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Utilization possibilities of antimicrobial biodegradable packaging produced by poly(butylene succinate) modified with zinc oxide nanoparticles in fresh-cut apple slices

Phisut Naknaen

Division of Food Science and Nutrition, Faculty of Agricultural Product Innovation and Technology, Srinakharinwirot University, Sukhumvit 23 Wattana, Bangkok 10110, Thailand

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Abstract

A novel nanocomposite-based packaging was prepared by blending poly(butylene succinate) (PBS) with various concentrations (0, 2, 4 and 6%, w/w) of nano-ZnO and was applied to prolong the shelf life of fresh-cut apples during cold storage. Results showed that, continuous increase in a^* values and browning index and decrease in L^* values, indicating that a trend of increase in brownness, were found during storage in all treatments. However, slight difference in browning parameters was observed among the treatments packed in different packages. Continuous decrease in firmness and increase in weight loss were also detected in all samples during storage. The decrease of sucrose was observed in all treatments, while fructose and glucose tended to increase within the first 6 days of storage and then decreased till the end of storage. During storage, the samples packed in the PBS/ZnO_0 and the PBS/ZnO_2 contained higher vitamin C and phenolic contents than those of the samples packed in the PBS/ZnO_4 and the PBS/ZnO_6. According to microbiological standard, the shelf life of samples was approximately 9 and 12 days for the samples packed in the PBS/ZnO_0 and the PBS/ZnO_2, respectively, while the samples packed the PBS/ZnO_4 and the PBS/ZnO_6 were assured for at least 18 days of storage. Thus, the incorporation of nano-ZnO (4%) in the PBS packaging could be used as an alternative way to extend shelf life of fresh-cut apples.

Introduction

Fresh-cut products are defined as fruits or vegetables that have been trimmed and/or peeled and/or cut in to 100% usable product and are bagged or pre-packaged while the freshness still maintains. The demand for fresh-cut fruits has increased due to health consciousness of consumers. Food marketplace evolves new products and changing trends, and fresh-cut products remain at the top of the list of products meeting the needs of busy consumers. The value of fresh-cut products lies on the primary characteristics of freshness and convenience (Supapvanich *et al*., 2011; Siddiq *et al.*, 2013). Fresh-cut apples which contain antioxidants and other nutrient components, have recently emerged as popular snacks in food service establishments, school lunch program, and for family consumption. Deterioration of the fruit after minimal processing from wound-induced biochemical and physiological changes associated with loss of mass, firmness, nutrition components and cut surface browning (Chiabrando and Giacalone, 2012). For these reasons, the preservative techniques, such as modified atmosphere (MA) packaging, storage at low temperature and chemical treatments, usually were applied to extend the shelf life of fresh-cut fruit (Chung and Moon, 2009). Currently, the application of antimicrobial packaging is another method to extend the shelf life of food. Antimicrobial packaging can be considered an emerging technology that could have a significant impact on shelf life extension and food safety. Use of antimicrobial agents in food packaging can control the microbial population and target specific microorganisms to provide higher safety and quality products. Many classes of antimicrobial compounds have been applied to polymer matrix for producing an antimicrobial packaging. As one of multifunctional inorganic nanoparticles, zinc oxide (ZnO) nanoparticles are known to inhibit microbial growth. The ZnO nanoparticles have selective toxicity to bacteria; at the same time, they have negligible negative effect on human cells. Because of their antimicrobial properties, ZnO nanoparticles have been increasingly applied in the food industry. Nano-ZnO has been listed as generally recognized as safe by the U.S. Food and Drug Administration (Ding *et al*., 2012; Espitia *et al.*, 2012). Emamifar *et al.* (2012) studied the use of nanocomposite low-density polyethylene (LDPE) films containing Ag and ZnO to extend the shelf life of orange juice. In addition, there were numerous studies related to the application of nano-composites LDPE films

*Corresponding author.
Email: *phisutx@gmail.com*
Tel: 66 2649 5000 ext. 18304; Fax : 66 2649 5000 ext. 15812

containing nano-ZnO and silver nanoparticles on the preservation of fresh-cut lotus root (Ding *et al*., 2012), kiwi (Hu *et al*., 2011) and fresh-cut apple (Zhou *et al*., 2012). However, LDPE is non-degradable. This plastic leads to the environmental problem such as global warning. Poly(butylene succinate) (PBS) is one of the most promising biodegradable aliphatic polyesters, with desirable properties such as biodegradability, melt processability, and both thermal and chemical resistance. In addition, PBS is a biodegradable material and can be used in food packaging. Some studies indicate that this material is equally suitable for such applications compared to the widely used low density polyethylene (Phua *et al.*, 2011). There are no publications currently reporting the application of PBS (containing ZnO nanoparticles) as an antimicrobial packaging in fresh fruits and vegetables. Therefore, the objective of this study is to develop nano-ZnO PBS film and evaluate the capabilities of nano-ZnO PBS film as an antimicrobial packaging to preserve and prolong the shelf life of fresh-cut apple.

Materials and Methods

Plastic film bag

The poly(butylenes succinate) (PBS) was obtained from Mitsubish Chemical Japan. The nano-ZnO (10-30 nm) was purchased from US research nanomaterial Inc, USA. The PBS/nano-ZnO composites with various concentration of nano-ZnO (0, 2, 4, 6%, w/w) were produced by co-rotating twin screw extruder (Lab Tech, Thailand) and single screw extruder film blown machine (Collin, Blown film line BL, Germany). Each plastic bag was determined for its oxygen permeation and water vapour permeation according to ASTM D3985 and ASTM F1249, respectively. Table 1 presents the properties of nano-ZnO PBS films.

Preparation of apple slices

Apples (Fuji) were obtained from a fresh produce detail market called Talad Thai. The fruit samples were delivered to Faculty of Agricultural Product Innovation and Technology, Srinakharinwirot University within 2 h. The fruit to be used for each experiment was selected on the basis of uniform size and colour, being free from diseases and physical damage. Each fruit was cleaned with tap water and then dipped in 100 µl/l sodium hypochloride for 5 min and then rinsed with cold boiled water. The fruit then air-dried at ambient temperature for 10 min. After that, the fruit samples were cored and sliced into 8 pieces with a stainless steel knife. To retard browning and maintain the texture of fruit, all apple slices were dipped in the solution containing 0.5% citric acid and 0.5% calcium lactate for 2 min and left for approximately 2 min until the excess of water was drained. Then, the apple slices were then packed into the PBS plastic bags containing different contents of nano-ZnO. The packages were completely sealed and stored at 10°C. They were labeled PBS/ZnO_0, PBS/ZnO_2, PBS/ZnO_4, and PBS/ZnO_6 according to the amount of nano-ZnO incorporated to the plastic bag 0, 2, 4 and 6%, respectively. All quality parameters were determined at three day intervals.

Measurement of colour

Colour values of the cut apple surface were directly measured with a colourimeter (Hunter Lab, USA).The results were expressed as CIELAB (L^* and a^*) colour space. L^* and a^* defines the lightness and red-greenness, respectively.

Measurement of browning index

Browning index was measured by colourimetric method as suggested by Saxena *et al.* (2009) with some modifications. Apple flesh (5 g) was homogenized in 50 ml of ethanol (95%), and the homogenate was centrifuged at 4000 rpm for 20 min. After that, the supernatant was collected to measure the absorbance at 420 nm using a spectrophotometer (Shimadzu, Japan) to assess browning index.

Measurement of weight loss

The weight loss was determined by calculating the difference between the initial and final weights of the apple slices outside storage bag. The value was expressed as a relative percentage and calculated as follows:

$$\text{Weight loss (\%)} = [(W_i - W_t) / W_i] \times 100$$

where W_i is the initial weight and W_t is the weight measured during storage (Song *et al*., 2013).

Measurement of firmness

The firmness was determined using a texture analyzer (TA-XT2, Stable Microsystem Ltd, Godalming, U.K.) as described by Song *et al.* (2013) with some modifications. A 5 mm diameter probes was used to penetrate apple slices (10×10×10 mm) to a depth of 5 mm. The samples were tested for the probe to penetrate a geometric center at a speed of 5.0 mm/s. Firmness was expressed in the unit of Newtons (N).

Determination of total soluble solid and total acidity

Total soluble solid was measured using a hand

refractometer (ATAGO, Japan). Juice was squeezed from the fresh-cut apple. The result was expressed as °Brix. A 5 ml of the apple juice prepared by direct homogenization was titrated with 0.1 N NaOH using phenophthalene as indicator. The end point of the reaction was pH 8.0. Total acidity of the juice was defined as % citric acid (Hu *et al*., 2011).

Determination of type and concentration of sugar

Type and concentration of sugars were determined by HPLC (Agilent 110 Series) with Zorbax Carbohydrate column, refractive index detector and 80% acetonitrile was used as a mobile phase. The type and concentration were quantified by comparing retention time and peak area of the samples with known standards.

Determination of ascorbic acid (vitamin C) content

Vitamin C was determined according to the method of Guimaraes *et al*. (2011). The sample (500 mg) was extracted with metaphosphoric acid (1%, 30 ml) for 45 min at room temperature and filtered through Whatman No. 4 filter paper. The filtrate (1 ml) was mixed with 2,6-dichloroindophenol (9 ml) and the absorbance was measured within 30 min at 515 nm against a blank. Content of vitamin C was calculated on the basis of the calibration curve of L-ascorbic acid, and the results were expressed as mg vitamin C per 100 grams sample.

Determination of phenolic content

Quantification of phenolic compounds in each sample was carried out according to the method of Balange and Benjakul (2009). The sample (10 g) was homogenised in 30 ml of distilled water and then made up to 50 ml. The homogenate was filtered and centrifuged at 5000 rpm for 10 min. The supernatant was used to measure phenolic content. The sample (0.5 ml) was mixed with 0.5 ml of distilled water. Thereafter, 0.5 ml of Folin-Ciocalteu reagent (1:1 with water) and 2.5 ml of 2% sodium carbonate solution were added. The mixture was mixed thoroughly and placed in dark for 40 min and the absorbance was recorded at 725 nm. The total phenolic content was calculated from the standard curve of gallic acid and expressed as mg gallic acid per 100 grams sample after blank substraction.

Microbiological analysis

The samples were homogenized using a Stomacher (MIX 2, AES Laboratoire, Combourg, France) for 3 min, filtered through a sterile cheesecloth, and diluted with peptone water to obtain the microbial count. Serial dilutions were performed in triplicate. The total aerobic bacteria counts were determined by plating appropriately diluted samples onto plate count agar (PCA, Difco Co., Detroit, Mich., U.S.A.). They were incubated at 37°C for 48 h. Each microbial count was acquired from the mean of 3 determinations and was expressed as log CFU/g (Kiss, 1984).

Statistical analysis

The experimental design used was a completely randomized design (CRD). The data were obtained from two batches. Each batch was measured all mentioned qualities in triplicate. Statistical analysis was run using one-way analysis of variance. The treatment means was compared using Duncan's new multiple range test (DMRT), with a level of significance of 0.05.

Results and Discussion

Colour and browning index

Colour and appearance are critical quality aspects for shopper when selecting fresh fruits. Discolouration usually occurs on the cut-surface of apple slices during storage, which degrades sensory properties and nutritional values and discourages the consumer purchase of products. Enzymatic browning is a major problem leading to colour change of apple slices due to the reaction of phenolic compounds with oxygen, catalysed by endogenous polyphenol oxidase (PPO). In addition, microbial spoilage has unfavorable colour effects, because it may result in the formation and accumulation of pigment on the cut-surface of apple slices (Supapvanich *et al.*, 2012; Song *et al*., 2013). According to the results, all apple slices showed a trend of increase in darkness, as L^* values decreased and a^* values increased over time (Figure 1a-1b). Type of packaging influenced L^* and a^* values of apple slices ($P \leq 0.05$). The samples packed in the packaging containing higher nano-ZnO content (4-6%) presented higher increase in a^* values and decrease in L^* values than those of the samples packed in the PBS/ZnO0 or the PBS/ZnO_2. This result could be explained by the addition of nano-ZnO caused an increase in the oxygen permeability of packaging as shown in Table 1. There were no significant differences in L^* and a^* values between the samples packed in the PBS/ZnO0 and the PBS/ZnO_2 ($P > 0.05$). In addition, no significant differences were also observed in the samples packed in either the PBS/ZnO_4 or the PBS/ZnO_6 ($P > 0.05$). The increase in a^* values and decrease in L^* values with increasing storage times were probably due to enzymatic browning reaction taking place. The PPO is responsible for this reaction. This enzyme catalyses

Table 1. Oxygen permeation and water vapor permeation of the nano-ZnO PBS film

Treatment	Oxygen permeation ($gm.mil/m^2.day.atm$)	Water vapor permeation ($cc.mil/m^2.day.atm$)
PBS/ZnO_0 %	189.34	147.96
PBS/ZnO_2 %	245.02	159.45
PBS/ZnO_4 %	247.68	183.65
PBS/ZnO_6 %	249.80	189.71

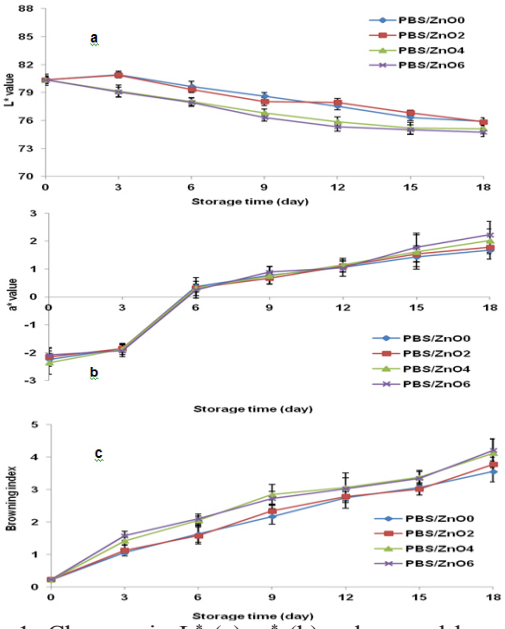

Figure 1. Changes in L^* (a), a^* (b) values and browning index (c) of apple slices packed in the PBS packages containing different contents of nano-ZnO during storage.

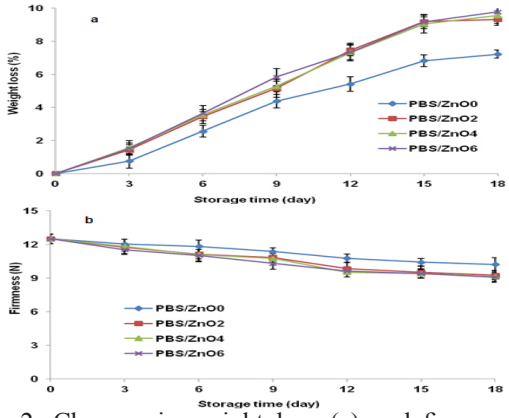

Figure 2. Changes in weight loss (a) and firmness (b) of apple slices packed in the PBS packages containing different contents of nano-ZnO during storage.

the hydroxylation of monophenols to o-diphenols and oxidation of o-diphenols to o-quinones. Quinones are very reactive compounds which strongly interact with other molecules, leading to a large pigment of high molecular weight with very red to brown colour. The enzymatic browning was normally promoted by high level of oxygen contacted of the surface of fruit (Chung and Moon, 2009). Lower oxygen permeability as detected in the PBS/ZnO_0 and the PBS/ZnO_2 could minimize the browning formation of apple slices when compared to those of the samples packed in the PBS/ZnO_4 and the PBS/ZnO_6. The colour of apple slices as evaluated in term of L^* and a^* values was correlated well with the browning index (Fig. 1c). An increase in the browning index of all samples was also found during storage. Higher browning index was also detected in the samples packed in the PBS/ZnO_4 and the PBS/ZnO_6 when compared to the samples packed in the PBS/ZnO_0 and the PBS/ZnO_2 ($P \leq 0.05$). However, small differences in the L^*, a^* values and browning index were found among the samples packed in various packages during storage.

Weight loss

The surface of the apple slices is exposed to the environment, which results in weight loss. Weight loss is mostly due to the evaporation of water facilitated by a water vapour pressure gradient. In addition, weight loss is a physiological event in fresh cut fruits that can be limited by using appropriate packaging and storage temperature. Thus, the moisture loss of packaged apple slices may be mainly attributed to two reasons. In the first case, moisture migration leads to water loss and, secondly, the respiration of living tissue consumes some nutrients (Zhou *et al*., 2011; Saleh *et al*., 2013; Song *et al.*, 2013). Fresh cut apple slices have high water content and reduction of water loss is most required to maintain their freshness. Therefore, the packaging material of apple slices should have proper permeability of respiration gases and water vapour. Figure 2a showed the weight loss of apple slices packed in the PBS/ZnO during storage. Generally, the weight loss of the apple slice samples increased during storage. The lowest weight loss was observed in the sample packed in the PBS/ZnO_0 ($P \leq 0.05$). This result could be due to the PBS/ZnO_0 exhibited the lowest water vapour permeability. In addition, low oxygen permeability of the PBS/ZnO_0 may reduce the product respiration rate and slow down the permeation of water vapour (Lucera *et al*., 2010). However, there was no significant difference in weight loss among the samples packed in the PBS packaging containing nano-ZnO (the $PBS/ZnO_{2\text{-}6}$). As note earlier, there are mainly two factors influencing the weight loss such as respiration rate of fruit and properties of packaging. Zhou *et al.* (2011) suggested that the packaging that behaved as good barrier to water vapour, should maintain the weight loss at low values (<2%). From the result, all packaging used in this study do not possess good barrier to water vapour. Thus, the improvement the water vapour permeability of the PBS/ZnO packaging should be further researched.

Firmness

Fresh-cut fruit is produced by minimal processing such as peeling and trimming. Wounding can induce several physiological changes of fresh-cut fruits

including hastening firmness loss by activities of pectic enzymes. Pectic enzymes have been attributed to play significant roles in the softening process during fruit ripening. Softening of cell wall components by actions of pectic enzyme relates with the progressive solubilization and depolymerization of pectic substances. In addition, water loss or transpiration is another factor that affects the quality of fresh-cut products. The loss of firmness is usually associated to water loss, which is expressed by decreased turgor and crispness (Rivera-Lopez *et al.*, 2005; Guan and Fan, 2010). Firmness is one of the quality attributes in apple slices. A decrease in the firmness was observed in all treatments during storage (Figure 2b). Greater decrease in the firmness was found in the samples packed in the PBS containing nano-ZnO compared to that of the sample packed in the PBS without nano-ZnO incorporated ($P \leq 0.05$). It is well known that pectins are major components of the cell wall and consist of chains of 1–4 linked D-galacturonic acid units which are esterified with methanol. Two major enzymes cause pectin degradation in fruit; pectin methylestrerase (PME) that catalyzes the de-estification of pectin and polygalacturonase (PG) that hydrolyze the glycodic bonds. Thus, the activity of pectinase leads to the loss of firmness. Based on the oxygen permeability of each packaging, high oxygen permeability could enhance the respiratory rate of fruit, leading to the increase in metabolism and the activity of enzyme. It is well known that PG is an ethylene regulated enzyme, thus high oxygen in the system might promote the ethylene production, resulting in the increment in the PG activity (Toivonen and Brummell, 2008; Song *et al*., 2013). In addition, the lowest reduction in the firmness for the sample packed in the PBs/ZnO_0 might be related to the lowest weight loss. However, a small difference in the firmness was observed among samples at the end of storage.

Total acidity, total soluble solid and type and concentration of sugar

Initial total acidity and total soluble solid in fresh-cut apples were 0.42% and 14.20°Brix, respectively. During storage, the reduction of total acidity and total soluble solid was found in all treatments, suggesting that these compounds were used as substrate in the respiration process. Higher decrease in total acidity and total soluble solid was observed in the samples packed in the PBS/ZnO_4 and the PBS/ZnO_6 when compared to those of the samples packed in the PBS/ZnO_0 and the PBS/ZnO_2 ($P \leq 0.05$). At the end of storage, the total acidity was approximately 0.36 (for the samples packed in the PBS/ZnO_0 and the PBS/ZnO_2) and 0.30 (for the samples packed in the PBS/ZnO_4 and the PBS/ZnO_6). In addition, the samples packed in the PBS/ZnO_0 and the PBS/ZnO_2 contained 12.50°Brix while the total soluble solid was 11.20°Brix for the samples packed in the PBS/ZnO_4 and the PBS/ZnO_6 at the 18th days of storage. Change in type and concentration of sugar were significantly ($p < 0.05$) effected by the packaging types. The decrease of sucrose was observed in all treatments during storage, while fructose and glucose tended to increase within the first 6 days of storage and decreased till the end of storage (Figure 3). It was found that the reduction of sugars with storage time was caused by oxidation of substrate in the respiration process. At the beginning of the storage time, fructose and glucose tended to increase due to each sucrose molecule was converted to produce monosaccharide (glucose and fructose) in order to use these sugars as substrate for respiration process. After that, the reduction in the percentage of reducing sugar in fruit is due to the quick consumption of sugars (Soliva-Fortuny *et al*., 2004; Hu *et al*., 2011).

Ascorbic acid content

Vitamin C (ascorbic acid) is one of the most important nutritional attributes in fruit and vegetables and has many biological activities in the human body. It is widely used as an antioxidant for preventing enzymatic browning in fresh cut processing and can be claimed as an important nutritional component of fruits. The physiological stress imposed upon fresh-cut commodities generally results in a significant reduction in vitamin C content (Saxena *et al.*, 2009; Hu *et al*., 2011). Change in vitamin C contents of sliced apples during storage was presented in Figure 4a. During storage, vitamin C content continuously declined in all treatments. The degradation of ascorbic acid associated with wounds occurred during minimally processing of apples. Deterioration of cellular integrity and enzymatic compartmentation results in releasing of inherent oxidative enzymes. Ascorbic acid oxidase (EC 1.10.3.3) is a Cu-containing enzyme that catalyses the oxidation reaction of ascorbic acid to dehydroascorbic acid with the concomitant reduction of molecular oxygen to water. It is thought to be the major enzyme responsible for the enzymatic degradation of ascorbic acid (Nakamura *et al.*, 1968). Manurakchinakorn *et al*. (2004) also found the reduction of vitamin C in fresh-cut mangosteens during storage and they suggested that ascorbic acid oxidase was responsible for the degradation of vitamin C. In addition, Yahia *et al.* (2001) found that a decrease of ascorbic acid in tomato and bell pepper fruits coincided with an

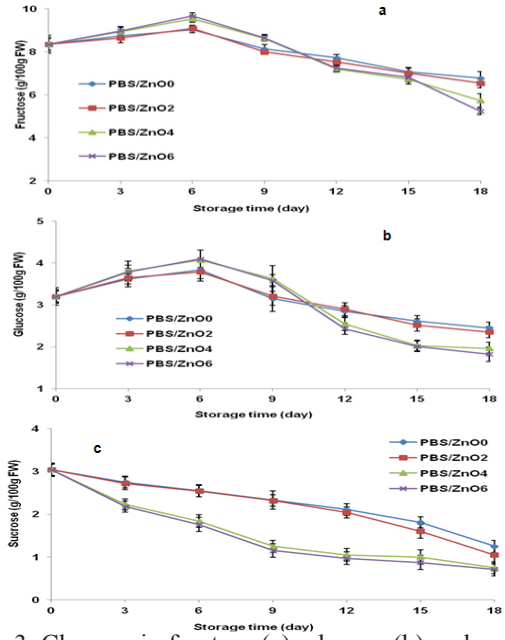

Figure 3. Changes in fructose (a), glucose (b) and sucrose (c) contents of apple slices packed in the PBS packages containing different contents of nano-ZnO during storage.

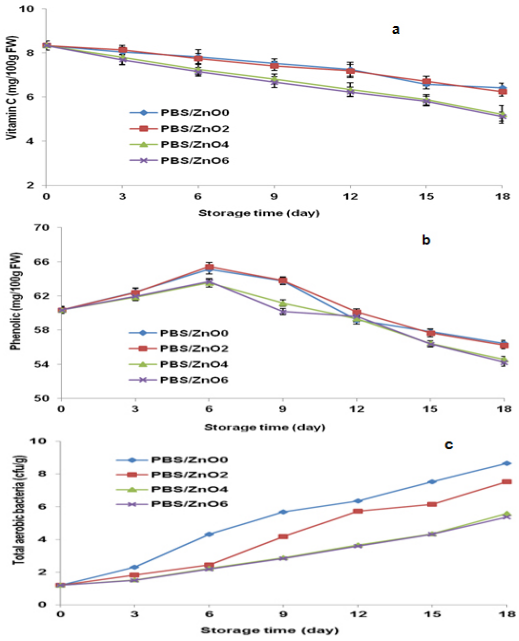

Figure 4. Changes in vitamin C (a), phenolic contents (b) and total aerobic count (c) of apple slices packed in the PBS packages containing different contents of nano-ZnO during storage.

increase in the activity of ascorbic acid oxidase. Furthermore, the lowest vitamin C content was found in the samples packed in the PBS/ZnO_4 and the PBS/ZnO_6 ($P \leq 0.05$). These results related to the degradation of ascorbic acid by non-enzymatic oxidation. High oxygen permeability of packaging resulted in a greater extent of ascorbic acid depletion. It appears that ascorbic acid is most likely converted to dehydroascorbic acid and further degraded to 2,3 diketo-gluconic acid. Thus, low oxygen atmosphere can slow ascorbic acid loss by inhibition its oxidation (Chung and Moon, 2009).

Phenolic content

The phenolic compounds, the substrates in enzymatic browning reactions, have many biological and functional activities for fruit quality and human health. The major phenolics found in apples are chlorogenic acid, catechin and epicatechin, these compounds are closely related with enzymatic browning (Carbone *et al*., 2011). Change in the phenolic content of sliced apple samples stored for 18 days was depicted in Figure 4b. Phenolic compounds of the slices tended to increase during the first 6 days of storage. Subsequently, a decrease in phenolic content was observed until the end of storage. The size reduction process during minimal processing is usually accredited with the accumulation of total phenolics compounds in many fruits and vegetables, where cells rapidly synthesis larger amount of phenolic acids as a defence for wound healing and to provide disease resistance. Thus, the increase in phenolic compounds could be directly associated with wound response. Wounding stimulates the cell adjacent to the injury to produce more phenolics in an attemps to initiate repair process (Saxena *et al*., 2009; Supapvanich *et al*., 2011). However, the reduction in phenolic content may occur during storage of fresh-cut fruits. The decrease in phenolic content upon fresh-cut treatment is due to the oxidation by polyphenoloxidase to give the coloured quinones. During storage, the enzymatic oxidation is continued, and the resulted quinones are polymerised non-enzymatically to give darker pigments, which explain the parallel consumption of phenols with the development of blackness throughout the storage period (Saleh *et al*., 2013). In addition, Saxena *et al.* (2009) and Hu *et al*. (2011) also found the decrease of total phenolic content in fresh-cut jackfruit and kiwifruit during low temperature storage, respectively. At the end of storage, the samples packed in the PBS/ZnO_4 and the PBS/ZnO_6 also exhibited the lowest phenolic content ($P \leq 0.05$). Browning of fresh cut fruit is mostly developed due to enzymatic oxidation of phenols. Thus, low phenolic content was concomitant with the high browning enzymatic activities resulting in higher brownness and lower whiteness of the fresh-cut fruit.

Microbiological analysis

The growth of total aerobic bacteria in the apple slices as affected by various packaging bags was evaluated during storage (Figure 4C). The initial population of total aerobic bacteria in apple slices was 1.20 log cfu/g. During storage, an increase in total aerobic bacteria was detected in all samples. At the end of storage of each sample, the total aerobic

bacteria in the sample packed in the PBS/ZnO0 was 8.65 log cfu/g whereas the population of the bacteria was 7.54, 5.58 and 5.37 log cfuU/g for the samples packed in the PBS/ZnO_2, PBS/ZnO_4 and PBS/ZnO_6, respectively. This result indicated that the use of nano-ZnO incorporated in the packaging could retard the growth of aerobic bacteria in the sample. In addition, the increase in level of nano-ZnO in the packaging caused a decrease in the population of aerobic bacteria in the fresh-cut. However, there was no difference in the total aerobic bacteria between the samples packed in the PBS/ZnO_4 and the PBS/ZnO_6 at the end of storage. As The USA and most European countries have regulations relating to fresh cut produce that limit the counts of aerobic microorganisms to 6 log cfu/g at expiry date (Odriozola-Serrano *et al*., 2008). According to this rule, the shelf life of samples as considered from microbiological standard was approximately 9 and 12 days for the samples packed in the PBS/ZnO_0 and PBS/ZnO_2, respectively, while the samples packed in the PBS/ZnO_4 and PBS/ZnO_6 were assured for at least 18 days of storage. Antimicrobials active packaging based on metal nanocomposites, which are made by incorporating some metal nanoparticles such as ZnO into the polymer films, are a new generation of nano food packaging. The high performance in nanoparticles is due to high surface area/volume ratio, which is the main reason for increasing antimicrobial activity of metal nanoparticles. Nanoparticles of ZnO are being used industrially for several purposes. ZnO has been used in many applications in daily life such as drug delivery, cosmetic and filling in medical, exhibited strong antimicrobial activity on a broad spectrum of microorganisms. Moreover, it is currently listed as a generally recognized as safe (GRAS) by the US Food and Drug Administration as mentioned previously. The antimicrobial activity of the ZnO-nanoparticles may be related to several mechanisms including, induction of oxidative stress because of the generation of reactive oxygen species (ROS), which may cause the degradation of the membrane structure of cell, the release of ions from the surface of nanoparticles that has been reported to lead bacterial death based on binding to cell membrane (Appendini and Hotchkiss, 2002; Jones *et al*., 2008; Ding *et al*., 2012; Espitia *et al*., 2012).

Conclusion

This study presents the preliminary research related to the possible use of the packaging prepared from the composite between the PBS and various concentrations of nano-ZnO as an antimicrobial packaging to extend the shelf life of fresh cut apple. The fresh cut apple packed in the PBS/ZnO_4 was assured for at least 18 days of storage, whereas microbial load was reduced below standard level. However, the samples packed in the PBS films containing higher content of nano-ZnO (4-6%) exhibited higher slightly browning than that of the sample packed in the PBS packaging without nano-ZnO incorporated. Thus, the results suggest that biodegradable nano-PBS package can provide an alternative to chemically replace package. However, the properties of films, such as oxygen and water vapour permeation, should be improved. In addition, the cost of package should be also considered. Further study related to off-flavor development and sensory evaluation during storage should be conducted. In addition, the comparative study between the PBS/ZnO packaging and other commercial packagings should be further investigated.

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