CHAPTER 4

Effect of bees-carnauba wax (mixed wax) coating on qualities and storage life of sweet orange fruit cv. Canh during storage period

The effect of bees - carnauba wax (mixed wax, MW) on postharvest qualities and storage life of Vietnamese sweet orange fruit cv. Canh was studied, by coating fruit with 4, 6, 8 and 10% MW and storing at ambient temperature (22°C±2) RH 80±5% for 20 days, and at low temperature (5±1°C) RH 80±5% for 50 days, while uncoated fruit were used as control. The results showed that 8% MW coating could reduced the percentage of fruit decay and weight loss, and rate of respiration; while ethanol concentration increased during the storage time. On the 20th day of storage at ambient temperature, the fruit maintained TSS content, total sugars, titrable acid, ascorbic acid, and sensory score higher than those of other treatments and control. At low temperature, the results shown that 8% MW coating could reduce the percentage of weight loss, rotten fruit, and rate of respiration for 50 days in storage and increase ethanol. Moreover, after 50 days in storage, the coated fruit still maintained their marketable properties expressed as titrable acid, TSS content, ascorbic acid, total sugars, ethanol and the sensory scores. For the control fruit at low temperature, the shelf-life was only 40 days.

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Coatings applied to the surfaces offruits and vegetables are commonly called 'waxes', whether or not any component thereof is actually a wax. Waxes prevent water loss thus reducing weight loss; inhibit post-harvest disease; decrease respiration rate; and maintain visual appearance in orange (Thirupathi *et al.*, 2006; Hung, 2008; Shahid and Abbasi, 2011), prolong storage life of orange, keeping freshness, and decreasing shrinkage (Hagenmaier and Shaw, 1992; Ladaniya and Wanjari, 2003). Carnauba wax (palm wax) is a wax of the leaves of the palm (*Copernicia prunifera*), a plant native to

and grown only in the northeastern Brazil. Bees wax is a natural wax produced in the bee hive of honey bees (*Apis* sp.).

The 'Delta Valencia' orange were coated with carnauba - based wax of refrigerated 5±1°C and 85±2% RH, extended storage life for 28 days (Pereira *et al.* 2013). Thang *et al.* (2013) showed that orange fruit cv. Vinh coating by polyethylene combination with bee wax reduced weight loss; and the fruits respiratory intensity, firmness, color and solid content changed slower than the control fruit. Coating 'Blood Red' orange fruit at concentration of bees wax along with benlate performed better results in improving the overall quality and extended the shelf life of fruit (Shahid and Abbasi, 2011).

The main purpose of this study was to evaluate effects of 4, 6, 8, and 10% MW on postharvest qualities and storage life of 'Canh' sweet orange during the storage period at ambient temperature and at low temperature storage.

4.2 Materials and Methods

4.2.1 Materials

Orange cv. Canh of a commercial orchard in ThanhOai district, Hanoi were harvested at 220-235 days after fruit set, were laid in spongy box (20 kg fruit/box) and transported to laboratory within 2-3 h. Fruit were then selected for uniformity of shape, size and non-defected fruit.

The bees wax and carnauba wax in the ratio of 7:3 was prepared following process of Thinh, (2013). The bees wax and carnauba wax was melted by a magnetic stirrerat 80-85°C. After that oleic acid, palmitic, water was added to the mixture during stirring and blending for 4, 6, 8 and 10% concentrations of mixed wax (MW) for 25-30 minutes. The MW of various concentrations were cooled down to ambient temperature before coated to the fruit.

Fruits were dipped in 4, 6, 8 and 10% MW for 1 min at room temperature. Then the coated fruit were stored under ceiling fan to let the MW coated dried, while uncoated fruit were used as control (Control). After that orange fruit were laid on trays and stored at ambient temperature (22±2°C) and RH 80±5% at each 5 days interval throughout for 20 days storage time.

Uniformity of orange fruit were dipped in 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) for 1 min at room temperature, after drying in a room by ceiling fan, coated fruit were laid on trays and stored at 5±1°C and RH 80±5% for 50 days, and sampled/analyzed at 10 day intervals. While uncoated fruit were store at 5°C (T0) for 40 days as control.

A completely randomized design was used for the experiment. All measurements of each treatment were the average of three replications.

4.2.1 Method

The titrable acid (TA) was determined as citric acid by titrating against 0.1NaOH by following the method given in AOAC (2000).

Total sugars were analyzed according to the procedure of Lane and Eynonas described in AOAC (2000).

Ascorbic acid (vitamin C) was estimated using the detective dye 2.6 dichlorophenol-indophenol by standardizing 0.1% standard 2.6 dichlorophenolindophenol dye solution against 0.1% ascorbic acid solution according to the method described in AOAC (2000).

Total soluble solids (TSS) content in fruit was determined in filtered juice by using a digital refractometer (RFM-80) (Atago, Tokyo, Japan)

Ethanol and acetaldehyde content in juice orange was estimated by using a Gas Solid Chromatography (GSC) Agilent 7890A equipped with a 30 m \times 0.25 mm \times 0.25 μm HP-5 column, and coupled with a Mass spectrometry (MS) 5975C detector (Agilent, Palo Alto, USA), according to the method described by Perez *et al.* (2002). The concentrations of the ethanol and acetaldehyde content were calculated base on (mg/L).

Fruit decay was assessed as the percentage of decay fruit as follows:

Percentage of weight loss was calculated by weighing the whole fruit putted into tray before and after storage, as follows:

Percentage of weight loss=
$$\frac{\text{Weight}_{\text{before}}\text{-Weight}_{\text{after}}}{\text{weight}_{\text{before}}} \times 100$$

Respiration rate was measured according to the method of Jiang and Li, 2001. The 1000 g fruit was sealed in a glass chamber for 2 hours at ambient temperature and 5°C, then a 5 ml gas sample was withdrawn with a gas-tight hypodermic syringe and analyzed by gas chromatography (Agilent Technology 7820A).

Sensory evaluation was tested on the day of harvest, on the day 5, 10, 15 and 20th at ambient temperature (22±2°C) and 0, 10, 20, 30, 40 and 50 days after harvesting at low temperatures (5°C). Fruit were peeled, and separated segments, and placed into disk. Each experiment sensory included a mixture of segments from five different fruit. Fruit taste was assessed by a committee consisting of 5 members. Each member evaluate the various attributes of samples, with points 'very good' and 'very bad' for each attribute, and sensory data were wrote point from 1 to 9 in the form. In addition, members were requested to rate overall: such as peel color, odor, taste, flavor on a hedonic scale from 1 to 9 point by a board of tasters as follows: 9=extremely like; 1= extremely dislike; and 5=neither like nor dislike (Hung, 2008). We also performed open discussion group in order and establish the sensory form.

Statistical analysis was carried out using Duncan's multiple range test was use to analyze the significant differences ($P \le 0.05$) between treatments and the control.

4.3 Results and Discussion

- 4.3.1 Effect of mixed wax coating on postharvest qualities and storage life of sweet orange fruit cv. Canh at ambient temperature
 - 1) Change in quality of fruit: titrable acid, total sugars, vitamin C, TSS, and ethanol

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Changes in titrable acid (TA) of mixed wax coated fruit and control at ambient temperature (22 ± 2 °C), 80 ± 5 %RH are shown in Fig. 4.1. The TA increased slightly

in all treatments and the control, and they did not significantly differ after 10 d in storage ($P \le 0.05$) (see appendix D). Maximum acidity (0.196%) was recorded in control. Least value (0.134%) was observed in 6% MW. Titrable acid of the fruit treated with 4, 8 and 10% MW were not different after 20 d in storage ($P \le 0.05$). After 20day of stored, the control had higher TA content (0.196%) than MW treatments (0.134-0.167%) (Fig. 4.1). Increase in acidity might be due to the water loss of the fruit which the control was the highest in water loss and it agree with Thang *et al.*, (2013), who reported that the total organic acids of coated 'Vinh' orange fruit decreased less than the control fruit after 7 weeks at 22-29°C.

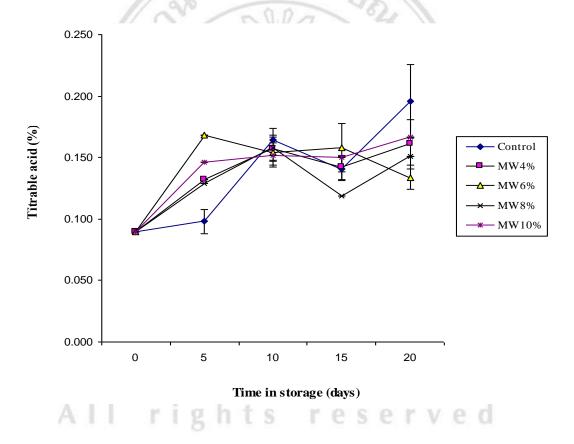


Figure 4.1 Effect of bees-carnauba wax (MW) coating on titrable acid of orange cv. Canh fruit stored at ambient temperature ($22 \pm 2^{\circ}$ C), $80 \pm 5\%$ RH

Changes in total sugars are indicated in the figure 4.2. After 10 d of storage, the total sugars content of all treatments tended to decrease slightly (Fig. 4.2). Total sugars content of control (4.86%) fruit decreased faster than other treatments. This should be due to the high respiration rate which sugars were used as the substrate of respiration.

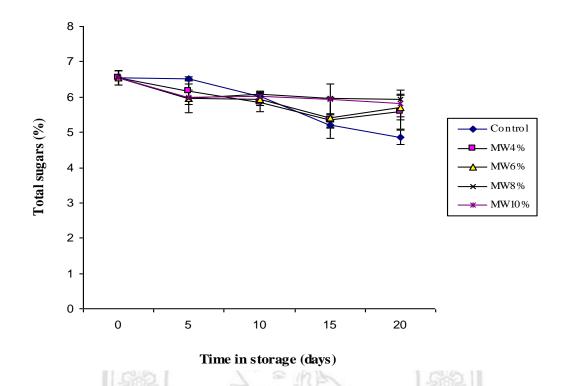


Figure 4.2 Effect of bees-carnauba wax (MW) coating on total sugars of orange cv. Canh fruit stored at ambient temperature $(22 \pm 2^{\circ}\text{C})$, $80 \pm 5\%\text{RH}$

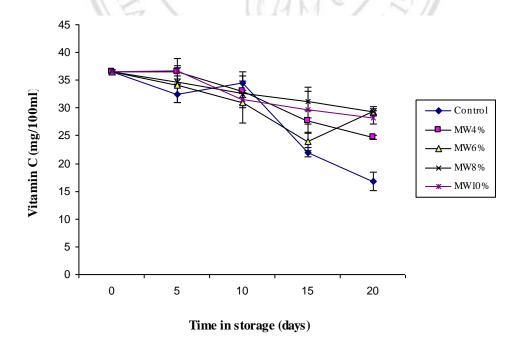


Figure 4.3 Effect of bees-carnauba wax (MW) coating on Vitamin C of orange cv. Canh fruit stored at ambient temperature (22 ± 2 °C), 80 ± 5 %RH

The changes in vitamin C content during storage period is shown in figure 4.3. After 20 d of storage, the control the lowest VitC content reduced from 36.46 to 16.78 mg/100 ml, while VitC content of the MW were reduced from 36.46 to 24.69-29.32 mg/100 ml. This result explains that MW could prevented in reduction VitC content due to the low respiration rate. This result agree with Thang *et al.*, (2013) who report that VitC of coated orange fruit was higher than control fruit during storage. The ascorbic acid of orange coated HPMC and *Moringa oleifera* extract treatment decreased gradually with prolonged storage time (Adetunji *et al.* 2012).

Changes in total soluble solids (TSS) content are presented in figure 4.4. TSS contents of all treatments tended to reduce during storage. TSS contents of the fruit were decreased from 11.60 to 10-11.33° brix after 20d of storage. TSS reduction should caused by fruit respiration but this situation TSS content of all treatments was not significant difference although the control had higher respiration rate than MW treatments. This should be caused by the water loss of the control fruit.

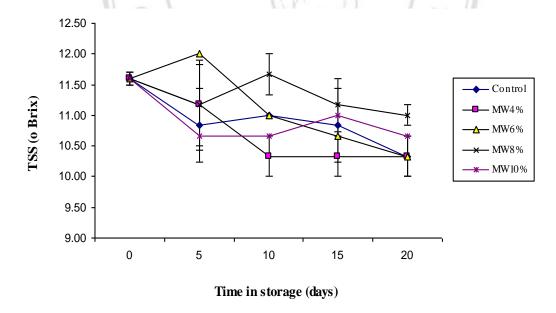


Figure 4.4 Effect of bees-carnauba wax (MW) coating on TSS of orange cv. Canh fruit stored at ambient temperature (22 ± 2 °C), 80 ± 5 %RH

Our results are consistent with the reported data on TSS content of oranges fruit of Shahid and Abbasi (2011) who reported that possible reason in reduction of TSS content in 'Blood Red' orange in 5% bee wax + 0.5% benlate was due to these retard

the hydrolysis of starch into sugars and also the conversion of polysaccharides in to disaccharides and monosaccharide by changing the biochemical activities. Ladaniya and Sonker (1997) concluded that maximum retention of TSS recorded when coating fruit were stored for up to 21days of storage of Nagpur mandarin. Baldwin *et al.* (1999) reported that polysaccharide and carnauba wax in mango created modified atmospheres, but only the polysaccharide coating delayed ripening and increased concentrations of vapor volatiles while the carnauba wax coating significantly reduced water loss compared to in the uncoated and polysaccharide coating treatments.

The changes in ethanol content in juice of the coated and control fruit during 20 days storage at ambient temperature are shown in figure 4.5. The lowest ethanol content in juice (88.44 mg/L) was observed in control fruit in other hand the highest ethanol content (110.4 mg/L) was found in 10% MW coated fruit. The ethanol should be a product of anaerobic respiration of the coated fruit which oxygen gas was blocked by the MW. Therefore, ethanol was accumulated inside the MW fruit more than the uncoated fruit (control). In contrast, acetaldehyde did not find in both coated and control fruit. This result was agree by Perez *et al.* (2002), and Hagenmaier (2000) who report that the coatings of mandarin and orangewith different emulsions have been combined with elevated levels of internal volatiles like ethanol, methanol, acetaldehyde and high internal carbon dioxide accumulation

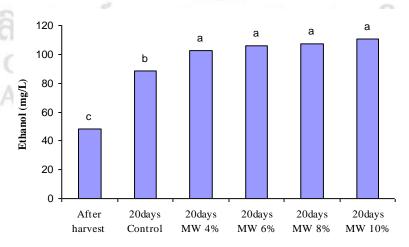


Figure. 4.5 Effect of bees-carnauba wax (MW) coating on ethanol of orange cv. Canh fruit stored at ambient temperature $(22 \pm 2^{\circ}\text{C})$, $80 \pm 5\%\text{RH}$

2) Change in fruit decay, weight loss, respiration, sensory values

The percentage of fruit decay in the treated and control fruit during the preservative time are illustrated in Figure 4.6. After stored for 10days, fruit decay about 3% were found in all treatments (Fig. 4.6). Percentage of fruit decay increased to 6-8% at 15th day of storage and increased to 11-14% day of storage which was not significant differences among all treatments. This result inferred that MW could not prevent fruit decay. In this study the fruit decay were caused by blue mold and green mold.

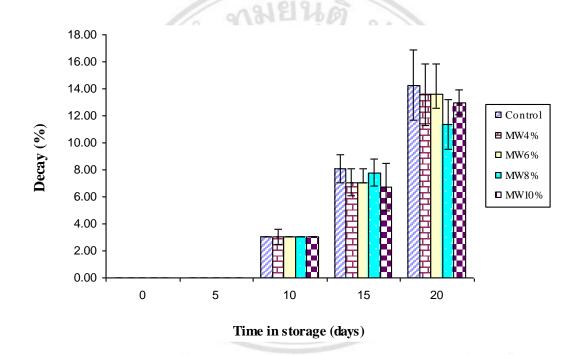


Figure 4.6 Effect of bees-carnauba wax (MW) coating on decay of orange cv. Canh fruit stored at ambient temperature (22 ± 2 °C), 80 ± 5 %RH

Changes in weight loss of treated and control fruit during the storage period are presented in figure 4.7. Weight loss percentages of all treatments were increased with the storage time. The uncoated fruit loosed weight higher and faster than the MW coated fruit (Fig. 4.7). The significant differences of weight loss percentage between the uncoated and coated fruit were found after 5 days storage. Among the MW coated treatments, only the 4% MW treatment had significant higher weight loss percentage than other MW treatment (13.29% and 8.37%-10.94%, respectively). This result show that MW coating could slow down the rate of water of the fruit.

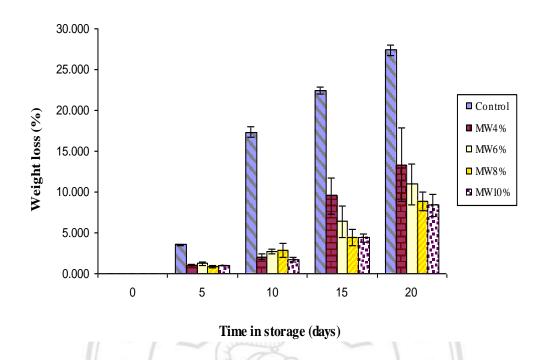


Figure 4.7 Effect of bees-carnauba wax (MW) coating on weight loss of orange cv. Canh fruit stored at ambient temperature $(22 \pm 2^{\circ}\text{C})$, $80 \pm 5\%\text{RH}$

Our results are consistent with the reported data on weight loss of oranges fruit of Ron et al. (2005) who showed that the weight loss from the 'Mor' mandarin coated either with the commercial 'Tag' wax and with the new modified 'Tag' wax was only half of that lost by the control fruit. Coating the orange fruit with wax could prevent water loss at ambient temperature (Binh and Dien, 1995; Po-Jung et al., 2005; Ron et al., 2005; Hoanet al., 2001; Thang et al., 2013, Ladaniya, 2008, and Farooqi et al., 1988). Oranges may lose up to 10% of the moisture in the peel after 3 wk at ambient temperature and relative humidity (Postharvest Handling Technical Bulletin 2003).

Figure 4.8 illustrates the changes in respiration rate of waxed and the control orange cv. Canh fruit during the storage time. The fruit surface coating was relatively impermeable to O2 and CO2 and water (Thirupathi *et al.*, 2006). This result was also confirmed that the MW had this properties. The respiration rate of the MW coated fruit were lower than the uncoated fruit (Fig. 4.9). Respiration rate pattern of the uncoated fruit tended to increase while the MW coated fruit tended to decrease. This should be the effect of MW which prevented O₂ to penetrate in to the fruit and prevented CO₂ to

penetrate out the fruit. Thus that low O_2 and high CO_2 content in the fruit could reduce fruit respiration rate.

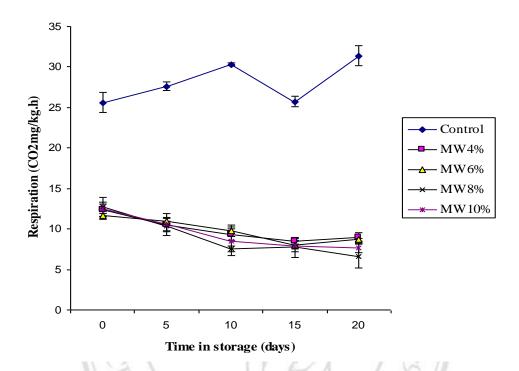


Figure 4.8 Effect of bees-carnauba wax (MW) coating on respiration of orange cv. Canh fruit stored at ambient temperature (22 ± 2 °C), 80 ± 5 %RH

Consistent with our result, Thang *et al.* (2013) showed that respiration rate of Vinh oranges coated with mixed bees and PE wax reduced during storage at 22-29°C. Our results are in accordance with the report of Ladaniya (1997) on respiration rate of 'Nagpur' mandarins. Respiration rate of TienGiang oranges coated with chitosan reduced after 25 days storage at 29°C (Binh and Dien, 1995). Commercial fruit wax has also been shown to reduce the respiration rate of coated fruit (Hagenmaier and Shaw, 1992).

Changes in sensory score of treated and control fruit during the storage period are indicated in Table 4.1. The overall fruit quality of MW coated fruit were higher than the uncoated fruit through the storage time (Table 4.1) while there were no significant differences among the MW coated fruit. The MW coated fruit still had high quality and got the high score (7.0-7.3) after 10 days of storage. After that the overall quality

reduced in all MW coated fruit and end up with neither like nor dislike quality (5.3-5.5) after 20d of storage. This result show that MW coating could preserve the fruit quality. This should be caused by the lower weight loss, respiration rate and fruit decay.

Table 4.1.Effect of mixed wax on sensory in orange cv. Canh at $22 \pm 2^{\circ}$ C, $80 \pm 5\%$ RH

	Time in storage (day)							
Treatments	0	5	10	15	20			
Control	8.5a	5.3a	5.3a	4.5a	3.5a			
4%MW	8.5a	7.3b	7.0b	6.5b	5.5b			
6%MW	8.5a	7.5b	7.2b	6.0b	5.3b			
8%MW	8.5a	7.8b	7.3b	6.5b	5.5b			
10%MW	8.5a	8.0b	7.2b	6.0b	5.5b			

Note: Means followed by the same letter(s) within a column are not significant different as determined by Duncan's multiple-range test P < 0.05.

9=extremely liked; 1=extremely disliked; and 5=neither liked nor disliked

Thirupathi *et al.*, (2006) showed that the fruit was left without waxy cuticle, the water quickly begins to evaporate, resulting in poor product shelf life to increase freshness and appearance. Our results are consistent with the reported data on sensory values of oranges fruit (Thang *et al.*, 2013) who reported that Vinh orange coated with bees and PE wax had better form, good quality scores and special taste than control. Hagenmaier (2000) reported that flavor scores of 'Valencia' oranges at all storage conditions (9-16 days at 15-25°C) were highest (8.0-10.5).

- 4.3.2 Effect of mixed wax coating on postharvest qualities and storage life of sweet orange fruit cv Canh at low temperature 5 ± 1 °C, 80 ± 5 %RH
 - 1) Change in quality of fruit: titrable acid, total sugars, vitamin C, TSS, ethanol content

The change in titrable acid of MW coated and control fruit is shown in figure 4.9. The TA of orange cv. Canh fruit increased slightly after 20 d and then decreased lightly after 50 d in storage at 5°C. TA of 6% MW coated fruit increased from 0.091 to

0.138% after 20 d in storage. In contrast, that of the control fruit increased by 0.118%. The TA of 8 and 10% MW coated fruit reached to 0.153 and 0.120% by day 40 in storage, respectively. By day 50 in storage, the TA of fruit coated with 6, 8 and 10% MW which decreased 0.092, 0.119 and 0.108%, respectively and they did not significantly differ ($P \le 0.05$) (see appendix D). This result shows that orange cv. Canh coated with MW increased lightly TA during preservation time.

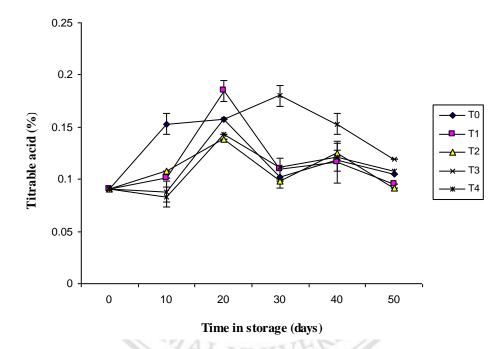


Figure 4.9 Effect of bees-carnauba wax (MW) coating on titrable acid of orange cv. Canh fruit stored at 5 ± 1 °C 80 ± 5 %RH

Note: Control (T0); 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) at 5°C

This finding is in accordance with the findings of Pereira *et al.* (2013); Rapisada, (2008). Pereira *et al.* (2013) reported that coating and un-coating 'Delta Valencia' orange fruit, titrable acid increase slowly during storage at 7°C, after 28 days (0 day was 0.38% and after 28 day was 0.43% for control, and for carnauba wax coating fruit increased 0.38 to 0.52%). Rapisada (2008) concluded that during storage at 6± 1°C for 65 days, five different orange genotype and citric acid content of 'Valencia' orange increased and decreased in the remaining varieties.

Figure 4.10 indicates the change in total sugars of control and MW coated orange cv.

Canh fruit during the storage period at 5°C. After 50 d in storage, total sugars of the fruit treated with 4, 8, 10% MW decreased quickly. Total sugars of 8% MW coated fruit decreased lightly from 6.54 to 6.30% when compared with the control was 5.52% after 20 d in storage. The total sugars of 4, 6, 8 and 10% MW coated fruit reduced lightly, and there was no significant difference (P≤0.05) in total sugars between treatmentsthroughout the preservative time. Total sugars of 10% MW coated fruit was found (5.66%). Total sugars loss may be due to the amount of sugar participate in respiration during the storage period.

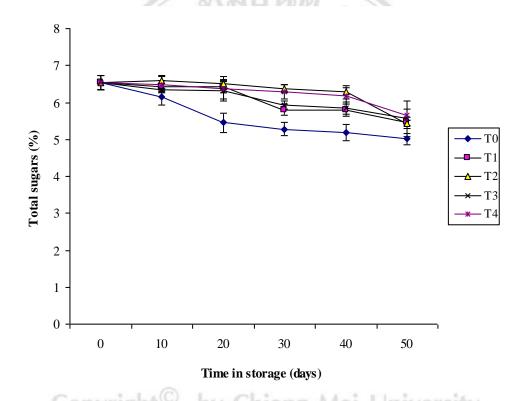


Figure 4.10 Effect of bees-carnauba wax (MW) coating on total sugars of orange cv. Canh fruit stored at $5 \pm 1^{\circ}$ C $80 \pm 5\%$ RH

Note: Control (T0); 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) at 5°C

Total sugars value of orange cv. Canh fruit in our study was similar to the reported data of David *et al.* (2008), who found that 'navel' orange coating with shellac and imazalil stored at 5°C for 6 weeks have sweet content reduced slightly from 111.2 to 108.2mg/100ml. The results of our study are in contrast to the results of Babar (2007), Pereira *et al.* (2013) who concluded that 'Kinnows' mandarin and 'Delta Valencia'

orange were treated with waxes and was stored having total sugars it increased throughout the storage period at low temperature.

Figure 4.11 illustrates the changes in vitamin C of coated and the control orange cv. Canh fruit during the storage period at 5°C. The amount of VitC decreased rapidly from 35.34 to 16.37 mg/100g in the control. Fruit coated with 6% MW that reduction VitC was 33.23 mg/100g after 20 d. VitC of the control (T0) treatment dropped rapidly (21.02mg/100g), which suggests that ascorbic acid is easy to decompose at 5°C and 50 d without wax. Overall, VitC tended to decrease in all coated fruit by day 50 in storage (Figure 4.11). Ascorbic acid of orange cv. Canh fruit was 24.94; 26.65 mg/100g in fruit was coated with 8 and 10% MW, respectively, and they did not differ (P≤0.05). This result shows that MW coating has an effective in maintaining VitC content in orange fruit cv. Canh.The results proves that there is no significant in VitC between MW different treatments of orange cv. Canh in the same condition storage (P≤0.05).

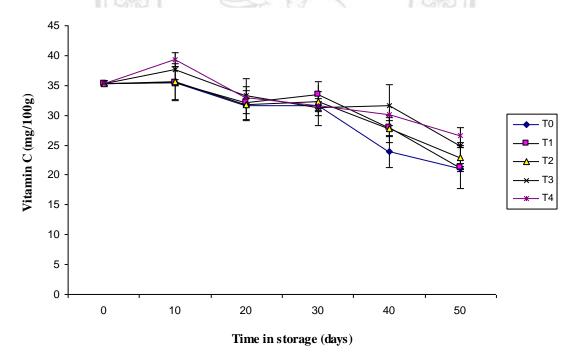


Figure 4.11 Effect of bees-carnauba wax (MW) coating on Vitamin C of orange cv. Canh fruit stored at $5 \pm 1^{\circ}$ C $80 \pm 5\%$ RH

Note: Control (T0); 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) at 5°C

Our results are in coincidence with the reported data on VitC content of orange fruit

(Mahajan*et al.* 2005, Babar, 2007, Kader, 2002) who reported that VitC of stored orange and mandarin reduced slightly after storage at low temperature. In contrast, Pereira *et al.* (2013) found that VitC levels were not affected by carnauba in 'Delta Valencia' orange.

Change in total soluble solids (TSS) content of orange cv. Canh fruit are reported in figure 4.12. After 20 d in storage, the TSS content of the control fruit decreased from 11.60 to 11.16 °brix. The TSS content of 4% MW coated fruit decreased from 11.60 to 9.33 °brix after 40 d in storage, and the TSS content of 8 and 10% MW coated fruit was similar (10.00 and 10.33 °brix, respectively) ($P \le 0.05$). The TSS content of treated and control orange slightly reduced with the raising of preservative time may be as a result of respiration and composing ethanol. Sala and Lafuente, (1999) showed that the hybrid nature of mandarin at low temperature storage could severely damage the fruit but holding fruit was most desired practice to maintain quality after storage and extend the marketing period.

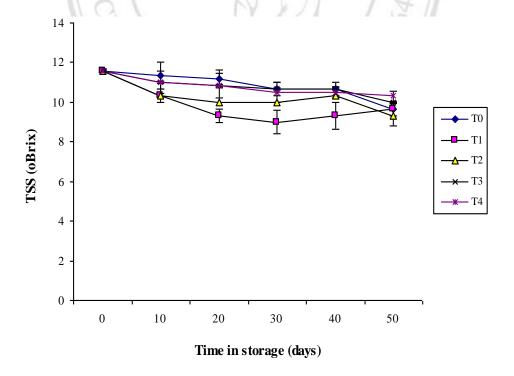


Figure 4.12 Effect of bees-carnauba wax (MW) coating on TSS of orange cv. Canh fruit stored at $5 \pm 1^{\circ}$ C $80 \pm 5\%$ RH

Control (T0); 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) at 5°C

The TSS contents of MW coated fruit was also in accordance with the reported date on TSS content of Thakur *et al.* (2002); Pereira *et al.* (2013) during cold temperature storage. Thakur *et al.* (2002) proved that higher TSS contents were found when Kinnows fruit was stored at ambient temperature, and lower TSS content is observed in Kinnows fruit when fruit was stored in evaporative cool chamber under ambient temperature might be due to higher rate of respiration and evaporation. TSS content was not affected by carnauba waxing after 28 days in storage, and TSS content reduced lightly from 9.63 to 9.38% (Pereira, 2013). On the other hand, Mahajan *et al.* (2005) and Babar (2007) concluded that TSS content of orange, mandarin in waxing increase during long term storage at low temperature.

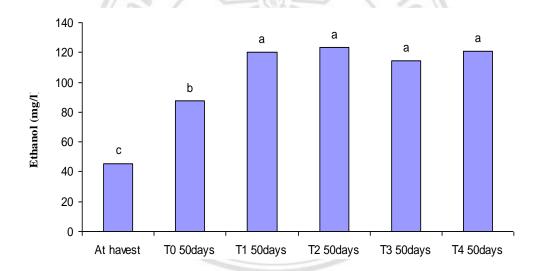


Figure 4.13 Effect of bees-carnauba wax (MW) coating on ethanol of orange cv. Canh fruit stored at 5 ± 1 °C 80 ± 5 %RH

Control (T0); 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) at 5°C

The changes in ethanol in juice of the coated and control fruit during the storage period at 5°C are shown in figure 4.13. After 50 d in storage, 8% MW coated fruit showed ethanol increasing higher than control. For control fruit, ethanol in juice was 87.4 mg/L. There were no significant differences in ethanol content among the treated fruit ($P \le 0.05$), which were all significantly different from the control fruit ($P \le 0.05$). The coating orange fruit with MW did not limit increasing of ethanol during low temperature. In contrast, acetaldehyde did not find in both coated and control fruit. The

main product of anaerobic respiration was ethanol and it could be seen that reducing the contents of both soluble solids and shellac in the wax, gradually reduced the accumulation of ethanol in the juice (Ron *et al.*, 2005). The chilling temperature also increased the production of ethylene and volatile components (ethanol and acetaldehyde) in fruit after a return to normal temperature (Eaks, 1980). In this research, ethanol in control more than other treatments perhaps due to respiration and metabolism sugar into alcohol

Levels of ethanol in oranges remain fairly constant with time at 20°C storage but the greatest change takes place after prolonged storage (>60d) at 9°C where ethanol levels reached values between 300 and 400mg/l00mL juice (Hagenmaier, 2000). David *et al.*(2008) assayed navel orange in California after 4 weeks storage, ethanol increased from 560 to 1488 μl/L, and very high levels of ethanol in orange would be expected to impart a fermented taste and strongly reduce the organoleptic properties of orange. Hagenmaier (2002) reported ethanol in mandarin hybrids was greater than 1500 μl/L. Hadar *et al.* (2014) showed that after 4 weeks in cold storage and 5 days at 20°C, ethanol of 'Mor' mandarin was high (3,700 μl/L) in juice of commercial wax and lilayer coated fruit. Babar (2007) reported that Kinnows stored at 2°C gave the lowest concentration of ethanol (224.33mg/kg) as compared to the 'Kinnows' stored at 5°C ethanol was found (297.83mg/kg).

2) Change in decay fruit and weight loss

The percentage of decay fruit of MW coated and control orange cv. Canh during the storage period at 5°C is shown in Figure 4.14. The rate of decay increased rapidly when fruit is stored for long time. After 40 d in storage, the control fruit had decayed by 8.58% and the minimum decay was found in MW coated fruit. For coated fruit, the highest fruit decay rate incidence was found (8.88%) in the 4% MW treatment and lowest decay rate was found (6.20%) in 10% MW coated fruit after 50 d in storage. Non significant variation existed in the decay fruit of orange cv. Canh fruit between bees and carnauba wax formulas ($P \le 0.05$), which were all significantly different from the control fruit ($P \le 0.05$). This result can assume that the preservative agents used in this study impacted the percentage decay of orange cv. Canh during preservative time.

Postharvest decay of oranges was also reduced by the use of appropriate prepostharvest and postharvest fungicides, proper sanitation of the wash water and appropriate storage temperature and RH condition (Postharvest Handling Technical Bulletin, 2003). This results are suitable for finding of Babar (2007) proved that the highest decay fruit (19.7%) occurred when (un-waxed) Kinnows mandarin when they stored under ambient conditions and the lowest incidence of decay (0.13%) when polyethylene based wax coated orange were stored at low temperature. Zhang *et al.*(2004) reported that rotting of 'Kinnows' took place due to moldattack during postharvest storage in cold chambers at different temperatures by green mold *Penicillium digitatum* which produces green filaments to cause rotting of citrus not subjected to any post harvest treatment. Waxing created a hydrophobic (non-water compatible) surface which was not conducive to pathogen growth and development (Postharvest Handling Technical Bulletin, 2004b)

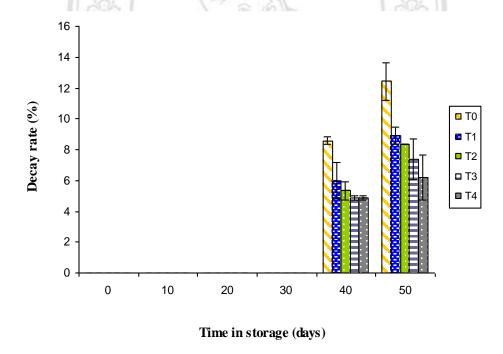


Figure 4.14 Effect of bees-carnauba wax (MW) coating on decay of orange cv. Canh fruit stored at $5 \pm 1^{\circ}$ C $80 \pm 5\%$ RH

Note: Control (T0); 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) at 5°C

Figure 4.15 shows the percentage of weight loss in the control and MW coated orange cv. Canh fruit during the storage period at 5°C. After 20 d in storage, weight loss

arranged from 1.95 to 3.89%, and there was no significant difference in the percentage of weight loss in 4, 6, 8, 10% MW coated fruit($P \le 0.05$), which were all significantly different from the control fruit ($P \le 0.05$). In contrast, the control fruit had 15.76% weight loss after 50 d. For coated fruit, the 4 and 6% MW coating treatments had the highest weight loss 11.15 and 10.39%, respectively after 50 d in storage, and the lowest weight loss in 8 and 10% MW were 7.41, 7.57%, respectively. Which were all treatments significantly different from the control fruit ($P \le 0.05$). Generally, percentage of weight loss of MW coated fruit and the control fruit increased with increasing of storage time and temperature storage. This study indicates that high weight loss appeared to be related to high decay and high respiratory (Figure 4.14 and 4.16). This result proved that 8% MW coating has the best effectiveness on reducing the weight loss in orange cv. Canh during storage at low temperature. Waxing reduced the severity of chilling injury and allows for storage of chilling injury sensitive commodities at slightly lower temperatures without incurring damage (Postharvest Handling Technical Bulletin 2004a).

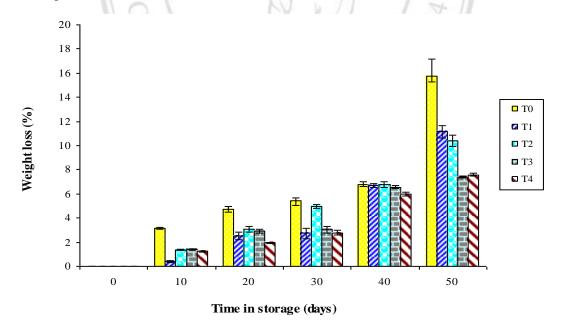


Figure 4.15 Effect of bees-carnauba wax (MW) coating on weight loss of orange cv. Canh fruit stored at $5 \pm 1^{\circ}$ C $80 \pm 5\%$ RH

Note: Control (T0); 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) at 5°C Our results are in accordance with the reported data on weight loss of orange cv. Canh, coating the orange, mandarin fruit with wax could prevent weight loss during storage at

low temperature (Pereira *et al.*, 2013, Zipora *et al.*, 2012, Mahajan *et al.*, 2005, and Ron *et al.* 2005). Wax emulsions Fruitex, Britex-561 and SB 65 coated on 'Kinnow' oranges, lemons and grape fruit reduced weight loss and kept the fruit firmer, thus maintaining their fresh look (Farooqi*et al.*, 1988). Mark (2013) illustrated that symptoms of chilling injury of Florida oranges included pitting, brown staining, increased decay, internal discoloration off-flavor, and watery breakdown that may take 60d to develop at 5 °C.

3) Change in respiration rate and sensory score

The changes in respiration rate of MW coated and control orange cv. Canh fruit during the storage period are measured and results are shown in Figure 4.16. The respiration rate of MW coated and control fruit tended to increase during the first 10 d at 5°C in storage, control increased strongly after 10 d. This study shows that the increasing of respiration rate appeared to be related with fruit decay (Figure 4.14 and 4.16).

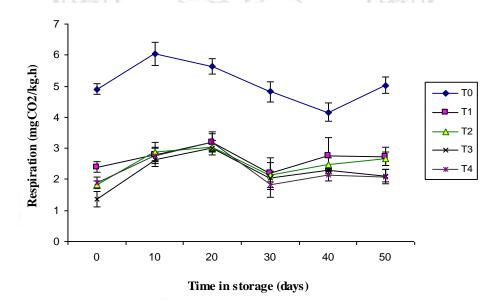


Figure 4.16 Effect of bees-carnauba wax (MW) coating on respiration of orange cv. Canh fruit stored at $5 \pm 1^{\circ}$ C $80 \pm 5\%$ RH

Note: Control (T0); 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) at 5°C

In Fig. 4.16, after 10 d, respiration rate of non-treated increased slightly at 5°C, that was 6.04 mg CO₂/kg.h - the highest after 10 d of storage. The respiration rate decreased at long time and respiration rate reduced rapidly in coated fruit. After that, respiration rate

of 10 and 8% MW tended to decrease after 50 d during the preservation (2.09 and 2.12 mgC0₂/kg.h, respectively). There were no significant differences in respiration rate among the treated fruit ($P \le 0.05$), which were all significantly different from the control fruit ($P \le 0.05$). This results concludes that MW coating limited respiration rate at low temperature. Respiration of citrus fruit was affected by temperature, humidity, air movement, atmospheric gases, and handling practices (Ladaniya, 2008). Increasing the temperature increased respiration rate; lowering the temperature restored the original respiration rate with no evidence whatsoever of a climacteric (Vines *et al.*, 1968).

Similar to the report of Farooqi *et al.*, (1988) who reported that wax emulsions Fruitex, Britex-561 and SB 65 coated on oranges, lemons and grape fruit reduced rate of respiration. Commercial fruit wax has also been shown to decreased the respiration rate of coated fruit (Hagenmaier and Shaw, 1992). Ni *et al.* (2008) concluded that respiration rate of mandarin cv. Sai Nam Peung Teva wax coating was lower than fruit did not wax. Postharvest development of chilling injury symptoms in citrus in Florida can reducedthrough temperature conditioning before storage, use of wax coating, use of high CO₂ atmosphere (Mark, 2013).

Table 4.2 Effect of mixed wax on sensory in orange cv. Canh at 5 ± 1 °C 80 ± 5 % RH

		7.1							
Treatments	Time in storage (days)								
	0	10	20	30	40	50			
T0	8.5a	7.5b	7.0ab	6.0a	5.5a	3.5a			
T1 C0	8.5a	7.3b	7.0b	6.0a	5.5a	4.2a			
T2	8.5a	7.5b	7.2b	6.0a	5.0a	4.2a			
Т3	8.5a	7.8b	7.3b	6.5a	5.5a	5.2b			
T4	8.5a	8.0b	7.2b	6.0a	5.7a	5.5b			

Note: Means followed by the same letter(s) within a column are not significant different as determined by Duncan's multiple-range test P < 0.05.

9 = extremely liked; 1= extremely disliked; and 5 = neither liked nor disliked

Note: Control (T0); 4% MW (T1), 6% MW (T2), 8% MW (T3) and 10% MW (T4) at 5°C

Table 4.4 shows the changes in sensory score of coated and the control orange cv. Canh

fruit during the storage period at 5°C. After 40 d, it was 5.5 in for the control, and 5.7 for 10% MW, and they were no significant difference in all MW treatments ($P \le 0.05$). Sensory score decreased slightly 5.5; 5.2 and 4.2 in fruit coated with 10, 8 and 6% MW, respectively and that was significant difference with MW treatments ($P \le 0.05$). After 50 d in storage at low temperatures, orange cv. Canh coated with 8% MW to have sensory score value that is accepted for marketable. This study indicated that high sensory score appeared to be related to low respiratory and low decay throughout the preservative time (Fig. 4.14 and 4.16). Ron *et al.* (2005) reported that although coating and un-coating 'Mor' mandarins have taste score it decrease slowly during storage at 5°C, and after 5 wk was 7.1 score in coating shellac. According to Hagenmaier (2000) an experimental polyethylene - candelilla wax coating formulation for 'Valencia' orange was evaluated high flavor score (8.9-10.4) even after 9-16 days in storage at 15-25°C.

Overall, results from this study proves the effectiveness of mixed wax at low temperature decreased respiration, slows pathogen growth, reduced water loss and extended shelf-life in orange cv. Canh throughout storage time.

4.4 Conclusion

Coating orange fruit cv. Canh in 8%MW (bees wax and carnauba wax) and storing at ambient temperature (22±2°C), 80±5%RH reduced weight loss, fruit decay and throughout the 20 d in storage. In addition, fruit maintained lower respiration rate, titrable acid, TSS content, total sugars, vitamin C, and ethanol content than other treatments and control.

Coating orange fruit cv. Canh in 8% MW (bees wax and carnauba wax) maintained a good postharvest qualities expressed as titrable acid, TSS content, total sugar, vitamin C, and sensory values for 50d at 5°C. Moreover, this treatment reduced respiration rate, weight loss, fruit decay, and increasing of ethanol. In addition, acetaldehyde did not find.

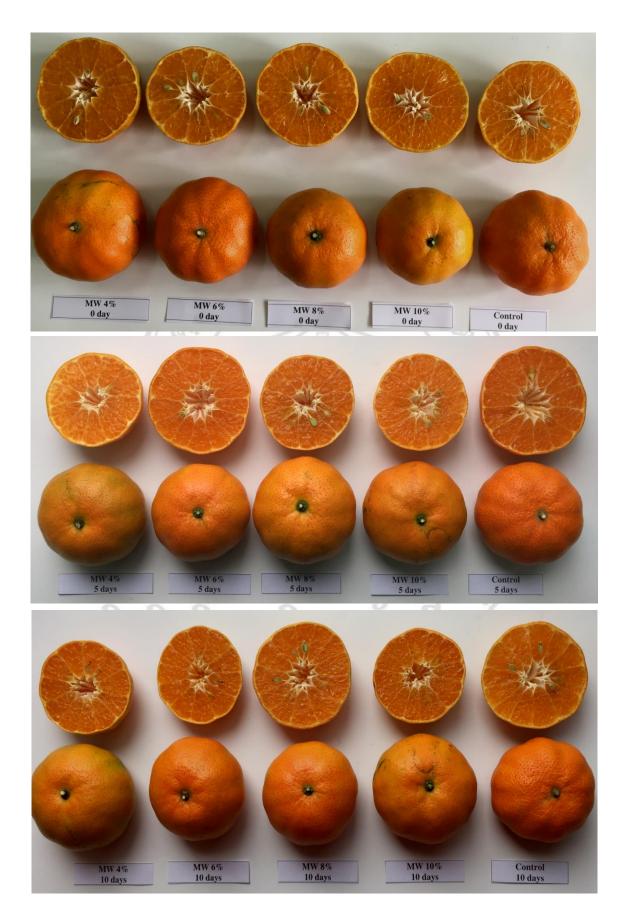






Figure 4.17 Effect of bees-carnauba wax (MW) coating on storage time of orange cv. Canh fruit stored at ambient temperature ($22 \pm 2^{\circ}$ C), $80 \pm 5\%$ RH