Quality Protein, Viscosity, Gel Strength and Structural Morphology of Sheepskin Gelatin Catalyzed HCl With Different Concentrations

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Abstract. Sheep skin has a high content of collagen and can be an alternative to gelatin making. Utilization of gelatin increasingly widespread so that it becomes lifestyle. Gelatin is often used for food or non-food. This study aims to determine the effect of HCl concentration on the quality of sheepskin gelatin. Sheep skin obtained from slaughterhouses in Brebes district. Parameters in this study were yield, protein, viscosity, gel strength and gelatin morphological structure. Different HCl concentrations (0.5%, 1% and 1.5%) are used to catalyze collagen from sheepskin. Hydrolysis of sheep skin collagen was performed for 4 hours with temperature 55-66°C. The results showed no significant effect on quality of protein and yield but significant effect on viscosity and gel strength. The observations by scanning electron microscopy (SEM) show a very fine morphological structure of gelatin produced with 1% and 1.5% HCl catheters. Gelatin catalyzed with HCl 1% and HCl 1.5% was appropriate standard GMIA.

1. Introduction
Gelatin is a type of collagen protein obtained by extracted from the skin. One of the alternative ingredients of gelatin processing is sheepskin. Sheepskin has a high content of collagen and can be an alternative to make gelatin. Gelatin includes a unique protein because it is capable of forming a thermo-reversible gel and is easily soluble in water [1]. Utilization of gelatin increasingly widespread to become a lifestyle. Gelatin is often used for food or non-food. Sheepskin gelatin should have the appropriate characteristics if it will be applied in the food or non food industry and should be in large quantities and the price is also cheap so it can be produced continuously. The use of chemical solutions to help the process of skin collagen extraction. Usually use acid or alkaline chemical solutions. The quality of gelatin catalyzed with an acidic chemical solution is preferable when compared to a gelatin catalyzed with alkali chemicals. The use of a solution of hydrochloric acid as a catalyst of sheep skin collagen protein can convert triple helices of collagen into a single chain in a short time and produce more collagen [2]. Immersion in an acid solution to collagen can produce gelatin polymers with glycine as the main constituent. Setting the concentration of hydrochloric acid solution in catalyzing collagen can affect the quality of gelatin produced. This study aims to determine the effect of HCl concentration on the quality of sheepskin gelatin.
2. Materials and Methods

The raw materials are prepared through a series of preliminary beam house operations from raw sheep skin, weighing, washing, soaking, discarding, discarding wool, disposing of fat to a clean sheep's skin (no hairs) and conditioned in a neutral atmosphere. Sheepskin without hair is cut into small pieces 2x2 cm. These pieces of sheepskin are used as raw materials of gelatin. Small cut sheepskin was immersed in 0.5%, 1% and 1.5% (v/v) HCl solutions for 4 hours. After the immersion process is complete, the skin is washed with running water repeated three times until the pH is neutral. The washed skin is then extracted temperature 55–60°C for 4 hours. Comparison of sheepskin: aqueous solution = 1: 3 for each treatment. Then do the filtration process to get collagen solution. The gelatin solution obtained for each ±300 ml was poured into a 30.5 cm x 30.5 cm container, then dried in a 60°C oven for 48 hours. The gelatin obtained is then mashed using a blender. The parameters used in this study were yield, protein content [3], gel strength and viscosity [4], and Scanning electron microscopy [5]. This research uses Completely Randomized Design (RAL) unidirectional pattern. The treatments were concentration of HCl solution 0.5%, 1% and 1.5% (v/v) and replication 5 times. If there is a marked difference between treatments, then a real difference test according to Duncan's Multiple Range Test [6].

3. Results and Discussions

3.1. Yield

Based on the result of data analysis in Table 1. showed the difference of HCl concentration in sheep skin catalysis there was no difference (P> 0.05) to sheep skin gelatin value. The average yield value generated in this study ranged from 23.10–23.33%. this shows the difference in HCl concentration having the same effectiveness in producing gelatin. In principle, the acid solvent causes the structure of collagen protein in sheepskin expands and opens so as to facilitate the process of collagen hydrolysis into gelatin [7]. Not the yield difference may be caused by long time soaking in an acid solution and the same 4-hour long extraction is also the same 4 hours. This process occurs due to the number of H⁺ ions hydrolyze more collagen, while the longer the extraction time can lead to the disintegration of collagen which is more into gelatin. Extraction serves as the advanced stages of the hydrogen bonds between molecules damage tropocollagen that at the time of preparation not previously been decomposed by the acid. H₂O molecules can denaturant hydrogen bonds in tropocolagen [2].

Table 1. Average of yield, protein content, gel strength, viscosity of sheepskin gelatin with different concentration HCl treatment

| Parameters                  | Concentration of HCl | Commercial Gelatin | GMIA Standard |
|-----------------------------|----------------------|--------------------|---------------|
|                             | 0.5%                 | 1%                 | 1.5%          |                |
| Yield (%)                   | 23.21 ± 0.21a        | 23.10 ± 0.11a      | 23.33 ± 0.15a | -             |
| Protein content (%)         | 90.93 ± 0.024        | 90.59 ± 0.034      | 90.98 ± 0.01b | 90.02 ± 0.01   |
| Gel strength (Bloom)        | 371.908 ± 0.637a     | 411.269 ± 0.586b   | 64.821 ± 0.750c | 191.416 ± 0.362 |
| Viscosity (cP)              | 13.55 ± 0.054        | 4.44 ± 0.066b      | 2.51 ± 0.01c  | 29.15 ± 0.06  |

Different letters within same column denote significant differences (P < 0.05). Mean ± SD from duplicate determinations

3.2. Protein Content

Different concentrations of HCl have the same effectiveness in producing protein content. This is seen in the results of statistical analysis showing that protein content in this study did not have differences (P> 0.05) between treatments, protein content of this study is directly proportional to yield. The average value of the resulting content proteins is 90.59% - 90.98% even still higher when compared with protein content of commercial gelatin. This shows the process of HCl catalysis of sheepskin
collagen is very good, so as to make the sheep skin to expand which ultimately facilitate the process of collagen hydrolysis. In principle, the quality of the gelatin protein is strongly influenced by the hydrolysis process, the extraction process and the use of chemical solutions as the main treatment [2].

3.3. Gel Strength
The gel strength of sheepskin gelatin produced in this study showed a significant difference (P>0.05) between treatments. Sheepskin gel treated with HCl 1.5% solution had a median strength of 64,821 Bloom and still met the GMIA standard, whereas sheepskin gel gelled with 0.5% HCl solution resulted in average gel strength 371,908 Bloom and sheepskin gelatin which catalyzed with a 1% HCl solution having an average gel strength of 411,269 Bloom. Gelatin gel strength of 0.5% HCl solution catalyst and 1% HCl solution did not meet GMIA standard. This phenomenon occurs due to HCl solution that is able to convert triple helical sheep skin collagen into single chain but display the amino acid chain different between treatments. The main factor that distinguishes the quality of gel strength is the length of its amino acid chain, and if its collagen condition has been perfectly hydrolyzed, resulting in a long polypeptide chain of hydroxyproline that increases gel strength [8].

![Figure 1](image.png)

Figure 1. Structural Morphology of sheepskin gelatin with Scanning Electron Microscopy (SEM) 5000x magnification

3.4. Viscosity
The results of viscosity analysis presented in Table 1. showed a significant difference (P <0.05) between treatments, which showed a decrease in the mean value of viscosity as the concentration of HCl solution as catalyst increased. Visible viscosity values of GMIA are sheepskin gelatin catalyzed with 1% HCl and 1.5% HCl. The mean value of sheep skin gelatin viscosity catalyzed by HCl solution in this study is still better when compared with the value of commercial gelatin viscosity. This event is
caused by the suspected decrease in the viscosity value of gelatin produced due to the decrease of pH in the addition of HCl concentration causing the amino acid chain to be shorter so that the viscosity of the gelatin becomes lower [9]. The high concentration of HCl in the acid process will have an effect on the sheep skin collagen change. Collagen structure is the main compiler is hydroxyproline swelling and become spread so causing the resulting viscosity value changes. Changing collagen chain structure causes a decrease in the molecular weight of gelatin [8].

3.5. Structural Morphology
Based on the results of Scanning electron microscopy (SEM) can be seen differences in morphological structure of sheepskin gelatin. The gelatin morphological structure in the catalyst with 1% HCl solution is noticeably smoother and denser without the presence of collagen protein clumps. In contrast to the gelatin SEM results categorized with 0.5% HCl there is still a lump of protein and there are still cracks. Results of gelatinized SEMs categorized with 1.5% HCl were still better than 0.5% catalytic gelate of HCl. The result of commercial SEM gelatin still shows the number of collagen protein clumps and uneven surfaces. The SEM results provide a reinforcing explanation that the sheepskin gelatin that catalyzes with 1% HCl solution is better.

Clumps of protein will make gelatin morphological structure becomes uneven, while cracks caused by not compact the collagen into gelatin protein binding. This morphological structure will describe the choice of using gelatin to be applied to the next stage.

4. Conclusion
Gelatin catalyzed with HCl 1% and HCl 1.5% was appropriate standard GMIA. Gelatin catalyzed with HCl 1% showed a finer morphological structure based on SEM results.

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