

Effect of Chitosan-based Edible Coating on Biological Properties and Quality of Fruit

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Abstract— The objective of this study is to develop chitosan based edible coating and to investigate the effect of chitosan edible coating with different concentration of turmeric oil (*curcuma longa*). The effectiveness of chitosan fruit coating with additional of *curcuma longa* to delay the ripening process and preserve the quality of strawberry was evaluated through several analyses include the appearance, weight loss, disk diffusion and total plate count. Different concentration of *curcuma longa* was used which were 5uL, 10uL, 15 uL and 20 uL and the storage period was 5 days under room temperature. Through the appearance analysis, chitosan with additional of 10 uL of *curcuma longa* had the best appearance for three days compares to the chitosan edible coating with other concentration of *curcuma longa*. The disk diffusion analysis proves that the chitosan with 10uL of *curcuma longa* had the highest inhibition zone which is 17 mm compares to other concentrations. Meanwhile, the weight loss analysis shows that the chitosan with 15 uL of *curcuma longa* had the lowest weight loss which is 3.35% after 5 days of leaving period. Thru the total plate count analysis, the chitosan edible coating added with 15 uL of *curcuma longa* has the lowest total colony forming unit (CFU) which is 4400 CFU/mL and the log CFU is 3.64. Through conducted analysis, the chitosan coating with additional turmeric oil can preserve the quality of the strawberry and prolong its shelf life.

Keywords— *Chitosan, edible coating, essential oil, strawberry.*

I. INTRODUCTION

In this technology era, a lot of methods in food technology emerge with the aims to preserve the quality of fruit and increase its shelf life. Researchers have done a lot of study on fruit coating and the most common process is fruit waxing. Fruit waxing is done by covering fruits with artificial waxing material. Usually, fruits have their own natural waxes but, due to washing process after the harvest, their waxes have been wash off. This intervention technology use waxes which can be come from natural source such as carnauba waxes and beeswax or petroleum based waxes such as polyethylene. This method is used to prevent water loss and slow down spoilage and shrinkage. It also able to slow the ripening process, inhibit mould growth and give shiny and glossy appearance as a good impression for customers. By waxing the fruit, it also decreases the rate of transpiration and retard the browning process of the fruit. Therefore,

the shelf life of fruit can be extended and quality and nutrients of the fruit can be retained.

Although gives many benefits in food engineering, it also has the drawbacks. Due to this problem, edible coating is invented to solve the rising problems. Edible coating is a thin layer of materials that offers a barrier to oxygen, microbes that come from environment, moisture loss and solute movement for food plus, it can be consumed by consumers (Raghav, Agarwal, Saini, Vidhyapeeth, & Vidhyapeeth, 2016). It also does not add unfavourable properties to the fruits and environmental friendly due to its natural biodegradable source. It works by delaying the rate of respiration, decrease weight loss and extend the shelf life of fruits and vegetable during the postharvest storage (Kerch, 2015).

Edible coating is classified into three major classes taking into account the nature of their component: hydrocolloid (which contains proteins, polysaccharide or alginate) lipids (formed from fatty acids, acylglycerols or waxes) and composited (combination substances from the two classes) (Skurtys O., Acevedo C., Pedreschi F., Enrione J., Osorio F., n.d.). In this study, chitosan coating in hydrocolloid categories is investigated. Chitosan is derivation of chitin component via deacetylation in alkaline media and it is a natural, nontoxic copolymer consist of β - (1-4)-2-acetamido-D-glucose and β -(1-4)-2-amino-D-glucose units. It is abundant as it naturally can be found in the exoskeleton of crustaceans, fungal cell walls and other biological materials (Conceição et al., 2015). Chitosan coating preserve the quality of fruit by regulating gas exchange and decrease the respiration. Furthermore, by lowering the respiration rate, it reduces the oxidative process due to reduction of reactive oxygen species accumulation. It has been proven by applying the chitosan treatment on postharvest fruit including logan, pear, plum and apple to maintain its quality (Liu et al., 2016).

Nowadays, researchers have developed new edible coatings with combination of numerous and edible herbs such as essential oil (such as turmeric oil) to enhance the edible coating. Essential oils integrated into the chitosan coating has the capability to improve the antimicrobial property for application on the storage of fruit. The addition of essential oil would give significant effect on antimicrobial activity against bacteria, yeast and moulds and higher safety to environment and consumer (Xing et al., 2015).

II. METHODOLOGY

A. Preparation of Chitosan Solution

The chitosan flakes were prepared by diluting it with acetic acid. First, the acetic acid was prepared by diluting the acetic acid with 100 mL distilled water thru dilution of 0.5% v/v. Next, 1g of chitosan flakes was mixed with the diluted acetic acid and the mixture was stirred for 24 hours at room temperature. After 24 hours of stirring, 2 mL of glycerol and 0.1 mL of Tween 80 were added to the mixture. Then, the pH of the chitosan solution was adjusted by using 0.1 M Sodium Hydroxide (NaOH) until it reached 5.6 while being stirred (Khaliq, Mohamed, Ding, Ghazali, & Ali, 2016).

B. Preparation of Tapioca Starch Solution

Start with, 8 g of tapioca powder was weighed by using electronic weight scale. The 8 g of tapioca starch powder was dissolved in 100 mL of distilled water for 40 minutes at 80°C. Next, 2 mL of glycerol is added to the tapioca solution as a plasticizer (Khaliq et al., 2016).

C. Preparation of Chitosan, Starch and Essential Oil

First, 100 mL of chitosan solution was taken and 2 mL of prepared starch solution was added into the chitosan solution. After that, 5 μ L of turmeric oil was added to the mixture. The process was repeated for different concentration of turmeric oil which is 10, 15 and 20 μ L for every 100 mL of prepared chitosan and 2 mL of starch solution (Khaliq et al., 2016).

D. Appearance Analysis

The strawberry samples appearance was observed and recorded for each day for 5 days

E. Weight loss Analysis

The weight loss of fruit samples were measured by weighting all of the fruit samples with precision weight balance. The weight reading was taken on the initial day and for every sampling days. The difference between both values would indicates as the weight loss of the fruit samples. The percentage of weight loss is evaluated based on the following equation (Vieira, 2014):

F. Disk Diffusion Analysis

By using aseptic technique, a sterile swab was used to take fungal culture from strawberry sample. Then, the culture was streak onto the Potato Dextrose Agar plate. In order to obtain uniform growth, the plate was steak with the swab in one direction and then rotate the 90° and streak the plate again in that direction. The rotation was repeated for 3 times. After that, antibiotic disc that had been immersed with the chitosan samples of different concentration of turmeric oil were dispensed onto the agar. The forceps were used to gently press each of the disc to the agar to ensure each of the disc was attached to the agar. The forceps were sterilized by using flame ("KIRBY - BAUER METHOD," 2017). The plates were incubated at 25°C for 48-72 hours (Guerreiro, Gago, Faleiro, Miguel, & Antunes, 2015).

G. Total Plate Count Analysis

Ten gram of each sample were transferred to 90 mL of peptone water and the mixture were homogenized at their designated sampling times. The incubation conditions for yeast and mold was $25 \pm 1^\circ\text{C}$ for 48 to 72 hours. Results were expressed as Log_{10} CFU (Colony Forming Unit) per gram of fresh weight (Guerreiro et al., 2015)

I. RESULTS AND DISCUSSION

A. Effect of chitosan coating with turmeric oil to the appearance of strawberry

The appearance of strawberry samples was observed as shown in figure 1 and figure 2 for five days. Based on the observation, the non-coated strawberry sample showed unsatisfactory of appearance after one day of storage period as juice leakage happened on the second day of storage. The strawberry samples coated with chitosan and addition of 10 ul of turmeric oil could last up to three days meanwhile, the rest of the strawberry samples, their appearance was satisfied for two days before juice leakage, wound and black spot could be seen on their surface on third day. From the appearance observation, chitosan coating can retain a good appearance of fruit.



Figure 1: The appearance observation for strawberry sample within 5 day of storage period

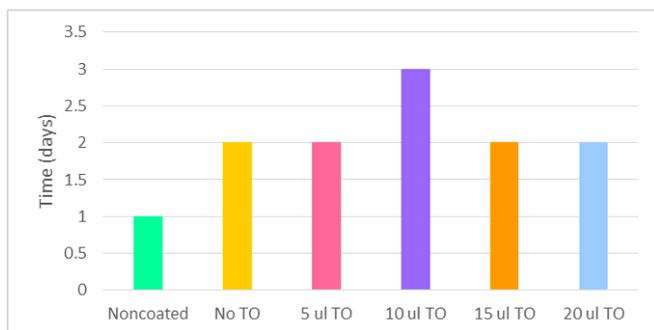


Figure 2: Effect of chitosan coating with different concentration turmeric oil on appearance of strawberry.

B. Effect of chitosan coating with turmeric oil to the weight loss of strawberry

The factor of weight loss for a fruit samples is due to process of transpiration that reduce the quality of strawberry samples. Through coating of chitosan, the weight loss of strawberry can be reduced. As shown in figure 3, the percentage weight loss of strawberry sample coated with chitosan is lower than non-coated sample. After five days, the percentage of weight loss for non-coated strawberry sample was 5.53% while the lowest of weight loss percentage is 3.35%, strawberry sample coated with chitosan and 15ul turmeric oil (15 ul TO). Thus, it shows that chitosan coating can reduce the transpiration and the leakage of juice which are the main factors of fruit weight loss.

According to the study by Jiang, Feng, & Zheng (2012), weight loss of shitake mushroom coated with chitosan-oil coating can be reduce significantly compares to the control samples thus, it delayed mushroom shriveling and quality deterioration. The edible coating on the fruit surface maintains the fluid contained in the fruit and interrupt the migration of moisture from escaping to the environment. This condition also decreases the respiration rate of the fruit. Based on the investigation of Petriccione et al. (2015), strawberry fruits are highly affected to rapid loss of water due to the physical features of thin skin thus, with chitosan coating which acts as semipermeable barrier against oxygen, carbon dioxide and moisture, water loss and respiration can be reduce. This resulting to prevention of fruit dehydration and shrinkage process.

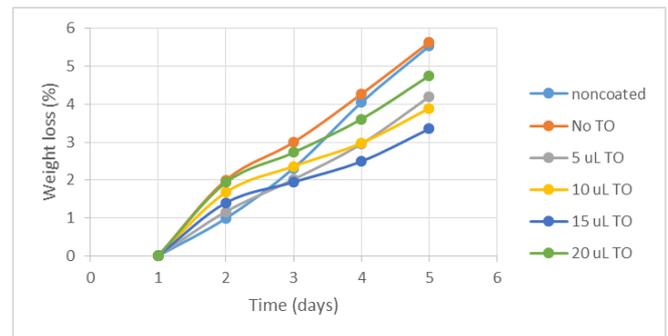


Figure 3: Effect of chitosan coating with different concentration turmeric oil on weight loss of strawberry.

C. Effect of chitosan coating with turmeric oil to the inhibition zone

Through the disk diffusion method, the inhibition zone for each strawberry sample were recorded as shown in the figure 4 and 5. The inhibition zone indicates the antifungal properties for each of the chitosan coating that had been added with different concentration of turmeric oil (TO). After three day of incubation period, the inhibition zone for chitosan coating added with 10 ul of turmeric oil showed the highest inhibition zone which is 17 mm while the chitosan coating with 5ul turmeric oil give 10mm. For the other samples, there were no sign of antifungal activity. From the result, chitosan coating added with 10ul of turmeric oil has the highest antifungal properties that can inhibit deterioration of fungal thus, prolong the shelf life of strawberry samples.

According to the investigation of (Xing et al., 2015), the inhibition zone of chitosan incorporated with cinnamon oil has better antimicrobial properties against *A. flavus*, *P. expansum*, *R.nigricans* and *P.citrimum*. It is also reported by Perdones, Sánchez-González, Chiralt, & Vargas (2012), the use of pure chitosan coating would result to reduction in growth of *Botrytis*

cinerea thus, it showed the degree of antifungal activity. Meanwhile, the chitosan film incorporated with lemon essential oil showed reduction in power of inhibition after two days storage period.

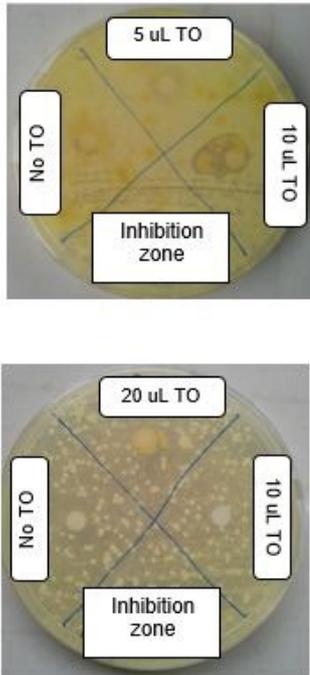


Figure 4: The inhibition zone was measured after 3 days of incubation period

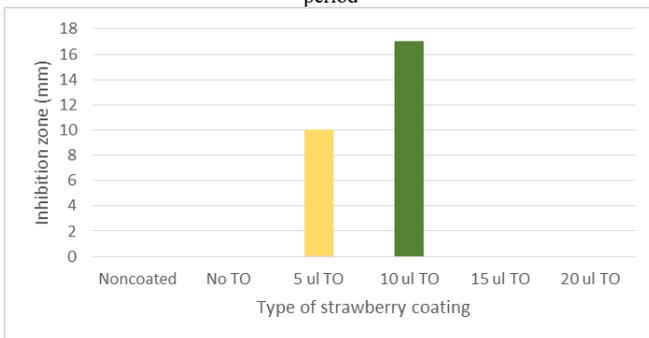


Figure 5: Effect of chitosan coating with different concentration turmeric oil on inhibition zone

D. Effect of chitosan coating with turmeric oil to the total plate count of microbe from strawberry samples.

The total plate count for each of strawberry samples were presented in the figure 4 below. From the graph, the non-coated strawberry samples have the highest value of log CFU which point out that the samples have the highest value of colony grew in the sample which is 4.13 meanwhile, the chitosan added with 15ul of turmeric oil has the lowest value of log CFU. From the data obtained, the chitosan coating with addition of turmeric oil inhibit the growth of microorganism that cause fruit spoilage.

According to Sessa, Ferrari, & Donsi (2015), the incorporation of Nano emulsified lemon essential oil into modified chitosan resulted to significant interaction which increase the property of antimicrobial activity by reduce the microbial load of approximately 4 Log in compare to the untreated samples. A study by Zhelyazkov, Zsivanovits, Brashlyanova, & Marudova-Zsivanovits (2014) also indicates increase of total plate count for untreated apple samples but, there is no significant change for the number pathogens during the storage period.

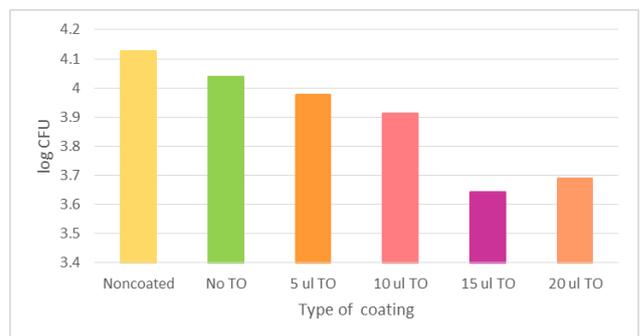


Figure 6: Effect of chitosan coating with different concentration turmeric oil on total plate count.

II. CONCLUSION

The application of chitosan coating gives positive effect on retarding the ripening process of the strawberry. By coating the strawberry with chitosan edible coating, the quality of the fruit can be maintaining and the shelf life of the strawberry can be prolong for a several days. The addition of turmeric essential oil improves the antimicrobial properties of the chitosan coating to inhibit the growth of spoilage microorganism. In overall, the chitosan coating with addition of 10 ul turmeric oil can preserve the appearance for the longest time and has the highest antimicrobial properties to inhibit fungal. Meanwhile, chitosan coating with 15 ul turmeric oil shows the lowest weight loss and lowest value of log CFU that indicates lowest colony forming unit.

Table 1: The overall result for each analysis on the effect of chitosan on biological properties and quality of strawberry.

Sample	Formulation						Analysis				
	Distilled water (ml)	Chitosan (g)	Starch (g)	Glycerol (ml)	Tween 80 (ml)	Turmeric oil (µl)	Weight loss (%)	Inhibition zone (mm)	Total plate count		Appearance (days)
									Total colony forming unit (CFU/g)	Log CFU	
EC 1	0	0	0	0	0	0	5.54	0	13400	4.1271	1
EC 2	100	1	8	2	0.1	0	5.64	0	10900	4.0374	2
	100		(2 ml)								
EC 3	100	1	8	2	0.1	5	4.20	10	9500	3.9778	2
	100		(2 ml)								
EC 4	100	1	8	2	0.1	10	3.89	17	8200	3.9138	3
	100		(2 ml)								
EC 5	100	1	8	2	0.1	15	3.35	0	4400	3.6434	2
	100		(2 ml)								
EC 6	100	1	8	2	0.1	20	4.75	0	4900	3.6902	2
	100		(2 ml)								

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