Efficacy of Carboxymethyl Chitosan Coating on Maintaining Quality of Sai Nam Pueng Tangerine Fruits Compared to Commercial Coating

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Abstract

The effect of carboxymethyl chitosan (CMCS) coating without and with konjac glucomannan (CMCS/KGM) on postharvest qualities of tangerine cv. 'Sai Nam Pueng' was intermittently evaluated and compared with the Commercial A coating during simulated commercial storage (at 5±1°C, 60-65%RH for 28 days followed by 25±2°C, 60-65% RH for 12 days). The qualities of uncoated and coated Sai Nam Pueng tangerines, including weight loss, firmness, internal gas composition, total soluble solids, titratable acidity, disease incidence, glossiness, and wilting, were investigated. It was found that Commercial A coatings outstandingly enhanced glossiness and firmness, as well as weight loss retardation, by 50% compared to the control (noncoating) and the polysaccharide coatings (CMCS and CMCS/KGM). Coatings, however, exerted no effect on the total soluble solids (TSS 11.2-13.4), titratable acidity (TA 0.65-0.86), and internal gas composition at elevated temperatures, with an increase of internal CO₂ from 2.5-4% (non-coated) to 4-7% (CMCS), 6-6.5% (CMCS/KGM), and 7-8% (Commercial A), and a decrease of internal O_2 from 18.5% (non-coated) to 10-14% (CMCS), 7-7.5% (CMCS/KGM), and 5-6% (Commercial A), respectively. Based on the average level of internal gas, CMCS coating permitted far better permeability to O_2 than CMCS/KGM coating and Commercial A. On the contrary, there were small differences in the level of internal CO₂ among various coatings, with

Commercial A having significantly less permeance to CO_2 than the polysaccharide coatings. It was also observed that KGM potentially increased the accumulation of CO_2 on the CMCS coating independently of the storage temperature. In this study, the CMCS/KGM coating delayed the disease incidence as effectively as the Commercial A coating.

Keywords: Polysaccharide-base Coating, Tangerine, Wax Coating, Fruit Qualities, Storage

Introduction

The steadily rising consumption of fruits, along with the indefinitely rising health awareness line, is due to the fact that fruits are an important source of multivital vitamins and antioxidants with proven anti- aging, anti- cancer, and heart- related diseases. Oranges of various cultivars are grown in many parts of Thailand, making them almost year-round available, including Sai Nam Pueng mandarin, which is widely grown in the northern part of Thailand. Sai Nam Pueng mandarin has been highly perceived owing not only to its availability but also to other factors such as its wellbalanced sweet and sour flavor with a good aroma, high vitamin C content, ease of peeling, and still being palatable at a high level of water loss. The exported value of 163 thousand tons in 2019 marked Sai Nam Pueng's popularity as one of the top ten most exported fruits from Thailand (Food and Agriculture Organization., 2021) Fruits and vegetables, including oranges, are very perishable due to their high-water content. Through respiration and transpiration, weight loss and senescence-related quality losses of products emphasize the importance of coatings. The 3-3.5 times higher respiration rate increase of mandarin compared to other oranges or grapefruit made mandarin the most difficult to handle among all citrus as they are very susceptible to off-flavor development and quality deterioration. The mandarin peel is denser in structure and more restrictive to oxygen and aroma volatile diffusion through the peel, hence one of the most likely reasons that mandarins are more prone to off-flavor compared to grapefruit. Based on the origin of the materials used, a coating is divided into synthetic coatings made from petroleum sources and natural coatings made from plants, animals, and microorganisms. Due to the rising concern over plastic pollution in the environment, the preference for edible coatings is thereby constantly increasing.

Sources of edible polymers are widely abundant, inexpensive, safe, and renewable. They can also be made from agricultural waste or sub-value agricultural products. Although varieties of edible coatings have been developed and applied, there is still a need to improve the coating properties, such as mechanical properties and water barrier, of the polysaccharide and protein-based edible coatings due to their hydrophilic nature. Moreover, many of the currently available coatings contain hazardous materials, which alarmingly raises concerns among consumers about seeking more safety alternatives. Due to the different inherent coating permeabilities, some of the coating can trigger the off-flavor potential of coated fruits. To improve shine and better weight loss control, shellac coating has long been widely used in the citrus industry, although it is at risk for off-flavor. The polysaccharide coating, on the contrary, does not provide the high gloss effect of shellac coatings but imparts an attractive sheen to the fruit, although it is not effective in retarding water loss. Mandarin fruits, which are more susceptible to anaerobic conditions (Cohen et al., 1990), could possibly benefit from a polysaccharide coating with improved weight loss control (Hagenmaier & Shaw, 2002).

Edible coatings should be stable and generally recognized as safe, tasteless, colorless, odorless, and with good mechanical properties. According to Liu et al. (2017), carboxymethyl chitosan (CMCS) is a secondary derivative of chitosan. Due to the presence of -NH2 and -COOH, the solubility of CMCS is not limited to some common solvents but includes water. This chitosan derivative is widely known for better biocompatibility, moisture absorption, antibacterial, and film-forming properties compared to chitosan (Pang et al., 2008). The gaining prominence and industrial attraction of fungal-sourced carboxymethyl chitosan is not only because of its superior functional and biological properties but also because it has a vegan appeal and is safe for consumers with hypersensitivity to crustacean food, which is estimated to be 5% of the world population. CMCS coating can delay the ripening-induced changes in Hami melons during transport vibration at 23°C (Zhou et al., 2020), and also delay ripening, reducing weight loss, and retaining fruit firmness of Moya tomatoes at 25-30 °C (Benhabiles et al., 2013). CMCS and gelatin maintained the quality and nutritional characteristics of four sweet cherry cultivars, reducing weight loss, maintaining skin color, peduncle freshness, higher fruit firmness, TA, AA, total phenolics content, total

anthocyanins concentration, and antioxidant capacity of sweet Cherries at 0 ± 0.5 °C, RH 85%–90% for 30 days (Zhang et al., 2021).

Konjac glucomannan (KGM) is extracted from the konjac corms after it has been sliced into chips and dried. The powerful absorption capacity and thermal resistance of non-toxic konjac glucomannan from the konjac tuber plant set it apart from other polysaccharides. The use of konjac glucomannan in combination with other polymers to make a film with an increased water barrier and mechanical properties has been reported (Wang et al., 2016). There are many researchers try to apply the KGM with another coating or varied the concentration of KGM to improve their properties such as Ratchawet and Joradol (2017) reported that 1.15% w/v was the optimum concentration of KGM to reduce weight loss and change of tomatoes color with no effect on soluble solids, the titratable acidity, and the firmness. Piriyaphattarakit et al. (2017) found that 0.5% konjac and GLK wax can extend the shelf life of tangerine orange when stored at 5°C and 90±5 % RH, notably customer acceptable and weight loss although no significant difference in peel color change.

However, no research related to the combination of CMCS and KGM application as a coating for tangerine was reported. Hence, in this study, the efficacy of carboxymethyl chitosan with and without Konjac glucomannan on the maintaining quality of Sai Nam Pueng tangerine was investigated and compared to the commercial polyethylene-shellac coating.

Methodology

1. Orange preparation

Late season Tangerine oranges cv. 'Sai Nam Pueng', size 4-5 (60.65 mm diameter) maturity 90%, were sorted from the Wang Ka Nil Orchard situated in Mae Ai, Chiang Mai District, Thailand. The oranges were packed in a cushion paper box of 3 layers and transported by air-conditioned car and precooled in a cold room at 5°C for 1 night before cleaning and coating.

2. Coating materials

Mushroom carboxymethyl chitosan (CMCS) was purchased from ChiBio (China) with % deacetylation \geq 98%, viscosity 10-80 mPa. s, pH 7-8, and molecular weight 10-90 kDa. Konjac glucomannan (KGM) is a water-soluble dietary fiber of high

molecular polysaccharide from Hubei Yizhi Konjac Biotechnology Co. Ltd. (China) with viscosity 23,700 mPa.s, molecular weight 20-200 kDa, and glucomannan >75%. Commercial A solution (ZivdarTM) was donated by Nature bright Co. Ltd. (Thailand).

3.1 Coating solutions preparation

Coating solution consisted of two polysaccharide formulations and a commercial coating 'Zivdar Wax' (Commercial A) were prepared as shown in table 1.

Composition	Zivdar	Polysaccharide 1	Polysaccharide 2
Material 1	Polyethylene	1.4% CMCS in	1.4% CMCS in
	+unknown wax	water	water
	(18%)		
Material 2	Shellac (3%)	-	0.1% KGM
Material 3	Ammonium	Tween 80	Tween 80
	hydroxide (1%)	(0.3%of dried	(0.3% of dried
	1652	coating material)	coating material)
Material 4	Oleic acid (1%)	Glycerol	Glycerol
	5/11	(20% of total vol.)	(20% of total vol.)
Material 5	Fungicide		2) -
	Imazalil		
Material 6	Water	Water	Water
рН	10-11	7.7	7.3
Odour	Strong	neutral	neutral
	ammonium smell	La.	1

Table 1 The Components and characteristics of coating solution

3.2 Tangerine coating and storage

Tangerines were coated with the coating solutions and into 4 groups: (1) non-coated fruits (control), (2) CMCS coated fruits, (3) CMCS/KGM coated fruits, and (4) Commercial A coated fruits. Five fruits were put on a plastic tray and packed in cardboard box; all treatments were done in 3 replications. During the first 28 days, the samples were stored in a cold room at $5\pm1^{\circ}$ C, 60-65 %RH. After 28 days, the samples

^{3.} Effect of Coating on Qualities of Sai Nam Pueng tangerines During Storage

were transferred to a new storage room at 25±2°C, 60-65%RH. Sampling was carried out intervalley every 7 days for quality assessment.

3.3 Quality Assessment

3.3.1 Weight Loss

The weight change was monitored by electric balance (Model PA4102, Ohaus Corporation, Pine Brook, USA). In this experiment, each treatment consisted of 3 replications where each replications contained 5 oranges arranged in a plastic basket before being placed in a cardboard box. Initial weight of oranges was recorded, and weight change of oranges was monitored weekly during 5 °C storage and every 3days during 25° C storage. The weight loss described in Angasu et al. (2014) was used for the calculation as simplified in equation (1).

Weight loss % = (Weight $_{D0}$ -Weight $_{Dn}$)/Weight $_{Dn}$ x100 (1) Weight $_{D0}$ = initial weight

Weight D_n = final weight at each sampling interval (Day $_n$)

3.3.2 Firmness

Firmness of oranges was taken as the force in newtons (N) required to compress the fruit by 19 mm between two flat surfaces closing together at the rate of 5 mm/min, Trigger Force 5g, Compression plate P100, Pre-test speed 1 mm/s. using the texture analyzer (Model TA-XT plus, Stable Micro System, Surrey, UK). Each reading consisted of 3 replications x 5 samples (N=15) (Singh & Reddy, 2006).

3.3.3 Internal Carbon Dioxide and Oxygen Content

The oranges were room temperate for 1 hour before the internal gas was measured by gas chromatography technique, Gas chromatography: model Agilent 7890N, made in China, Column: MolSieve 5A and HP-PLOT M with thermal conductivity detector 200 °C, Carrier gas: Helium: flow rate 52.2 ml/min. Using the micro syringe 0.5mm, approx.5 ml of gas was carefully withdrawn from the inner core of oranges prior to an immediate insertion of the syringe through the injector. Care must be taken so the needle does not get into the orange flesh.

The sampling of internal gas was taken at day12 and day 26 during cold storage. After the transition of sample to ambient storage, the sampling was taken after day3, day6, day10 and day12 respectively.

3.3.4 Total Soluble Solids (TSS)

The sample of oranges after texture analysis were peeled and cut through. Juice of all 5 fruits were collected in the same container and stirred well before each measurement. Total soluble solid (Brix) was measured by dropping a drop of well stirred juice to a digital hand-held refractometer (model PALI, Atago,Japan). Each treatment consisted of 3 replications x 3 readings (N=9).

3.3.5 Total Titratable Acidity (TA)

Total titratable acidity was perform using a hand-held TA analyzer (Atago, Tokyo, Japan). The orange juice sample from the same beaker prepared for total soluble solids was diluted with a distilled water at weight ratio 1:49 (juice: water). Each treatment consisted of 3 replications x 3 readings (N=9).

3.3.6 Disease Incidence

Disease Incidence was calculated as number of fruits with defect / total number of fruits \times 100; fruits with defects included any signs of pathological, physiological disorders or chilling injury.

3.3.7 Measurement of Gloss

Gloss level from matt (1) to glossy (5) was recorded weekly with naked eyes during cold storage (28 days at $5\pm1^{\circ}$ C, $60\pm5^{\circ}$ RH) and 3 days interval during ambient storage (12 days at $25\pm2^{\circ}$ C, $60\pm5^{\circ}$ RH).



GLOSS SCALE 1-3-5 Matt-satin/sneen-glossy

Figure 1 Standard gloss level score of orange from matt (score 1) to glossy (score 5)

3.3.8 Measurement of Wilting

Degree of wilting level from low /wilting-free (5) to high/serious wilting (1) was recorded weekly during cold storage (28 days at $5\pm1^{\circ}C$ $60\pm5^{\circ}RH$) and 3 days interval during ambient storage (12 days at $25\pm2^{\circ}C$ $60\pm5^{\circ}RH$).



WILTING SCALE 1-3-5 (high – no wilting)

Figure 2 Standard degree of wilting level of orange from low or no wilting (score 5) to high or serious wilting (score 1)

3.4 Statistical Analysis

The data will be presented as mean \pm SD at least triplicate determinations. Analysis of variance (ANOVA) and significant tests of the mean by Tukey HSD Multiple Range Tests. The results will be measured at p<0.05 statistical significance. The analysis was conducted by using SPSS 17 program.

Results and discussion

1. Weight Loss

A constant rise in weight loss during storage reduces fruit quality and market value by changing firmness, turgidity, crispiness, aroma, color, gloss, nutritional content, and disease resistance (Boonyakiat et al., 2012). Even 5-6% water loss in orange and mandarin could affect fruit look and firmness, reducing marketability. Figure 3 shows weight loss of tangerines cv. 'Sai Nam Pueng' at 5±1°C, 60-65% RH for 28 days, followed by 12 days at 25±2°C, 60-65% RH for distribution temperature simulation. Weight loss increased with storage time regardless of coating type or temperature. In storage, Commercial A coating could reduce weight loss more than CMCS/ KGM coating, CMCS coating, and non- coating (control), respectively. Comparing tangerines coated with polysaccharide CMCS and CMCS/KGM to the control showed no significant weight loss. Weight loss is related to coating water vapor permeability (WVTR), according to Mannheim and Soffer (1996). Lower WVTR coatings impede weight loss better. Compared to Commercial A (Zivdar), wax, or other shellac composites, carboxymethyl chitosan and KGM were more hydrophilic, causing the weight loss at 4°C. Zivdar coating using polyethylene wax and shellac is hydrophobic compared to CMCS and CMCS/KGM. A similar weight loss was found on CMCS/KGM coated oranges and CMCS-coated oranges compared to non-coated oranges, possibly due to reduced film cohesiveness over time.



Figure 3 Weight Loss of Tangerines cv. 'Sai Nam Pueng' During Combined Storage at 5±1°C, 60-65% RH (28 days) Followed by 25±2 °C, 60-65% RH (12 days)

2. Firmness

Orange softening is one of the first signals consumers use to assess postharvest quality and shelf life (Kader, 1992; Singh & Reddy, 2006). Firmness value gradually declines during storage due to fruit softening or crispiness loss following water evaporation, cell wall pectin degradation, nutritional substrate depletion, over maturity, or microbial attack.

Coated and untreated fruits attained a textural peak and retained firmness before and after (Figure 4). Unlike Commercial A, CMCS, CMCS/KGM coated oranges, and non-coated oranges increased in firmness from 121 N to 133 N, 136 N, and 140 N in the first week of cold storage and remained steady for 21 and 28 days. Non-coated, CMCS, and CMCS/KGM-coated fruits were firm on days 7 and 21. CMCS/KGM firmness increased from day 21 (133 N) to day 28 (148 N), while non-coated orange firmness fell from 140.6 N to 132.5 N. On day 3 of ambient storage, non-coated oranges were softest and CMCS/KGM coated fruits were firmest. At ambient, CMCS/KGM was firmer than non-coated, CMCS-coated, and Commercial A-coated fruits only on day 3. After ambient storage, firmness dropped slightly and became insignificant for non-coated oranges (130.8N), CMCS-coated oranges (130 N), and CMCS/KGM coated oranges (132 N) after day 6. Commercial A-coated oranges showed no hardness increase in the first week but a second week (from 212.8 N to 136 N) before a considerable decline from day 14 (136 N) to day 21 (119 N) and stayed consistent throughout cold storage (125 N) CMCS and CMCS/KGM coating may have kept fruits firmer than Commercial A throughout combined storage, but not after day 6 at ambient shelf life. Commercial A firmness was the lowest. The decline of firmness in mature fruits is an early sign of quality index (Chang et al., 2017; Salvador et al., 2008; Xue et al., 2020), as raw pectin, tightly attached to cellulose, disintegrates (Khademi et al., 2014) and becomes soft water-soluble pectin. This study found that firmness reduced with storage time, contrary to prior findings on Kinnow mandarin, Korla Fragrant pear, tomatoes, and strawberry. The hardness of CMCS or CMCS/KGM coating layer may be the cause of firmness increases. Coating has been shown to retard cell wall degradation enzymes in wolfberries (Ban et al., 2015), tomatoes (Reddy et al., 2000), apples (Atkinson et al., 2012), strawberries (He et al., 2018), date fruits (El-Zoghbi, 1994; Mehyar et al., 2014), and bananas. This study found that, at the end of storage, coated oranges were firmer than non-coated. Polysaccharide-coated fruits (CMCS and CMCS/KGM) were firmer than Commercial A-coated fruits possibly due to the hardness of CMCS and CNCS/ KGM coating layer. Higher firmness at lower temperatures was observed in non-coated fruits, CMCS, and CMCS/KGM, aligning with Satsuma mandarin research by Won and Min (2018) and the findings of Olmo et (2000) that low temperatures slow fruit ripening by slowing respiration, al. transpiration, and softening. Zivdar-coated fruits were firmer at room temperature than in cold storage, unlike polysaccharides. Reduced CO₂ solubility at high temperatures may limit carbonic acid production, causing peel softening at higher temperatures.



Figure 4 Firmness of Tangerines cv. 'Sai Nam Pueng' During Combined Storage at 5±1°C, 60-65% RH (28 days) Followed by 25±2°C, 60-65% RH (12 days)

3. Internal Gas Composition

The internal gas composition of non-coated and coated oranges was determined and shown in Figure 5. Increasing of internal CO_2 and decreasing of O_2 were observed in every coating during storage due to the gas barrier properties of coating materials. Thus, the highest CO₂ levels in Commercial A-coated fruits, CMCS coated fruits, and CMCS/KGM were found at day 14, 28, and 31, respectively (correlated to the highest firmness of the coated fruits). According to Brummell and Harpster (2001), CO₂ increase during respiration reduces structural degradation enzyme activity, causing firmness loss. This study found that coated fruits increased CO₂ during storage at both Commercial A or Zivdar- coated oranges kept their immaculate temperatures. appearance after 42 days, satisfying the features described in earlier references about shellac's exceptional effect on weight loss retention, gas barrier effect, and gloss The CMCS- based and water- soluble shellac- based coatings were attribution. significantly affected by poor relative humidity (Hagenmaier & Shaw, 1991). All coated fruits were permeable during cold storage in this study (60% RH), but Commercial A coated fruits accumulated more CO₂ than polysaccharide coated fruits, as shown in the internal gas discussion that CMCS is impermeable to CO₂ and O₂ gases below 70% RH. Acidic CO₂ may have softened and swelled Commercial A-coated orange peels, which improved in humid conditions. This experiment shows that different coatings affect internal gas levels (Banks et al., 1993; Hagenmaier & Baker, 1994; Mannheim & Soffer, 1996; Navarro-Tarazaga et al., 2007). Coating thickness is less important than surface pore blocking, even with a modest deposit or partial blocking (Amarante et al., 2001). Most likely, CMCS-coated fruits had the maximum pores blocking and peel permeance after coating, followed by CMCS/KGM and Commercial A coated fruits. Thus, CMCS-coated fruits had greater internal CO₂ levels.

4. Total Soluble Solids (TSS)

Figure 6 shows TSS levels of tangerines cv. 'Sai Nam Pueng' stored at 5±1°C 60-65% RH (28 days) and $25\pm2^{\circ}$ C 60-65% RH (12 days). Coated oranges had lower TSS than non-coated ones, especially at ambient shelf life, but both showed a modest insignificant increase as storage time advanced throughout coating types. Lower TSS of all coated oranges compared to non-coated may be due to the semipermeable effect of coating to gas and moisture. Ben-Yehoshua et al. (1985) recommended to plug the stomatal pores to form an intermittent cracked layer over the fruit's surface, restricting ethylene, O₂, and CO₂ transport but not water transport. TSS may have increased due to starch metabolism during storage and its conversion to sugar and other soluble solids (El-Zeftawi, 1976). The increase of TSS at higher temperatures is consistent with Hassan et al. (2014), who showed that tangerine citrus TSS increased progressively with storage time. Research suggests that oranges covered with various emulsions have lower TSS loss than controls (Toğrul & Arslan, 2004). On day 21 and 28, commercial A coated oranges had a lower TSS than the control and polysaccharides, demonstrating the improved efficacy of this polyethylene coating's hydrophobicity and waterrepellency in humid circumstances. Commercial A coated oranges had less TSS variation than polysaccharides. This may indicate improved homogeneity and fewer faults in Commercial A coating, as discussed under flavor and internal gas. Soonthornvipat (2003) found that Zivdar reduced tangerine cv weight loss and transpiration. Storing 'Sai Nam Pheung' at 21±2°C, 67% RH did not affect its peel color, total soluble solids, titratable acidity, or sensory score. Shein et al. (2008)) found no significant changes in TSS/TA after 1 month of cold storage of Sai Nam Pueng coated with Teva wax, a comparable composition to Zivdar (18% food grade shellac, polyethylene). Soranacom and Khamsee (2011) found that the total soluble solid of





Figure 5 Internal Gas Concentrations: (A) carbon dioxide and (B) oxygen of Tangerines cv. 'Sai Nam Pueng' During Combined Storage at 5±1°C, 60±5% RH (28 days) Followed by 25±2°C, 60-65% RH (12 days)

5. Titratable Acidity (TA)

Titratable acidity (TA) is made up of organic acids that help plants respire, and it is used to assess horticulture crop respiration (Chen et al., 2013). All coated oranges had far lower TA than non-coated oranges in the first week. After one week, CMCS- coated oranges had a higher TA than CMCS/KGM and Commercial A. The total titratable acidity of CMCS/KGM-coated oranges was a bit higher than that of CMCS and Commercial A coated oranges at higher storage temperatures (Figure 7). Coated fruit reduced organic acid loss by reducing oxygen permeability, respiration rate, and acid oxidation (Ahmed et al., 2009; Yaman & Bayoundurl, 2002). At both storage temperatures, coated fruits with higher TA and internal CO₂ were softer. The higher TA in CMCS-coated fruits corresponded to the trend of the highest internal CO2 during cold storage of the coating, while the higher TA in CMCS/KGM- coated fruits corresponded to the trend of the distinctively lower internal O2 at ambient shelf life. Since coatings accumulate carbon dioxide and oxygen inside the fruit during respiration, they slow structural breakdown and firmness loss.



Figure 6 Total Soluble Solids (TSS) of Tangerines cv. 'Sai Nam Pueng' During Combined Storage at 5±1°C, 60-65% RH (28 days) Followed by 25±2°C, 60-65% RH (12 days)



Figure 7 Total Titratable Acidity of Tangerines cv. 'Sai Nam Pueng' During Combined Storage at 5±1°C, 60-65% RH (28 days) Followed by 25±2 °C, 60-65% RH (12 days)

6. Disease Incidence

Low temperature is the most common way to extend postharvest produce storage and shelf life by delaying ripening, fruit respiration, enzymatic activities, and pathogen infections. This experiment found the disease incidence (Figure 8) in noncoated and CMCS coated Sai Nam Pueng oranges during cold storage after 2 weeks and 4 weeks, respectively. While CMCS/KGM and Commercial A coated oranges showed no disease incidence throughout the experiment. This result was similar to the results of Feygenberg et al. (2004) who found waxes prevented decay in avocado after 3 weeks at 5°C and 8 d at 20°C, similar to coated guava (McGuire & Hallman, 1995), and mango (Baldwin et al., 1999). Due to their intrinsic antibacterial and antioxidant properties, CMCS-based and chitosan-based coatings could reduce pathogen growth in Sai Nam Pueng compared to non-coated fruits. In contrast, Commercial A or Zivdar contains fungicide (Imazaril) which can inhibit fungi growth (Gassner et al., 1969). Surjyatem et al. (2018) found that CMCS film with propolis extract inhibited Grampositive bacteria (S. aureus and B. cereus) but not Gram-negative bacteria. On peach coatings, CMCS's antioxidant activity increased with decreasing molecular weight, suggesting its use as an antioxidant and preservative coating (Elbarbary & Mostafa, 2014; Jung & Zhao, 2016). The antioxidant nature of the hydroxyl groups in KGM and CMCS can also explain the superior green mold resistance of CMCS/KGM coating compared to CMCS (Wang et al., 2016).



Figure 8 Disease Incidence in Tangerines cv. 'Sai Nam Pueng' During Combined Storage at 5±1°C 60±5%RH (28 days) Followed by 25±2°C 60±5%RH (12 days)

7. Glossiness

Variable coating glossiness is seen in Figure 9. From day 0, coated oranges presented the higher glossiness than non-coated fruit and could maintain during 28-days in cold storage. After 28 days, Commercial A coated fruits still showed the highest glossiness followed by CMCS/KGM coated, CMCS coated, and non-coated fruits. CMCS/ KGM coated fruits lost glossiness after three days in ambient storage. Semipermeable coating barriers make goods shiny. This experiment found that the CMCS/KGM coating lost gloss on day 3 at 25°C, which increased wilting and weight loss faster than CMCS from the prior week of cold storage. Coating type determines gloss. Chitosan, alginate, xanthan, aloe-vera, and caraway oil give fruits a glossy appearance and extend their shelf life (Ganiari et al., 2017; Tiwari et al., 2022), as do carnauba wax, shellac, and resin coatings. Valencia oranges coated with carnauba wax (1.5/1 by weight) (Hagenmaier, 2000) yielded similar results. According to Boonyakiat et al. (2012), Sai Nam Pueng oranges coated with Zivdar had a better visual appearance than the control and Fomesa (polyethylene and wood resin) after 10 days of storage at

 24 ± 3 °C, $59\pm6\%$ RH. Zivdar, an oxidized polyethylene-based commercial wax, contains shellac to improve gloss and lower polyethylene melting point (Roongruangsri et al., 2013). Its capacity to keep covered fruits glossy and water-resistant is widely known.



Figure 9 Gloss Development of Tangerines cv. 'Sai Nam Pueng' During Combined Storage at 5±1°C, 60±5% RH (28 days) Followed by 25±2 °C, 60±5% RH (12 days)

8. Wilting (Shriveling of the Peel)

All fruits had no wilting (score = 5) during 7 days of cold storage as shown in Figure 10. From day 14, wilting of fruits appeared in non-coating, CMCS coating, and CMCS/KGM treatments as the reducing of wilting score. After day 3 stored at 25°C storage, wilting acceptance score of CMCS/KGM-coated fruits were less than CMCS-coated fruits and non-coated. Most CMCS-coated, CMCS/KGM-coated, and uncoated fruits looked good. By day 9, wilting increased (Figure 10), and the same amount of stem-end aberrant fruits were uneatable. Non-coated fruits were better than CMCS and CMCS/KGM-coated fruits, even when they were old. Wilting is linked to weight loss, as Holcroft (2015) found. More weight loss means more withering. Wilting appears after rapid weight loss and generally precedes off-flavor. Wilting of Sai Nam Pueng, both coated and uncoated, increased throughout storage Leven 5-6% water loss in orange and mandarin could affect fruit beauty and firmness, reducing marketability (Ladaniya, 2008). The investigation found that fruits stored at $5\pm1^{\circ}$ C, $60\pm5^{\circ}$ RH saw

a weight loss of 5-6% for non-coated, CMCS-coated, and CMCS/KGM-coated fruits after 2-3 weeks. Commercial A could retain weight more than polysaccharide because Commercial A coating was more hydrophobic than CMCS and CMCS/KGM coatings. This result was the same trend with the result of weight loss.



Figure 10 Wilting Score of Tangerines cv. 'Sai Nam Pueng' During a Combined Storage at 5±1°C, 60±5% RH (28 days) Followed by 25±2°C, 60±5% RH (12 days)

Conclusion

In this study, polysaccharide- base coatings from carboxymethyl chitosan (CMCS) and CMCS mixed with konjac glucomannan (CMCS/KGM) were developed and applied in Sai Nam Pueng tangerine compared to the commercial tangerine coating agent (Commercial A). From the results, all coatings exerted no effect on the total soluble solids (TSS 11.2-13.4) or titratable acidity (TA 0.65-0.86), but affected internal gas composition with an increase in CO₂ and a decrease in O₂. Commercial A coatings outstandingly prevented weight loss by 50% compared to the control and the experimental polysaccharide coatings (CMCS and CMCS/KGM), enhanced the shine and firmness, but gave the permeability of O₂ and CO₂ similar to CMCS and CMCS/KGM coatings. With the increase in storage at ambient temperature, Sai Nam Pueng oranges with a polysaccharide coating were inevitably less prone to off-flavor than Commercial A coating, although they had a less attractive appearance.

Commercial A and CMCS/KGM coatings also presented the good effect on delaying of disease incidence in tangerines.

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