



## Effect of chitosan/methyl cellulose films on microbial and quality characteristics of fresh-cut cantaloupe and pineapple

Jurmkwan Sangsuwan<sup>a,\*</sup>, Nithiya Rattanapanone<sup>b</sup>, Pornchai Rachtanapun<sup>c</sup>

<sup>a</sup> Postharvest Technology Institute, Chiang Mai University, Chiang Mai 50200, Thailand

<sup>b</sup> Department of Food Science and Technology, Faculty of Agro-Industry, Chiang Mai University, Chiang Mai 50100, Thailand

<sup>c</sup> Department of Packaging Technology, Faculty of Agro-Industry, Chiang Mai University, Chiang Mai 50100, Thailand

### ARTICLE INFO

#### Article history:

Received 16 September 2007

Accepted 18 February 2008

#### Keywords:

Chitosan  
Methyl cellulose  
Fresh-cut  
Cantaloupe  
Pineapple  
Vanillin  
Antimicrobial film

### ABSTRACT

Two experimental films were applied on fresh-cut cantaloupe and pineapple and their effects on microbial control and fruit quality were investigated during storage at 10 °C. Three types of films were used in this study: a commercial stretch film, an experimental chitosan/methyl cellulose film, and a chitosan/methyl cellulose film incorporating vanillin (vanillin film) as a natural antimicrobial agent. Fresh-cut fruit without any film wrapping served as controls. Chitosan/methyl cellulose film and vanillin film provided an inhibitory effect against *Escherichia coli* on fresh-cut cantaloupe. The chitosan/methyl cellulose film rapidly reduced the number of *Saccharomyces cerevisiae* yeast inoculated on cantaloupe and pineapple. Vanillin film was more efficient than chitosan/methyl cellulose in reducing the number of yeast, which decreased by 4 logs in fresh-cut pineapple on day 6. Vanillin film increased the intensity of yellow color of pineapple. Pineapple removed from stretch film had higher respiration rates and ethanol contents than other treatments. Unsurprisingly, the stretch film maintained the moisture content in fruit better than other treatments. However, vanillin film reduced the ascorbic acid content in pineapple. At the end of storage, ascorbic acid in pineapple wrapped with vanillin film was only 10% of its original concentration.

© 2008 Elsevier B.V. All rights reserved.

### 1. Introduction

Chitosan is the second most abundant polysaccharide on earth and is inherently antimicrobial (Goldberg et al., 1990). Furthermore, it provides films with good mechanical and oxygen barrier properties (Caner et al., 1998; Chen et al., 1996). Chitosan, however, has poor tensile strength when wet. It is rigid and has poor elongation properties. Blending cellulose with chitosan can be expected to correct these weaknesses; film flexibility has been shown to increase with increasing methyl cellulose content (Garcia et al., 2004).

The growth of microorganisms on the cut surfaces is a main cause of food spoilage for fresh-cut produce. The application of antibacterial substances directly onto a food has some limitations because the active substances can be neutralized, evaporated or they may inadequately diffuse into the bulk of the food (Torres et al., 1985; Siragusa and Dickson, 1992). The incorporation of antimicrobial agents into packaging can create an environment inside the package that may delay or prevent the growth of microorganisms on the product's surface and, hence, lead to an extension of its shelf-life. Antimicrobial packaging has attracted much attention from

the food industry because of the increase in consumer demand for minimally processed and preservative-free products. Reflecting this demand, preservative agents (preferably natural preservatives) must be applied at the lowest effective level possible (Cha and Chinnan, 2004). According to Brody et al. (2001), the antimicrobial effect of chitosan occurs when organisms are in direct contact with the active sites of chitosan. When antimicrobial agents are incorporated into film, they diffuse out of the film, thus improving its antimicrobial efficacy. Zivanovic et al. (2005) applied chitosan-oregano essential oil (EO) in comparison with chitosan films on inoculated bologna meat samples stored for 5 d at 10 °C. Pure chitosan films reduced *Listeria monocytogenes* by 2 logs, whereas the films with 1 and 2% oregano EO decreased the numbers of *L. monocytogenes* by 3.6 to 4 logs and *Escherichia coli* by 3 logs. Pranoto et al. (2005) incorporated garlic oil, potassium sorbate and nisin in chitosan films. The activity of the antimicrobial films was tested against the food pathogenic bacteria, *E. coli*, *Staphylococcus aureus*, *Salmonella typhimurium*, *L. monocytogenes*, and *Bacillus cereus*. They found that the pure chitosan film had no inhibitory effect. Incorporation of 100 µL of garlic oil/g, 100 mg potassium sorbate/g or nisin at 51,000 IU/g of chitosan had antimicrobial activity against *S. aureus*, *L. monocytogenes*, and *B. cereus*.

Many consumers have concerns over the addition of chemical additives to food, and this has driven the food industry

\* Corresponding author. Tel.: +66 53948226; fax: +66 53948201.

E-mail address: [jurmkwan@chiangmai.ac.th](mailto:jurmkwan@chiangmai.ac.th) (J. Sangsuwan).

**Table 1**  
Numbers of *Escherichia coli* on inoculated cantaloupe (log cfu/piece) during storage at 10 °C

	No film	Stretch film	Chitosan/methyl cellulose film	Vanillin film
Day 0	5.18 ± 0.00 f	5.18 ± 0.00 f	5.18 ± 0.00 f	5.18 ± 0.00 f
Day 1	4.50 ± 0.23 e	5.45 ± 0.19 fg	3.88 ± 0.52 de	4.43 ± 0.20 e
Day 2	3.94 ± 0.01 de	5.95 ± 0.05 g	2.29 ± 0.83 c	3.54 ± 0.58 d
Day 4	2.86 ± 0.15 c	7.19 ± 0.22 h	1.00 ± 0.00 b	1.00 ± 0.00 b
Day 6	2.56 ± 0.15 c	8.98 ± 0.05 i	0.74 ± 0.00 b	0.74 ± 0.00 b
Day 8	1.00 ± 0.00 b	9.27 ± 0.05 i	0 a	0 a

Means with different letters are significantly different at  $p=0.05$ .

and food research towards the search for natural antimicrobial compounds (Devlieghere et al., 2004). Vanillin (4-hydroxy-3-methoxybenzaldehyde) is the major constituent of vanilla beans and is a flavor compound used in many baked or processed foods. Prindle and Wright (1977) found that the effect of phenolic compounds was concentration dependent. At low concentrations, phenols affected enzyme activity, especially those enzymes associated with energy production, while at greater concentrations, they caused proteins to denature. The antimicrobial activity of vanillin depended on the time of exposure, concentration and the target organism. Recent reports have shown that vanillin can be effective in inhibiting bacteria, yeasts and molds (Jay and Rivers, 1984; Cerrutti and Alzamora, 1996; Matamoros-Leon et al., 1999; Fitzgerald et al., 2004). Vanillin has been used to inhibit *E. coli* O157:H7 and *L. monocytogenes* in 'Granny Smith' apple juice (Moon et al., 2006). Rupasinghe et al. (2006) reported that total aerobic counts of fresh-cut apple slices decreased from 4.3 log cfu/g fresh weight (untreated) to 1.6 log/cfu by using NatureSeal (an anti-browning agent) plus 12 mM vanillin after 19 d at 4 °C. Cerrutti et al. (1997) treated strawberry puree with a mild heat treatment combined with 3000 mg/L vanillin and 500 mg/L ascorbic acid. They found that the inhibition of native and inoculated flora growth for at least 60 d storage at room temperature. Penney et al. (2004) found that vanillin at 2000 mg/L suppressed fungal and total microbial growth in yoghurt significantly over the 3-week period.

Research on the application of antimicrobial biodegradable films on fresh-cut fruit is limited. The objectives of this work were to evaluate the inhibitory effect of chitosan/methyl cellulose film and chitosan/methyl cellulose film with vanillin against *E. coli* and *Saccharomyces cerevisiae*, and to determine the effect of these films on fresh-cut cantaloupe and pineapple quality.

## 2. Material and methods

### 2.1. Film preparation

Chitosan with a degree of deacetylation of 90% and purity >99.75% (Bannawach Bio-line Co. Ltd., Thailand) was prepared by dissolving 1.5 g of chitosan in 100 mL of 1% acetic acid solution. One half grams of methyl cellulose, 1.5 g, (M-043, BENECEL®) was dis-

solved in 50% ethanol. One gram of polyethylene glycol (PEG) 400 was used as a plasticizer. Solutions of chitosan and methyl cellulose were mixed in a beaker with a stir bar and heated to 72 °C. Stearic acid, 0.075 g was added to improve the water barrier properties of the film. Vanillin, 0.9 g (Sigma, St. Louis, USA) was incorporated after the temperature of the solution reached its melting point (83 °C). The film-forming solution was filtered through a cheese cloth to remove undissolved parts, homogenized with a homogenizer, degassed, cast onto glass plates, and dried at 40 °C for 42 h. Dried films were peeled off and conditioned at 25 ± 2 °C, 50 ± 5% RH for at least 48 h prior to use. Film thickness was measured with a gauge micrometer GT-313-A (Taiwan) with an accuracy of 0.01 mm.

### 2.2. Fruit preparation

Cantaloupe (*Cucumis melo*) and pineapple (*Ananas comosus*) fruit were purchased from a wholesale market in Chiang Mai province, Thailand. Total soluble solids were measured to indicate fruit maturity. Cantaloupe and pineapple used in this study had total soluble solids in the range of 7.0–8.2 and 17.0–19.4%, respectively. Whole fruit were washed with 500 mg/L chlorine solution. The blossom and stem ends were discarded. Cantaloupe and pineapple were sliced longitudinally into 12 wedges and 8 wedges, respectively using a sanitized sharp knife and cutting board. Then, the seeds or core, and peel were removed. All knives, cutting boards and other equipment which come into contact with the fruit were sanitized by immersion in 1000 mg/L chlorine solution for 30 min before cutting.

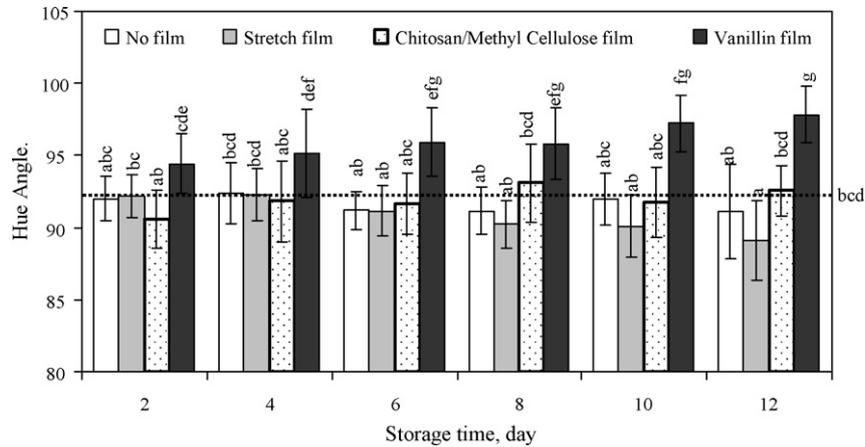
### 2.3. *E. coli* and *S. cerevisiae* inoculation and determination of *E. coli* and *S. cerevisiae* number through incubation

Cantaloupe and pineapple wedges were cut into 2.5 cm × 2.5 cm × 0.5 cm pieces. They were then inoculated with 20 µL of approximately 10<sup>5</sup> cfu/mL *E. coli* (TISTR 780) or *S. cerevisiae* (TISTR 5240) suspensions on the top surface of each piece (Zivanovic et al., 2005). Then, commercial stretch film, M wrap®, chitosan/methyl cellulose film and chitosan/methyl cellulose film with vanillin were wrapped around each piece. Wrapped fruit were placed on polystyrene trays and stored at 10 °C up to 20 d. Inoculated fruit without any wrapping served

**Table 2**  
Numbers of *Saccharomyces cerevisiae* on inoculated cantaloupe (log cfu/piece) during storage at 10 °C

	No film	Stretch film	Chitosan/methyl cellulose film	Vanillin film
Day 0	5.36 ± 0.00 bc	5.36 ± 0.00 bc	5.36 ± 0.00 bc	5.36 ± 0.00 bc
Day 1	5.07 ± 0.06 bc	5.26 ± 0.04 bc	2.83 ± 0.95 a	4.81 ± 0.19 bc
Day 2	5.11 ± 0.03 bc	5.38 ± 0.04 bc	3.00 ± 0.00 a	4.85 ± 0.22 bc
Day 4	5.39 ± 0.16 bc	6.81 ± 0.10 de	2.26 ± 1.16 a	4.31 ± 0.12 bc
Day 8	7.07 ± 0.49 e	8.47 ± 0.23 fg	4.24 ± 1.29 b	4.85 ± 0.11 bc
Day 12	6.72 ± 0.85 de	9.03 ± 0.11 gh	5.65 ± 0.58 bcd	4.45 ± 0.11 bc
Day 16	7.45 ± 0.38 ef	8.87 ± 0.14 gh	5.68 ± 0.55 cd	4.75 ± 0.05 bc
Day 20	7.27 ± 0.63 e	9.72 ± 0.16 h	5.20 ± 0.62 bc	4.71 ± 0.12 bc

Means with different letters are significantly different at  $p=0.05$ .

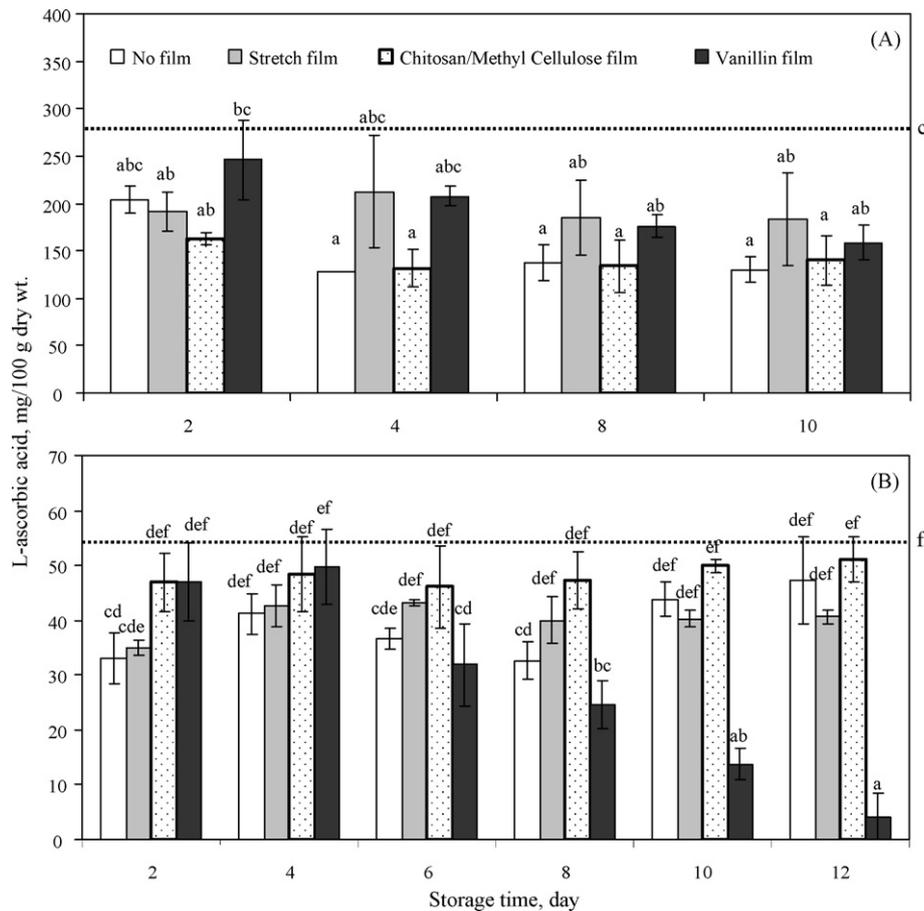


**Fig. 1.** Hue angle of fresh-cut pineapple during storage at 10 °C. Dash line represents hue angle on day 0. Bars with the same letters are not significantly different according to the Tukey's-*b* test ( $p > 0.05$ ).

as controls. At specific time intervals, fruit pieces and films were washed with sterile 0.1% peptone. The plate counts of *E. coli* were performed on violet red bile agar with MUG (Criterion, USA) after incubated at 37 °C for 48 h. MUG generally permits the rapid detection of *E. coli* when the medium is observed for fluorescence under long wavelength UV light. The numbers of yeast cells were determined by surface plating of 0.1 mL washed peptone solution on Sabouraud agar (MERCK, Germany) with 1% yeast extract. They were incubated at 25 °C for 48 h prior to counting.

2.4. Quality evaluation of fruit wrapped with antimicrobial film

Fresh-cut cantaloupe and pineapple wedges were wrapped with the three types of films (Stretch film, chitosan/methyl cellulose film and vanillin film), and unwrapped fruit served as controls. All fruit pieces were placed on polystyrene trays and stored at 10 °C for up to 20 d. Since Thailand is a tropical country and fresh-cut fruit are stored in open chiller displays or even placed on ice-cubes, 10 °C was selected in this study. Measurements of all attributes were done every 2–4 d until the end of storage:



**Fig. 2.** L-Ascorbic acid content of fresh-cut cantaloupe (A) and pineapple (B) during storage at 10 °C. Dash line represents L-ascorbic acid content on day 0. Bars with the same letters are not significantly different according to the Tukey's-*b* test ( $p > 0.05$ ).

Flesh color ( $L^*$ ,  $a^*$ ,  $b^*$ ) of both fruit species was measured on both longitudinally cut surfaces after removal from the wraps using the Hunterlab color meter ColorQuest XE (The Color Management Company, Virginia, USA) calibrated with a white tile. Readings were taken from six wedges of each treatment.

Firmness was measured as the maximum force required to penetrate into fruit wedges using a Texture Analyser TA.XT2i (Texture Technologies Corp., Scarsdale, NY, USA) with a 50-kg load cell. A 6-mm diameter flat head stainless steel cylindrical probe was set to penetrate into the fruit at a speed of 0.5 mm/min.

L-Ascorbic acid (AA) was determined using an Agilent 1100 HPLC equipped with a quaternary pump system, an autosampler and a UV detector at 254 nm (Agilent Technologies, Palo Alto, CA). The analytical column used was a Restek Ultra Aqueous C18 (150 mm × 4.6 mm, 5- $\mu$ m particle size). The sample preparation was done at cold temperature and under reduced lighting. Fifteen grams of each flesh was blended with cold 0.4% oxalic acid solution and adjusted to a final volume of 50 mL. The homogenate was filtered through a filter paper and then a 0.45- $\mu$ m syringe filter. The filtrate, 20  $\mu$ L, was injected into the HPLC system. Isocratic separation was carried out using a mobile phase of Milli-Q water with 0.1% (v/v) acetic acid. The eluent flow rate was 0.7 mL/min and the column temperature was 25 °C (Romeu-Nadal et al., 2006). HPLC grade L-ascorbic acid (MERCK, Germany) was used to make the calibration curve. Calibration curve was created by diluting L-ascorbic acid in the concentration range from 5 to 100 mg/L. Standard solutions were prepared fresh under cold and dark conditions to avoid AA degradation in samples. The relative retention time was 4.5 min. The determination of linearity ( $R^2$ ) of the standard curve was 0.9993.

Fruit respiration rate was determined using a static method. Each unwrapped cantaloupe and pineapple wedges were put into a 710-mL airtight glass container. CO<sub>2</sub> released from the products was absorbed by calibrated 0.01N NaOH solution for 1 h at 10 °C. The solution was then titrated with 0.005N oxalic acid. The respiration rate was expressed as mg CO<sub>2</sub>/kg h (Zhang et al., 2005).

Ethanol content of the fruit was determined by gas chromatographic analysis of the headspace according to a method developed by Davis and Chace (1969) with some modification. Five grams of flesh were placed in a 10-mL amber glass bottle with rubber cap and incubated in a water bath at 60 °C for 45 min. Headspace gas was withdrawn using a 1-mL syringe and injected into a TRACE GC gas chromatograph (ThermoQuest Italia S.p.A., Italy) equipped with a flame ionization detector. The temperature of the oven, injector and detector were 150, 175 and 200 °C, respectively. The column used was a 30 m × 0.53 mm I.D. × 1  $\mu$ m OV-1 (100% dimethylpolysiloxane) capillary column. Retention times and a standard curve of absolute ethanol (31–2000 mg/L) in water solution were used for peak identification and quantification.

Total soluble solids (TSS) and pH were measured. TSS was determined using a digital refractometer (Pocket PAL-1, Japan). Ten

grams of flesh were blended with 40 mL distilled water in a blender. The pH was measured at 25 °C with a pH meter (Consort C831, Belgium).

Weight loss of fresh-cut cantaloupe and pineapple was determined by weighing the samples at specific time intervals and plotting weight losses against time.

### 2.5. Statistical analysis

All experiments were conducted by triplicate determinations and data were subjected to analysis of variance and Tukey's-*b* multiple range test ( $p < 0.05$ ).

## 3. Results and discussion

Chitosan/methyl cellulose film was colorless and transparent while the film containing vanillin (vanillin film) was more opaque and yellow. Film thickness varied from 40 to 50  $\mu$ m. Chitosan/methyl cellulose film was apparently more hydrophilic than vanillin film because it absorbed some water from the fruit wedges, causing it to swell and its surface to become rough. It also adhered to the surface of the fruit and was difficult to remove. Vanillin film adhered to fruit wedges like the other synthetic plastic films. Unlike the chitosan/methyl cellulose film, it was easy to remove from the fruit surface. Mold incidence was visually observed on day 12 in all cantaloupe treatments except with the vanillin film. At the same time, even though there was no mold on fresh-cut pineapple, an off-odor was detected.

### 3.1. Inhibitory effect of antimicrobial film against *E. coli* on fruit at 10 °C

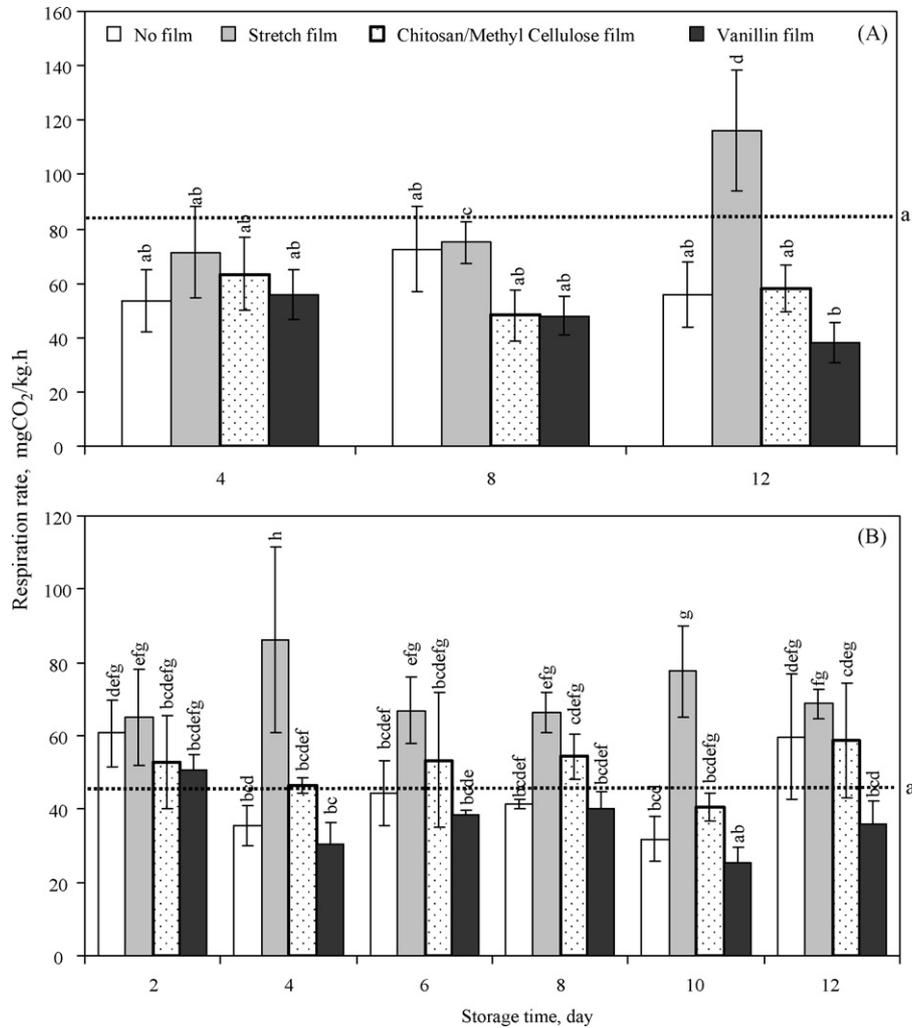
The number of *E. coli* on each cantaloupe piece on day 0 was  $1.5 \times 10^5$  cfu/piece. To disregard the difference in weight loss of each treatment during storage, the microbiological counts were expressed per piece instead of per gram. As storage time increased, the number of *E. coli* on cantaloupe wrapped with stretch film increased, while the populations of *E. coli* on cantaloupe without film, wrapped over with chitosan/methyl cellulose film and vanillin film decreased (Table 1). The reduction of *E. coli* populations might be due to the loss of water content on fruit during storage. The decline rate was faster on fruit wrapped with chitosan/methyl cellulose film during the first 2 d. After that, the populations of *E. coli* on cantaloupe wrapped with chitosan/methyl cellulose film and vanillin film were not different. After 4 d storage, cantaloupe in chitosan/methyl cellulose and vanillin films had a significantly lower number of *E. coli* than fruit without wrapping.

The initial number of *E. coli* on pineapple pieces was equivalent to that on cantaloupe. However, *E. coli* populations of all treatments decreased over time. This seemed to be the effect of fruit pH which was too low for this microorganism. Presser et al. (1997) reported that *E. coli* grew at pH 4.0 but not at pH 3.7. Pineapple used in this

**Table 3**  
Numbers of *S. cerevisiae* on inoculated pineapple (log cfu/piece) during storage at 10 °C

	No film	Stretch film	Chitosan/methyl cellulose film	Vanillin film
Day 0	5.08 ± 0.00 <sup>efgh</sup>	5.08 ± 0.00 <sup>efgh</sup>	5.08 ± 0.00 <sup>efgh</sup>	5.08 ± 0.00 <sup>efgh</sup>
Day 1	4.73 ± 0.03 <sup>ef</sup>	4.93 ± 0.17 <sup>efg</sup>	2.93 ± 0.99 <sup>bc</sup>	4.89 ± 0.15 <sup>efg</sup>
Day 2	5.30 ± 0.14 <sup>fgh</sup>	5.11 ± 0.18 <sup>efgh</sup>	3.00 ± 0.00 <sup>bc</sup>	4.27 ± 0.21 <sup>de</sup>
Day 4	5.53 ± 0.17 <sup>fghi</sup>	5.98 ± 0.31 <sup>hi</sup>	3.15 ± 0.12 <sup>bc</sup>	3.20 ± 0.44 <sup>bc</sup>
Day 6	6.36 ± 0.33 <sup>ij</sup>	7.06 ± 0.21 <sup>jk</sup>	3.56 ± 0.21 <sup>cd</sup>	1.00 ± 0.00 <sup>a</sup>
Day 8	5.82 ± 0.49 <sup>ghi</sup>	7.36 ± 0.05 <sup>k</sup>	3.08 ± 0.16 <sup>bc</sup>	1.43 ± 0.75 <sup>a</sup>
Day 10	5.39 ± 0.31 <sup>fghi</sup>	7.33 ± 0.36 <sup>k</sup>	3.10 ± 0.61 <sup>bc</sup>	1.00 ± 0.00 <sup>a</sup>
Day 12	5.82 ± 0.35 <sup>ghi</sup>	7.96 ± 0.19 <sup>k</sup>	2.52 ± 0.50 <sup>b</sup>	1.23 ± 0.40 <sup>a</sup>

Means with different letters are significantly different at  $p = 0.05$ .



**Fig. 3.** Respiration rate of fresh-cut cantaloupe (A) and pineapple (B) during storage at 10°C. Dash line represents respiration rate on day 0. Bars with the same letters are not significantly different according to the Tukey's-*b* test ( $p > 0.05$ ).

study had pH values of 3.3–3.8. Therefore, the inhibitory effect of the antimicrobial films studied was not relevant (data not shown).

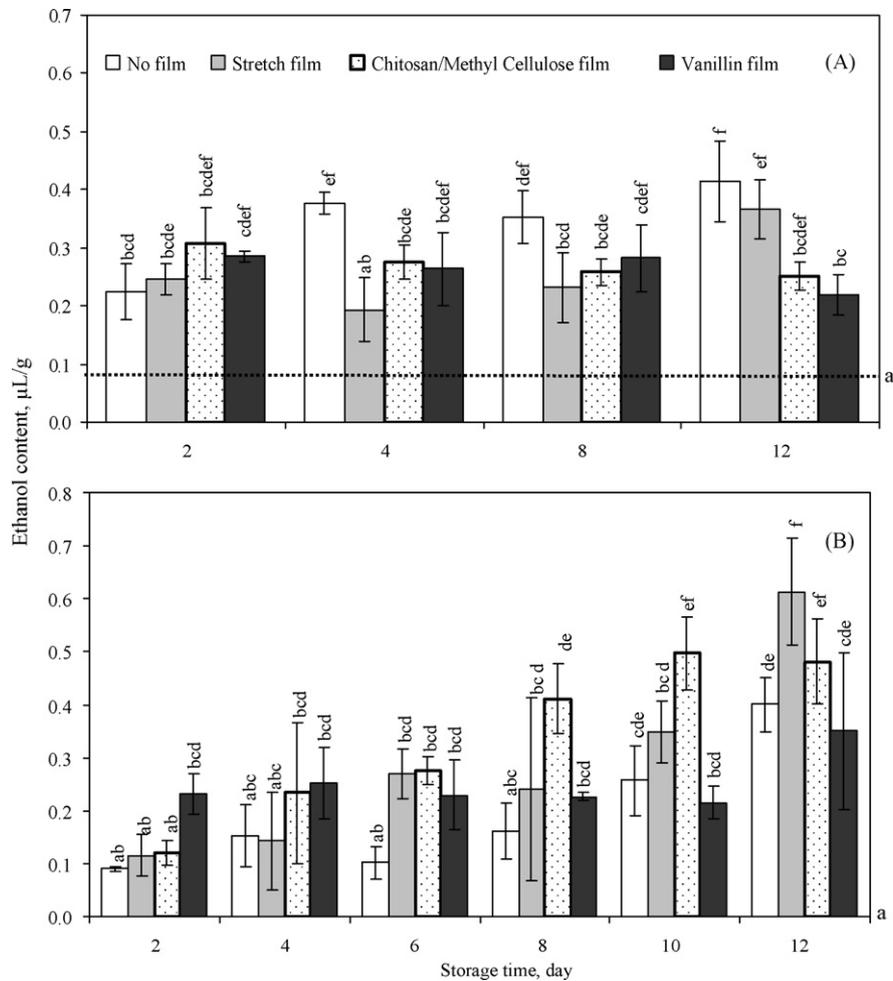
### 3.2. Inhibitory effect of antimicrobial film against *S. cerevisiae* on fruit at 10°C

The initial numbers of yeast on cantaloupe and pineapple pieces after inoculation with *S. cerevisiae* were  $2.3 \times 10^5$  and  $1.2 \times 10^5$  cfu/piece, respectively. After 2 d, the numbers began to increase on the control cantaloupe and cantaloupe wrapped with commercial stretch film (Table 2). The increase in the latter film was faster because it maintained higher moisture contents and lower oxygen concentrations. On the other hand, the numbers of yeast on cantaloupe in the vanillin film remained constant over time, while those wrapped with chitosan/methyl cellulose film decreased over the first 4 d and then increased afterwards. After that, chitosan/methyl cellulose and vanillin films provided the same inhibition. Similar to cantaloupe, the numbers of yeast on pineapple in stretch film increased (Table 3). The yeast populations on pineapple wrapped with chitosan/methyl cellulose film decreased almost 2 logs cfu/piece on the first day and remained constant afterwards. Film containing vanillin resulted in a decrease of 4 logs more than the other films. Vanillin film was more effective but took a longer time to show the effect than chitosan/methyl cellulose film. Vanillin

film may be useful for food with longer storage life. Application of vanillin film on cantaloupe and pineapple showed a different behavior. Vanillin film maintained the amount of yeast or inhibited cell division of yeast on cantaloupe while it decreased the number of yeast on pineapple. These results agreed with Lopez-malo et al. (1998) and Matamoros-Leon et al. (1999) who reported that vanillin was more effective in inhibiting microorganisms in lower pH foods. Visual observation showed that the yellowness of vanillin film used to wrap pineapple decreased. Therefore, the greater inhibition might be the result of a higher release rate of vanillin out of the film.

### 3.3. Flesh color

Color of fresh-cut cantaloupe in all treatments remained unchanged over the storage period. Hue angle of fresh cantaloupe was 67.13. However, the hue angle of pineapple flesh wrapped with stretch film tended to decrease over time while that of tissue wrapped with vanillin film increased significantly from 92.3 to 97.9 (Fig. 1). This may be the result of the yellow vanillin completely migrating from the film. On the contrary, the color of vanillin film was still yellow after removed from the cantaloupe flesh. The sensory changes could possibly be expected as a result of vanillin migration from film to fruit surface. However, sensory analysis by taste panel was not performed in this study.



**Fig. 4.** Ethanol content of fresh-cut cantaloupe (A) and pineapple (B) during storage at 10°C. Dash line represents ethanol content on day 0. Bars with the same letters are not significantly different according to the Tukey's-*b* test ( $p > 0.05$ ).

### 3.4. Firmness

Firmness of cantaloupe flesh in all treatments remained unchanged over time. Even though the force measured did not show any difference, it is possible that the mouth-feel of the fruit is completely different due to water loss. None of the film had an effect on pineapple firmness, which decreased by approximately 20% over the first 2 d and was almost unchanged after that (data not shown).

### 3.5. L-Ascorbic acid (AA)

Initial AA content of fresh-cut cantaloupe was 17.6 mg/100 g fresh weight or 281.3 mg/100 g dry weight. The values were in the range of 14–19.8 mg/100 g fresh weight published by Saftner et al. (2006). AA content decreased over the first 2 d (Fig. 2A). The reduction in AA content at the end of storage was about half of the initial concentration.

Initial AA content of fresh-cut pineapple was only 8.6 mg/100 g fresh weight or 54.3 mg/100 g dry weight (Fig. 2B). This seemed to be low compared with that found by Vinci et al. (1995) who reported that the AA contents of fresh and artificially ripened pineapple were 30.6 and 18.1 mg/100 g fresh weight. The low AA content might be due to a higher storage temperature at the market before purchasing. In a preliminary study, it was found that AA rapidly degraded in pineapple samples. Therefore, the sample

must be carefully prepared and immediately injected into the HPLC system. From Fig. 2B, the AA content in pineapple wrapped with chitosan/methyl cellulose film, stretch film and no film slightly decreased during storage. Unexpectedly, the AA content in pineapple wrapped with vanillin film diminished drastically. Only 10% of the initial concentration was left in the fruit after 12 d storage. The reaction of vanillin and AA should be investigated. Any hydrogen exchange or bonding could be considered. However, Burri et al. (1989) reported that vanillin also acts as an antioxidant.

### 3.6. Respiration rate

The respiration rate of fresh-cut cantaloupe was unchanged during storage except in cantaloupe wrapped with stretch film on day 12, where a spike in respiration was observed (Fig. 3A). Similarly, the respiration rate of fresh-cut pineapple wrapped with stretch film was generally higher than that for other treatments, while that wrapped with vanillin film was the lowest (Fig. 3B). In general, chitosan/methyl cellulose and vanillin films were better gas barriers than the stretch film in dry conditions (Sangsuwan et al., unpublished data), but the barrier properties decline with moisture absorption. Thus, the gas protection of these films is limited. Too high a gas barrier in stretch film might result in depletion of oxygen, resulting in fermentation of the product. Therefore, films should have an appropriate oxygen permeability which is very important

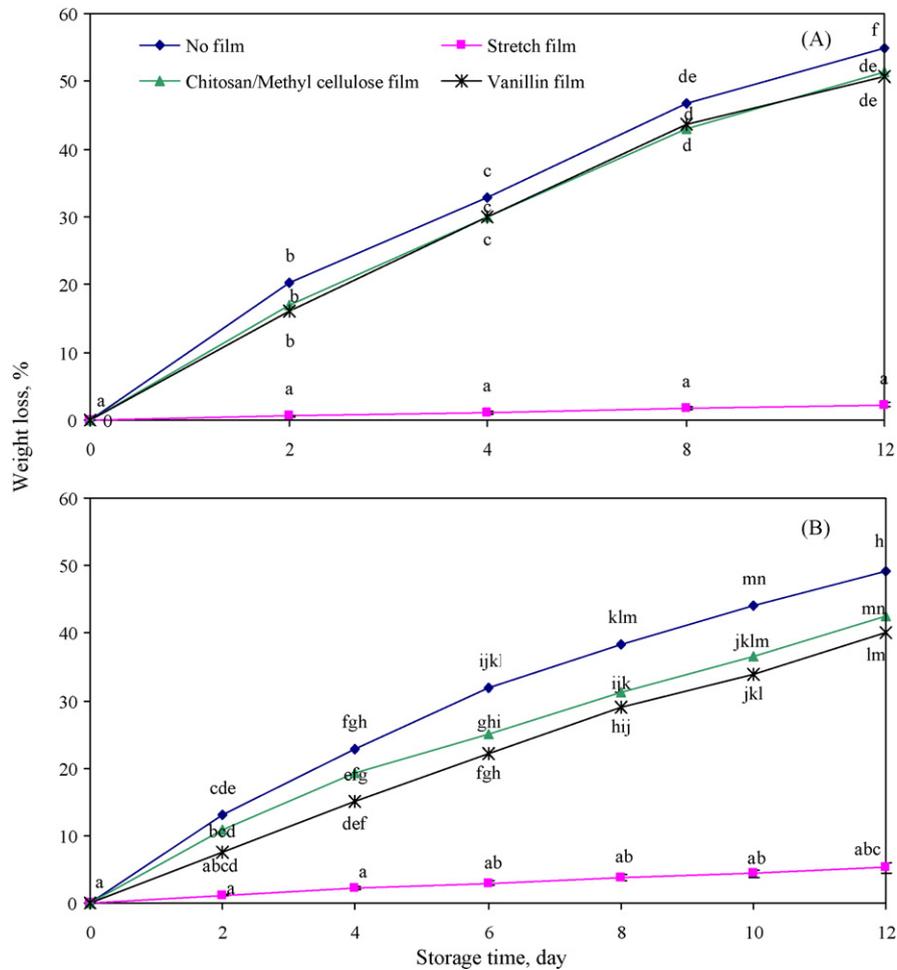


Fig. 5. Weight loss of fresh-cut cantaloupe (A) and pineapple (B) during storage at 10 °C.

for respiring products. The burst of CO<sub>2</sub> production in fruit wrapped with stretch film could also be the result of microbial activity, since these fruit had higher microbial counts.

### 3.7. Ethanol content

The initial ethanol content of fresh-cut cantaloupe, immediately after processing, was 0.08 μL/g. Ethanol contents increased in all treatments and varied from 0.19 to 0.41 μL/g afterwards (Fig. 4A). In pineapple, ethanol increased over time, indicating fermentative metabolism associated with senescence (Fig. 4B). Ethanol content of pineapple wrapped with vanillin film increased on day 2 and remained unchanged until day 12. The final content was slightly lower than for other treatments.

### 3.8. Total soluble solids and pH

Initial TSS of cantaloupe and pineapple was 7.8 and 18.0%, respectively. TSS in all treatments, except in the stretch film, increased with time because the fruit lost approximately 50% water content. Therefore, fruit in the stretch film maintained the best TSS. The pHs of cantaloupe and pineapple flesh were in the range of 5.3–6.1 and 3.3–3.8, respectively (data not shown).

### 3.9. Weight loss

Weight loss of both cantaloupe and pineapple in the commercial stretch film was significantly lower than that in fruit

tissue wrapped with vanillin or chitosan/methyl cellulose film, or without film (Fig. 5). Pineapple without any wrapping lost the most moisture (Fig. 5B). Stretch film provided a better barrier to water than vanillin and chitosan/methyl cellulose film. Water vapor permeabilities at 23 °C, 53% RH of vanillin film and chitosan/methyl cellulose film were 1.03 and 1.09 ng cm/cm<sup>2</sup> s cmHg, respectively (Sangsuwan et al., unpublished data). The water barrier of a biopolymer was impaired in a higher relative humidity environment or with higher moisture content of the food (Garcia et al., 2004). A poor water vapor barrier property allows the movement of water vapor across the film, thus, preventing water condensation that can be a potential source of microbial spoilage in horticultural commodities (Park et al., 1994). With respect to synthetic polymers, chitosan/methyl cellulose films had WVP values similar to those of cellophane, as expected due to the similar chemical structure of the components. However, chitosan/methyl cellulose films are better water vapor barriers than hydrophilic films based on starch, casein and wheat gluten (Greener and Fennema, 1989; Kester and Fennema, 1989; Aydt et al., 1991; Gontard and Guilbert, 1994).

## 4. Conclusion

Chitosan/methyl cellulose and vanillin films provided an inhibitory effect against *E. coli* bacteria and *S. cerevisiae* yeast. Vanillin film reduced microorganisms levels (higher log reduction) to a greater extent, but over a longer time. Use of vanillin film on cantaloupe and pineapple showed different responses.

In a low pH fruit, vanillin was more effective at inhibiting microorganisms. Quality attributes of fresh-cut cantaloupe and pineapple were generally acceptable. An extreme reduction of L-ascorbic acid, which represents vitamin C, of pineapple wrapped in vanillin film, was observed, and would be worth further investigation.

## Acknowledgements

Financial support from the Thailand Research Fund through the Royal Golden Jubilee Ph.D. Program (Grant number PHD/0187/2004) and Postharvest Technology Institute, Chiang Mai University are acknowledged. The authors would like to thank Dr. Anne Plotto (Citrus and subtropical products laboratory, ARS, USDA, Winter Haven, FL.) and Prof. Dr. Bruce Harte (School of Packaging, Michigan State University) for reviewing the manuscript.

## References

- Aydt, T.P., Weller, C.L., Testin, R.F., 1991. Mechanical and barrier properties of edible corn and wheat protein films. *Trans. ASAE* 34, 207–211.
- Brody, A.L., Strupinsky, E.R., Kline, L.R., 2001. *Active Packaging for Food Applications*. Technomic Publishing, Inc., Lancaster, p. 218.
- Burri, J., Graf, M., Lambelet, P., Loliger, J., 1989. Vanillin: more than a flavouring agent—a potent antioxidant. *J. Food Sci. Agric.* 48, 49–56.
- Caner, C., Vergano, P., Wiles, J., 1998. Chitosan film mechanical and permeation properties as affected by acid, plasticizer, and storage. *J. Food Sci.* 63, 1049–1053.
- Cerrutti, P., Alzamora, S.M., 1996. Inhibitory effects of vanillin on some food spoilage yeasts in laboratory media and fruit purees. *Int. J. Food Microbiol.* 29, 379–386.
- Cerrutti, P., Alzamora, S.M., Vidales, S.L., 1997. Vanillin as an antimicrobial for producing shelf-stable strawberry puree. *J. Food Sci.* 62, 608–610.
- Cha, D.S., Chinnan, M.S., 2004. Biopolymer-based antimicrobial packaging: a review. *Crit. Rev. Food Sci.* 44, 223–237.
- Chen, M.C., Yeh, G.H.C., Chiang, B.H., 1996. Antimicrobial and physicochemical properties of methyl cellulose and chitosan films containing a preservative. *J. Food Process. Preserv.* 20, 279–390.
- Davis, P.L., Chace, W.G., 1969. Determination of alcohol in citrus juice by gas chromatographic analysis of headspace. *HortScience* 4, 117–119.
- Devlieghere, F., Vermeiren, L., Debevere, J., 2004. Chitosan: antimicrobial activity, interactions with food components and applicability as a coating on fruit and vegetables. *Food Microbiol.* 21, 703–714.
- Fitzgerald, D.J., Stratford, M., Gasson, M.J., Ueckert, J., Bos, A., Narbad, A., 2004. Mode of antimicrobial action of vanillin against *Escherichia coli*, *Lactobacillus plantarum* and *Listeria innocua*. *J. Appl. Microbiol.* 97, 104–113.
- Garcia, M.A., Pinottia, A., Martino, M.N., Zaritzky, N.E., 2004. Characterization of composite hydrocolloid films. *Carbohydr. Polym.* 56, 339–345.
- Goldberg, S., Doyle, R., Rosenberg, M., 1990. Mechanism of enhancement of microbial cell hydrophobicity by cationic polymers. *J. Bacteriol.* 172, 5650–5654.
- Gontard, N., Guilbert, S., 1994. Bio-packaging: technology and properties of edible and/or biodegradable material of agricultural origin. In: Mathlouthi, M. (Ed.), *Food Packaging and Preservation*. Elsevier, New York, pp. 159–179.
- Greener, I.K., Fennema, O., 1989. Evaluation of edible, bilayer films for use as moisture barriers for food. *J. Food Sci.* 54, 1400–1406.
- Jay, J.M., Rivers, G.M., 1984. Antimicrobial activity of some food flavoring compounds. *J. Food Safety* 6, 129–139.
- Kester, J.J., Fennema, O., 1989. An edible of lipids and cellulose ethers: barrier properties to moisture vapor transmission and structural evaluation. *J. Food Sci.* 54, 1383–1389.
- Lopez-malo, A., Alzamora, S.M., Argai, A., 1998. Vanillin and pH synergistic effects on mold growth. *J. Food Sci.* 63, 143–146.
- Matamoros-Leon, B., Argai, A., Lopez-Malo, A., 1999. Individual and combined effects of vanillin and potassium sorbate on *Penicillium digitatum*, *Penicillium glabrum*, and *Penicillium italicum* growth. *J. Food Prot.* 62, 540–542.
- Moon, K.D., Delaquis, P., Toivonen, P., Stanich, K., 2006. Effect of vanillin on the fate of *Listeria monocytogenes* and *Escherichia coli* O157:H7 in a model apple juice medium and in apple juice. *Food Microbiol.* 23, 169–174.
- Park, H.J., Chinnan, M.S., Shewfelt, R.L., 1994. Edible coating effects on storage life and quality of tomatoes. *J. Food Sci.* 59, 568–570.
- Penney, V., Henderson, G., Blumb, C., Johnson-Green, P., 2004. The potential of phytopreservatives and nisin to control microbial spoilage of minimally processed fruit yogurts. *Innov. Food Sci. Emerg.* 5, 69–75.
- Pranoto, Y., Rakshit, S.K., Salokhe, V.M., 2005. Enhancing antimicrobial activity if chitosan films by incorporating garlic oil, potassium sorbate and nisin. *Food Sci. Technol.—LEB* 38, 859–865.
- Presser, K.A., Ratkowsky, D.A., Ross, T., 1997. Modelling the growth rate of *Escherichia coli* as a function of pH and lactic acid concentration. *Appl. Environ. Microbiol.* 63, 2355–2360.
- Prindle, R.F., Wright, E.S., 1977. Phenolic compounds. In: Block, S.S. (Ed.), *Disinfection, Sterilization and Preservation*. Lea & Febiger, Philadelphia, p. 1049.
- Romeu-Nadal, M., Morera-Pons, S., Castellote, A.I., Lopez-Sabater, M.C., 2006. Rapid high-performance liquid chromatographic method for vitamin C determination in human milk versus an enzymatic method. *J. Chromatogr. B* 830, 41–46.
- Rupasinghe, H.P.V., Boulter-Bitzer, J., Ahn, T., Odumeru, J.A., 2006. Vanillin inhibits pathogenic and spoilage micro-organisms *in vitro* and aerobic microbial growth in fresh-cut apples. *Food Res. Int.* 39, 575–580.
- Saftner, R., Abbott, J.A., Lester, G., Vinyard, B., 2006. Sensory and analytical comparison of orange-fleshed honeydew to cantaloupe and green-fleshed honeydew for fresh-cut chunks. *Postharvest Biol. Technol.* 42, 150–160.
- Siragusa, G.R., Dickson, J.S., 1992. Inhibition of *Listeria monocytogenes* on beef tissue by application of organic acids immobilized in a calcium alginate gel. *J. Food Sci.* 57, 293–296.
- Torres, J.A., Motoki, M., Karel, M., 1985. Microbial stabilization of intermediate moisture food surfaces. Part I. Control of surface preservative concentration. *J. Food Process. Pres.* 9, 75–92.
- Vinci, G., Botre, F., Mele, G., 1995. Ascorbic acid in exotic fruit: a liquid chromatographic investigation. *Food Chem.* 53, 211–214.
- Zhang, L., Lu, Z., Yu, Z., Gao, X., 2005. Preservation of fresh-cut cereals by treatment of ozonated water. *Food Control* 16, 279–283.
- Zivanovic, S., Chi, S., Draughon, A.F., 2005. Antimicrobial activity of chitosan films enriched with essential oils. *J. Food Sci.* 70, M45–M51.