The effect of native chicken legskin gelatin concentration on physical characteristics and molecular weight of edible film

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Abstract. Edible film is a thin layer made from polysaccharides, proteins, and lipids. These research was aimed to determine the effect of gelatin concentration on physical characteristics and molecular weight distribution of edible film produced from native chicken legskin gelatin. This research materials were used native chicken legskin gelatin and plasticizer gliserol. This study used Completely Randomized Design (CRD) with different concentration of gelatin (T1 = 5%, T2 = 10%, T3 = 15% and T4 = 20%) and five replications. The result of study described that the different gelatin concentrations had significant effect (P<0.05) on tensile strength, elongation and thickness of edible film. SDS-PAGE result showed that the band patterns of the molecules was dominated by the protein molecules of gelatin, SDS-PAGE pattern showed that do not changed of bands the type α1 and α2 of edible films. The molecular weight distribution of edible film from chicken legskin gelatin had ranges from 140 -148 kDa. The molecular weight distribution of edible film from chicken legskin gelatin had ranges from 140 -148 kDa.

1. Introduction
The development of edible films as coating and packaging materials is increasing, due to the higher human awareness of packaging materials that can be degraded to replace plastic materials that cannot be degraded [10]. Packaging is one way to maintain food quality, protect products from chemical and biological contamination, as a protector of food because it can prevent the migration of water vapor, gas, fat and aroma from ingredients to the environment and prevent damage caused by microbes [8]. One type of packaging that is environmentally friendly is edible packaging, it can protect food products, be edible and safe for the environment. In the most recent years, food and packaging industries have been joining efforts to reduce the amount of food packaging materials. An edible film could be defined as primary packaging made from edible components. Edible film is a thin layer of edible coatings and are often used as food, and able to be a barrier of moisture, oxygen, mechanical properties, sensory, convenience, and prolong the shelf life of various products [3, 7, 11].

Edible polymers such as polysaccharide, protein, and lipid are the three main ingredients used to produce edible films [12]. Edible films have the same properties as packaging films such as plastics, which must have water retaining properties so they can prevent product moisture, control the transfer
of dissolved solids to maintain color, natural pigments and nutrients and improves food quality. The applications of edible films and coatings include fresh produce coatings of sausage casings from collagen [10]. The main ingredient for making edible films is gelatin [1, 21, 22, 27]. Gelatin is a protein obtained by boiling skin, tendons, ligaments, and bones with water [16], that can be obtained from collagen. Its functional properties depend on processing conditions as well as the raw material [17, 21, 24]. The gelatin quality based on its physical and chemical properties and rheological properties [20, 23]. Gelatin-based films are thin. The addition of plasticizer, make changes some of the functional and physical properties of these films. Edible films from hydrocolloid have a good ability to protect products from oxygen, carbon dioxide and fat and have mechanical properties that can improve the structural integrity of products that are easily damaged. Edible films from gelatin or collagen have good potential [26]. Plasticizer from the group of polyols are widely used to improve the mechanical. This polyol gliserol as plasticizer can lower the modulus properties of tensile strength and increase elongation film [9]. Effects of different concentration of gelatin to produce edible film from native chicken legskin was limited information. If viewed from the chemical composition, native chicken legs skin has a protein content of 22.98%, 5.60% fat content and 3.49% ash content [9] This study was investigated the effect of native chicken legskin gelatin concentration on the physical characteristics and molecular weight of edible film.

2. Materials and methods
Three thousand grams of native chicken legskin gelatin were used as a raw material, plasticizer glycerol and distilled water.

2.1. Process of gelatin
Gelatine was prepared by the acid extraction method with 3% acetic acid (v/v) and distilled water [4]. 100 grams native chicken legskin as raw material were soaked for 24 hours. After soaked, samples were neutralized to pH 6 and extracted [5]. The extraction temperature were performed at 55°C. Solubilized gelatin was separated from residual skin fragments by filtration through a filter. The extracted gelatin was concentrated at 70°C for 5 hours then dried at 60°C for 24-48 hours until the gelatin sheet was dried and solid. Gelatin sheets were milled and packaged in vacuum plastic, stored in a desiccator for subsequent process [9, 22].

2.2. Preparation of edible film
The process of edible film made from native chicken leg skin gelatin according to the method of Sobral et al. (2001) as follows: the concentration of film-forming solution was prepared based on treatment by dissolving 5, 10, 15 and 20 grams gelatin in 100 ml distilled water in accordance with the treatment specified, then added plasticizer glycerol with concentrations 10% (w / w) then dissolved in a water bath at a temperature of 50 °C while stirring for 30 minutes until the gelatin granules and homogeneously mixed plasticizer [2]. Homogenized for 5 minutes at room temperature. The solution was subsequently cast films formed in a Teflon and dried using an oven at a temperature of 55°C for 18-20 hours. Sheet edible film formed was wrapped with clear plastic and stored in a container with silica gel, then analyzed according to the treatment [18].

2.3. Experimental Design.
This research were determined by analysis of Completely Randomized Design (CRD) 4x5 with different concentration of gelatin (T1 = 5%, T2 = 10%, T3 = 15% and T4 = 20%) and five replications. The significant differences were determined using Duncan’s new multiple range test [25].
2.4. Parameters.
The characteristics parameters of this study were film thickness, tensile strength, elongation at break and molecular weight distribution. The thickness film was measured at five different points for each sample to be tested. Tensile strength is the maximum pull that can be achieved through film to survive before breaking, the elongation percentage is calculated based on the length of the film.

The molecular weight distribution of gelatin was determined according to Carvalho [4] using Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis (SDS-PAGE). Edible film 0.01 g was added with 1 ml of distilled water then homogenized. Then dilution of dissolved protein was carried out as much as 500 g at every 0.1 ml of the solution which was added with 0.4 ml of distilled water, then continued with a diluted sample of centrifuged at 3000 rpm at room temperature for 10 minutes. The supernatant 0.015 ml which was added with 0.015 ml of the buffer sample then heated at 100°C for 2 minutes and cooled at room temperature. The sample was taken 0.01 ml, and running ends after the sample dye reaches the bottom of the gel.

Molecular weight was identified by means of gel soaked with commasie brilliant blue dye solution for 12 hours, slowly agitated, then the gel was dried and pressed with plastic until it became stiff and could be stored for a long time.

3. Results and discussion

3.1. Thickness
The film thickness will affect the physical properties and the rate of water vapor edible film (Bergo and Sobral, 2007). Statistical analysis showed that different gelatin concentration had significant effect (P<0.05) on edible film from native chicken legskin gelatin. This structure film is formed by the matrix protein interactions proteins catalyzed by heat that is hydrophobic bonds, hydrogen bonds and disulfide bonds [6]. Factors affecting edible film thickness is the concentration of dissolved solids in solution films. The higher concentration of dissolved solids, the film thickness increases.

| Gelatin concentration (%) | thickness (%) | Tensile Strength (MPa) | Elongation at Break(%) |
|---------------------------|---------------|------------------------|-----------------------|
| 5                         | 0.112±0.02"a" | 5.713±0.218"a"        | 57.110±0.230"a"      |
| 10                        | 0.123±0.05"b" | 4.512±0.018"b"        | 59.210±0.210"b"      |
| 15                        | 0.127±0.01"b" | 4.271±0.005"b"        | 61.310±0.061"c"      |
| 20                        | 0.128±0.01"b" | 4.751±0.203"b"        | 61.712±0.170"c"      |

Different letters indicated the significant differences (P<0.05), Sd = standard deviation

3.2. Tensile Strength
The tensile strength is the ability of a material to resist breaking under tensile stress. The average tensile strength of edible film from native chicken legskin gelatin was displayed in Table 1. Statistical analysis showed that the different concentration of native chicken legskin gelatin had significant effect (P<0.05) on edible film. An increase in the concentration of gelatin in a solution decreases the tensile strength of the film. This is due to the destability of the film matrix by an increase in the components of hydrophilic proteins in the edible film structure [2].

3.3. Elongation at break
The elongation at break of an engineering material is the length increase, expressed as a percentage, which occurs before it breaks under tension. Edible film with gelatin 20% (61.712±0.170) was higher EB than edible film with gelatin 15% (61.310±0.061), gelatin 10% (59.210±0.210) and gelatin 5% (57.110±0.230). The more powerful the film is formed, the more difficult for the film to elongate, that
it will reduce the value of percentage elongation, increasing TS is accompanied by a decreases elongation at break.

3.4. Molecular Weight Distribution

Based on Figures 1, it can be seen that in general the protein molecular pattern is clearly visible for the four samples (A,B,C,D and M as marker). It can be seen that some bands have not been able to provide real line information, but in general the band's appearance can already illustrate the pattern of molecular weight distribution produced by gelatin. The whole band showed that from the treatment applied, the molecular weight distribution of edible film from native chicken legskin gelatin had ranges 140-148 kDa. Color thickening in some bands can be caused by a less than optimal protein preparation process, but at least that information on the BM distribution pattern of the tested product can still be estimated based on the comparison band of marker (M). SDS-PAGE result showed the band pattern of the molecule was dominated by the protein molecules of gelatin, SDS-PAGE pattern showed no changed of bands the type α1 and α2 of edible films. The distribution of molecular weight was related to the length of the amino acid bond chain and the gel strength ([15, 19]). The dark colored ribbon shows the molecular weight of edible film was good. The magnitude of this molecular weight will affect all the physical characteristics of edible film. The bigger molecular weight had a longer amino acid bond chain and higher gel strength [19, 20].

![Figure 1. SDS-PAGE (Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis) edible film from native chicken legskin gelatin](image)

M = Marker  
A = gelatin concentration 5%  
B = gelatin concentration 10%  
C = gelatin concentration 15%  
D = gelatin concentration 20%

4. Conclusion

It was concluded that the use of native chicken legskin gelatin 10% had the best physical characteristics of edible film (thickness mm 0.123±0.05, tensile strength 4.512±0.018, elongation at break 59.210±0.210% and molecular weight distribution 148 kDa).
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6. References
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