

Effects of Cinnamon Bark Essential Oil (*Cinnamomum burmannii*) on Characteristics of Edible Film and Quality of Fresh Beef

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ABSTRACT

The effects of cinnamon bark essential oil on the characteristics of edible film and the quality of fresh beef were investigated to determine its ability to extend fresh beef shelf life. Films were prepared by incorporating cinnamon bark essential oil (0, 0.5, 1.0, 1.5 and 2%) into a film mixture of tapioca starch and glycerol. The selected film and control solution were applied to evaluate fresh beef quality. The concentration of cinnamon bark essential oil did not affect the thickness but significantly affected the water vapor transmission rate, tensile strength, elongation and antibacterial activity of edible film. The water vapor transmission rate was found to be lowered with the increasing cinnamon bark oil concentration. An increase of tensile strength was observed with increasing oil concentration up to 1.5%. The elongation of film was significantly reduced to 58.56% while an improved antibacterial activity of edible film resulted by oil addition. Overall, results indicate that 2% cinnamon bark oil enriched film preserves freshness of beef. The addition of cinnamon bark oil to an edible coating is effective in reducing microbial growth and lipid oxidation while the edible film enriched with 2% cinnamon

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bark oil would maintain the freshness of beef as long as 15 days during storage.

Keywords: Beef, cinnamon, edible film, essential oil, preservation

INTRODUCTION

Beef is one of the meat types most commonly consumed as a protein source globally. However, beef has a fairly short shelf life due to its high moisture content and high fat content (68% and 11%) (Gaman & Sherrington, 1994) which can facilitate the proliferation of microbial spoilage and rancidity due to oxidation reactions.

Edible films can increase the shelf life and maintain the quality of food through selective permeability to moisture and oxygen transfer, and by inhibiting the oxidation of fat and additives into the carrier material (Kester & Fennema, 1986). Additives which are usually added to the edible film are antimicrobial substances derived from spices. Spices that can be used as antimicrobial substances in the edible film are ginger essential oil (Miksusanti & Masril, 2013), nutmeg essential oil (Matan, 2012) and cinnamon essential oil, which have been applied to edible film made from whey protein (Bahram et al., 2013), apple puree (Du et al., 2009), chitosan (Hosseini et al., 2009) or gelatin-chitosan (Gómez-Estaca et al., 2010). Some other antimicrobial substances that have been used as preservatives in meat are β -CD-citral which has been added to a chitosan edible

coating (Chen et al., 2014), oregano oil in whey protein isolate films (Zinoviadou et al., 2009), and clove-oregano essential oil in a film made from milk protein (Oussalah et al., 2004).

Cinnamomum burmannii (cinnamon) is a subtropical plant from the *Lauraceae* family and reported as an important export commodity of Indonesia. Cinnamon contains active substances in the form of trans-cinnamaldehyde (60.72%), eugenol (17.62%), and coumarin (13.39%) (Wang et al., 2009). The processing of cinnamon bark into essential oils by a steam distillation resulted in active substances such as cinnamaldehyde (92.84%), cinnamyl acetate (2.34%), α -copaene (1.56%), coumarin (1.01%), delta-cadinene (0.70%), 1.8-cineol (0.66%), isopropyl acetate (0.57%), and α -terpineol (0.32%) (Khasanah et al., 2013).

The objective of this study was to investigate the effects of cinnamon bark essential oil's addition on the characteristics of edible film and the quality of fresh beef.

MATERIALS AND METHODS

Cinnamon Bark Essential Oil

Cinnamon barks were obtained from Bubakan village, Girimarto, Wonogiri (Indonesia). Essential oil was distilled from cinnamon bark using a steam distillation method. The cinnamon bark was ground in advance using a grinding machine into particle sizes of 7-15 mesh. The ground cinnamon bark was distilled for four hours (Khasanah et al., 2013).

Preparation of Edible Film

Edible films were prepared by dissolving tapioca starch (5 g) in distilled water (100 mL) at 70°C for 30 min. The solution was cooled down to 60°C and then glycerol (2 mL) was added. The solution was then heated at 60°C and stirred on a hotplate (Heidolph MR 3001 K) with a magnetic stirrer for 30 min. The solution was cooled to 30°C and cinnamon bark essential oil (0; 0.5; 1; 1.5 and 2%) and Tween 80 (1:1) were added. The solution was cast onto a 20 x 20 x 2 cm³ plastic plate followed by cabinet drying at 75°C for 5 h. The dried films were cooled for 10 minutes in ambient conditions and peeled from the casting surface. The films were stored in a plastic container containing silica gel for further analysis. Each film was prepared in duplicate.

Analyses of Edible Film Characteristics

The characteristics of edible film were analyzed based on its thickness (McHugh et al., 1994), tensile strength (Gontard et al., 1993), water vapor transmission rate (Gontard et al., 1993), elongation (Gontard et al., 1993), and antibacterial activity (Manab et al., 2011). A micrometer (Krisbow 0.001 mm) was used to determine the thickness of films at five different spots of films. Tensile strength and elongation of the films were determined with Zwicki I Z 0.5 Universal Testing Machine. For water vapor transmission rate analysis, films were sealed in test cells with silica gel inside. The test cells were then stored in jars (70% RH by 27% w/v NaCl) at 28±2°C and weighed at 1 h interval during 8 hours. The water vapor

transmission rate was calculated from the slope of the weight gain vs. time plot. Agar diffusion method was used to determine the antibacterial activity of films against *Pseudomonas fluorescens* FNCC 0071 as meat spoilage indicator bacterium. Films (5 mm diameter) were placed on nutrient agar plate containing 10⁶ CFU/mL bacteria, and incubated at 37°C for 24 h. The inhibition zones were measured in mm.

Application of Edible Film on Fresh Beef

The selected concentration (2%) of cinnamon bark oil edible films that performed the best characteristics was applied to beef. An edible film solution without oil addition was used to coat the control beef sample. The tenderloin part of beef was obtained from Jagalan Slaughterhouse (Surakarta, Indonesia) and cut into 30-45 g pieces (4 x 2.5 x 2.5 cm). The beef samples were dipped into edible film solutions and allowed to drip off and were then dried at 70°C for 1 hour. The coated samples were stored in styrofoam and wrapped with plastic in cold storage at 4 ± 1°C. Analyses were carried out periodically at 0, 5, 10, 15 and 20 days of storage. Each beef sample was prepared in duplicate.

Quality Analyses of Fresh Beef

The quality of fresh beef was assessed by considering total plate count (Utami et al., 2014), total volatile bases (Min et al., 2007) and thiobarbituric acid (Tokur et al., 2006).

Data Analysis

The data on edible film characteristics were subjected to one-way analysis of variance (ANOVA) at 0.05 significance level and differences in the mean values were determined by Duncan's test ($p < 0.05$) by SPSS Statistics 16 program. The paired-sample T-test was conducted to determine the significant difference between control and treatment beef samples.

RESULTS AND DISCUSSION

Thickness

The addition of cinnamon bark essential oil did not affect the thickness of the films (Table 1). Edible film thicknesses ranged from 0.114 to 0.176 mm. Similar thicknesses were shown by the previous research on edible films made from apple puree incorporated with cinnamon oil, ranging from 0.127 to 0.137 mm (Du et al., 2009). However, Friedman et al. (2000) reported that increasing the concentration

of cinnamon oil affected the film thickness, causing the total solids in the films to increase.

Water Vapor Transmission Rate

Water vapor transmission rate indicates the water vapor barrier properties of the edible films (Bahram et al., 2013). The results of water vapor transmission rate analysis are shown in Table 1. The water vapor transmission rate of the edible film without the addition of essential oil was 23.52 g/h.m². At a concentration of 2%, cinnamon bark oil was able to reduce the water vapor transmission rate to 21.38 g/h.m². Lower value of water vapor transmission rate indicates better protection from moisture of food. The addition of essential oils can enhance the film's hydrophobic compounds and thus reducing the water vapor transmission rate (Sánchez-González et al., 2011). This is because water vapor absorption occurs only in the hydrophilic portion of the molecule.

Table 1

Characteristics of edible film incorporated with various concentrations of cinnamon bark essential oil

Concentration of essential oil (%)	Mean thickness (mm) ^{ns}	Mean water vapor transmission rate (g/h.m ²) [*]	Mean tensile strength (MPa) [*]	Mean elongation percent (%) [*]	Mean inhibition zone (mm)
0	0.114±0.024	23.52 ^b ±0.45	0.69 ^a ±0.06	159.91 ^c ±2.44	13.75 ^a ±3.18
0.5	0.130±0.028	22.24 ^{ab} ±0.32	1.65 ^b ±0.08	96.99 ^b ±1.66	16.81 ^a ±1.86
1	0.170±0.001	22.11 ^{ab} ±0.65	1.57 ^b ±0.24	89.25 ^{ab} ±1.44	18.62 ^{ab} ±1.59
1.5	0.175±0.046	22.20 ^{ab} ±0.65	1.46 ^b ±0.07	83.29 ^{ab} ±30.93	22.65 ^b ±0.49
2	0.176±0.001	21.38 ^{ab} ±1.29	1.23 ^c ±0.09	58.56 ^a ±1.23	28.6 ^c ±0.85

Note. ^{*} Significant; ^{ns} Not Significant; ^{a-c} Means in the same column with different letters are significantly different at $p < 0.05$; ± SD values

Tween 80 also affects the water vapor transmission rate of the films. The addition of Tween 80 can increase the permeability of the film (Carneiro-da-Cunha et al., 2009). However, the results of this study are not in accordance with the opinion of Carneiro-da-Cunha et al. (2009). The water vapor transmission rate of edible film with the addition of cinnamon bark oil decreased with an increasing concentration of essential oil. Another factor that affected water vapor transmission rate is the film thickness. According to McHugh et al. (1994), in hydrophilic films there is a positive relationship between the film thickness and the water vapor transmission rate. The addition of cinnamon bark essential oil also reduced the water vapor transmission rate of edible film made of cheese whey from 22.20 g.mm/kpa.day.m² to 17.56 g.mm/kpa.day.m² and further improved the film barrier properties against water vapor (Bahram et al., 2013).

Tensile Strength

Table 1 shows the tensile strength values of the films. Tensile strength values were increased by up to 1% after the addition of essential oils but then showed a decrease with 2% additional treatment. The negative control had 0.69 MPa of tensile strength value, whereas edible films with the addition of cinnamon bark essential oils had tensile strength values between 1.23 MPa and 1.65 MPa. The improved tensile strength of the film after the addition of cinnamon oil might be caused by changes in the water

content of the film. Hosseini et al. (2009) reported that water content reduction of film made of chitosan with incorporated cinnamon oil led to a decreased tension and an increased tensile strength. The higher the concentration of essential oils in the edible film, the lower the tensile strength of the resultant film. A higher concentration of fat and *Zataria multiflora* essential oil added to film made from sodium-casein caused a decrease in the tensile strength of the film (Broumand et al., 2011). Weakening film tensile strength can be attributed to essential oil being added to the film solution, which induced the development of a heterogeneous film structure and influenced the tensile strength of the film (Zinoviadou et al., 2009). Carneiro-da-Cunha et al. (2009) reported that the addition of Tween 80 could reduce the tensile strength of film. In addition, Du et al. (2009) found out that the addition of essential oil of cinnamon could lower the tensile strength of film significantly.

Elongation

Edible film elongation percentages are shown in Table 1. The addition of cinnamon bark essential oil lowered the value of the elongation of the film significantly, from the original 159.91% to 58.56%. The addition of essential oil is able to create a compact film structure, thereby improving continuity in a network of polysaccharides, which leads to a decrease in the elongation. Cross-linker effect was produced by a strong interaction of polymer and cinnamon oil which reduced the free volume and the molecular mobility

of the polymer (Hosseini et al., 2009). However, the addition of Tween 80 affected the elongation percentage of the film negatively. Increasing the concentration of Tween 80 could cause a decrease in the elongation percentage of the film (Carneiro-da-Cunha et al., 2009). Rojas-Grau et al. (2006) reported that edible film made from apple puree with the addition of cinnamon bark essential oil had a decreased elongation percentage. Furthermore, Hosseini et al. (2009) also reported that the elongation percentage of edible film made of chitosan also decreased with the addition of cinnamon bark essential oil.

Antibacterial Activity

Antibacterial activity analysis was performed using the agar diffusion method. In an experiment by Manab et al. (2011), the analysis was performed using *Pseudomonas fluorescens* as spoilage indicator bacterium. The inhibition zones of the edible films are shown in Table 1. The inhibition zone of the edible film containing 2% of essential oils was 28.6 mm, which was higher than the negative control, with only 13.75 mm. Comparing with the control film, the addition of cinnamon bark essential oil in the edible film increased the inhibition zone between 3.06 and 14.85 mm. The higher the concentration of cinnamon bark essential oil in the edible film, the larger the inhibition zones. A higher concentration of essential oil can significantly increase the inhibition zones (Hosseini et al., 2009). Essential oil is well known for its antimicrobial compounds and its ability to control food spoilage

and the growth of pathogenic bacteria (Du et al., 2009). Cinnamon oil contains about 85% of the active antimicrobial compound cinnamaldehyde (Friedman et al., 2004). According to El-Baroty et al. (2010), cinnamaldehyde can penetrate the membrane of microorganisms and react with enzymes and proteins as well as the membrane phospholipid bilayer, which causes disruption of microbial and enzyme systems or interferes with the function of the genetic materials. Bahram et al. (2013) reported that edible films made from whey protein when added with cinnamon bark essential oil and using *Pseudomonas putida* as spoilage indicator bacterium, had increased inhibition zones from 0 mm to 22.18 mm.

Based on the analysis of the characteristics of edible films, the best essential oil concentration is 2% because the treated film exhibited higher tensile strength and antibacterial activity compared with the films applied with the other essential oil concentrations.

Total Plate Count

The TPC of beef ranged from 4.17 to 4.23 log CFU/g (Figure 1) on the first day of storage and then increased significantly over 20 days of storage. The TPC of the negative control sample rose to 7.13 log CFU/g on the 20th day of storage and beef coated with edible coating enriched with cinnamon bark oil rose to 6.03 log CFU/g on the 20th day. The TPC of beef coated with edible coating enriched with cinnamon bark oil was significantly lower compared with the

control. Chen et al. (2014) reported that in the control sample and beef coated with a chitosan solution enriched with β -CD-citral, the TPC increased during storage, but the treatment samples showed a lower TPC than the control sample.

After 20 days of storage, the TPC of all beef samples (Figure 1) exceeded the limit of microbiological quality standard of fresh beef established by the National Standardization Agency of Indonesia (Standard Nasional Indonesia [SNI] 3932:2008, 2008), which is equal to 6 log CFU/g. The control beef exceeded the limit of microbiological quality standard on day 10, with a TPC of 6.26 log CFU/g, while beef coated with edible coating enriched with essential oil exceeded the limit of microbiological quality standard on day 20, with a TPC of 6.03 log CFU/g.

Total Volatile Bases (TVB) Content

The TVB values of the meat samples are shown in Figure 2. The TVB value of the control was 12.68 mg N/100 g and then rose to 46.69 mg N/100 g on the 20th day of storage. The TVB value of the beef coated with edible coating enriched with cinnamon bark essential oil was 12.63 mg N/100 g on the first day of storage and then rose to 30.53 mg N/100 g on the 20th day of storage. The TVB value of the control was significantly higher than the TVB value of the beef coated with edible film enriched with cinnamon bark essential oil. Chen et al. (2014) reported that the TVB value of the control beef and beef with chitosan coating and β -CD-citral increased during storage, but beef with chitosan coating treatment and β -CD-citral showed lower TVB values compared with the controls. Thus, citral addition may decrease the TVB value of beef.

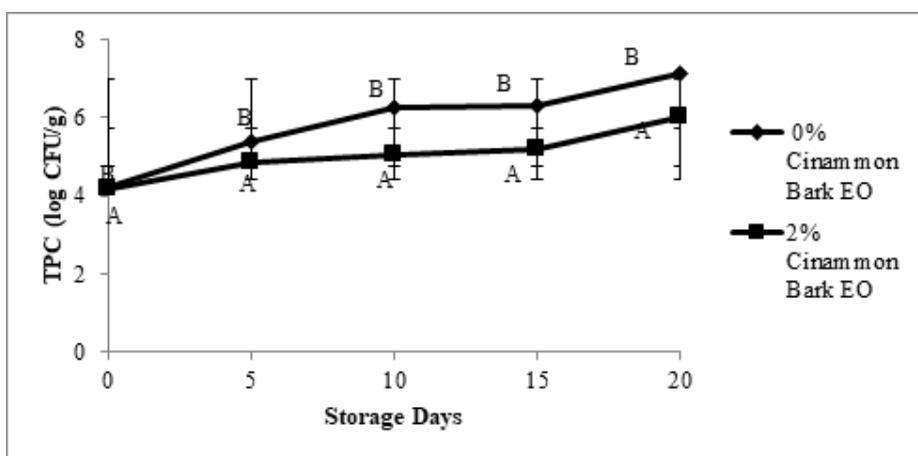


Figure 1. Total Plate Count (TPC) values of the control beef and beef coated with edible film enriched with 2% cinnamon bark essential oil during storage at low temperature ($4 \pm 1^{\circ}\text{C}$) (Means with different letters are significantly different at $p < 0.05$; \pm SD values)

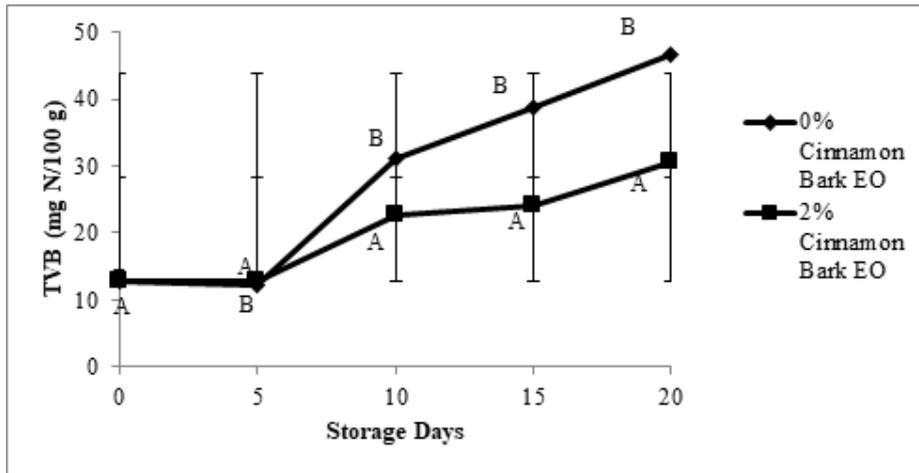


Figure 2. Total Volatile Bases (TVB) values of the control beef and beef coated with edible film enriched with 2% cinnamon bark essential oil during storage at low temperature ($4 \pm 1^{\circ}\text{C}$) (Means with different letters are significantly different at $p < 0.05$; \pm SD values)

An indication of livestock meat quality deterioration is an increase of TVB content. Xiao et al. (2014) indicated that the maximum TVB value for food products was 30 mg N/100 g. The TVB value of the beef control had exceeded the pre-defined maximum limit, which was equal to 31.10 mg N/100 g on day 10, and then to 46.69 mg N/100 g on day 20. Beef coated with edible film enriched with cinnamon bark essential oil passed the limit on day 20, with a TVB value of 30.53 mg N/100 g.

Thiobarbituric Acid

Measuring the thiobarbituric acid (TBA) value is one of the methods used to detect fat oxidation. The test is related to the level of aldehydes present in the oil. The TBA values of the control and beef coated with edible film enriched with cinnamon bark essential oil are shown in Figure 3. The TBA value of the control sample was 0.05 mg

malonaldehyde/kg on day 1 and then rose to 0.68 mg malonaldehyde/kg on the 20th day of storage. The TBA value of the beef coated with edible film enriched with essential oil was originally 0.04 mg malonaldehyde/kg and then rose to 0.47 mg malonaldehyde/kg on day 20. The TBA value of the control was significantly higher compared with the TBA value of beef coated with edible film enriched with cinnamon bark essential oil. Chidanandaiah et al. (2009) also reported that the TBA values of the control and beef with a sodium alginate coating increased during storage, but beef with sodium alginate coating treatment had a lower TBA value compared with the controls.

The maximum TBA value that is still acceptable is 2.0 mg malonaldehyde per kg of meat. If the value exceeds this level, then the meat is considered rancid and unsafe for human consumption (Campo et al., 2005). Meanwhile, according to Kuo and Chu (2003), meat products with a TBA value

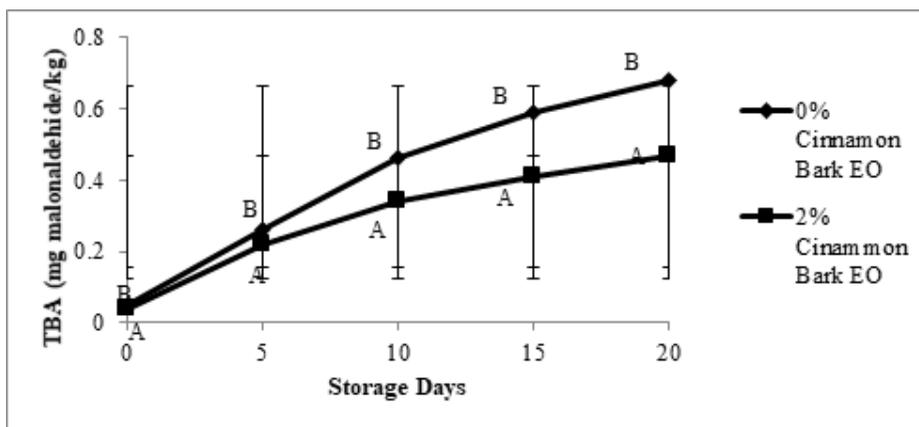


Figure 3. Thiobarbituric Acid (TBA) values of the control beef and beef coated with edible film enriched with 2% cinnamon bark essential oil during storage at low temperature ($4 \pm 1^{\circ}\text{C}$) (Means with different letters are significantly different at $p < 0.05$; \pm SD values)

between 0.5 and 2.0 malonaldehyde/kg have a rancid flavor. In the current experiment, the TBA value of the control exceeded the predetermined limit, which was equal to 0.59 mg malonaldehyde/kg on day 15 and 0.68 mg malonaldehyde/kg on day 20. These results indicate that the control beef is not suitable for consumption on day 15 of storage and onwards. However, based on TBA value, the beef coated with edible film enriched with 2% cinnamon bark essential oil is still suitable for human consumption because the TBA value has not exceeded the prescribed limit, which is equal to 0.47 mg malonaldehyde/kg.

Based on TPC and TVB analyses, the edible film enriched with 2% cinnamon bark essential oil is not suitable to preserve fresh beef at refrigerated temperature ($4 \pm 1^{\circ}\text{C}$) for 20 days. However, the quality of beef coated with edible film enriched with cinnamon bark essential oil is still maintained after 15 days storage at refrigerated temperature.

Edible film probably is more suitable to be used for processed meat rather than raw meat which could be stored at refrigerated temperature. The shelf life of cooked pork sausages coated with edible coating enriched with clove oil under refrigeration storage is 20 days (Lekjing, 2016).

CONCLUSION

Addition of cinnamon bark essential oil on the edible film does not affect the thickness of the film but it lowers water vapor transmission rate and results to elongation of the film. Also, increasing cinnamon bark essential oil improves tensile strength and antibacterial activity of the film. Overall, based on the film characteristics, a 2% cinnamon bark essential oil enriched film formula will improve preservation of fresh beef. The addition of 2% cinnamon bark oil to an edible coating is effective in reducing microbial growth and lipid oxidation as reflected in total plate count, total volatile

bases and thiobarbituric acid values of the beef samples. Thus, the application of edible film enriched with 2% cinnamon bark oil would maintain the freshness of beef as long as 15 days during storage.

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