

## The efficacy of combined coatings of chitosan and cinnamic acid on tomatoes

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### Abbreviation

Ch - chitosan

CA - cinnamic acid

TSS - total soluble solid

### Abstract

Tomatoes require appropriate environment to stay sturdy due to earlier decay process. Deterioration causes short shelf life of tomatoes with unfavourable quality, resulting in potential rejection by customers. The objective of the study is to observe the effect of combined coatings of chitosan (Ch) and cinnamic acid (CA) in extending the tomato shelf life. Layer by layer coating of chitosan prior to the cinnamic acid (single coating for each) were applied on fresh graded tomato at two maturity stages; breaker and turning. Twelve days observations at ambient temperature with three-day intervals were recorded. Combined coating of chitosan and cinnamic acid were expected to influence firmness, TSS value, hue angle and weight loss. Results showed that a combined coating of 1.0% Ch + 3 mM CA has significant increment at breaker stage to firmness (8.26 N), hue angle (60.42%) and weight loss value (6.51%) compared to untreated tomato whereas for turning stage, the results showed there were no significant different in all parameters observed except the changes of fruit sweetness (TSS). 1.0% Ch + 4 mM CA show highest TSS value, 3.48% indicating 21% difference than untreated tomato (3.27%). Cinnamic acid helped chitosan in improving coating ability by serving better barrier from pathogen and oxidative gas penetration to prevent earlier spoilage problem.

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### Introduction

*Solanum lycopersicum* L. comes from the Solanaceae family that has different appearance in shape and size based on growth region (USDA, 2014). Wild tomato underwent evolution to adapt and thrive in different climates and soil at specific regions, which appeared to be one of the characteristics to be accepted by consumers. Tomato is rich in lutein, vitamin C,  $\alpha$ -carotene,  $\beta$ -carotene, and lycopene which help to improve eyesight, blood circulation and digestive system (Pochelli, 2014).

Tomatoes cultivated at Lojing, Gua Musang, Kelantan and Cameron Highland, Pahang are exported to Singapore (Islam *et al.*, 2012). In 2016, DOA reported 242,946.4 Mt of tomato production worth RM425,156,000. Tomatoes are significantly invading processing industry, providing employment opportunities and also contribute as much as the way paste and sauce is vital to make bolognaise.

Tomato is a climacteric fruit that continues to ripen after harvesting through ethylene that causes quick deterioration (Cara and Giovannoni, 2008). Earlier spoilage limits the shelf life of tomatoes. Postharvest activities before reaching target market

including packing, storage, and transportation may be wasted if the tomatoes get rejected by consumers eventually. In addition, aging tomato favour pathogen which causes a further loss. Diseases control measure to treat infected tomatoes is through usage of chemical fungicides. Then, scientists came up with the solution of using genetically modified (GM) crop. For example, Appleton (1999) had grown genetically modified organism (GMO) tomatoes that were purple in colour and proven to be resistant towards fungus due to the *Bacillus thuringiensis* (Bt) action (insertion of insecticidal toxin). Unfortunately, prolonged usage of GM crop results in increased resistivity. Pathogen outbreak defined as microbial imbalance comes from rising antimicrobial resistivity to treat diseases (Pham and Lawley, 2014). Emerging infectious diseases (EIDs) caused by this causative agent of diseases (pathogen) are unlikely to be treated by synthetic chemicals (Anderson, 2004; BMJ, 2017).

Thus, usage of edible coating seemed to be a good substitution to extend the shelf life of food product. Edible coating and edible film is different in terms of bio-based materials. Edible coating is a thin material solution coating food product surface, whereas edible film is a material placed between food

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components (McHugh, 2000; Falguera *et al.*, 2011; Luo *et al.*, 2015). Edible coating provides semi-permeable properties that provide barrier between oxygen, carbon dioxide as well as pathogen and the host (Fundazioa, 2006).

Chitosan coating is used in agriculture industry and in fact is seeing a rise in interest in pharmaceutical world. Del Giudice *et al.* (2015) proved that certain maturity tomato stage (breakers) had the disastrous effect on cancer cells due to the  $\alpha$ -tomatine content.  $\alpha$ -tomatine of tomato fruit implying necrotic status of cancerous cells by binding to the cholesterol on damaged plasma membrane. Chitosan is a biodegradable material derived from chitin (Sabaghi *et al.*, 2015). Chitosan has an antifungal property that influences internal metabolic reaction of pathogen (Bautista-Banos *et al.*, 2006). Chitosan is not a new thing in agriculture. El Ghaouth *et al.* (1992) extensive previous studies on the promising properties of chitosan believed that chitosan is a safe replacement on the fruit shelf life extension. Cinnamic acid is known as the agent of antimicrobial (Muche *et al.*, 2011). Cinnamic acid is rarely used in single coating but tend to be used in combination coating. Coating is used to protect the food and conserve their shelf life.

Hence, the present study aims to investigate the efficiency of combined coating of chitosan and cinnamic acid using layer by layer dipping process in prolonging shelf life of tomato through optimum concentration comparing two maturity stages of tomato; breakers and turning stage.

## Material and method

### Fruit selection

Raw and fresh tomatoes were bought from tomato farm at Lojing, Gua Musang, Kelantan and Cameron Highland, Pahang. Tomatoes with uniform size, colour, stages (breaker and turning), weight (60 g - 90 g) with no physical damage were selected. Selection of tomato stage was based on Federal Agricultural Marketing Authority (FAMA) tomato characteristics, Index 2/breaker (<10% of pink or red on surface) and Index 3/turning (10%-30% of pink or red on surface).

### Preparation of chitosan

Commercial chitosan and acetic acid were purchased from Sigma-Aldrich, Kuala Lumpur. Chitosan was dissolved in 0.1% (w/v) in 1.0% (v/v) acetic acid. The solution was stirred overnight at ambient temperature. The solution was filtered using muslin cloth and its volume was adjusted to 1000 ml with distilled water. The coating formulation resulted

in several sets of treatment: a) Control (no coating), b) 0.5% chitosan acetate solution, c) 0.75% chitosan acetate solution, and d) 1.0% chitosan acetate solution.

### Preparation of cinnamic acid

Cinnamic acid was purchased from Sigma-Aldrich, Kuala Lumpur. Cinnamic acid was dissolved in distilled water and stirred until dissolved. The coating formulation resulted in several sets of treatment: a) 2 mM cinnamic acid solution, b) 3 mM cinnamic acid solution, and c) 4 mM cinnamic acid solution.

### Coating application

Tomato fruits were divided into breaker and turning batch. Each batch was divided into three different coating treatments and each batch of coating treatment was divided into five different batches for the 0<sup>th</sup>, 3<sup>rd</sup>, 6<sup>th</sup>, 9<sup>th</sup> and 12<sup>th</sup> observational days. Tomato fruits were rinsed using distilled water and air-dried before treatment. Tomato was dipped prior into chitosan for 30 s and then allowed dry for two hours at ambient temperature, followed by layer by layer coating of 3 min in cinnamic acid, and then dried for two hours at ambient/room temperature on tissue of permeable paper or simply in a tray to remove excess liquid solution. Two factorial experimental design had been used with nine experimental units of three replications (every three days interval) using SPSS of Tukey. The coating formulation resulted in several sets of treatment: a) control (uncoated tomatoes/dipping in distilled water), b) 0.5% chitosan acetate solution + 2 mM cinnamic acid, c) 0.5% chitosan acetate solution + 3 mM cinnamic acid, d) 0.5% chitosan acetate solution + 4 mM cinnamic acid, e) 0.75% chitosan acetate solution + 2 mM cinnamic acid, f) 0.75% chitosan acetate solution + 3 mM cinnamic acid, g) 0.75% chitosan acetate solution + 4 mM cinnamic acid, h) 1.0% chitosan acetate solution + 2 mM cinnamic acid, i) 1.0% chitosan acetate solution, and j) 1.0% chitosan acetate solution + 4 mM cinnamic acid.

### Assessment firmness

By using Brookly Texture Analyser, firmness of tomato was determined by converting the force in gram into Newton. Puncture method using TA 39/100 (probe TA39 of TA-MTP) was performed automatically through remote control by directing the machine to the computer to compute graft of firmness. Reading was taken twice at opposite points (4 or 5 cm) apart. Texture pressure analyser (TPA) was set to the speed of 10 mm/s. Firmness of first

peak using surface penetration was noted and average reading was calculated.

#### Total soluble solid (TSS)

The flesh of tomato without seeds was taken through vertical cutting before put on the hand refractometer (Atago, USA) to document suspended solid ( $^{\circ}$ Brix, 20% sucrose) in percentage which was observed on 0<sup>th</sup>, 3<sup>rd</sup>, 6<sup>th</sup>, 9<sup>th</sup> and 12<sup>th</sup> days.

#### Colour

Minolta chromameter (model CR-400X Minolta Camera Co. Ltd., Japan) was used to determine the colour changes at three-day intervals. The chromameter was calibrated using white tiles ( $L^*=98.15$ ,  $a^*=0.13$ ,  $b^*=1.92$ ). The colour was determined from three light pulses points of equatorial area without spots and tissue discolouration from nine fruits per treatment. The values of hue angle were calculated using ( $h = \tan^{-1}[b^*/a^*]$ ). Chromameter read L as lightness (black [ $L^* = 0$ ]); (white [ $L^* = 100$ ]),  $a^*$  indicated redness to greenness (red [ $a^* = 100$ ]); (green [ $a^* = -100$ ]),  $b^*$  indicated yellowness to blueness (yellow [ $b^* = 100$ ]); (blue [ $b^* = -100$ ]).

#### Weight loss

Tomato weight was calculated at the beginning of the experiment just after coating and air-dried using analytical balance (Kern EMB 2200-00). Result was reported as weight loss percentage.

$$\text{Weight loss} = \frac{\text{Final Weight} - \text{Initial Weight}}{\text{Initial Weight}} \times 100$$

## Result

#### Statistical analysis

The data of differences between treatments were analysed using SPSS of mean comparison through Tukey's multiple range test at significant level of  $p < 0.05$ .

#### Weight loss

Table 1 shows weight loss of tomato at layer by layer coating of chitosan and cinnamic acid stored at ambient temperature. The highest weight loss value was observed for 0.75% Ch + 4 mM CA at turning stage, approximately 8.59% compared to control after 12 days storage (8.12%) whereas 0.5% Ch + 3 mM CA at turning stage seemed to show approximately 4.87% of weight loss, the lowest among treated tomato at turning stage. 0.5% Ch + 2 mM CA of breaker stage was observed to experience less weight reduction than untreated tomatoes, acknowledged to be the lowest loss, at 4.68%.

Table 1: Weight loss of tomato after 12 days storage

Treatment	Weight loss (%)	
	Breaker	Turning
0.75% Ch + 2mM CA	7.30±0.176 <sup>bcd</sup>	7.33±0.122 <sup>c</sup>
0.75% Ch + 3mM CA	7.16±0.204 <sup>bcd</sup>	6.72±0.172 <sup>bc</sup>
0.75% Ch + 4mM CA	7.54±0.204 <sup>cd</sup>	8.59±0.149 <sup>d</sup>
1.0% Ch + 2mM CA	7.25±0.204 <sup>bcd</sup>	6.76±0.149 <sup>bc</sup>
1.0% Ch + 3mM CA	6.51±0.204 <sup>b</sup>	6.73±0.172 <sup>bc</sup>
1.0% Ch + 4mM CA	6.63±0.204 <sup>bc</sup>	6.62±0.149 <sup>bc</sup>
0.5% Ch + 2mM CA	4.68±0.204 <sup>a</sup>	6.42±0.133 <sup>b</sup>
0.5% Ch + 3mM CA	5.44±0.204 <sup>a</sup>	4.87±0.172 <sup>a</sup>
0.5% Ch + 4mM CA	5.33±0.176 <sup>a</sup>	5.32±0.172 <sup>a</sup>
Control	8.12±0.204 <sup>d</sup>	6.49±0.172 <sup>b</sup>

\*± represent standard error of the mean

#### Firmness

The turning stage showed more firmness reduction rather than breaker stage (Table 2). At turning stage, 0.5% Ch + 2 mM CA was observed to be abnormally increased in firmness value at 3<sup>rd</sup> day storage before gradually reduced until the last day storage, and showed the lowest firmness value observed, approximately 1.8%. Few treatments of coated tomato including 1.0% Ch + 3 mM CA, 0.5% Ch + 2 mM CA and 0.5% Ch + 3 Mm CA at breaker stage were observed to be eventually increased in firmness value after certain day storage (9<sup>th</sup> day), supporting uncoated tomato of turning stage that was observed to be abnormally fluctuated and control tomato at breaker stage that was increased in firmness value after 6<sup>th</sup> day storage. Firmness at turning stage underwent decrement in value until it reached less than 2 N respectively.

#### TSS

There was no significant difference and influence between TSS values with control, maturity stages and storage period. Total soluble solid indicated the sweetness of tomatoes. This TSS table showed constantly decreasing TSS value for both stages but it was still the highest compared to control. The TSS value range is 2.5 - 4.5%. While other treatments showed a decreasing trend of TSS value, 0.75% Ch + 4 mM CA at breaker stage showed constant low increment from day 6 until the end of storage days. 0.75% Ch + 2 mM CA and 1% Ch + 3 mM CA of breaker stage had decreased with respect to each other before increasing after 6 days. All treatments including control for both maturity stages showed a certain peak value of TSS along the storage period. However, 0.5% Ch + 2 mM CA showed approximately 92% reduction in TSS value, which indicated the highest loss.

Table 2: Effect of combined coating chitosan and cinnamic acid on TSS, Firmness, and Hue angle after 12 days application

Treatment	TSS (%)			Hue angle (%)			Firmness (N)		
	Breaker	Turning	Breaker	Breaker	Turning	Breaker	Turning	Breaker	Turning
Day 0									
0.75% Ch + 2mM CA	4.00±0.097 <sup>ij</sup>	3.96±0.088 <sup>hi-l</sup>	59.14±0.863 <sup>q-s</sup>	46.98±0.717 <sup>o</sup>	6.53±0.184 <sup>st</sup>	7.85±0.249 <sup>st</sup>			
0.75% Ch + 3mM CA	3.90±0.088 <sup>hij</sup>	3.73±0.114 <sup>fi</sup>	54.61±0.863 <sup>q-r</sup>	54.72±0.717 <sup>o-t</sup>	6.13±0.184 <sup>pt</sup>	6.37±0.249 <sup>o-s</sup>			
0.75% Ch + 4mM CA	3.85±0.088 <sup>sj</sup>	4.00±0.080 <sup>hl</sup>	52.17±0.863 <sup>mp</sup>	54.39±0.717 <sup>o-t</sup>	8.25±0.184 <sup>u</sup>	6.70±0.249 <sup>o-s</sup>			
1.0% Ch + 2mM CA	3.76±0.018 <sup>ij</sup>	4.08±0.088 <sup>h-l</sup>	45.39±0.863 <sup>d-j</sup>	50.86±0.717 <sup>o-r</sup>	4.72±0.160 <sup>o</sup>	5.49±0.193 <sup>m-s</sup>			
1.0% Ch + 3mM CA	4.00±0.097 <sup>ij</sup>	4.00±0.114 <sup>bl-l</sup>	60.42±0.863 <sup>rs</sup>	58.90±0.717 <sup>uv</sup>	8.26±0.184 <sup>u</sup>	5.47±0.249 <sup>m-s</sup>			
1.0% Ch + 4mM CA	3.87±0.088 <sup>sj</sup>	3.72±0.088 <sup>dk</sup>	67.61±0.863 <sup>w</sup>	51.49±0.717 <sup>p-s</sup>	4.78±0.160 <sup>bc-o</sup>	5.64±0.215 <sup>n-s</sup>			
0.5% Ch + 2mM CA	3.52±0.097 <sup>bj</sup>	4.00±0.098 <sup>bl-l</sup>	62.42±0.863 <sup>st</sup>	53.25±0.717 <sup>q-s</sup>	5.03±0.160 <sup>tp</sup>	1.88±0.249 <sup>ab</sup>			
0.5% Ch + 3mM CA	3.48±0.097 <sup>bj</sup>	3.93±0.080 <sup>hl</sup>	59.86±0.863 <sup>rs</sup>	46.06±0.717 <sup>h-n</sup>	3.35±0.184 <sup>b-c</sup>	2.65±0.193 <sup>a-c</sup>			
0.5% Ch + 4mM CA	4.00±0.125 <sup>ij</sup>	3.90±0.080 <sup>hl</sup>	52.58±0.863 <sup>mp</sup>	67.51±0.717 <sup>w</sup>	4.66±0.184 <sup>n</sup>	6.21±0.249 <sup>p-s</sup>			
Control	3.60±0.097 <sup>sj</sup>	4.00±0.114 <sup>bl-l</sup>	72.64 ±0.863 <sup>u</sup>	55.16 ±0.717 <sup>su</sup>	3.80±0.160 <sup>dk</sup>	4.49±0.249 <sup>dk</sup>			
Day 3									
0.75% Ch + 2mM CA	4.00±0.097 <sup>ij</sup>	3.87±0.088 <sup>hi-l</sup>	61.02±0.863 <sup>rs</sup>	40.69±0.717 <sup>ab</sup>	6.70±0.184 <sup>t</sup>	6.07±0.215 <sup>o-s</sup>			
0.75% Ch + 3mM CA	3.52±0.097 <sup>sj</sup>	3.90±0.098 <sup>hl</sup>	46.23±0.863 <sup>fi-l</sup>	41.52±0.717 <sup>af</sup>	5.64±0.160 <sup>o-s</sup>	6.25±0.215 <sup>ps</sup>			
0.75% Ch + 4mM CA	3.65±0.108 <sup>sj</sup>	4.00±0.114 <sup>bl-l</sup>	44.81±0.863 <sup>ci</sup>	49.24±0.717 <sup>q</sup>	6.54±0.184 <sup>st</sup>	5.39±0.215 <sup>l-s</sup>			
1.0% Ch + 2mM CA	3.60±0.018 <sup>sj</sup>	4.30±0.098 <sup>l</sup>	51.23±0.863 <sup>o</sup>	49.07±0.717 <sup>q</sup>	4.49±0.184 <sup>g-n</sup>	5.39±0.249 <sup>l-s</sup>			
1.0% Ch + 3mM CA	4.07±0.125 <sup>j</sup>	4.00±0.098 <sup>hl</sup>	43.73±0.863 <sup>bi</sup>	43.92±0.717 <sup>bk</sup>	6.43±0.160 <sup>l</sup>	4.25±0.249 <sup>fm</sup>			
1.0% Ch + 4mM CA	3.70±0.108 <sup>d-j</sup>	3.68±0.088 <sup>dk</sup>	53.86±0.863 <sup>op</sup>	45.80±0.717 <sup>g-m</sup>	5.07±0.184 <sup>tp</sup>	4.54±0.215 <sup>g-n</sup>			
0.5% Ch + 2mM CA	4.03±0.088 <sup>j</sup>	3.80±0.088 <sup>hl</sup>	52.40±0.863 <sup>mp</sup>	46.13±0.717 <sup>h-n</sup>	3.86±0.160 <sup>pk</sup>	4.08±0.249 <sup>kl</sup>			
0.5% Ch + 3mM CA	3.20±0.108 <sup>sf</sup>	3.83±0.080 <sup>fl</sup>	47.64±0.863 <sup>bm</sup>	48.78±0.717 <sup>p</sup>	3.51±0.184 <sup>fg</sup>	2.94±0.249 <sup>af</sup>			
0.5% Ch + 4mM CA	3.60±0.125 <sup>sj</sup>	3.68±0.088 <sup>dk</sup>	59.19±0.863 <sup>rs</sup>	43.53±0.717 <sup>nk</sup>	4.41±0.160 <sup>fm</sup>	4.66±0.193 <sup>bn</sup>			
Control	3.90±0.088 <sup>bj</sup>	4.12±0.080 <sup>kl</sup>	52.22±0.863 <sup>mp</sup>	50.04±0.717 <sup>n-q</sup>	3.68±0.160 <sup>bi</sup>	3.02±0.249 <sup>af</sup>			
Day 6									
0.75% Ch + 2mM CA	3.07±0.097 <sup>ad</sup>	3.80±0.144 <sup>fi</sup>	41.43±0.863 <sup>bf</sup>	45.54±0.717 <sup>o-m</sup>	6.53±0.184 <sup>st</sup>	5.31±0.249 <sup>kr</sup>			
0.75% Ch + 3mM CA	3.32±0.097 <sup>a-h</sup>	3.60±0.144 <sup>ck</sup>	40.98±0.863 <sup>b-e</sup>	45.35±0.717 <sup>d-l</sup>	5.97±0.184 <sup>pt</sup>	4.47±0.215 <sup>g-n</sup>			
0.75% Ch + 4mM CA	3.92±0.108 <sup>bj</sup>	3.96±0.088 <sup>hl</sup>	50.20±0.863 <sup>o</sup>	46.00±0.717 <sup>h-n</sup>	5.64±0.184 <sup>o-s</sup>	3.76±0.249 <sup>d-i</sup>			
1.0% Ch + 2mM CA	3.50±0.108 <sup>bj</sup>	4.13±0.114 <sup>kl</sup>	50.99±0.863 <sup>k-o</sup>	42.13±0.717 <sup>a-i</sup>	4.90±0.184 <sup>o</sup>	5.15±0.249 <sup>h-n</sup>			
1.0% Ch + 3mM CA	3.08±0.125 <sup>bj</sup>	3.67±0.114 <sup>dk</sup>	48.65±0.863 <sup>fn</sup>	41.10±0.717 <sup>abc</sup>	5.31±0.184 <sup>q</sup>	4.74±0.249 <sup>h-n</sup>			
1.0% Ch + 4mM CA	3.80±0.125 <sup>ij</sup>	3.27±0.114 <sup>af</sup>	52.87±0.863 <sup>mp</sup>	51.33±0.717 <sup>ps</sup>	4.53±0.160 <sup>bn</sup>	4.66±0.249 <sup>bn</sup>			
0.5% Ch + 2mM CA	4.00±0.125 <sup>ij</sup>	3.17±0.080 <sup>cd</sup>	44.25±0.863 <sup>ci</sup>	43.15±0.717 <sup>aj</sup>	3.04±0.143 <sup>d</sup>	3.92±0.249 <sup>ej</sup>			

Table 2: (Cont.)

0.5% Ch + 3mM CA	3.20±0.088 <sup>u-f</sup>	3.45±0.098 <sup>e-i</sup>	45.79±0.863 <sup>e-j</sup>	44.58±0.717 <sup>b-k</sup>	2.61±0.184 <sup>a</sup>	2.70±0.249 <sup>a-e</sup>
0.5% Ch + 4mM CA	3.40±0.097 <sup>u-i</sup>	3.25±0.098 <sup>a-f</sup>	45.94±0.863 <sup>e-j</sup>	47.39±0.717 <sup>k-p</sup>	3.56±0.160 <sup>a-h</sup>	4.66±0.249 <sup>b-h</sup>
Control	3.70±0.108 <sup>d-j</sup>	3.53±0.114 <sup>b-j</sup>	50.33±0.863 <sup>o</sup>	47.07±0.717 <sup>o</sup>	3.43±0.160 <sup>a-f</sup>	4.49±0.249 <sup>g-n</sup>
Day 9						
0.75% Ch + 2mM CA	3.47±0.088 <sup>b-j</sup>	3.93±0.144 <sup>h-l</sup>	43.39±0.863 <sup>b-h</sup>	41.25±0.717 <sup>a-d</sup>	6.21±0.184 <sup>q-t</sup>	4.90±0.215 <sup>l-n</sup>
0.75% Ch + 3mM CA	3.20±0.088 <sup>u-f</sup>	3.55±0.098 <sup>b-j</sup>	42.24±0.863 <sup>b-g</sup>	48.93±0.717 <sup>p</sup>	5.47±0.184 <sup>b-r</sup>	3.60±0.249 <sup>d-i</sup>
0.75% Ch + 4mM CA	3.93±0.125 <sup>l-j</sup>	3.07±0.080 <sup>abc</sup>	41.05±0.863 <sup>b-e</sup>	45.60±0.717 <sup>f-m</sup>	5.64±0.184 <sup>b-s</sup>	3.02±0.249 <sup>a-f</sup>
1.0% Ch + 2mM CA	3.04±0.097 <sup>abc</sup>	3.93±0.114 <sup>h-l</sup>	41.63±0.863 <sup>b-f</sup>	43.31±0.717 <sup>a-k</sup>	3.59±0.184 <sup>a-h</sup>	4.58±0.249 <sup>b-h</sup>
1.0% Ch + 3mM CA	3.08±0.097 <sup>a-d</sup>	3.00±0.114 <sup>ab</sup>	42.82±0.863 <sup>b-h</sup>	42.17±0.717 <sup>a-i</sup>	3.86±0.160 <sup>h-k</sup>	4.84±0.215 <sup>h-n</sup>
1.0% Ch + 4mM CA	3.50±0.108 <sup>b-j</sup>	3.33±0.114 <sup>a-h</sup>	43.61±0.863 <sup>e-j</sup>	43.97±0.717 <sup>b-k</sup>	4.35±0.160 <sup>e-l</sup>	3.51±0.249 <sup>c-h</sup>
0.5% Ch + 2mM CA	3.08±0.097 <sup>a-d</sup>	3.10±0.098 <sup>abc</sup>	59.86±0.863 <sup>st</sup>	43.66±0.717 <sup>b-k</sup>	2.70±0.184 <sup>ab</sup>	2.70±0.249 <sup>a-e</sup>
0.5% Ch + 3mM CA	3.13±0.125 <sup>a-e</sup>	3.60±0.098 <sup>a-k</sup>	45.95±0.863 <sup>a-k</sup>	49.71±0.717 <sup>m-q</sup>	2.78±0.184 <sup>abc</sup>	2.21±0.249 <sup>abc</sup>
0.5% Ch + 4mM CA	3.13±0.088 <sup>a-e</sup>	3.07±0.114 <sup>abc</sup>	58.95±0.863 <sup>st</sup>	46.10±0.717 <sup>n</sup>	3.60±0.184 <sup>a-h</sup>	4.46±0.193 <sup>g-n</sup>
Control	3.67±0.125 <sup>e-j</sup>	3.72±0.088 <sup>d-k</sup>	46.79±0.863 <sup>g-i</sup>	475.21±0.717 <sup>c-l</sup>	4.53±0.160 <sup>b-n</sup>	1.96±0.215 <sup>ab</sup>
Day 12						
0.75% Ch + 2mM CA	3.10±0.108 <sup>a-e</sup>	3.07±0.144 <sup>abc</sup>	42.47±0.863 <sup>b-g</sup>	41.16±0.717 <sup>abc</sup>	3.76±0.184 <sup>e-j</sup>	4.26±0.193 <sup>f-n</sup>
0.75% Ch + 3mM CA	2.93±0.125 <sup>ab</sup>	3.17±0.080 <sup>a-d</sup>	38.90±0.863 <sup>ab</sup>	42.42±0.717 <sup>a-i</sup>	4.66±0.184 <sup>f-o</sup>	3.19±0.249 <sup>b-g</sup>
0.75% Ch + 4mM CA	4.00±0.125 <sup>l-j</sup>	3.60±0.098 <sup>h-k</sup>	40.41±0.863 <sup>bed</sup>	39.47±0.717 <sup>a</sup>	3.72±0.143 <sup>e-j</sup>	3.92±0.249 <sup>e-j</sup>
1.0% Ch + 2mM CA	3.13±0.125 <sup>a-e</sup>	3.20±0.098 <sup>a-e</sup>	43.87±0.863 <sup>b-i</sup>	41.39±0.717 <sup>a-e</sup>	2.70±0.184 <sup>ab</sup>	4.00±0.249 <sup>a-k</sup>
1.0% Ch + 3mM CA	3.13±0.097 <sup>a-e</sup>	2.96±0.088 <sup>a</sup>	43.61±0.863 <sup>b-h</sup>	41.88±0.717 <sup>a-h</sup>	5.39±0.184 <sup>m-q</sup>	3.92±0.215 <sup>e-j</sup>
1.0% Ch + 4mM CA	3.48±0.108 <sup>b-j</sup>	2.97±0.080 <sup>a</sup>	34.87±0.863 <sup>a</sup>	41.65±0.717 <sup>g</sup>	4.49±0.184 <sup>g-n</sup>	3.92±0.249 <sup>e-j</sup>
0.5% Ch + 2mM CA	3.07±0.097 <sup>a-d</sup>	3.08±0.088 <sup>abc</sup>	56.61±0.863 <sup>p-r</sup>	61.00±0.717 <sup>v</sup>	3.68±0.143 <sup>b-i</sup>	1.80±0.249 <sup>a</sup>
0.5% Ch + 3mM CA	2.80±0.125 <sup>a</sup>	3.20±0.088 <sup>a-g</sup>	62.06±0.863 <sup>s</sup>	61.22±0.717 <sup>v</sup>	2.62±0.160 <sup>a-h</sup>	1.96±0.249 <sup>ab</sup>
0.5% Ch + 4mM CA	3.24±0.088 <sup>a-f</sup>	2.97±0.080 <sup>a</sup>	62.74±0.863 <sup>s</sup>	58.52±0.717 <sup>uv</sup>	2.78±0.184 <sup>abc</sup>	3.68±0.193 <sup>d-i</sup>
Control	3.27±0.088 <sup>a-g</sup>	3.50±0.098 <sup>e-i</sup>	40.28±0.863 <sup>bc</sup>	62.46±0.717 <sup>v</sup>	4.49±0.184 <sup>g-n</sup>	2.53±0.249 <sup>a-d</sup>

\*± represent standard error of the mean

### Hue angle

Table 2 shows the increasing hue angle value for both stages for some of the combination treatments. At breaker stage, untreated tomato fruits rapidly decreased by 32% of initial hue angle value after 12 days storage. Normally, hue angle value decreased due to the accumulation of lycopene content because of the ripening. A significant difference ( $p < 0.05$ ) of edible coating on colour changes was observed. Hue angle value decreased during storage period. For breaker, 0.75% Ch + 4 mM CA whereas for turning, 0.5% Ch + 3 mM CA treated tomatoes showed the highest hue angle compared to untreated tomatoes after 12 days, less green but not too red. Lowest hue value was recorded for 1.0% Ch + 2 mM CA at breaker stage at 34.87%. Increasing value of hue angle was observed particularly on usage of 0.5% chitosan for both maturity stages.

### Discussion

Usage of edible coating is appropriate to preserve and protect tomato fruit from pathogen infestation. In addition, they cause no harm to the tomato due to the edible properties as compared to synthetic chemical to treat postharvest problem. Cherry tomato was proven to be preserved for 14 days at 2°C plus 3 days at 24°C when coated with 0.5% chitosan (Petriccione *et al.*, 2015). When coated with 1.5% chitosan, tomatoes were preserved for 37 days at 10°C with the aid of zeolite without improving the weight loss of tomato (García *et al.*, 2014). Benhabiles *et al.* (2013) succeeded in prolonging tomato shelf life up to a week using 2.0% chitosan. Usage of chitosan in combination with cinnamic acid has not been widely implemented. Nevertheless, chitosan is an effective rising star. Chitosan as an antifungal agent (Cheba, 2011) and cinnamic acid as an antimicrobial agent (Muche *et al.*, 2011) are believed to be capable of helping and maximizing efficiency towards tomato shelf life extension.

The result showed no fungal infection after 12 days storage unless for normal physical changes such as wrinkles and aging process. Treatment using 1.0% Ch + 3 mM CA on tomato had significantly delayed early spoilage at weight loss, firmness and hue angle (Fig. 1). The physico-chemical properties had influenced tomato shelf life extension, excluding TSS unfortunately. Utilization of cinnamic acid is not widely studied by researchers apart from Alkan and Yemenicioğlu (2016) who had studied effectiveness of cinnamic acid on plum tree stem, which showed the microbial inhibition (*Xanthomonas vesicatoria*) and agreed that hydroxyl factor was vital for the

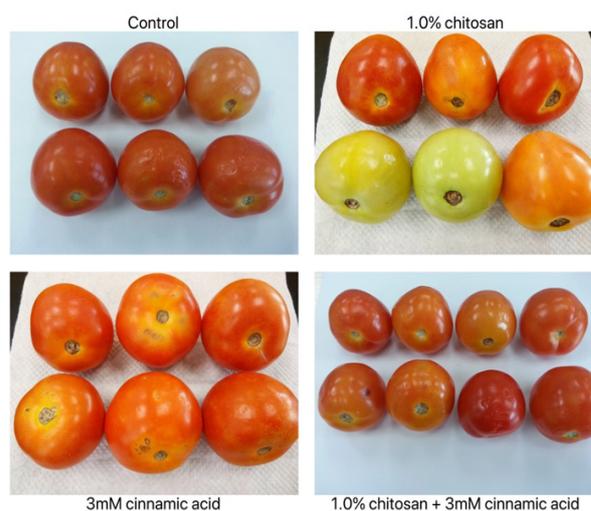


Fig. 1: Comparing untreated and treated tomato with 1.0% Ch, 3 mM CA and combined coating of 1.0% Ch + 3 mM CA

antimicrobial capability. Most of the postharvest handling studied the fresh material at low temperature before being transferred at ambient temperature. Yet, commercial production was performed at ambient temperature.

### Weight loss

Soft tissue was destructed as the tomato ripens (Cantu *et al.*, 2009). Tissue maceration due to pathogen curative action leads to progressive deterioration (Ahmed *et al.*, 2017). This causes the inside of tomatoes to be watery which produced an undesirable smell. In addition, ripening process allows respiration and transpiration of tomato which reduced the weight of the tomato. Losing weight cause changes to firmness alteration. All of the parameters used influence each other. Thus, usage of edible coating help in facilitating surface feature by avoiding shrivelling and moisture loss which is similar to the result achieved by Chauhan *et al.* (2015) by using aloe vera. Similar trend was also observed in research conducted by Benhabiles *et al.* (2013) and Guerra *et al.* (2015), who studied about chitosan efficiency on tomato. Benhabiles concluded that 2.0% of chitosan effectively preserved the tomato fruits (11% weight loss) than at 0.5% (12% weight loss) compared to uncoated tomato (19%) after 29 days storage. Guerra *et al.* (2015) observed the antifungal efficiency of chitosan which infected only 36% of tomato by *B. cinerea* after 12 days storage at room temperature. Aside from the water vapour pressure that influences transpiration rate as studied by Bautista-Banos *et al.* (2006), Limchoowong *et al.* (2016) also believed the effect of coating thickness in reducing the loss of weight of tomato. The higher the

thickness (concentration) of chitosan, the lower the loss of weight. On the other hand, Souza *et al.* (2009) found in his work that higher concentration of chitosan formed larger pore size of poor surfactant instead of smooth coating layer. This hydrophilic-lipophilic imbalance (HLiB) resulted in impaired barrier. Even though acetic acid had been used, their usage did not support the coating structure as adhesives even on steel (Kotsev *et al.*, 1987; Popović *et al.*, 2015). However, in this study, the coating had significantly enhanced weight loss of tomato due to the efficient layer by layer barrier of chitosan and cinnamic acid. Mustafa *et al.* (2014) and Chien *et al.* (2007) assumed that coating enhanced water retention which provides a secure barrier from external damage.

#### *Firmness and softening*

The higher firmness value demonstrated reduction in fruit softening after storage days, in the cases of the coated samples. Comparing breaker and turning stage, breaker showed the higher end firmness value than turning stage, approximately 2.7 N and 1.8 N. 1.0% Ch + 3 mM CA showed the least aging process throughout the 12 days. This result was not so impressive since mature-green tomato treated with salicylic acid showed more favourable firmness after 20 days storage, with a range of 4.0 - 4.1 N (Baninaiem *et al.*, 2016). The composite wall of fruit is a fence-like blockage to the external penetration. Any left bruises due to the mechanical damage accelerated pectinmethylesterase (PME) activity, affecting organoleptic properties of tomato (Sila *et al.*, 2008). The chitosan effectiveness supported by Cissé *et al.* (2015) that concluded usage of 1.0% chitosan in retaining firmness value of mango due to the low respiration and water loss. Apart from Cissé, Dovale-Rosabal *et al.* (2015) agreed that low concentration of chitosan affect the turgidity of tomato fruit tissue. The higher concentration distributed to the loss of physical appearance although previous research by Benhabiles *et al.* (2013) succeeded. The properties of tomato might change over time. The diversification of firmness value related to the ripening stage. Even though the 1.0% chitosan + 3 mM cinnamic acid corresponded to the results from previous study, some of the coating obviously did not enhance the physico-chemical properties desired. They did not compromise the quality loss of tomato which concurred with Mustafa *et al.* (2014) who had exposed the higher reduction in firmness value of treated tomato compared to untreated tomato. With retention 3.73 N firmness value compared to uncoated fruit, a combination of 1.0% chitosan and 3 mM cinnamic acid seemed to contribute to the

lowering of PME and polygalacturonase cell wall degrading enzymes (Zapata *et al.*, 2008). Firmness value significantly reduced weight loss of tomato.

#### *TSS*

From the aspect of the sweetness of tomato or total soluble solid (TSS), lower TSS value was detected in breaker stage than at turning stage. This was due to the greener stage of breaker which contained less ethylene to speed up the ripening process. Reduction of TSS value as the number of the storage days increased was verified in a study conducted by Barreto *et al.* (2016) that showed a decrease of TSS value (20% sucrose) on coated or uncoated fruit at both cold and room temperature. However, a slight reduction was normal for the climacteric fruit that continues to ripen. A study conducted by Al-Juhaimi (2014) showed a decrement of TSS value from 3.5 - 4.5 when treated with a combination of arabic gum and chitosan. Degradation of cell wall and sugar accumulation happen during ripening (Hossain *et al.*, 2014). Coating combination inhibits respiration process by minimising starch conversion into sugar, resulting in lower sugar (TSS) content. Lower respiration rate minimised acidity loss, preserve quality of tomato (Petriccione *et al.*, 2015). Slower ripening process delayed the carbohydrate hydrolysis into sugar (Elsabee and Abdou, 2013). Elsabee and Abdou (2013) studied the sugar content in tomato after coating process, and concluded that sugar might have increased due to their predominant properties in biological process except for sucrose. Glucose produced faster reduction in water activity than sucrose, which led to weight loss (Ortega-Rivas, 2007) based on Monsalve-Gonzales's study on water activity reduction by glucose immersion curve rising in 1993.

#### *Colour changes*

Colour was also indicated by higher hue angle (colour percentage) value. Usage of greener stage for coating process delayed and lowered fruit ability to undergo decay. Chitosan was helped by cinnamic acid to retain high L\*, inhibited colour changes and did not influence lycopene level in breaker stage (Sharma and Rao, 2014; Dávila-Aviña *et al.*, 2014). Chitosan deactivated polyphenol oxidase (PPO) to suppress phenylalanine ammonia-lyase (PAL) activity (Zhang *et al.*, 2017). Chitosan controlled activity of PAL, primary enzyme in phenolic compound biosynthesis. Decrease in PAL activity prevented increment of phenolic compound, a substrate for oxidative enzymes of PPO (Vitti *et al.*, 2011). PPO catalysed quinones oxidation which was associated with

browning (Queiroz *et al.*, 2008; Yamane *et al.*, 2010) in organelle prevented by chitosan as reducing agent [NH<sub>2</sub>] (Nicolas *et al.*, 2003). Chitosan suppressed PAL, prevented browning, lycopene degradation and retained the tomato colour. PPO activity was associated with the browning and lycopene degradation that caused the colour changes (Spagna *et al.*, 2005; Silva *et al.*, 2017). Sibozza *et al.* (2014) agreed that activation of PPO was responsible for tissue browning in lemon fruit. Ethylene synthesis inhibition due to the poor O<sub>2</sub> level restricted the rapid changes of colour before reaching consumers. During ripening of tomato, carotenoids were synthesized to lycopene (red) from colourless, whereas chlorophyll was degraded to colourless from green (Giuliano *et al.*, 1993; Fagundes *et al.*, 2014). 1.0% Ch + 3 mM CA and untreated tomato significantly influenced each other. The result showed red colouration increment (lycopene accumulation) along the storage days at ambient temperature as reported in Dumas *et al.* (2003) study which concluded that the optimum lycopene biosynthesis occurred at room temperature. Too much lower or higher than 32°C interrupted the synthesis. However, the changes of colour occurred at desirable and accepted level to indicate the maturity of tomato (Fagundes *et al.*, 2015).

## Conclusion

Tomatoes' physico-chemical qualities maintenance in term of lowering colour degradation, weight loss minimization, softening reduction and changes in TSS value has been proven by using layer by layer combined coating of 1.0% Ch + 3 mM CA. Favourable qualities preservation encourage the long lasting shelf life of tomato at the market. Usage of coating for commercial purposes are recommended due to the low cost and workmanship, yet provide a huge impact to agricultural practitioners. Chitosan and cinnamic acid can be a safe replacement for synthetic chemicals in protecting the condition of tomatoes before reaching target markets.

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