วารสารสมาคมวิศวกรรมเกษตรแห่งประเทศไทย
ปีที่ 19 ฉบับที่ 1 (2556) 38-51
Available online at www.tsae.asia

บทความวิจัย

ISSN 1685-408X

The Production Conditions of Biodegradable Film Containing Pomelo Peel Extract for *Staphylococcus aureus* Inhibition

Sawanit Aichayawanich[1]*

[1]Department of Design and Production Technology of Agricultural industrial Machinery, Faculty of Industrial Technology and Management, King Mongkut's University of Technology North Bangkok Prachinburi Campus, Prachinburi, 25250

*Corresponding author: Tel: +66-37-217-327, Fax: +66-37-217-327, E-mail: sawanit.a@fitm.kmutnb.ac.th

Abstract

This research aimed at studying the production conditions of biodegradable film containing pomelo peel extract for *Staphylococcus aureus* inhibition. The effects of extracting solvents (95% ethanol, dichloromethane, hexane, and ethyl acetate) on yield and *Staphylococcus aureus* inhibiting activity of the pomelo peel extract were evaluated. Then, the influences of film process conditions; including drying temperatures (40, 45, 50, 55, and 60°C) and concentrations of pomelo peel extract (0.050, 0.075, 0.100, 0.125, and 0.150% w/w) on properties of the film were studied. The results showed that pomelo peel extract exhibited *Staphylococcus aureus* inhibition. Yield and *Staphylococcus aureus* inhibiting activity of the extract extracting by dichloromethane were the highest. The concentrations of the extract and drying temperatures had significant effect on the film properties. The highest tensile strength, percent elongation at break, and *Staphylococcus aureus* inhibiting activity were obtained from 0.150% (w/w) pomelo peel extract film that was dried at high temperature (60°C).

Keywords: Biodegradable film, Pomelo peel extract, *Staphylococcus aureus*

1 Introduction

Pomelo (*Citrus maxima* Merr.) is one of the most famous fruits in Asia (Burana-osot et al., 2010). It is primarily eaten fresh and used as a component in main dishes and desserts. Before processing, the thick peels of pomelo are peeled. This peel has some essential oils such as flavonoid, geraniol, linolool, citral, and methylantranilate (Deans and Ritchie, 1987; Mexis et al., 2012; Naradisorn and Ruenkum, 2009; Soffer and Mannheim, 1994; Tim Cushnie and Lamb, 2005). The essential oil include in the pomelo peel extract. Many extracting solvents were used because type, yield, and microbial activity of these essential oils in pomelo peel extract depend on type of extracting solvent (Naradisorn and Ruenkum, 2009). Typically, the pomelo peel extract can be used for medical purposes (Burana-osot et al., 2010; Tim Cushnie and Lamb, 2005). Moreover, in agricultural industry, the pomelo peel extract can be used to inhibit some microbial such as *Colletotrichum gloeosporioides* that causes anthracnose disease in some plants (Naradisorn and Ruenkum, 2009).

Staphylococcus aureas (*S. aureas*) is one of the microbial of gastro enteritis resulting from the consumption of contaminated foods. It is an important microbial due to a combination of toxin-mediated virulence, invasiveness, and antibiotic resistance. It can be found normally in warm-blooded animal nose, on warm-blooded animal hair and skin, especially human nose, hair, and skin. Therefore, *S. aureas* has an opportunity to infect in fresh and processed foods that

were provided or contacted by human (Jablonski and Bohach, 2001). In order to keep foods products from *S. aureas* contamination, it is necessary to control sanitation of corresponding human or select correct antimicrobial packaging technologies.

In the last decade, the use of biodegradable film as food packaging continuously increased due to advantages of the film over other traditional materials such as glass, plastic, and tinplate (Souza et al., 2012). Many natural materials, especially starches that are bio-plastic, have been extensively used to produce the film because of the attractive combination of performance and price. Moreover, these materials do not contribute to environmental pollution (Paes et al., 2008). However, these materials normally cannot inhibit microbial resulting in limitation of biodegradable film application. Therefore, many types of antimicrobial biodegradable films have been developed and were used to inhibit grown or activity of microbial (Appendini and Hotchkiss, 2002; Sanla-Ead et al., 2012; Suppakul et al. 2008). Most of them always contain antimicrobial agent, especially the antimicrobial agent of plant extracts which not affect human health. For example, Mayachiew et al. (2010) who study effect of drying methods and conditions on antimicrobial activity of biodegradable films enriched with galangal extract found that film enriched with galangal extract can be used to inhibit growth of *S. aureas*. Chana-Thaworn et al. (2011) studied antimicrobial activity of hydroxypropyl methylcellulose (HPMC) films containing kiam wood extract against *Escherichia coli*, *Listeria monocytogenes*, and *S. aureas*. The results suggested that the film containing kiam wood extract exhibited antimicrobial activity. Moreover, Maizura et al. (2007) found that antimicrobial hydrolyzed sago starch films containing lemongrass oil were effective in inhibiting the growth of *Escherichia coli* based on clear zone inhibition method.

The production of antimicrobial biodegradable films containing plant extracts starts with plant extracting process. Then, the extract is mixed with other film components such as cassava starch, glycerin, and natural plasticizers (Paes et al., 2008). After mixing, the film solution is casted and dried to obtain the antimicrobial biodegradable film. The application properties of film such as tensile strength, percent elongation at break, and microbial inhibition activity are then evaluated (Maizura et al., 2007; Sivarooban et al., 2008; Seydim and Sarikus, 2006).

From the recent research, many researchers proposed that properties of some antimicrobial biodegradable films depended on process conditions such as concentrations of plant extract and drying conditions. The concentrations of the plant extract have strong effect on antimicrobial activity of the film (Mayachiew et al., 2010; Kechichian et al., 2010; Srinivasa et al., 2004). Moreover, it is well known that drying temperature significantly affect mechanical properties and antimicrobial activity of some antimicrobial biodegradable films (Mayachiew et al., 2010). For example, Jiang et al. (2007) reported that the mechanical properties of transglutaminase-treated soy protein isolate films such as tensile strength and percent elongation at break were significantly varied with drying temperature. Mayachiew et al. (2010) found that vacuum and low pressure superheated steam drying temperatures (70-90°C) had profound effect on antimicrobial activity against *S. aureas* of chitosan films containing galangal extract.

As mention above, this research aimed at producing antimicrobial biodegradable film containing pomelo peel extract. The effects of extracting solvent type on yield and antimicrobial activity of pomelo peel extract against *S. aureus* were determined. Moreover, the effects of concentrations of pomelo peel extract and drying temperature on tensile strength, percent elongation at break, and

antimicrobial activity against *S. aureus* of the films were proposed.

2 Material and Methods

2.1 Material

Pomelo (*Citrus maxima* Merr.), Thong Dee cultivar was collected at the mature stage from fruit orchards around Prachinburi province in Thailand.

The extracting solvents were 95% ethanol (Mallinckrodt, USA), dichlorometane (Burdick and Jackson, Korea), hexane (Mallinckrodt, USA), and ethyl acetate (Mallinckrodt, USA).

2.2 Material preparation

Pomelo was first washed thoroughly to remove impurities. After washing, the pomelo was peeled. The green pomelo peels (flavedo) were cut into small pieces and dried overnight in a hot air dryer (ULM 600II, Memmert, Germany) at 40°C. The dried peels were collected in desiccator.

2.3 Pomelo peel extraction procedures

To prepare pomelo peel extract, 250 g of dried pomelo peels was extracted overnight with 750 ml of 95% ethanol, dichlorometane, hexane, and ethyl acetate at 35°C. The extract was filtered through a filter paper (ø110 mm, Cat. No. 1001 110, Schleicher and Schuell GmbH, Dassel, Germany). The filtrate was concentrated in a rotary evaporator (Resona Technics Labo Rota 300, Gossau, Switzerland) at 40°C for 30 min. Yield of the pomelo peel extract was determined. Then, the pomelo peel extract was kept at 4°C prior to determination of clear zone of inhibition and antimicrobial index (AI).

2.4 Determination of yield of pomelo peel extract

Yield of pomelo peel extract was calculated as the following equation:

$$Yield \quad (\%) \quad = \frac{W_1}{W_2} \times 100$$

where, W_1 is weight of extract recovered (g) and W_2 is weight of fresh pomelo peel (g).

2.5 Evaluation of clear zone of inhibition and antimicrobial index (AI) of pomelo peel extract

S. aureas (ATCC 25923) was obtained from the Department of Medical Sciences, Ministry of Public Health, Thailand. The microbial was maintained in TSA at 5°C. Stock culture of *S. aureas* was grown in TSB at 37°C for 18 h at 160 rpm. The maximum level of the microbial was 1,010 CFU/ml. The concentration was subsequently adjusted to 108 CFU/ml using buffer peptone water. A suspension of the tested microbial was spreaded on the MHA plate. Then, 6 mm in diameter of filter paper which was soaked into 15 µl of the pomelo peel extract was placed on the inoculated plates. After keeping at 4°C for 2 h, the plate was incubated at 37°C for 24 h. After that, the diameter of clear zone of inhibition was evaluated in millimeters. The filter paper that was not soaked into the pomelo peel extract was used as a control sample. Then, the antimicrobial index (AI) was calculated as (diameter of clear zone of inhibition – diameter of filter paper) / (diameter of filter paper).

2.6 Preparation of biodegradable film containing pomelo peel extract

To determine the effects of concentrations of the extract on properties and antimicrobial activity of biodegradable film, pomelo peel extract was mixed with 5.0 g of cassava starch (Cholcharoen Co., Ltd., Thailand), 0.75 g of glycerin, 0.7 g of sucrose, and 1.4 g of inverted sugar at concentrations of 0.050, 0.075, 0.100, 0.125, and 0.150% w/w. The specific content of film mixture was homogenized and poured on an acrylic plate with dimensions of 10×10 cm to cast a film at thickness of 100 ± 10 µm (Figure 1). A micrometer was used for measuring film thickness. The film was hot air dried at 40°C until moisture content of the film equal to 16% (wet basis). The film which not

contained pomelo peel extract was used as a control sample.

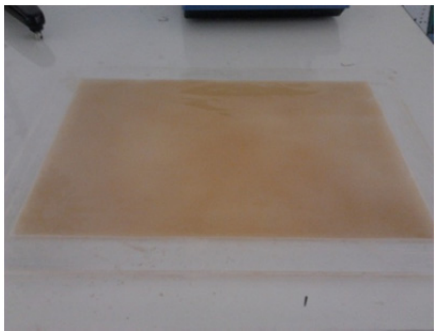

Figure 1 Biodegradable film in acrylic plate before drying.

To determine the effects of film drying temperatures on properties and antimicrobial activity of biodegradable film containing pomelo peel extract, pomelo peel extract at selected concentration was mixed in another film component. The film mixture was prepared following above method. After casting the film mixture to an acrylic plate, hot air drying of the film was performed by five temperatures, which are 40, 45, 50, 55, and 60°C, until moisture content of the film equal to 16% (wet basis).

2.7 Evaluation of clear zone of inhibition and antimicrobial index (AI) of biodegradable film containing pomelo peel extract

To determine antimicrobial activity of biodegradable film containing pomelo peel extract, the method was similar to the method of evaluate clear zone of inhibition of pomelo peel extract as mention above, but 6 mm in diameter of filter paper which was soaked into 15 µl of the extract was changed to 6 mm in diameter of the biodegradable film wall containing pomelo peel extract. Then, the antimicrobial index (AI) was calculated as (diameter of clear zone of inhibition – diameter of biodegradable film wall) / (diameter of biodegradable film wall).

2.8 Determination of tensile strength and percent elongation at break of biodegradable film containing pomelo peel extract

The tensile strength of biodegradable film containing pomelo peel extract was carried out according to ASTM Standard Method D 882-09 (ASTM, 2009). Texture analyzer (TA-TX Plus, Stable Micro Systems, UK) was used. 100×25 mm of the film was prepared before testing. The film specimens were mounted in the grips of the texture analyzer and stretched at a rate of 50 mm min^{-1} until breaking. Tensile strength was calculated by dividing the maximum load by original cross-sectional area of the film. Percent elongation at break was calculated by dividing the extension at the moment of rupture of the film by its initial gage length and multiplying by 100.

2.9 Statistical analysis

All experiments were performed in triplicate. The variance was determined by ANOVA and the difference of mean values was determined by Duncan's multiple range tests at a statistically significant level of 0.05.

3 Results and Discussions

Yield values of pomelo peel extract extracting by 95% ethanol, dichlorometane, hexane, and ethyl acetate are shown in Figure 2. The result showed that the values of pomelo peel extract were in range of 0.19-0.57%. The amount of the extract that extracted by dichlorometane has the highest. The reason may be the solubility of the extract in each extracting solvent and degree of polarity of extracting solvents (Zhao and Hall III, 2008).

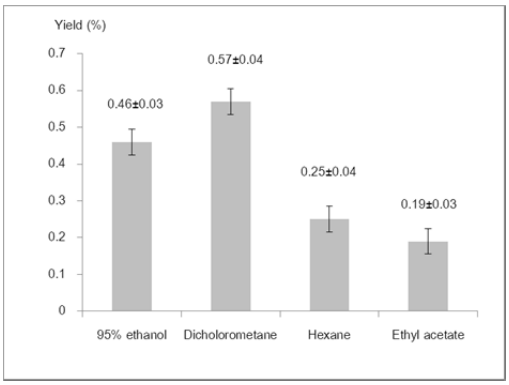

Figure 2 Yield of pomelo peel extract extracting by 95% ethanol, dichlorometane, hexane and ethyl acetate.

The antimicrobial activity of pomelo peel extract from pomelo peel against *S. aureas* was proposed to determine clear zone of inhibition around the filter paper. Figure 3 shows the filter paper that was soaked dichlorometane pomelo peel extract. The result showed that clear zone of inhibition can be generated around the filter paper. This experiment indicated that dichlorometane pomelo peel extract can be used to inhibit *S. aureus* growth. Similar result was obtained in 95% ethanol, hexane, and ethyl acetate pomelo peel extract but the figures are not shown. This result is corresponding to result of Naradisorn and Ruenkum (2009) who studied antimicrobial activity of pomelo peel extract against *Colletotrichum gloeosporioides*. They found that pomelo peel extract can inhibit this microbial that causes a anthracnose disease. Normally, pomelo peel extract contains a lot of essential oils (Deans and Ritchie, 1987; Mexis et al., 2012; Naradisorn and Ruenkum, 2009; Soffer and Mannheim, 1994; Tim Cushnie and Lamb, 2005). Some essential oils that are lipophilic compounds might destroy the phopholipid cell membrane of *S. aureus*, causing increased permeability and leakage of cytoplasm. Moreover, the essential oils might inhibit production of essential enzyme of *S. aureus* or coagulate of *S. aureus* cell content (Sanla-Ead et al., 2012). This action is typically used to explain the microbial inhibition activity of essential oils from many plant extracts (Burt, 2004; Cowan, 1999; Tassou et al., 2000).

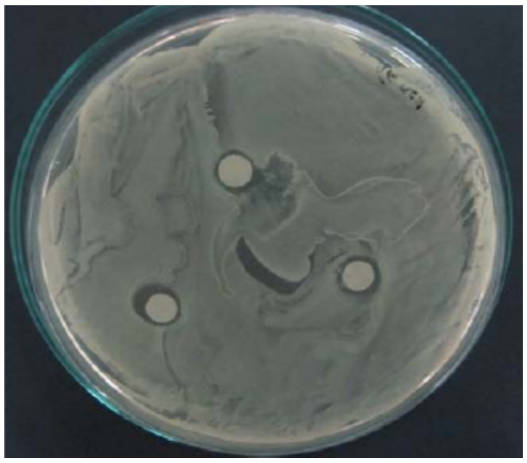

Figure 3 Clear zones of inhibition of the filter paper that was soaked dichlorometane pomelo peel extract.

Table 1 shows the values of antimicrobial index (AI) of the pomelo peel extract that were extracted by different extracting solvents. The result showed that the antimicrobial index (AI) of inhibition of pomelo peel extracts were in the range of 0.26-0.47. The values were significant different among the extracting solvents. The reason may be the difference of components in pomelo peel extracts that were extracted from different extracting solvents. Moreover, the result showed that pomelo peel extract that extract by dichlorometane is the best inhibitor. Srisajjalertwaja (1996) who investigated antifungal compounds from pomelo peel extract that extract by dichlorometane suggested that the pomelo peel extract can be used to inhibit fungi that is *Cladosporium cladosporioides*. The antifungal essential oils in the extract (extracting by dichlorometane) that were determined using Gas cromatrograpy-Mass spectroscopy (GC-MS), IR-spectroscopy and UV-spectroscopy contained some functional groups of hydroxyl and ester. These essential oils might affect on *S. aureas* similar to affect on *Cladosporium cladosporioides*.

Table 1 Antimicrobial index (AI) of the filter paper that were soaked 95% ethanol, dichlorometane, hexane and ethyl acetate pomelo peel extract.

Solvents	AI* (mm)
Control	$0.00^{d} \pm 0.00$
95% ethanol	$0.31^{b} \pm 0.01$
Dichlorometane	$0.47^{a} \pm 0.01$
Hexane	$0.31^{b} \pm 0.01$
Ethyl acetate	$0.26^{c} \pm 0.01$

*Values followed by the same letter are not significantly different ($P \geq 0.05$)

Effects of concentrations of pomelo peel extract on tensile strength, percent elongation at break, and antimicrobial index (AI) of biodegradable films containing pomelo peel extract were studied. Due to high yield and *S. aureus* inhibition activity, pomelo peel extract that extract by dichlorometane was selected to prepare antimicrobial biodegradable film. The effect of concentrations of pomelo peel extract that was mixed in the film was investigated. Tensile strength, percent elongation at break, and antimicrobial index (AI) of the films were determined. In case of mechanical properties of the film, high tensile strength and percent elongation at break are generally required because structure that is strength and elastic is the desired properties of film for application (Gontard et al., 1992). The values of tensile strength and percent elongation at break of the biodegradable film containing pomelo peel extract are showed in Table 2. The results showed that tensile strength and percent elongation at break of the film were approximately 17.37-17.38 MPa and 15.7-16.0%, respectively. The values did not significant different between the film samples. This result indicated that pomelo peel extract did not influence structure of the film. The result agreed with the result of Kechichian (2010) who studied natural antimicrobial ingredients incorporated in biodegradable films based on cassava starch. The researcher suggested that some natural antimicrobial ingredients such as cinnamon powder did not influence tensile strength of the film. Moreover, clove powder did not significantly change the percent elongation at break of the film. This may due to a little particle size and less amount of the natural antimicrobial ingredients in the film.

Table 2 Effects of concentrations of pomelo peel extract on tensile strength, percent elongation at break, and antimicrobial index (AI) of biodegradable films containing pomelo peel extract.

Concentration of pomelo peel extract (% w/w)	Tensile strength* (MPa)	Percent elongation at break* (%)	AI*
0.000(Control)	$17.37^{NS} \pm 0.07$	$15.8^{NS} \pm 0.2$	$0.000^{e} \pm 0.000$
0.050	$17.38^{NS} \pm 0.08$	$16.0^{NS} \pm 0.1$	$0.000^{e} \pm 0.000$
0.075	$17.38^{NS} \pm 0.05$	$16.1^{NS} \pm 0.3$	$0.018^{d} \pm 0.003$
0.100	$17.38^{NS} \pm 0.06$	$15.7^{NS} \pm 0.5$	$0.050^{c} \pm 0.007$
0.125	$17.38^{NS} \pm 0.03$	$15.8^{NS} \pm 0.4$	$0.105^{b} \pm 0.015$
0.150	$17.37^{NS} \pm 0.02$	$16.0^{NS} \pm 0.2$	$0.167^{a} \pm 0.016$

*Values followed by the same letter are not significantly different ($P \geq 0.05$)

As in Table 2, *S. aureus* inhibiting activity of biodegradable film containing pomelo peel extract was also summarized. The antimicrobial index (AI) of the film containing 0.050, 0.075, 0.100, 0.125, and 0.150% (w/w) of pomelo peel extract equal to 0.000, 0.018, 0.050, 0.105, and 0167, respectively. The result revealed that *S. aureus* inhibiting activity of pomelo peel extract active when its concentration in the film was higher than 0.050% (w/w). Moreover, when comparing this result with the antimicrobial index (AI) result of pure pomelo peel extract extracting by dichlorometane in Table 1, the result suggested that antimicrobial index (AI) of the film containing pomelo peel extract is lower than that of the pure pomelo peel extract. This may due to concentration of the extract in the film.

The result in Table 2 also showed that antimicrobial index (AI) of the film increased with increasing of concentration of pomelo peel extract. antimicrobial index (AI) of the films mixed with 0.150% (w/w) pomelo peel extract were higher than that of the films that were mixed with pomelo peel extract at lower concentration. This result agreed with the result of Mayachiew et al. (2010). They suggested that the concentration of galangal extract significantly affected the cell viability of the *S. aureus*. High concentration of galangal extract exhibited high cell reduction number of tested microbial.

To obtained biodegradable film containing 0.150% (w/w) pomelo peel extract at moisture content of 16% (wet basis), the film mixture were hot air dried for 14.43, 13.35, 12.87, 12.11, and 11.84 h at 40, 45, 50, 55, and 60°C, respectively. After drying, tensile strength, percent elongation at break, and antimicrobial index (AI) of the films were determined. The obtained values are presented in Table 3. The values were approximately 17.37-19.90 MPa, 16.0-24.4%, and 0.167-0.292, respectively. When comparing with the result of Chana-Thaworn et al. (2011) who studied properties of antimicrobial activity of hydroxypropyl methylcellulose (HPMC) films containing kiam wood extract, the values of tensile strength and percent elongation at break of film containing pomelo peel extract closed to those of HPMC films containing kiam wood at 1,500 mg l^{-1} (tensile strength is 18.48 MPa and percent elongation at break is 11.19%). However, the antimicrobial activity against *S. aureus* of HPMC films containing kiam wood at 1,500 mg l^{-1} (diameter of clear zone inhibition is 25.33 mm) is higher than that of film containing the highest concentration of pomelo peel extract in this research (diameter of clear zone inhibition is 1.75 mm). Possible reason could be the different concentration and functional properties of both extracts.

Table 3 Effects of drying temperatures on tensile strength, percent elongation at break, and antimicrobial index (AI) of biodegradable films containing pomelo peel extract.

Drying temperature ($^{\circ}$C)	Tensile strength* (MPa)	Percent elongation at break* (%)	AI*
40	17.37^{e}±0.02	16.0^{e}±0.2	0.167^{e}±0.016
45	18.66^{d}±0.07	17.3^{d}±0.7	0.182^{d}±0.011
50	19.30^{c}±0.01	18.6^{c}±0.2	0.200^{c}±0.013
55	19.63^{b}±0.02	20.1^{b}±0.8	0.247^{b}±0.008
60	19.90^{a}±0.03	24.4^{a}±1.2	0.292^{a}±0.003

*Values followed by the same letter are not significantly different (P≥0.05)

The results in Table 3 also revealed that tensile strength, percent elongation at break, and antimicrobial index (AI) of the film significantly increased when drying temperature increased. The tensile strength, percent elongation at break, and antimicrobial index (AI) of the films dried at 60°C were higher than those of the films that were dried at lower temperature. Due to the lowest drying time, the film dried at the highest temperature had the highest quality. The result suggested that heat could affect some properties of essential oils in the pomelo peel extract such as water solubility and rate of vapourization during drying (Sanla-Ead et al., 2012). Moreover, the structure of the biodegradable film containing pomelo peel extract may depend on drying time. This result agreed with the result of Denavi et al. (2009) who studied effects of drying conditions (drying temperature at 34, 40, 55, 70, and 76°C) on some physical properties of soy protein films. These authors demonstrated that the best properties such as tensile strength, percent elongation at break, and barrier property of the film were obtained at higher temperature which allows a little longer time of drying process. However, the result disagreed with the result

of Mayachiew and Devahastin (2008) who demonstrated that low-pressure superheated stream drying at high temperature (70°C) led to chitosan film with higher tensile strength and percent elongation. The reason may involve drying methods and film materials.

4 Conclusions

From the experimental results, it was indicated that pomelo peel extract can be used to inhibit *S. aureus* that is a food pathogen. Yield and *S. aureus* inhibiting activity of the extract extracting by dichloromethane were the highest. After film production, the results of film properties testing appeared that the tensile strength, percent elongation at break and *S. aureus* inhibiting activity of the film depend on concentration of pomelo peel extract and film drying temperature. The edible film mixed with 0.15% w/w pomelo peel extract and dried at 60°C were higher than those of the films that were produced using another condition.

5 Acknowledgement

This work was financially supported by Faculty of Industrial Technology and Management, King Mongkut's University of Technology North Bangkok Foundation.

6 References

Appendini, P., Hotchkiss, J. H. 2002. Review of antimicrobial food packaging. Innovative Food Science and Emerging Technologies 3, 113-126.

ASTM standard D 882-09. 2009. Satandard test method for tensile properties of thin plastic sheeting. Annual book of American Standard Testing Methods. Philadelphia. PA.

Burana-osot, J., Soonthornchareonnon, N., Chaidedgumjorn, A., Hosoyama, S., Toida, T. 2010. Determination of galacturonic acid from pomelo pectin in term of galactose by HPAEC with fluorescence detection. Carbohydrate Polymers 81, 461-465.

Burt, S. 2004. Essential oils: Their antibacterial properties and potential applications in food-A review. International Journal of Food Microbiology 94, 223-253.

Chana-Thaworn, J., Chanthachum, S., Wittaya, T. 2011. Properties and antimicrobial activity of edible films incorporated with kiam wood (Cotyleobium lanceotatum) extract. LWT 44, 284-292.

Cowan, M.M. 1999. Plant productions as antimicrobial agents. Clinical Microbiology Review 12, 564-582.

Deans, S.G., Ritchie, G. 1987. Antibacterial properties of plant essentail oils. International Journal of Food Microbiology 5, 165-180.

Denavi, G., Tapia-Blácido, D.R., Añón, M.C., Sorbral, P.J.A., Mauri, A.N., Menegalli, F.C. 2009. Effect of drying conditions on some physical properties of soy protein films. Journal of Food Engineering 90, 341-349.

Gontard, N., Guilbert, S., Cuq, J.L. 1992. Edible wheat gluten film: influence of the main process variable on film properties using response surface methodology. Journal of Food Science 57, 190-195.

Jablonski, L.M., Bohach, G.E. 2001. Food microbiology: Fundamentals and frontiers. In: Doyle, M.P., Beuchat, L.R., Montville, T.J. (Eds.), (pp. 1-59). Washington DC: ASM Press.

Jiang, Y., Tang, C.H., Wen, Q.B., Li, L., Yang, X.Q. 2007. Effect of processing parameters on the properties of transglutamminase-treated soy protein isolate films. Innovation Food Science and Engineering Technology 8, 218-225.

Kechichian, V., Ditchfield, C., Veiga-Santos, P., Tadini, C.C. 2010. Natural antimicrobial ingredients incorporated in biodegradable films based on cassava starch. LWT 43, 1088-1094.

Maizura, M., Fazilah, A., Norziah, M.H., Karim, A.A. 2007. Antibacterial activity and mechanical properties of

partially hydrolyzed sago starch-alginate edible film containing lemongrass oil. Journal of Food Science 72, C324-C330.

Mayachiew, P., Devahastin, S. 2008. CoMParative evaluation of physical properties of edible chitosan films prepared by different drying methods. Drying Technology 26, 176-185.

Mayachiew, P., Devahastin, S., Mackey, B.M., Niranjan, K. 2010. Effects of drying methods and conditions on antimicrobial activity of edible chitosan films enriched with galangal extract. Food Research International 43, 125-132.

Mexis, S.F., Chouliara, E., Kontominas, M.G. 2012. Shelf life extension of ground chicken meat using an oxygen absorber and a citus extract. LWT 49, 21-27.

Naradisorn, M., Ruenkum, A. 2009. Preliminary study on antimicrobial activity of crude extracts of pomelo albedo against Colletotrichum gloeosporioides. Journal of Food Agricultural-industrial Special issue, S138-S142.

Paes, S.S., Yakimets, I., Mitchell, J.R. 2008. Influence of gelatinization process on functional properties of cassava starch films. Food Hydrocolloid 22, 788-797.

Sanla-Ead, N., Jangchud, A., Chonhenchob, V., Suppakul, P. 2012. Antimicrobial activity of cinnamaldehyde and eugenol and their activity after incorporation into cellulosed-based packaging film. Packaging Technology and Science 25(1), 7-17.

Seydim, A.C., Sarikus, G. 2006. Antimicrobial activity of whey protein based edible films incorporated with oregano, rosemary and garlic essential oils. Research International 39, 639-644.

Sivarooban, T., Hettiarachchy, N.S., Johnson, M.G. 2008. Physical and antimicrobial properties of grape seed extract, nisin, and EDTA incorporated soy protein edible films. Food Research International 41, 781-785.

Soffer, T., Mannheim, C.H. 1994. Optimization of Enzymatic Peeling of Oranges and Pomelo. LWT 27, 245-248.

Souza, A.C., Benze, R., Ferrão, E.S., Ditchfield, C., Coelho, A.C.V., Tadini, C.C. 2012. Cassava starch biodegradable films: Influence of glycerol and clay nanoparticles content on tensile and barrier properties and glass transition temperature. LWT 46, 110-117.

Srinivasa, P.C., Ramesh, M.N., Kumar, K.R., Tharanathan, R.N. 2004. Properties of chitosan films prepared under different drying conditions. Jouranl of Food Engineering 63, 79-85.

Srisajjalertwaja, S. 1996. Antifungal compounds from pomelo ZCitrus grandis, L. Osbeck) peel. MSD thesis. Chiang Mai: Department of Chemistry, Chiang Mai University.

Suppakul, P., Miltz, J., Sonneveld, K., Bigger, S. W. 2003. Active packaging technologies with an emphasis on antimicrobial packaging and its applications. Journal of Food Science 68, 408-420.

Tassou, C., Koutsoumanis, K., Nychas, G.J.E. 2000. Inhibition of Salmonella enteritidis ans Staphylococcus aureus in nutrient broth by mint essential oil. Food Research International 33, 273-280.

Tim Cushnie, T.P., Lamb, A. 2005. Antimicrobial activity of flavonoids. International Journal of Antimicrobial Agent 26, 343-356.

Zhao, B. Hall III, C.A. 2008. Composition and antioxidant activity of raisin extracts obtained from various solvents. Food Chemistry 108, 511-518.