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The Effect of Lemon (*Citrus limon*) as a Coagulant on Fresh Cheese Chemical Composition and Storage

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Abstract. Demand for fresh cheese increases along with welfare. As Indonesia has a muslim population around 200 millions therefore there is a demand for halal cheese starter. The purpose of this research was to prove that the use of lemom juice as a replacement rennet is able to coagulate milk while produce fresh cheese which is also a halal because it comes from plants.This study aims to determine the lemon juice (*citrus limon*) in the process of milk coagulation. This research method used a 4 x 4 Factorial Completely Randomized Design with 2 replications of factor A, levels of lemon juice, namely A0 = 0.01% rennet, A1 = 5% lemon juice, A2 = 10% lemon juice, A3 = 15% lemon juice, and factor B storage time B0 = 0 days, B1 = 5 days, B2 = 10 days and B3 = 15 days. The variables measured were chemical compositions including moisture content, dry matter content, fat content, protein content. Based on the research results, it was found that from the addition of four levels of lemon juice used and four variations of storage time, giving 10% lemon juice (A2) and storage time of 15 days (B3) was the optimal treatment to increase moisture content, dry matter content, fat content, protein content in cheese and can be used as an alternative of halal cheese starter.

Keywords: fresh cheese, goat's milk, lemon juice

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1. Introduction

One of the livestock commodity products which has quite a demand is milk. Milk is one of the energy sources needed by the body and is a highly nutritious food ingredient. The main composition of milk is often defined as the content of fat, protein, lactose, ash and total solids (total solid). Milk also contains small amounts of mineral salts, pigments, enzymes and vitamins. Milk plasma is milk that does not contain fat globules with a composition almost the same as skim milk meanwhile skim milk contains fat resulting from incomplete separation processes.

Milk serum is milk plasma without casein micelles with a composition almost the same as whey with the difference that whey still contains various proteolytic products from the chymosin enzyme [1]. Among the various types of milk produced by mammals, goat milk is milk that has the most benefits for human health. Goat milk is very good for people who have lactose intolerance disorders, because the lactose in goat milk is lower (10%) than cow milk [2]. In addition, goat milk fat globules have small particle diameters compared to cow milk [3], so that

goat milk is easily digested and absorbed by the body to produce energy compared to cow's milk. One of the alternative products produced by goat milk is cheese.

Cheese is one of the dairy products known by the Indonesian people which is useful because it is long lasting and contains high fat, protein, calcium and phosphorus. The price for cheese until now is relatively expensive but the need is quite large and continues to increase. In 2002, the national cheese consumption was 8000 tons per year, an increase of 20% compared to 2001 [4]. Although it is expensive, the amount of use of cheese by the community is quite large, both for the components of the mixture in making cakes, as well as for sprinkling bread.

Based on cheese making, cheese is made from fresh cow milk as raw material, however some people cannot consume cow milk because cow milk contains higher lactose than other animal milk. Unlike cow milk, goat milk does not contain agglutinins. As a result, goat milk fat globules are not clustered so that they are easier to digest. Goat milk contains lactose which is slightly lower than cow milk. This condition is very good for people who are lactose-intolerant [5].

One of the important things in the cheese industry is the availability of a coagulant, namely rennet. Rennet is a casein clot in the cheese making process which contains the enzyme rennin protease. The rennet enzyme used is a proteolytic enzyme that can coagulate milk in the initial process of making cheese. According to its development, the rennet enzyme is divided into 4 main types, namely: rennet from ruminant stomachs, microbial rennet, fermentation produced chymosin (FPC) with genetic engineering, and vegetable rennet.

Because the ingredient for making cheese (rennet) is quite expensive and production is scarce, some dairy nutritionists are trying to make vegetable rennet as a substitute for commercial rennet. Rennet enzymes from the stomach of ruminants have an impact on the high price of cheese. Rennet is generally produced from non muslim countries and in the process of making rennet, there is a possibility for ruminant to be slaughter without halal process.

An alternative to rennet enzyme is the use of lemon juice (Citrus limon). Lemon is a type of citrus fruit that tastes sour with a high content of vitamin C. Lemon juice contains 5% acid which has many uses in everyday life in the household. The lemon component that plays a role in the coagulation process of milk is pectin. In making cheese, it is hoped that it can be used as an alternative to coagulate in cheese making because the addition of acid will cause the protein coagulation process and can reduce costs, and improve cheese storage time.

2. Materials and Methods

2.1. Materials

The research used fresh goat milk and lemon juice fas coagulant for making fresh cheese. "Figure 1" shows fresh cheese production process.

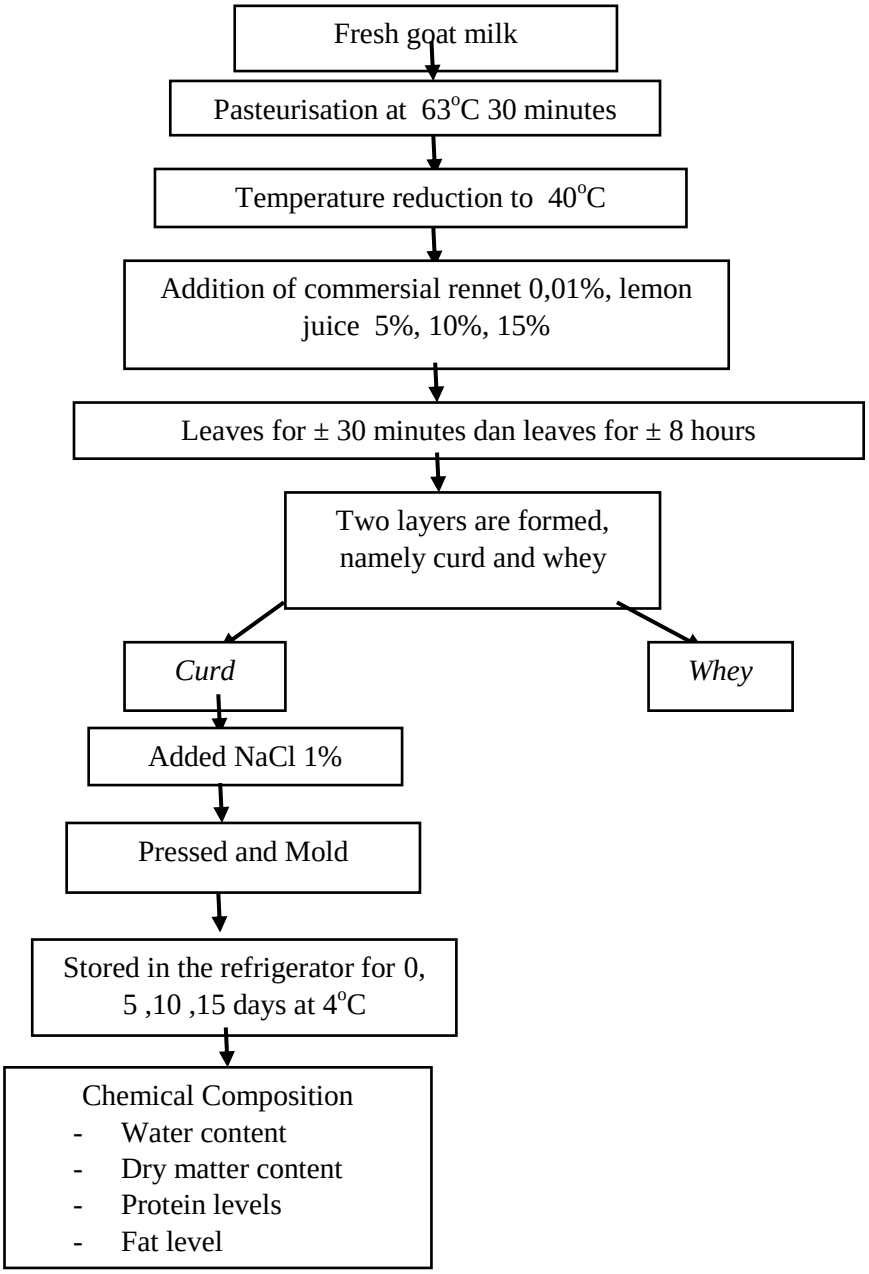

Figure 1. Scheme of Fresh Cheese Production Process

2.2. Methods

The research design used was a Completely Randomized Design (CRD) Factorial pattern with two factors, which were tested with the following treatments :

Factor A (lemon juice level)

A0 = 0.01% (using commercial rennet)

A1 = 5% of the level lemon juice

A2 = 10% of the level lemon juice

A3 = 15% of the level lemon juice

Factor B (storage time)

B0 = 0 day storage

B1 = 5 days storage

B 2 = 10 days storage

B3 = 15 days storage

Research Parameters

Chemical Content Test

1. *Water Content*

The water content of the sample was calculated by the formula:

$$M = \frac{A - B}{A} \times 100\%$$

Note:

M: Sample water content

A: Sample weight

B: Sample weight after drying

2. Dry matter content

The dry matter content was calculated by using the formula:

Dry matter content = 100% - Water content

3. Protein Levels content using the Kjedhal method.

The percentage was calculated by using the formula:

$$\% N = \frac{(\text{ml NaOH blank} - \text{ml sample NaOH}) \times 14.008 \times N\ NaOH}{\text{Sample weight}} \times 100\%$$

Protein levels =% N x 6.38

Note:

% N: Percentage of nitrogen

KP: Protein content

14.008: Molecular weight of nitrogen

N NaOH: Normality of NaOH

6.38: Cheese conversion factor

4. Fat level content by extraction according to Soxhlet.

The sample was weighed as much as 5 g and dried in an oven at 100ºC until constant weight, then put into a tube of filter paper and put into a Soxhlet containing petroleum solvent. The material was then extracted for 3 hours, then the material was dried in an oven for 45 minutes

and put in a desiccator for 15 minutes, weighed until its weight was balanced. The difference in weight before and after extraction per sample weight was the percentage of fat extracted.

$$\% \text{ fat} = \frac{a - b}{} \times 100\%$$

Note : a: Weight of fat flask + extracted fat (g)

b: Weight of empty fat flask (g)

c: Weight of sample (g)

3. Result and Discussion

Tabel 1. Water Content, Dry Matter, Protein Level and Fat Level of Fresh Cheese by Using Lemon Juice as Coagulant

Treatmeant	Water content	Dry matter	Protein level	Fat level
A0B0	$32,53^{dD}$	$67,47^{aA}$	$20,38^{cD}$	$23,22^{cC}$
A0B1	$34,72^{dC}$	$65,28^{aB}$	$21,56^{cC}$	$24,37^{cB}$
A0B2	$38,94^{dB}$	$61,06^{aC}$	$22,40^{cB}$	$25,98^{cA}$
A0B3	$41,17^{dA}$	$58,83^{aD}$	$23,04^{cA}$	$20,19^{cD}$
A1B0	$34,98^{bD}$	$65,03^{cA}$	$22,93^{bD}$	$28,87^{aC}$
A1B1	$38,62^{bC}$	$61,38^{cB}$	$24,03^{bC}$	$29,92^{aB}$
A1B2	$42,07^{bB}$	$57,94^{cC}$	$25,05^{bB}$	$30,56^{aA}$
A1B3	$46,12^{bA}$	$53,88^{cD}$	$26,21^{bA}$	$26,10^{aD}$
A2B0	$34,25^{cD}$	$65,75^{bA}$	$24,44^{aD}$	$19,95^{dC}$
A2B1	$36,56^{cC}$	$63,45^{bB}$	$26,16^{aC}$	$21,06^{dB}$
A2B2	$39,54^{cB}$	$60,46^{bC}$	$27,60^{aB}$	$23,33^{dA}$
A2B3	$41,71^{cA}$	$58,29^{bD}$	$27,01^{aA}$	$19,36^{dD}$
A3B0	$35,64^{aD}$	$64,37^{dA}$	$21,59^{cD}$	$26,49^{bC}$
A3B1	$39,61^{aC}$	$60,39^{dB}$	$21,95^{cC}$	$27,65^{bB}$
A3B2	$43,81^{aB}$	$56,09^{dC}$	$22,41^{cB}$	$28,86^{bA}$
A3B3	$49,12^{aA}$	$50,88^{dD}$	$22,88^{cA}$	$24,61^{bB}$

Note : Different super scripts in the same table show significant differences ($P < 0,01$). Lowercase notation indicates lemon level, uppercase notation indicates storage period

3.1. Water Content

The levels of lemon juice 5%, 10% 15% on goat cheese showed a very significant effect ($P < 0.01$) on water content. The moisture content in cheese with commercial rennet coagulation (0.01%) ranged from 32.53 - 41.17% while the protein in cheese with lemon juice coagulation ranged from 34.25 to 49.12%. In the manufacture of fresh cheese the expected moisture content is a maximum of 80% [6].

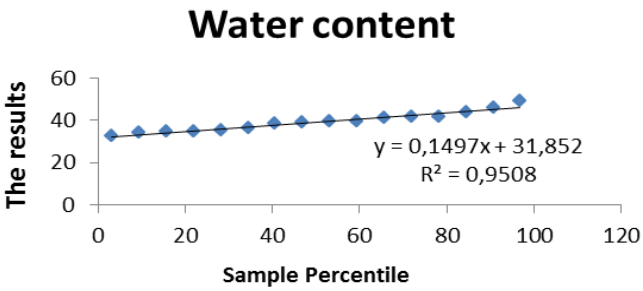

Figure 2. Water Content Regression Graph of Fresh Cheese with Lemon Juice Coagulant

From "Figure 1", it can be seen that the most significant increase in water content occurred in treatment A3, namely 49.12%, but in fresh cheese the expected water content was low water content found in treatment A2, namely 34.25%. The difference in water content of the cheese was thought to be caused by the presence of water in the cheese which were in three levels, namely 1) bound to the curd component structure, 2) retained in the curd particles which are hygroscopic and 3) free water. The presence of free water in the curd was influenced by the degree of emphasis on whey removal. The lower the water content, most likely because the greater the acid concentration used, the pH will decrease. Acidic conditions will help the process of particle shrinkage to remove more whey, so that more water is released [7]. The interaction between 5%, 10%, 15%, lemon juice and storage time had a significant effect (P <0.01) on the water content of the cheese. As mentioned by [8], classifying types of cheese based on the amount or water content contained in cheese, namely cheese is very soft (55-80%), soft (45- 55%), hard (34- 45%) , and very hard (13- 34%). The factor of adding 10% lemon juice and 15 days of storage time wass considered to reduce the water content of fresh cheese.

3.2. Dry Matter Content

Application of lemon juice 5%, 10% 15% to cheese from goat's milk showed a very significant effect (P <0.01) on dry matter content. Dry matter content in cheese with commercial rennet clotting with storage time variants ranged from 58.83 - 67.47% while the dry matter content in cheese with lemon juice coagulation with storage time variants ranged from 50.88 to 65.75%. The same thing also happened in research by [9] that the dry matter content of cheese produced in the study ranged from 39.82 to 49.72%, the less water content in the cheese caused the dry matter content high.

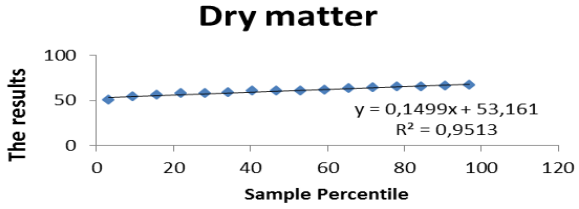

Figure 3. Dry Matter Regression Graph of Fresh Cheese with Lemon Juice Coagulant

From “Figure 2” it is known that the most significant increase in dry matter content occurred in treatment A2, namely 65.75%. Low water content means high dry matter content. Dry matter is one way of measuring what is contained in food ingredients. Dry matter contains organic compounds such as fat, protein, fiber, carbohydrates, and inorganic compounds such as minerals [10]. The interaction between lemon juice level and storage time was very significant (P <0.01) on dry matter content in cheese. It is also suspected by Scoot (1981) that the presence of free water in cheese was influenced by the degree of pressure when removing whey. The factor of adding 10% lemon juice and 15 days of storage time was considered to increase dry matter content in fresh cheese.

3.3. Protein Level

The results of protein analysis with the administration of 0.01% commercial rennet and several levels of lemon juice 5%, 10%, 15% and storage time of 0, 5, 10, 15 days showed that the administration of lemon juice and commercial rennet to goat milk showed the results were very significant differences (P <0.01) on protein content.

According to [11], one of the main components in milk is protein, which is 3.25%. In addition, [12] mentioned that the composition of cheese was very much influenced by the type of milk and the composition of the milk used, the better the composition of the milk contained, the better the quality of the cheese produced.

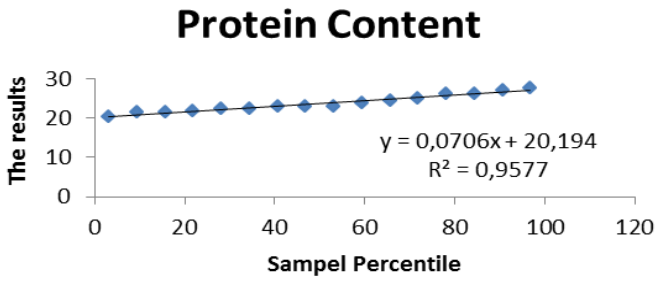

Figure 4. Regression Graph of Protein Content of Fresh Cheese with Lemon Juice Coagulant

From “Figure 3”, the most significant increase in protein content occurred in treatment A2, namely 27.60%. The factor of adding 10% lemon juice and 15 days of storage time was considered to increase protein content in fresh cheese. The interaction between lemon juice level and storage time had a significant effect (P> 0.01) on the protein in cheese. This was because the microbial activity was still working during storage which in turn will change the composition of the cheese. [13] stated that during storage the microbes change the composition of cheese.[14] explained, during storage there was a biochemical change in cheese.

3.4. Fat Level

Fat is a second energy-rich food source for humans. Fat consists of fatty acids which combine with glycerol molecules to form triglycerides which are encased in a phospholipid-protein membrane, forming fat globules that cannot combine with one another. Acid and proteolytic activity which act on the coagulation process of milk causes the destruction of the phospholipid-protein layer so that the fat globules will be trapped when the protein is packed and finally unite with the *curd* [15].

The results of the analysis of fat content with the administration of 0.01% commercial rennet and several levels of lemon juice 5%, 10%, 15% and storage time of 0, 5, 10, 15 obtained the fat content in cheese with commercial rennet coagulant with variants of storage time ranging 20.19 - 25.98% while the fat content in cheese with lemon juice thickener with storage time variants ranged from 19.36 - 30.56%.

The level of lemon juice administration had a very significant effect (P <0.01) on the fat in cheese. The fat content of cheese in the treatment was higher than cheese with variations in the level of lemon juice. This was due to the acid from the lemon juice which can break down fat. Acids in local lime (*Citrus aurantifolia swingle*) has the highest citric acid content compared to other types of citrus. Citric acid in local limes was 55.6 g / kg, while lemons were 48.9 g / kg and Bangkok lime 38.6 g / kg. Citric acid is a water-soluble organic acid. Citric acid is believed to dissolve fat. Much of what is used can affect the value of fat content, because the citric acid in oranges can bind fat.

According to [16], hydrolysis of fat produces volatile fatty acids, volatile fatty acids which result in reduced fat in cheese. Volatile fatty acids are volatile fatty acids, such as butyric caproate, caprichate and caprate. Volatile fatty acids play a role in the formation of cheese aroma. The amount of fat trapped in cheese affects the fat content of the milk used as its raw material [17].

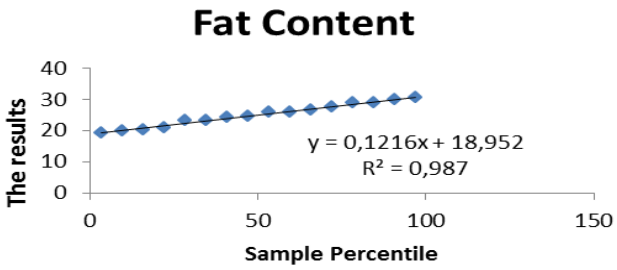

Figure 5. Regression of Fat Content of Fresh Cheese with Lemon Juice Coagulant

From "Figure 4", that the most significant increase in fat content occurred in treatment A1, namely 30.56%, but in fresh cheese the expected fat was low fat content found in treatment A2, namely 19.36%. The factor of adding 10% lemon juice and 15 days of storage time is considered to reduce the fat content in fresh cheese. and the interaction between the dose of

lemon juice and the length of storage had a very significant effect (P <0.01) on the fat in cheese. This was because microbial activity is still working during storage, which in turn will change the composition of the cheese.

The fat content of processed cheese depends on the natural milk fat content used, but in the process of making processed cheese there is a possibility that fat will come out of the processed cheese during the processing process. It is suspected that adding 10% lemon juice causes a lot of whey to come out. Generally whey not only contains protein but also contains fat**.**

3.5. Recapitulation of Research Results

Based on the results of the study, it was found that the making of fresh cheese with lemon juice coagulants had an effect on moisture, dry matter, protein and fat content.The effect of using 0.01% rennet (A0) on the content of fresh cheese were as follows: the moisture content 36.84%, dry matter 63.16%, protein 21.84% and fat 23.44%. The effect of using lemon juice level 5% (A1) on fresh cheese content were dry matter 40.45% , water content 59.56%, protein 24.55% and fat 28.86%. The effect of using lemon juice level 10% (A2) on fresh cheese content were water content 38.01%, dry matter 61.99%, protein 26.30% and fat 20.92%. The effect of using the 15% (A3) level of lemon juice on the content of fresh cheese were water content of 42.04%, dry matter 57.93%, protein 22.21% and fat 26.90%.

The storage time by giving commercial rennet (0.01) and giving lemon juice levels (5%, 10%, 15%) for 0 days (B0) to the content of fresh cheese, namely the moisture content of 34.35%, 65 dry matter 65%, 22.33% protein and 24.63% fat. In the storage time for 5 days (B1) the content of fresh cheese were water content 37.38%, dry matter 62.62%, protein 23.43% and fat 25.75%. In the storage time for 10 days (B2) the content of fresh cheese were water content 41.09%, dry matter 58.89%, protein 24.36% and fat 27.18%. At the storage time for 15 days (B3) the content of fresh cheese were water coontent 44.53%, dry matter 57, 93%, protein 24.78% and fat 22.57%.

The use of commercial rennet coagulants and lemon juice coagulants has a very significant effect on the chemical composition of moisture content, dry matter content, protein, and fat content in cheese. And the interaction of storage time has a very significant effect on the chemical composition of moisture content, dry matter content, protein and fat content.

4. Conclusion

The dosage of 10% lemon juice and 15 days storage time were the optimum result in increasing protein and dry matter content and reduced fat and moisture content in fresh cheese from goat milk. Lemon juice can be a coagulant and can be another rennet alternative for making halal fresh cheese.

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