

Characterization of cassava starch-based edible film enriched with lemongrass oil (*Cymbopogon citratus*)

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Abstract. Resianingrum R, Atmaka W, Khasanah LU, Kawiji, Utami R, Praseptianga D. 2016. Characterization of cassava starch-based edible film enriched with lemongrass oil (*Cymbopogon citratus*). *Nusantara Bioscience* 8: 278-282. Good packaging is necessary to prevent food spoilage before and after processing. The edible film maintains food quality and eco-friendly packaging. Cassava starch-based edible film is capable of being developed because of the approaching appearance of plastic packaging. The addition of essential oils can be used as an alternative to improve the antimicrobial properties of the edible film. Lemongrass oil consists of active compounds that shown antimicrobial activity. Therefore, the characteristics of cassava starch-based edible film incorporated with lemongrass oil were investigated. Concentration of lemongrass oils varied at 0%, 0.25%, 0.5%, 0.75% and 1%. The analyzed edible film characteristics include thickness, water vapor transmission rate, tensile strength, elongation, and antimicrobial activity. The results showed that the addition of lemongrass oil increased the thickness, tensile strength, and antimicrobial activity but decreased the elongation of edible films. The water vapor transmission rate of edible films was not affected by the addition of lemongrass oil.

Keywords: Cassava-starch, edible film, essential oil, lemongrass

INTRODUCTION

Food products may encounter several undesirable characteristics change before and after processing. One of the preventions of food damage is packaging. According to Buckle et al. (1982), the packaging is a way to provide the right surrounding conditions of food. Edible film packaging is a relatively new and environmentally friendly food preservation technique compared to the use of plastic packaging that tends to damage the environment. Edible film is a thin layer that is used to coat food, or placed between components that function as a barrier to mass transfer such as water, oxygen, and fat. Edible films can be combined with a food additive to enhance the quality of color, aroma, and texture of the product, as well as to control microbial growth (Sinaga 2013). An edible film with antimicrobial characteristics has a potency to prevent pathogen contamination in various textured food products (meat, fruits, and vegetables). The combination of antimicrobial compounds in film packaging may be used to control microbial growth in food products and to extend the shelf life, as well as to improve the quality of food (Quintavalla 2002).

The edible film can be made from various raw materials which have a relatively high starch composition. The processing of cassava starch starch-based edible film has quite good characteristics, even though the water vapor transmission rate is quite high. Cassava starch-based edible film has similar mechanical properties with plastic and also its transparent appearance (Arinda 2009). Among the wide variety of edible coating materials, starch has been

regarded as one of the promising material because of its similar physical characteristics compared to the synthetic polymers such as transparent, odorless, tasteless, semi-permeable to carbon dioxide and oxygen proof (Vásconez et al. 2009).

Lemongrass (*Cymbopogon citratus*) is an annual plant that lives in the wild-with a quasi trunk that forms thick clumps with up to 1-2 meters height and has a strong aroma and fragrance. Lemongrass oil is one of the important essential oils because it contains citral component between 70-80% (Guenther 1987). Citral is the raw material for the synthesis of vitamin A. Lemongrass essential oil showed antifungal and antibacterial characteristics, especially for gram-positive bacteria. It also demonstrated a synergistic effect when combined with an antimicrobial agent against *Staphylococcus aureus* in agar medium. Antioxidant activity in the form of citral components and phenol group was also found in the lemongrass essential oil (Leung 1980).

According to Pramani (2010), shredding process with 4 hours of distillation time was a treatment with the most optimal time to produce a better lemongrass oil. The utilization of lemongrass essential oil in the food sector is still small. Related research about lemongrass essential oils in the edible film was found on research conducted by Grau et al. (2006), concerning the addition of lemongrass essential oil into the edible films of apple puree which has pretty good characteristics and inhibition properties at a concentration of 0.5%. Ahmad et al. (2011) also noted that the highest antimicrobial effect on the addition of lemongrass essential oils and gelatin derived from animal protein in edible film was 0.75%.

Thus, this study is needed to determine the effect of lemongrass essential oil (*Cymbopogon citratus*) addition in varied concentrations (0%; 0.25%; 0.5%; 0.75% and 1%) on the characteristics of cassava starch-based edible film, including thickness, water vapor transmission rate, tensile strength, percent elongation, and antimicrobial activity.

MATERIALS AND METHODS

Lemongrass essential oil processing

Lemongrass essential oil was obtained by steam distillation for four hours. Before the distillation, the lemongrass was shredded with a length of about 1 cm. The shredded lemongrass was put into the steam distillation unit and distilled for four hours, starting from the first droplet.

Edible film processing

The processing of edible film was done by dissolving 5 grams of cassava starch with 150 ml of aquadest until it diffused (Hasyim 2008). Furthermore, based on research by Firdaus et al. (2008), cassava starch solution was heated at 68°C until the solution gelatinized. 2 ml of gelatinized glycerol was added to the solution, mixed, and heated at 60°C for 30 minutes. After the heating process, cooling process was carried out until the temperature reached 30°C. Essential oil of lemongrass was put into the solution after the cooling process, and stirred with a magnetic stirrer. The concentration of essential oil used varied from 0%; 0.25%; 0.5%; 0.75% and 1% by the volume of aquadest used. Edible film solution was poured on the top of plastic trays and dried for 5 hours of drying at a temperature of 75°C (Wigati 2014).

Edible film characterization

Characterization of the edible film includes thickness based on Wafiroh et al. (2011) method, water vapor transmission rate and percent elongation based on ASTM method (1993), tensile strength based on Murdianto (2005) method, and also antimicrobial activity based on agar diffusion method (Manab et al. 2011).

Data analysis

The research design used a completely randomized design (CRD), which consists of a single factor, which is the variation of lemongrass essential oil concentration. Data were analyzed using ANOVA to determine whether there is a difference at a significance level of 5%, followed by the test of Duncan Multiple Range Test (DMRT) at the 5% significance level.

RESULT AND DISCUSSION

Thickness analysis

The addition of lemongrass essential oil on cassava starch-based edible film solution shown a significant effect on film thickness Table 1. The higher oil concentrations addition generated the increase of the thickness.

According to Nugroho et al. (2013), thickness increasing was occurred due to the differences in the film material concentration, while the same volume of solution was poured at each plate. This resulted in an increase in total solids of the film after the drying process, as well as an increase in film matrix constituent polymers. Pramani (2010) also stated that the °Brix value of distilled lemongrass oil ranged between 79.066 to 80.533 °Brix, which exhibited a dense fluid property. It is expected, which the addition of high viscous lemongrass essential oil will lead to an increase in the thickness properties, which was added to the cassava starch-based edible film. This is in accordance with research from Pramadita and Sutrisno (2012), which stated that the large number addition of cinnamon essential oil would increase the amount of total solids, so that the film thickness increased.

In the study by Ghassemloo et al. (2011), the thickness of corn starch-based film with the addition of essential oil ranged between 0.17 to 0.24 mm. Reported by Ayuningrum (2015), an increase in the cassava starch-based film thickness with cinnamon bark essential oil occurred in the study. Cassava starch-based edible film enriched with lemongrass essential oil showed the thickness of 0.144 to 0.189 mm and fulfilled the standard of Japanese Industrial Standards (1975), which is not thicker than 0.25 mm.

Water vapor transmission rate analysis

According to Table 2, the addition of lemongrass essential oil had no significant effect on the water vapor transmission rate of cassava starch-based edible film. Garcia et al. (2000) mentioned that the main factor is causing the high value of water vapor transmission rate on edible film because of the hydrophilic component was higher than the hydrophobic one. According to Bourtoom (2008), the water vapor permeability of edible film reduced if the hydrophobic component on edible film increased. With the addition of hydrophobic compounds into a solution of edible film, the hydrophobic properties will increase and hydrophilic properties will decrease. Thus, it is suspected that the hydrophobic properties in cassava starch-based edible film with lemongrass essential oil addition did not change significantly, so that the addition of lemongrass essential oils did not give a significant change on the rate of water vapor transmission on the film. This is in accordance with the research conducted by Grau et al. (2006) about lemongrass essential oil addition on the apple puree edible film, which showed no significant effect because it was suspected that essential oil contains more terpene than fat. So that the hydrophilic ratio inside edible film was still higher than its hydrophobic ratio.

Estiningtyas (2010), reported that the addition of 10% ginger extract on corn starch-based edible film showed lower the water vapor permeability of edible film than an edible film with no ginger extract addition, but still did not give a significantly different result between the two. It was suspected that the ginger extract still contained starch compound, which could add more matrix to the film.

Table 1. Thickness of cassava starch-based edible film with lemongrass essential oil addition

Essential oil concentration	Edible film thickness (mm)
0%	0.143 ^a ±0.003
0.25%	0.175 ^b ±0.010
0.5%	0.177 ^{bc} ±0.003
0.75%	0.188 ^c ±0.009
1%	0.189 ^c ±0.005

Note: Different letter in the same column shown a significantly different result ($\alpha=0.05$)

Table 2. Water vapor transmission rate of cassava starch-based edible film with lemongrass essential oil addition

Essential oil concentration	Water vapor transmission rate of edible film (gram/hour m ²)
0%	24.60 ^a ±1.60
0.25%	24.40 ^a ±1.07
0.5%	24.37 ^a ±0.35
0.75%	23.41 ^a ±1.72
1%	23.28 ^a ±1.17

Note: Different letter in the same column shown a significantly different result ($\alpha=0.05$)

Table 3. Tensile strength of cassava starch-based edible film with lemongrass edible film addition

Essential oil concentration	Tensile strength elongation (MPa)
0%	0.16 ^a ±0.019
0.25%	0.52 ^b ±0.018
0.5%	0.52 ^b ±0.022
0.75%	0.47 ^b ±0.025
1%	0.48 ^b ±0.101

Note: Different letter in the same column shown a significantly different result ($\alpha=0.05$)

Table 4. Percent elongation of cassava starch-based edible film with lemongrass edible film addition

Essential oil concentration	Percent elongation of edible film (%)
0%	68.31 ^a ±2.64
0.25%	31.75 ^{cd} ±3.66
0.5%	30.02 ^d ±1.46
0.75%	36.40 ^b ±3.64
1%	41.60 ^b ±3.62

Note: Different letter in the same column shown a significantly different result ($\alpha=0.05$)

Table 5. *Pseudomonas fluorescens* FNCC 0071 and *Aspergillus niger* FNCC 6081 inhibition zone of cassava starch-based edible film with lemongrass edible film addition

Essential oil Concentration	<i>Pseudomonas fluorescens</i> (mm)	<i>Aspergillus niger</i> (mm)
0%	9.08 ^a ±0.04	10.25 ^a ±0.193
0.25%	25.98 ^b ±0.02	11.74 ^{ab} ±0.196
0.5%	25.89 ^b ±0.12	11.97 ^{ab} ±0.176
0.75%	25.97 ^b ±0.04	12.16 ^{ab} ±0.212
1%	26.05 ^b ±0.06	14.320 ^b ±0.134

Note: Different letter in the same column shown a significantly different result ($\alpha=0.05$)

Tensile strength analysis

Based on Table 3, the tensile strength of cassava starch based-edible film with lemongrass essential film addition significantly higher than that of the control sample, but there was no significant difference between the other cassava starch based-edible film with different concentrations of lemongrass essential oil. The tensile strength of cassava starch based-edible film with lemongrass essential oil addition ranged from 0.16 to 0.52 MPa.

Amalia et al. (2014), mention that increasing number of solid granular in a polymer will minimize the voids between the cells on the formed gel. Lemongrass essential oil contains solid compound that allegedly reduced inter-cell cavities in the edible film, which then formed a more compact matrix. Film formation in a more compact matrix will influence the increasing tensile strength of cassava starch-based edible film.

Ojagh et al. (2010) reported that the addition of essential oil in the edible film caused different effects. The addition of cinnamon essential oils in chitosan-based edible film increased tensile strength and decreased elongation. A strong interaction between the polymer and cinnamon essential oil resulted in a cross-linking effect, which lowered the free volume and molecular mobility of the polymer. This was presumably the same as the effect of lemongrass essential oil addition to cassava starch-based edible film, which caused an increase in the tensile strength ability. Peng and Li (2014) also stated that the addition of several kinds of essential oils in chitosan-based edible film chitosan increased tensile strength.

Percent elongation analysis

According to Table 4 percent elongation of the cassava starch-based edible film with lemongrass essential oil addition significantly lower than edible film with no oil addition. Percent elongation of samples with 0,25% and 0,5% addition of lemongrass essential oil resulted in no significant difference but showed a significant difference when compared to samples with 0,75% and 1% addition of lemongrass addition.

Stated by Sinaga et al. (2013), the reduction of intermolecular interaction on protein chain was caused by the addition of glycerol. Plasticizer molecules disrupted the cohesiveness of the starch, lowered the intermolecular interactions and increased the mobility of the polymer. Furthermore, this increased elongation at the separating attempt due to the increase of glycerol concentration. Siswanti et al. (2011) mentioned that mechanical properties of the film depended on the material strength used in the making of the film, which was to form a strong molecular bond in such significant amounts. Based on Sinaga et al. (2013) and Siswanti et al. (2011), percent elongation decreasing of cassava starch-based edible film with lemongrass essential oil was suspected by unlike plasticizer, lemongrass essential oil in edible film strengthened the molecular properties of edible film that will lead to the increase of starch cohesiveness and reducing polymer mobility. Addition compactness of starch increased tensile strength while decreasing polymer mobility decreased percent elongation value of the edible film.

According to Pramadita and Sutrisno (2012), the higher the concentration of cinnamon essential oils, the lower the elongation of edible film. Two factors affecting the value of elongation at edible film were the type of polysaccharide and the addition of glycerol as plasticizer. The addition of cinnamon essential oils tends to lower down the value of elongation. The addition of cinnamon essential oil weakens the network of the film that the more oil was added, the matrix film formed brittle because the oil has weak bonds between the compounds. Pramadita and Sutrisno (2012) also suggested that the addition of essential oil of lemongrass on an edible film formed a dense matrix of the film and a rigid network. So that the cassava starch-based edible film has a low percent elongation. This is also supported statement of Hosseini et al. (2009) that the addition of essential oils can cause the films to have more compact structure, thus leading to an elongation decreasing. Ayuningrum (2015) stated that percent elongation of cassava starch-based edible film decreased by the addition of cinnamon essential oil to 58.56%. Grau et al. (2006), a chitosan-based edible film with essential oil addition also showed percent elongation value decreasing.

Antimicrobial activity analysis

Antimicrobial activity analysis was conducted to determine the level of inhibition of cassava starch-based edible film with lemongrass essential oil addition against *Pseudomonas fluorescence* FNCC 0071 and *Aspergillus niger* FNCC 6081 because they are common microbes that attack food products. Inhibition of microbial was shown by the clear zone, which was formed around the samples that were placed in the media. The wider the clear zone formed, the stronger the inhibition power of the sample.

Davis and Stout (1971) stated that inhibition power criteria were 20 mm or more area of a clear zone represented a very strong inhibition, 10-20 mm of clear zone area represented a strong category, 5-10 mm clear zone represented a medium category, and the less area of clear zone represented a weak category.

Table 5 show that the cassava starch-based edible film with lemongrass essential oil addition was able to inhibit *Pseudomonas fluorescence*. The inhibition level of the edible film with lemongrass essential oil addition was significantly different from the control sample. The control sample without lemongrass essential oil addition resulted in a clear zone of 9.08 mm, which was included in the medium category in inhibiting *Pseudomonas fluorescence*. While cassava starch-based edible film with lemongrass essential oil addition ranged from 25.89 to 26.05 mm, which was included in the very strong category in inhibiting *Pseudomonas fluorescence*.

Besides, cassava starch-based edible film with lemongrass essential oil addition also able to inhibit *Aspergillus niger*. The range of clear zone against *Aspergillus niger* was 10.25 to 13.68 mm, which then included in the strong category in inhibiting *Aspergillus niger*. The level of inhibition produced by cassava starch-based edible film with the highest concentration of lemongrass essential oil addition (1%) showed a significant difference result compared with the control. According to

Utami et al. (2012), incorporation of red ginger and red galangal essential oils into cassava starch-based edible film caused a microbial growth inhibition activity. When essential oils are combined into the edible film, essential oils diffused to the agar media and generated a clear zone on the microbial growth medium. It is expected that the higher the concentration of essential oil was added, the more citral active compounds diffused inside the agar to inhibit microbes.

According to Guenther (1987), lemongrass (*Cymbopogon citratus* Stapf.) essential oil has a distinct aroma, which is similar to the original plant. The aroma comes from compounds of aldehyde function groups, namely citral as the main compound of the essential oil. Lemongrass contains 0.4% of essential oil with the main component in the form of citral as much as 70-80%. Manvitha and Bidya (2014) stated that lemongrass has several active compounds such as citronellal, citronellol, and geraniol. Meanwhile, lemongrass essential oil contained a main active compound, which named citral. Citral is a mixture of two stereoisomeric monoterpene aldehydes.

The antifungal compound has various mechanisms of fungal cells inhibition. Djunaedy (2008) stated that antifungal compound has a working mechanism by neutralizing the enzymes involved in the invasion of fungi, damaging the fungal cell membranes, inhibiting the fungal enzyme systems that disrupt the formation of hyphae tip, and affecting the nucleic acids and proteins synthesis. Ganiswarna (2004) reported that the antifungal activity of lemongrass plant is originated from its essential oil compounds. It also has a common mechanism with the antifungal from azoles group, which inhibits demethylation lanosterol into ergosterol, which is an essential sterol membrane for the growth of fungi. This inhibitory process would interfere with the function of the fungus and improve the permeability. Therefore, the citral active compound is thought to inhibit fungal as well as this azoles group antifungal.

Reported by Rahman et al. (2013), the inhibition mechanism of the antibacterial compound was explained by issuing disturbances in cell walls, increasing the permeability of cells that may cause loss of the cell components, activating enzymes, and also destructing or damaging the genetic material in cells. In the study by Fazilah et al. (2006), alginate sago starch-based edible film with lemongrass essential oil addition was able to inhibit the *Escherichia coli* bacteria at all concentrations, which varied from 0.1%; 0.2%; 0.3% to 0.4% (v/v). Inhibition zone formed on the edible film by glycerol was higher than the edible film without glycerol.

From this study, it can be concluded that the addition of lemongrass essential oil on the cassava starch-based edible film was able to increase the film thickness, increase the tensile strength and antimicrobial activity, lower the percent elongation of the film, and water vapor transmission rate of edible films were not affected by the addition of lemongrass oil.

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