium molybdate as reagent [20]. Firstly, stock solution of vitamin C (1000 ppm) was prepeared by dissolved ascorbic acid in deionized water containing 0.4% oxalic acid solution. Maximum absorption was determined by scanning of incubated solution (30 min) containing 0.8 mL vitamin C stock solution (1000 ppm), 4 mL H_2SO_4 solution 5% and sufficent ammonium molibdate 5% to give a total volume of 10 mL. Scanning was done in the wavelength range of 400-800 nm. Calibration curve was constructed by using standart solution of vitamin C at various concentration (0-10 ppm) with same reagents of maximum wavelength determination. Absorbance was measured at maximum wavelength against a reagent blank prepared in the same manner but without vitamin C. Calculation of vitamin C levels was carried out by extrapolating absorbance data in a linear regression equation from the calibration curve of vitamin C.

Characterization of Edible Film and Biodegradation Test

The biodegredability test was carried out by burying the alginate-chitosan edible film in the soil (Eq. 1) [21] film had been prepared previously on a 10×10 cm glass plate and dried at room temperature overnight for 2×24 hours. The functional groups on the edible film before and after biodegradation were characterized by using an FTIR spectrophotometer with the KBr pellet method.

$$\% \text{ biodegredability} = \frac{W-W_0}{W_0} \times 100\% \quad (1)$$

W_0 is the initial weight and W is the final weight of alginate-chitosan edible film.

RESULTS AND DISCUSSION

The molybdenum blue complex (blue colour) formed on reduction of ammonium molybdate with ascorbic acid in the presence of sulphate [20] at a maximum wavelength of 660 nm in this work. The equation of calibration curve is Y=0.0175x + 0.0013 with R value of 0.9914, indicated linear relationship of absorbance and ascorbic acid concentration. This equation is used to calculate vitamin C levels in pineapples coated with edible film (sample) and uncoated (control). Then, level of vitamin C in various temperature storage at 6 days storage was determined, as described in Fig. 1.

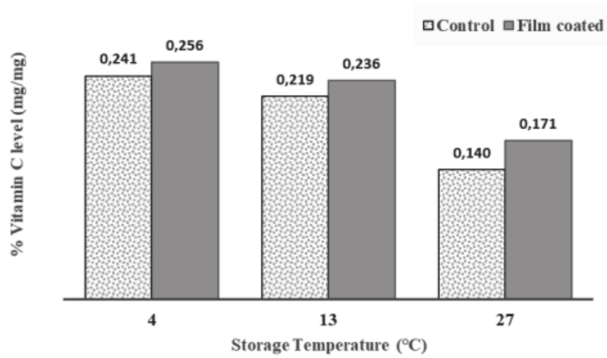

FIGURE 1. Effect of storage temperature on % vitamin C level in fresh cut sliced pineapple

FIGURE 1. show that samples coated with edible film had higher vitamin C level than the control for all storage temperature, indicate that alginate-chitosan edible film coating is able to inhibit the loss of vitamin C levels in fresh cut sliced pineapple. Interaction of alginate and chitosan via electrostatic in the film induce the distance between molecules getting closer produce tight structure in film [22], then the oxygen permeablity is decreased. Oxygen contributed in many degradation in food including vitamin loss via oxidation of vitamin C [23], hence coating alginate-chitosan film as barrier of oxygen could reduce the loss of vitamin C in fruit. Storage temperature also affect delaying vitamin C in fruit [24]. Fig. 1 shows the relationship between storage temperature and vitamin C levels in fresh cut sliced pineapple, the higher the storage temperature, the lower the vitamin C content, both in the sample and in the control. This is because kinetics of vitamin C oxidation is corelated with temperature. At room temperature, vitamin C in pineapple is oxidized faster than at 4 °C and 13 °C. Moreover, low humadity also slowed down chemical and biological process as well as vitamin C loss in fruit [25]. Interaction of amina protonated of chitosan with carboxylate groups of alginate in alginate-chitosan film increase the hydrophobicity of film and greater resistance to water has achived [22,26].

The stability of vitamin C level in fresh cut sliced pineapple coated with alginate-chitosan edible film was carried out at various storage time of 1, 5, 24 hours, 3 and 7 days at storage temperature of 4 °C, as shown in FIGURE 2. The stability of vitamin C during storage period 7 days decreased by 8.81%. It can be seen that the alginate-chitosan edible film coating was able to mantain levels of vitamin C in fresh cut sliced pineapple due to its properties as barier of oxygen and water.

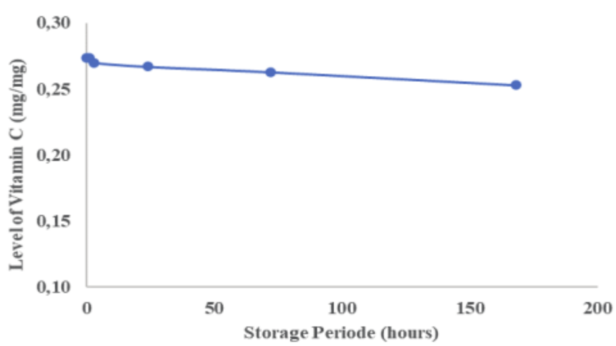

FIGURE 2. Stability of vitamin C level in various storage periode

Edible film alginate-chitosan were characterized using FTIR, as well as the degraded edible films, as shown in FIGURE 3. While, the interpretation of spectra was described in TABLE 1.

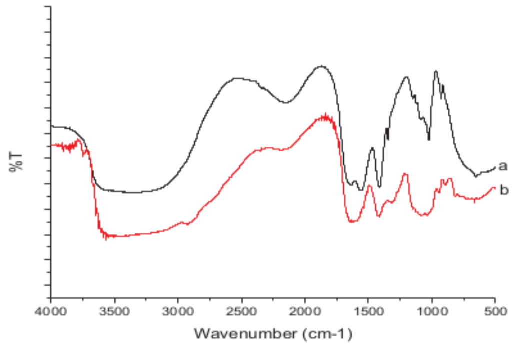

FIGURE 3. FTIR spectra of alginate-chitosan edible film before (a) and after (b) biodegradablity test

TABLE 1. Interpretation of alginate-chitosan FTIR spectra before (a) and after (b) biodegradablity test

Vibration	Alginate-Chitosan Film (cm^{-1})	Degraded Alginate-Chitosan Film (cm^{-1})
N-H and O-H	3354	3584.95
CC(sp) and =C-N	2154.69	-
C=C, CN and C=O	1639.16	1650,83
NH_3^+	1559.9	-
C-H bending	1414.6	1415,92
$-COO^-$	1346.08	-
C-O-C	1023.7	1080.38
Aromatic C-H	928.22	893.22
C-S, S-S	654.05	666.37

Alginate-chitosan edible film spectra showed absorption at wave number 3354 cm^{-1} which indicat the presence of –OH group from alginate and –NH group from chitosan. Absorption at 1559.9 cm^{-1} was the vibration of protonated amine group. Absorption in the 1639.16 cm^{-1} indicates the presence of C=C, and C=O groups, absorption in the 1400-1300 cm^{-1} indicates C-H bending. The presence of carboxylate groups in film was indicated by absorption at 1346.08 cm^{-1}. This is in accordance with previous studies which indicated protonated amine groups and COO- groups on alginate-chitosan edible films that appeared in the 1550 and 1398 cm^{-1} [17,18].

Biodegradablity of alginate-chitosn edible film was evaluated by soil burried test for 7, 14, 21 and 28 days. The weight loss of film during soil burried was performed in percentage, as shown in FIGURE 4.

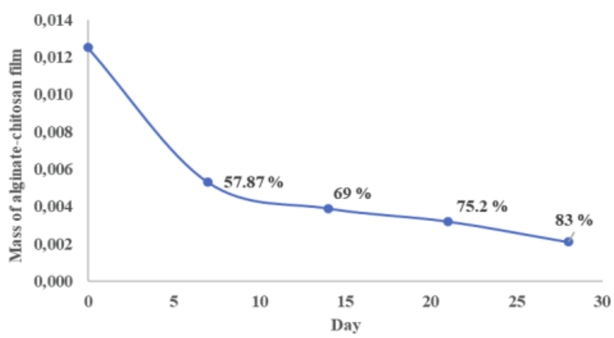

FIGURE 4. Weight loss of alginate-chitosan film during soil burrial test

The percentage of biodegradability obtained after being buried for 28 days was 83% with a loss of protonated amine, carboxylate groups and =CN functional groups. This indicates that functional groups containing oxygen and nitrogen is well degraded via soil burried, hence these groups is responsible for the degradablity of the film.

CONCLUSION

Chitosan alginate film was able inhibit the loss of vitamin C in fresh cut sliced pineapple at various temperatures. The reduction of vitamin C content up to 7 days of storage period was 8.81%. The biodegradability test on the soil burried after 28 days showed 83% weight loss of the film. The functional groups containing oxygen and nitrogen such us protonated amine, carboxylate and =CN groups is responsible for the biodagradability of the film according to FTIR spectra. Good characteristics of alginate-chitosan film make it an excellent coating for mantaining vitamin C in fresh cut fruit.

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