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The Effects of the Types of Milk (Cow, Goat, Soya) and Enzymes (Rennet, Papain, Bromelain) Toward Cheddar Cheese Production

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The Effects of the Types of Milk (Cow, Goat, Soya) and Enzymes (Rennet, Papain, Bromelain) Toward Cheddar Cheese Production

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Abstract

The objectives of this research are to study the effects of different types of milk and enzymes toward the yield and quality (moisture, ash, protein, fat content, and texture) of cheddar cheese and the interaction between those two variables during the process. The types of milk are cow, goat, and soya milk, while the types of enzymes are rennet, papain, and bromelain enzymes. Regarding the procedure, the milk is first pasteurized before $CaCl_2$ and *Lactobacillus lactis* that acts as the acidifier starter as much as 0.2% (w/v) and 0.5% of the milk volume are added respectively. The amount of enzyme added is appropriate for the determination of enzyme dose. The curd is separated from the whey and then 2.5 grams of salt is added to 100 grams of curd. Afterwards, the curd is pressed until the water content decreases (cheese), then ripened for 1 month. The analyses conducted are moisture, ash, protein, fat content, and texture (hardness). The conclusion is the goat milk and the rennet enzyme are the suitable raw material for cheddar cheese production. Furthermore, different types of milk and enzymes affect the yield. However, there is no interaction between the types of milk and enzymes to the yield.

Abstrak

Pengaruh Jenis Susu (Sapi, Kambing, Kedelai) dan Jenis Enzim (Rennet, Papain, Bromelin) terhadap Pembuatan Keju. Tujuan penelitian ini adalah untuk mempelajari pengaruh jenis susu, jenis enzim, dan interaksi antara jenis susu dan jenis enzim terhadap perolehan dan kualitas (kadar air, kadar abu, kadar protein, kadar lemak, dan tekstur) dari keju *cheddar*. Jenis susu yang divariasikan adalah susu sapi, susu kambing, dan susu kedelai sedangkan jenis enzim yang divariasikan adalah enzim rennet, papain, dan bromelin. Pertama-tama, susu dipasteurisasi kemudian ditambahkan $CaCl_2$ sebanyak 0,2% (b/v) dan *Lactobacillus lactis* sebagai starter pengasam juga ditambahkan sebanyak 0,5% dari volume susu. Jumlah enzim yang ditambahkan sesuai dengan hasil penentuan dosis enzim. *Curd* dipisahkan dari *whey*, kemudian ditambahkan garam dapur pada *curd* dengan perbandingan 2,5 gram garam dalam 100 gram *curd*. Kemudian *curd* ditekan hingga kadar air berkurang (keju). Keju akan diperam selama 1 bulan. Analisis yang dilakukan adalah analisis kadar air, kadar abu, kadar protein, kadar lemak, dan tekstur (kekerasan). Kesimpulan yang dapat diambil adalah susu kambing dan enzim rennet merupakan bahan baku yang cocok untuk pembuatan keju *cheddar*. Selain itu, jenis susu dan jenis enzim mempengaruhi perolehan. Namun, tidak ada interaksi antara jenis susu dan jenis enzim terhadap perolehan.

Keywords: cheddar, cheese, enzyme, Lactobacillus lactis, ripeness

1. Introduction

Cheese is a food product made of the coagulation of milk proteins. One type of cheese which is well-known among Indonesians is the cheddar cheese. According to *Badan Pusat Statistik*, in 2002 the consumption of cheese reached 8,000 ton per year and increased 20% since 2001. Meanwhile, cheese imports kept increasing about 5.96% each year [1]. Cheddar cheese is a relatively solid cheese with a pale yellow to ivory white colour and a strong flavor. It also has a dense texture and gentle nature.

The nutrient content of the different types of milk are presented in Table 1. The protein content in milk is the most important factor for making cheese. The protein

Table 1. Nutrient Content of Milk

Composition/100 g milk	Types of milk		
	Cow	Goat	Soy bean milk
Protein (g)	2.82	3.48	4.90
Fat (g)	3.42	5.23	2.65
Ash (g)	0.65	0.75	0.31
Lactose (g)	4.47	4.11	-

content of cow milk is proven to be adequate as a raw material for making cheese [2]. Therefore, the protein content of goat milk and soya milk is higher than that of cow milk thus making it eligible for cheese. The type of protein which is of great influence to the yield of cheese is casein [3]. The casein coagulates and forms the curds.

In the making of cheddar cheese, the coagulation process of milk occurs by adding the rennet enzyme [2]. The protease enzyme from calf abomasum (rennet), which is usually used as a coagulant [3], is available in limited quantities and has a relatively expensive price. On this account, further research about alternative coagulants is required. The papain enzyme is an enzyme derived from papaya latex and can be used as a substitute for rennet. The papain is also a proteolytic enzyme and is able to break down molecules from amino acid. The papain enzyme is available in large quantities, more resistant to acidic conditions or base conditions and high temperatures, and also has a reasonable price [4]. An alternative enzyme is a bromelain enzyme. Bromelain enzyme is also a proteolytic enzyme. This enzyme comes from pineapple fruit [5]. Pineapple is one of the fruits produced in Indonesia. According to *Badan Pusat Statistik* (2010), from the year 2007 to 2009 there was an increase of 1,558,196 tons [6]. Thus, this enzyme is a fine alternative for making cheese.

2. Methods

Preliminary experiments. The preliminary experiments consist of the pasteurization of animal milk, milk manufacturing from soybeans, bromelain enzyme isolation, and dosage determination of the enzyme. The cow milk and goat milk which are used are purchased directly from a ranch in Lembang, Bandung. Meanwhile, for soya milk, the soybeans are purchased at the Sederhana Market, Bandung. The pasteurization of animal milk is conducted for 30 minutes at a temperature of 62.80 °C, and then the milk is cooled to 31-32 °C. The rennet enzyme (under the brand name Fromase) is purchased from Depok, and the papain enzyme (under the brand name Paya) is purchased from a local market in Bandung.

The manufacturing of soya milk refers to the journal of Agricultural Economics Faculty of Agricultural Social Sriwijaya University in 2009 [9]. One kg of soybeans are cleaned and washed, then boiled for 15 minutes. After that, soybeans are soaked for 8 hours. After 8 hours, they are filtered and peeled, and then drained and blended. Boiled water is added to the soybeans with a ratio of 1 liter of water to 250 grams of soybeans. This mixture is filtered with a cloth filter and soymilk is thus obtained. The milk is pasteurized for 30 minutes at a temperature of 62.80 °C, and then the milk is cooled to 31-32 °C.

The bromelain isolation procedure is performed in accordance with Wuryanti's procedure from the Faculty of Mathematics and Natural Sciences Diponegoro in 2004 [10]. The pineapple fruit is cut into pieces and then blended. The mixture is homogenized with phosphate buffer and 7.5 pH for 15 minutes. The ratio of fruit to phosphate buffer is 1:1. This mixture is centrifuged for 15 minutes at 3000 rpm using Hettich Zentrifugen EBA 20 The supernatant is separated. This supernatant is the crude extract of the bromelain enzyme.

Dosage determination of the enzyme refers to the journal of Food and Agro-Industry Techno Bogor Agricultural University [11]. As much as 100 mL of milk is heated to the temperature of 35 °C. The enzyme is then added and stirred until it becomes homogeneous. This step is done simultaneously when the stopwatch is turned on. The minute the stirring process starts to feel heavy, the stopwatch is turned off and the time is recorded.

Cheddar cheese making. Variations of this research are milk variations (cow milk, goat milk, and soya milk) and the types of enzymes (rennet enzyme, papain enzyme and bromelain enzyme). The cheddar cheese manufacturing process, as referred in Handbook of Food Products Manufacturing [12], is as follows: The volume of the milk is 2 L. Calcium chloride 0.2% of the weight of the milk is added to the pasteurized milk. When the temperature reaches 30 °C, *Lactobacillus lactis*, as the starter, is added as much as 0.5% of the volume of milk. The milk is allowed to stand for 30 minutes, and the coagulation occurs in this process. The appropriate enzyme dose which has been determined in the preliminary experiment is added and this mixture is incubated for 30 minutes. The curd is formed after this process. The curd is cut into 3 cm x 3 cm and left for 10 minutes. The mixture is heated slowly to 38 °C temperature (the rising of the temperature is 2 °C in 5 minutes) and then filtered. The curd is sprinkled with salt (NaCl) of 2.5 g in 100 g of curd. The curd is put in a container and pressed with a pressure of 1.4 atm for 12 hours at room temperature. The pressed curd is then ripened in a refrigerator for 1 month. This process is called cheddaring. The cheese is formed after this process.

Analysis of the milk and the product. The analysis of the milk consists of analysis of the water content, ash

content, protein content, fat content, and calcium content. Meanwhile, the analysis of the product consists of yield analysis, water content, ash content, protein content, fat content, calcium content, and texture analysis, which are performed both before and after the ripeness process. The water content analysis is conducted using the oven method in accordance with SNI 01-2980-1992 [13]. The analysis of ash content refers to SNI 01-2980-1992 [13] and uses ThermoLine 1300 electric furnace. The analysis of protein content uses the semi micro Kjeldhal method with the Kjeltec 8100 instrument. The analysis of fat content uses Soxhlet method. The analysis of texture uses the texture analyzer instrument Brookfield CT 3 4500. The calcium content analysis refers to Standard Methods for the Examination of Water and Wastewater Part 3500-Ca [14].

3. Results and Discussion

The analysis of milk. The composition of milk as a raw material, which is obtained from the analysis, is shown in Table 2. The different values between the analysis and the literature in Table 1 are caused by the different sources of milk (for cow and goat milk) and the different procedures in making soya milk.

The determination of enzyme dosage. The dosage determination of rennet, papain, and bromelain enzymes is carried out on each type of milk. Based on the results obtained, the highest dose is the bromelain enzyme. The enzyme dose for each type of milk can be seen in Figure 1.

The highest enzyme dosage is for soya milk. This is due to the fat and protein contents of soya milk which are lesser than cow milk and goat milk, thus requires a greater amount of enzymes in the coagulation process.

For the three types of milk, the highest dosage of enzyme is the bromelain enzyme. This type of enzyme works by breaking the peptide bonds in the molecules of the protein. The optimum temperature for bromelain enzyme is 55 °C [15]. This value has significant difference for an optimum temperature for the rennet and papain enzymes. The optimum temperature for rennet is 40 °C [16], while for papain it is 50 °C [17].

Meanwhile, the temperature of the milk when the enzyme is added is 26-30 °C. Therefore, the huge difference in temperature between the milk and the optimum temperature of the enzyme can cause a decrease in enzyme activities. A higher amount of bromelain enzyme is needed to achieve the same activity of the other enzymes.

The yield of cheese. The yield of the mass is calculated by dividing the mass of cheese with the mass of milk. The yield percentage of cheese from goat milk is greater than that of the cheese from soya and cow milk. This is due to the fat and protein contents of goat milk which is higher than cow and soya milk. The yield of cheese can be seen in Figure 2. The higher the fat and protein contents are, the higher the amount of curd is. Thus, the higher the amount of cheese will also be. From the experimental design, it can be concluded that the types of milk influence the yield of the cheese. The highest yield is obtained at the rennet enzyme variation. This means that the curd produced at the rennet enzyme variation is the highest compared to other enzymes. This is due to the enzyme rennet that works closer to the optimum temperature than other enzymes.

Rennet contains an acid protease enzyme and mushroom Rhyzomucor miehei as well as the lipase enzyme, which is secreted through the external membrane into the culture medium. Lipase enzyme breaks down fat and converts it into simpler components, such as fatty acids and glycerol. Thus, the protein can catch the simpler components easier to form the curd and cause the yield of the curd to be higher [18].

Table 2. Composition of Milk

Analysis	Cow milk	Goat milk	Soya Milk
Water (%)	87.00	81.50	90.50
Ash (%)	2.00	2.50	3.50
Protein (%)	1.97	3.80	1.03
Fat (%)	3.50	5.00	2.00
Calcium (g/L)	0.92	1.24	0.44

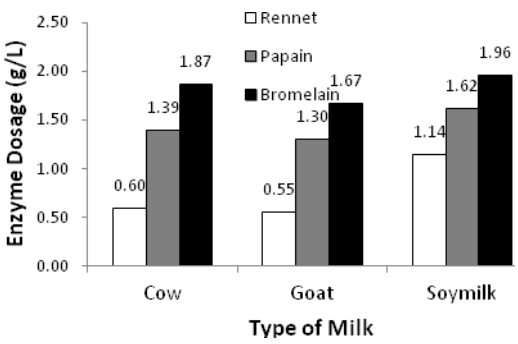

Figure 1. The Enzyme Dosage

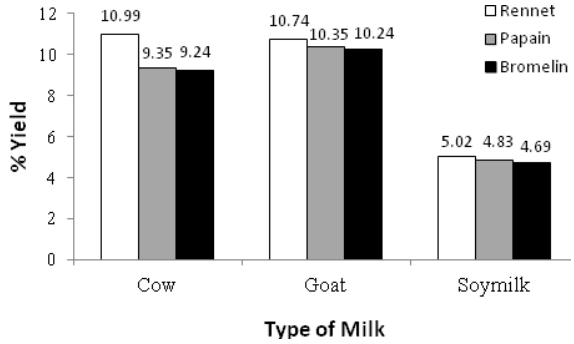

Figure 2. Yield of Cheese

The high acid content in bromelain enzyme, which results in a faster release of whey, thus can cause casein molecules to form a globule of casein (curd). When the milk is mixed with acid components, it releases hydrogen ions and thus leads to the decrease of pH. This can cause the casein micelles to dissolve and form calcium ions (Ca^{+}). These ions will penetrate other structures of the micelles casein and then form an internal strong chain of calcium. Thus, the micelles of the casein change because of the aggregation and end in the curd formation [10].

From the experimental design, it can be concluded that the types of milk and enzymes have an effect on the yield percentage of cheddar cheese. Meanwhile, there is no interaction between the types of milk and enzymes to the yield percentage.

The water content of cheese. The loss of water content in the cheese occurs in the pressing and cheddaring process. Cheddar cheese is a type of hard cheese with maximum moisture content of 45%. The results of analysis of the water content can be seen in Figure 3.

The water content for all types of cheese after ripeness is no more than 45% and appropriate to the SNI standard. The cheese from soya milk has higher water content than that of cow and goat milk. The high water content in soybean milk cheese is due to the hydrophilic and hydrophobic nature of soy protein. The characteristic of hydrophilic causes the protein to bind water and hold it. Meanwhile, the characteristic of hydrophilic causes the protein to bind fat [19]. However, in animal-based dairy products, there are three casein fractions, namely alpha, betta, and gamma. Fraction consists of kappa casein alpha has a molecular weight 19000-31000.

Kappa casein is an important component whose micelle structures are broken by rennet and becomes the para-kappa-casein and soluble glycopeptide. The bond dis-attachment between fenilalanin-mentionin of the kappa casein, releases macrogycopeptide molecules which cause the surface of the hydrophilic micelle to disappear. The result of this broken bond is hidrofobic chain proteins which form insoluble curd in the cheese production [20].

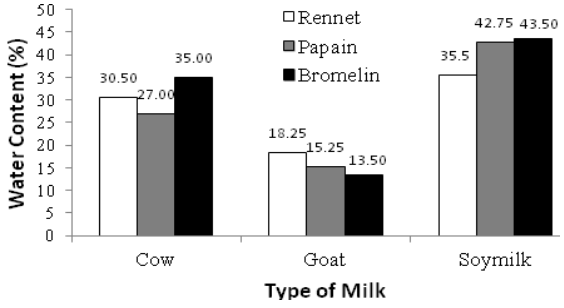

Figure 3. The Water Content of Cheese after Ripeness

The cheese made of the bromelain enzyme has higher water content than the papain and rennet enzymes. This is due to the effect of the non-optimal performance of the bromelain enzyme in the heating process which causes the milk clotting process not to produce a compact curd and which results in the high moisture content.

The low calcium content in the raw milk can cause the water content of the cheese to be higher. The reduction of water content of cheese when ripeness is caused by the decomposition material by proteolytic activity. The activity is the breaking down of long chains of protein molecules into smaller molecules because of *Lactobacillus lactis* and enzymes. The decreasing percentage in water content before and after ripeness is 4.44%.

Based on the result of two-factor factorial design, it can be concluded that the types of milk affects the water content. On the other hand, the types of enzyme do not affect the water content. There is an interaction between the types of milk and types of enzyme towards the water content.

The ash content analysis. The maximum ash content of SNI standard is 5.5% [13]. The result of ash content analysis can be seen in Figure 4. The cheese from soymilk has higher ash content than that of cow and goat and does not meet the SNI standard. Nevertheless, the ash content of cheddar cheese from cow and goat milk are under 5.5% and meet the SNI standard.

The ash content in cheese made of rennet and papain enzymes tend to be higher than that of the bromelain enzyme. This is due to the phase of the rennet and papain in the form of solid and also the enzyme dosage lower than bromelain enzymes (Figure 1).

Based on the result of the two-factor factorial design, it can be concluded that the different types of milk has an effect on the ash content. On the other hand, the types of enzymes do not affect the ash content. There is interaction between the types of milk and of enzymes towards the ash content. During the ripeness process of the cheddar cheese, the hydrolysis reaction occurs between the peptide bonds in protein or polypeptide and then become smaller molecules, amino acids.

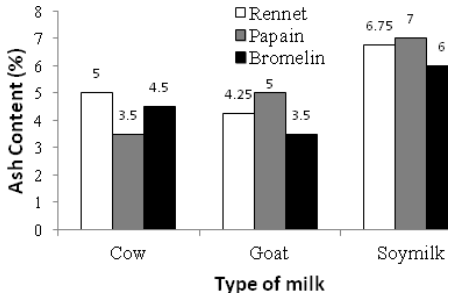

Figure 4. Cheese's Ash Content after Ripeness

There is also the breakage of fat, lactose, and organic compounds into simpler molecules.This hydrolysis reaction and breakage of the components can also give effects on the ash content. Therefore, the ash content after ripeness process is smaller than before process [16]. The less percentage of the ash content before and after ripeness is 0.89%.

Protein content analysis. The protein content of SNI standard of cheddar cheese at a minimum is 19.5% w/w [13]. The protein content in the cheese from cows and goat milk meets the SNI standard and higher than that of the cheese from soya milk. However, the protein content of soya milk cheese is under 19.5% w/w. The result of analysis of protein content can be seen in Figure 5.

The cheese from cow and goat milk tend to have higher protein content than others. This is due to the protein content of goat milk which is greater than cow milk and soya milk. Meanwhile, the cheese from soya milk has the least protein content because of the protein and fat contents in the soya milk is also the lowest than all the others.

The cheese which uses the papain and rennet enzyme gives higher yields than bromelain enzymes. The enzymes cause the proteolytic activity that breaks the peptide bonds and coagulates the milk protein. The bromelain enzyme does not give satisfying results, because the cheese production process does not reach its optimum condition.

The optimum temperature for bromelain enzyme is 55 °C [15] while the maximum temperature of the process is 38 °C.

The protein content increases after one month of the ripeness process with an increasing value of protein content percentage of 1.38%. The increasing protein content is due to the hydrolysis of proteins by protease enzyme into peptones, and amino acids during ripeness. Enzyme activity in the cheese ripeness process is derived not only from the enzymes, but also the starter, *Lactobacillus lactis* bacteria.

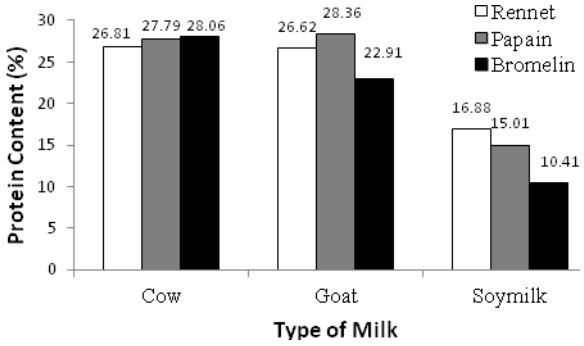

Figure 5. The Protein Content of the Cheese after Ripeness

The hydrolysis of proteins is a breakage of the peptide bonds of proteins into simpler molecules. The hydrolysis of peptide bonds will cause some changes in the protein and increase the solubility due to the increase of NH_3^+ and COO^- content and the reduction of molecular weight proteins [22].

The protease enzymes are divided into four categories, namely serine proteases, sulfihidril protease, metal proteases and acid proteases. The rennet enzyme is included in the acid protease group, which has two carboxyl groups on the active side. Carboxyl group consists of a carbonyl group (-CO-) and hydroxyl (-OH). The hydroxyl group (-OH) interacts with the H^+ from the amino acids group and form water. However, the carbonyl groups will interact with water through hydrogen bonds. Papain and bromelain enzymes are included in proteases sulfihidril group, an enzyme which is able to hydrolise the peptide bonds in proteins or polypeptides into smaller molecules; that is amino acids [18]. The papain and bromelain enzymes are also proteolytic enzymes which have the ability to break the peptide bonds and coagulate the milk proteins [23].

Based on the result of the two-factor factorial design, it can be concluded that the types of milk affects the protein content. On the other hand, the types of enzyme do not affect the protein content. There is no interaction between the types of milk and the types of enzyme towards the protein content.

Fat content analysis. The amount of fat in the curd is influenced by the content of fat in milk and the temperature during the cheese production. The higher the processing temperature is, the lesser the fat content in the curd will be. This is because the heat makes the fat easier to dissolve in the water and in the whey. The results of fat content analysis can be seen in Figure 6. The minimum fat content of cheddar cheese according to the SNI standard is 25% w/w [13]. Nevertheless, the fat content of the cheddar cheese does not reach the minimum fat content of SNI standard.

The fat content in cheddar cheese made of goat milk tends to be the highest because its fat content is also the

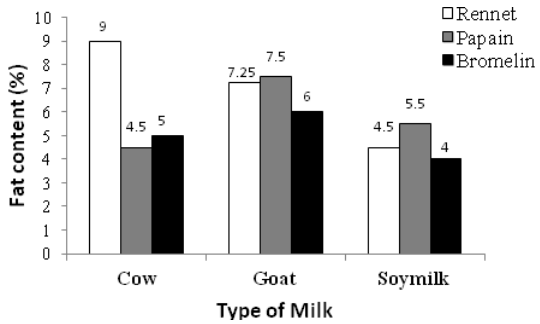

Figure 6. Fat Content Analysis of the Cheese afterRipeness

highest compared to the others. However, the fat content in cheddar cheeses made of soya milk is the lowest due to the lowest fat content of the soya milk.

The cheese made of rennet enzyme has the highest fat content, because the rennet enzyme contains an acid protease enzyme and *Rhyzomucor miehei* mushroom, as well as the lipase enzyme, which is produced from the external membrane into the culture medium. Lipase enzyme breaks down the fat and converts fat into simpler components, such as fatty acids and glycerol. Thus, the protein can attach the simpler components and allows the process to be easier form the curd and increases the yield of the curd [18].

The fat in the cheese during the ripeness process has been changed into fatty acids such as acetic, butyric, caproic, stearic, oleic, and others. The fatty acid turns into various esters which cause the scent, flavor, taste and texture. Fat degradation occurs because of the lipase enzyme derived from bacterial activity of *Lactobacillus lactis* [8].

Even though the cheese does not meet the SNI standard, the cheese can potentially be low fat cheese. The high fat content can lead to heart disease, stroke, and obesity. Fat component in the form of cholesterol is believed to be the major cause of heart disease. Excessive cholesterol can form deposits on the blood vessel walls, which results in the narrowing of the blood vessels [16].

Based on the result of the two-factor factorial design, it can be concluded that the types of milk affects the fat content but the types of enzymes do not. There is no interaction between the types of milk and those of enzyme towards the fat content.

Texture analysis. The ripeness process is conducted within 1 month. During the process, the cheese texture will be changed, especially its hardness. The hardness of the cheese before and after the process can be seen in Figure 7 and 8. The higher the value of hardness is, the harder the cheese will be. The different value of hardness is due to the enzyme hydrolysis which causes the protein casein to become more soluble, thus the cheese becomes softer [13]. The higher the activity of proteolytic enzymes is, the softer the cheese will be.

However, the hardness of the soymilk cheese decreases after ripeness because of the texture of the cheese, which also depends on the calcium content in raw milk and the cheese. The calcium content of the raw milk and the cheese can be seen in Table 2 and Figure 9 respectively. The calcium affects the fuse process between the casein micelle and forms a strong internal calcium chain, which is eventually finished with a coagulation process. The lower the calcium content is, the softer and stickier the cheese will be.

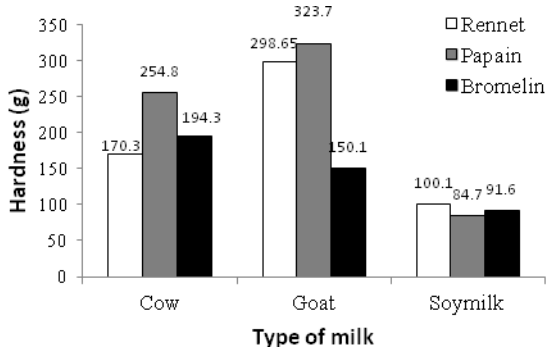

Figure 7. Hardness of Cheese before Ripeness

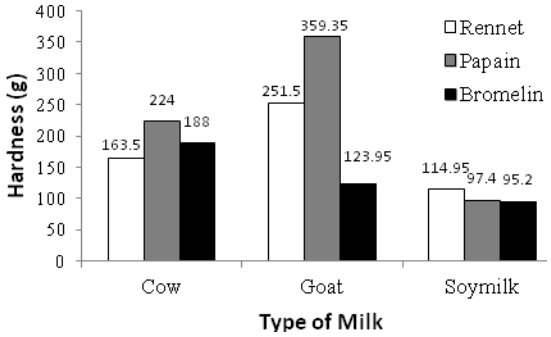

Figure 8. Hardness of Cheese after Ripeness

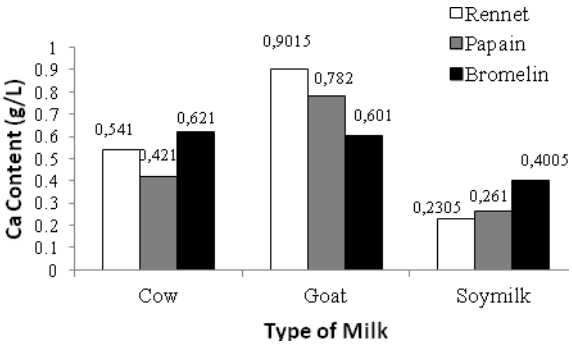

Figure 9. Calcium Content of Cheddar Cheese

The cheddar cheese from goat milk has the highest level of hardness due to the highest calcium content in the raw milk and cheese. The calcium content in soya milk is the lowest, thus the level of hardness is the lowest.

The hardness of the cheese made of rennet and papain is higher than bromelain enzyme due to the phase of the rennet and papain in the form of solid, thus the water content of these enzymes is lower than bromelain. The lower the water content is, the higher the hardness will be.

Based on the result of the two-factor factorial design, it can be concluded that the types of milk and those of enzymes affect the texture. There is an interaction

between the types of milk and the types of enzyme towards the texture.

4. Conclusions

The goat milk and the rennet enzyme are the suitable raw material for cheddar cheese production. Furthermore, different types of milk have different effects on the yield, water, ash, protein, fat, and texture of the cheese. The types of enzymes affect the yield and texture of the cheese. There is an interaction between the types of the milk and the enzymes on water, ash, and texture of the cheese. However, no interaction of those two variables on the yield, the protein and fat content. The cheddar cheese from this experiment meets the SNI standard, except for the cheese made of soya milk because the ash and fat contents do not meet the SNI standard.

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References

[1] Anon., Konsumsi Keju Nasional, Badan Pusat Statistik Indonesia, *http://www.bps.go.id/*, 2003. [In Indonesia]
[2] R. Sathya, B.V. Pradeep, J. Angayarkanni, M. Palaniswamy, Asian Pac. J. Trop. Biomed. 14/6 (2009) 788.
[3] F.V. Kosikowski, V.V. Mistry, Cheese and Fermented Milk Foods, Vol. 1, Origins and Principles, 3rd ed., Westport, 1997, p.711.
[4] E. Amri, F. Mamboya, Am. J. Biochem. Biotechnol. 8/2 (2012) 99.
[5] S. Ketnawa, S. Sai-Ut, T. Theppakorn, P. Chaiwut, S. Rawdkuen, As. J. Food Ag-Ind. 2/04 (2009) 457.
[6] Anon., Produksi Nanas di Indonesia, Badan Pusat Statistik Indonesia, *http://www.bps.go.id/*, 2010. [In Indonesia]
[7] L.S. Ceballos, E.R. Morales, G. de la T. Adarve, J.D. Castro, L.P. Martínes, R.D.S. Sampelay, J. Food Compos. Anal. 22 (2009) 322.
[8] H. Chairunnisa, J. Ilmu Ternak 1/1 (2007) 16. [In Indonesia]
[9] I.R. Setiawan, Magister Thesis, Puslit KIM-LIPI, Kawasan Puspiptek Serpong, Tangerang, 2006. [In Indonesia]
[10] Wuryanti, Magister Thesis, Universitas Diponegoro, Semarang, 2004. [In Indonesia]
[11] Park, Jurnal Teknologi Pangan dan Agroindustri, 1/5 (1990). [In Indonesia]
[12] Y.H. Hui, Principles, Bakery, Beverages, Cereals, Cheese, Confectionary, Fats, Fruits, and Functional Foods, Handbook of Food Products Manufacturing, Vol. 1, John Wiley & Sons, Inc., New Jersey, 2007, p.1131.
[13] Anon., Keju Cheddar Olahan, Badan Standardisasi Nasional, *http://www.bsn.go.id*, 1992. [In Indonesia]
[14] L.S. Clesceri, A.E. Greenberg, A.D. Eaton, Standard Methods For The Examination of Water and Wastewater 20th edition, Part 3500-Ca, USA, 1999.
[15] R.P. Anggraini, A.H.D. Rahardjo, R.S.S. Santosa, J. Ilmiah Peternakan, 1/2 (2013) 508. [In Indonesia]
[16] H. Herawati, Prosiding Seminar Nasional Teknologi Peternakan dan Veteriner, 2011, p.413. [In Indonesia]
[17] A.P. Kusumadjaja, R.P. Dewi, Indonesian J. Chem. 5/2 (2005) 147.
[18] D.R.D. Kurnia, Magister Thesis, Universitas Diponegoro, Semarang, 2010. [In Indonesia]
[19] P. Grace, Undergraduate Thesis, Universitas Hasanuddin, Makasar, 2011. [In Indonesia]
[20] D. Makfoeld, D.W. Marseno, P. Hastuti, S. Anggrahini, S. Raharjo, S. Sastrosuwignyo, Suhardi, S. Martoharsono, S. Hadiwiyoto, Tranggono, Kamus Istilah Pangan & Nutrisi, Universitas Gajah Mada, Kanisius, Yogyakarta, 2002, p.76. [In Indonesia]
[21] A. Triyono, Prosiding Seminar Rekayasa Kimia dan Proses, 2010. [In Indonesia]
[22] B.S. Hartley, Ann. Rev. Biochem. 29 (1960) 45.