

## Physicochemical and Microbiological Appearance of Sapera Goat's Milk on Frozen Storage

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### ABSTRACT

This study aims to examine the effect of the storage time of milk at a temperature of  $-18^{\circ}\text{C}$  on the chemical, physical and microbiological content due to different storage times. The goat's milk studied was the milk of the Sannen Crossbreed of the Sapera goat. Twenty samples were taken from the milking results on the same day. The design used in this study was a completely randomized design (CRD) with five treatments and four replications. Samples were grouped according to treatment, namely 0d (control) and 10d, 20d, 30d, 40d; stored for 10, 20, 30 and 40 days, respectively. The milk storage process is carried out using a freezer at a temperature of  $-18^{\circ}\text{C}$ . The physicochemical and microbiological parameters observed were protein, fat, lactose, solid-non-fat (SNF), total solid (TS), specific gravity, pH, total plate count (TPC), and coliform. Evaluation of milk quality is based on the Indonesian National Standard (SNI) and Thai Agricultural Standard (TAS). The results showed that storage time did not significantly affect the components of fat, protein, lactose, total plate count and coliform ( $p > 0.05$ ). In milk stored for 40 days, there were differences in the values of SNF, TS, specific gravity and pH compared to control ( $p < 0.05$ ). In terms of chemical, physical and microbiological quality, Sapera goat's milk stored at  $-18^{\circ}\text{C}$  for 40 days still complies with SNI and TAS. The process of storing and freezing milk can be an alternative for preservation to ensure the physical and chemical quality of Sapera goat's milk.

**Keywords:** Coliform, fat, goat, milk, protein, Sapera.

### INTRODUCTION

Goat's milk is the third most consumed milk worldwide. The world's total goat milk production was estimated at 18.7 million tonnes in 2017, increasing 16% from 2007 to 2017 (FAOSTAT, 2019). The goat population in Indonesia since 2013 has continued to increase to reach 15.3 million heads in 2020, with the highest goat population on the island of Java. On the other hand, data on the total production of goat milk and the number of dairy goats in Indonesia are not known in detail (BPS, 2020). Through the technical implementation unit (UPT) of the Baturraden Center for Superior Livestock Breeding and Forage (BBPTU HPT) Baturraden, the Indonesian government has brought in Saanen goats at the end of 2019 to meet the need for quality goat breeds. Saanen is a subtropical goat from the Swiss mountains and is cultivated as a dairy goat in various countries. Saanen goats can produce up to 3.8 liters of milk per day with a fat content of up to 2.5-3% (DJPKH, 2020). Saanen goats are currently increasingly popular being developed in Indonesia. Breeders bred Saanen goats with local Ettawa Peranakan (PE) goats to produce Saanen crossbreed offspring commonly called "Sapera" goats.

Although in Indonesia, goat's milk is not as popular as cow's milk, goat's milk is known to

have better nutritional value than cow's milk and is believed to be efficacious for health. Several studies have shown that goat's milk is able to improve digestive tract disorders (gastro-intestinal disorders), is hypoallergenic (does not cause allergies), has high digestibility, helps growth and bone density and is able to increase levels of Vitamin A, Ca, thiamine, riboflavin, niacin content in blood (Haenlein, 2004); (Stergiadis et al., 2019). In line with the increasing public awareness of the health and benefits of goat's milk, the demand for goat's milk is increasing. The goat's milk business has spread widely, especially in the provinces of East Java, West Java, and Central Java, which are areas with the largest goat population in Indonesia.

Goat's milk is a perishable food that contains easily damagable nutritional components. This is a challenge for farmers in storing and marketing the goat's milk they produce. Milk quality must be protected from chemical, physical and microbiological damage in order to meet the standards. Freezing is one way that can be done to maintain the quality and extend the shelf life of milk. It was stated by Faridah (2018) that frozen food and stored at a temperature of  $-12^{\circ}\text{C}$  to  $-24^{\circ}\text{C}$  with stable temperature conditions can prevent microbiological damage.

Therefore, this research is important to determine the impact of the freezing process and storage time of goat's milk. It can be used as a reference for the community, especially dairy goat farmers, to store goat's milk.

## MATERIALS AND METHODS

The material used in this study was 20 liters of fresh milk from Sapera goat from UD. Mitra Agro Abadi Farm, Salatiga. The materials used were 0.1% peptone water (BPW), plate count agar (PCA), 70% alcohol and distilled water. The equipment used in the research were 1 L sample bottle, measuring cup, freezer with a temperature of -18°C, coolbox to temporarily store milk during the trip to the laboratory, Milk analyzer, test tube, petri dish, Bunsen, 1000 L micropipette, vortex and incubator. Milk samples were taken from the total yield of milking in the morning. All milk obtained was collected and homogenized and then 20 samples were taken with a volume of 1 liter per sample. The process of freezing and storing milk samples was carried out using a freezer with a temperature of -18°C. Storage time is carried out according to the treatment applied. Samples stored frozen are thawed again before analysis in the laboratory; by immersing it in water at a temperature of 30°C for 25 minutes.

This study used a completely randomized design with five treatments and four replications. The treatment applied was the difference in storage time in frozen conditions at -18°C. The types of treatment were as follows 0d=fresh milk (control), 10d=10 days storage, 20d=20days storage, 30d=30days storage and 40d=40 days storage.

The parameters observed were milk's chemical and physical components, including fat, protein, lactose, solids non-fat, total solids, pH, and specific gravity. Microbiological parameters observed included the total number of microorganisms / Total Plate Count (TPC) and Coliform. Physicochemical measurements of goat's milk using the Funke Gerber No Cat Milk Analyzer Brand: FG-3510. The analysis was repeated three times for each sample. Testing and calculating the total number of microorganisms based on the Indonesian National Standard (SNI) 2897-2008 concerning the method of testing for microbial contamination in meat, eggs, and milk, as well as their products; the total number of microorganisms was tested using the cup count method by pouring (pour plate method) (BSN, 2008). Differences in the physicochemical and microbiological appearance of goat's milk due to differences in storage time were analyzed using analysis of variance (ANOVA) with a significance level of 5%. If there is a significant effect of the treatment, it is continued with Duncan's Multiple Region Test to determine the differences between treatments (Steell and Torie, 1991).

## RESULTS AND DISCUSSION

The Indonesian National Standard (SNI) on goat's milk is not yet available, so the determination of the quality of goat's milk in this study is based on the standard of fresh cow's milk (SNI-3141.1:2011) and the Thai Agricultural Standard (TAS) No.6006:2008 for the standard of goat's milk. The chemical test results of goat's milk content can be seen in Table 1.

Table. 1 Milk chemistry test

Chemical Composition (%)	Storage Time					SNI <sup>1</sup>	Standardisation		
	0h	10h	20h	30h	40h		TAS <sup>2</sup>		
							Premium	Goods	Standard
Fat content	4,81	4,87	4,93	4,38	5,02	≥3,0	> 4,0	3,5-4,0	3,25-3,5
Protein content	3,64	3,61	3,58	3,48	3,37	≥2,8	> 3,7	3,4-3,7	3,1-3,4
Lactose content	5,39	5,07	4,71	4,69	4,43	n.a	n.a	n.a	n.a
<i>Solid non fat</i> (SnF)	9,04 <sup>a</sup>	8,97 <sup>a</sup>	8,30 <sup>ab</sup>	8,17 <sup>ab</sup>	7,80 <sup>b</sup>	≥7,8	> 8,25	>8,25	> 8,25
<i>Total Solid</i> (TS)	14,06 <sup>a</sup>	13,87 <sup>a</sup>	13,29 <sup>ab</sup>	12,90 <sup>ab</sup>	12,26 <sup>b</sup>	n.a	> 13	12-13	11,7 -12

Description: - Different superscripts on the same line show significantly different at the 5% level (P<0.05).

- 1: standard of fresh cow's milk (SNI-3141.1:2011)

- 2: Thai Agricultural Standard (TAS) No.6006:2008

- n.a = not available; The SNI and TAS have not set the standard for lactose content in milk.

## Fat content

The lowest percentage of the average milk fat content is 4.38% and the highest is 5.02%. This value met the standards of SNI and TAS (Fig. 1), where the fat content value meets at least 3% and can be categorized as a premium quality based on TAS, namely fat content >4%. The fat content in the 0d milk sample (control) was 4.8%. The fat content of the Sapera goat in this study was lower than the fat content of the Ettawa Peranakan goat's milk in the Arifin et al. (2016) study, which was 5.53%, but slightly higher than the fat content of the Saanen goat's milk in the study of Zurriyati et al., (2011) which is 4.59%. The difference in milk fat content is influenced by the type of goat used. This is by the opinion of Ljutovac et al. (2008), which states that the fat content of goat's milk

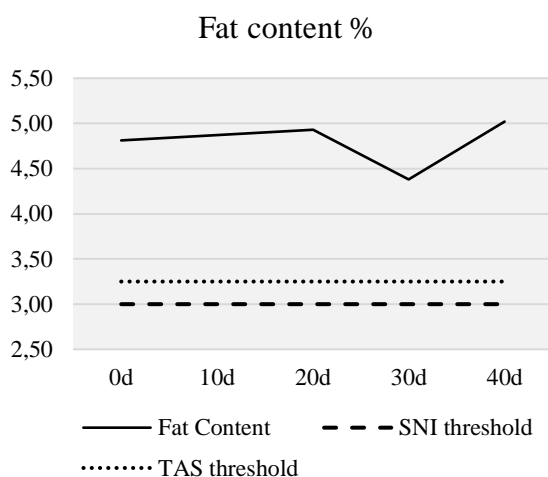


Figure 1. Fat content

## Protein content

The highest protein content of goat's milk in this study was 3.64% and the lowest was 3.37%, which was considered to have met the SNI criteria, namely the protein content of a minimum of 2.8% and a minimum of 3.1% for TAS. Statistical analysis found that the research treatment had no significant effect on protein content ( $p > 0.05$ ). This shows that storage in frozen conditions can maintain the protein content of milk from damage. The average milk protein content at 10d, 20d, 30d, and 40d was not statistically significantly different. Still, it tended to be lower than 0d (Fig. 2). This is because in samples 10d, 20d, 30d, and 40d, there is a thawing process or re-thawing from storage freezing which may cause denaturation of milk proteins. This is by Wulandari (2020) opinion that frozen and thawed food will experience protein denaturation

varies and many factors influence, including nation, feed quality, lactation period and season. The statistical analysis of the effect of storage time of frozen milk on fat content obtained results that were not significantly different ( $p > 0.05$ ). Storage at a temperature of  $-18^{\circ}\text{C}$  causes the rate of chemical and enzymatic reactions in milk to run slower to maintain milk fat content so that it does not suffer damage. This is by Wulandari (2020) opinion that at the freezing point, the fluidity of water is at its minimum so that it cannot act as a medium for a chemical reaction. Michal (2010) added that the storage temperature ( $-18$ ) was able to inhibit the growth of lactic acid bacteria producing lipase enzymes so that the hydrolyzed fat by the lipase enzyme could be minimized.

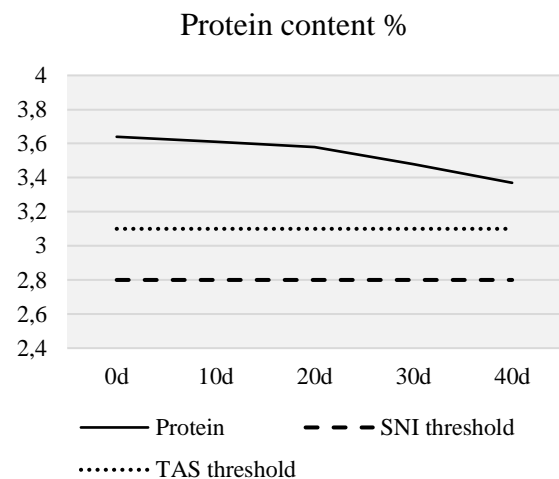


Figure 2. Protein content

and will not be able to absorb water again because of the enlargement of the molecules.

## Lactose content

Data on the lactose content of goat's milk in this study can be seen in Table 1. Statistical analysis showed that the length of frozen storage had no significant effect on lactose content ( $p > 0,05$ ). The frozen storage process in Sapera goat milk causes a decrease in chemical processes and microbiological activity associated with spoilage and spoilage of milk. Frozen storage can keep lactose from microbial degradation, where lactose is an energy source for the growth of lactic acid bacteria. This is under the opinion of Buckle et al. (1985), which states that the effect of cooling results in a decrease in chemical, microbiological

and biochemical processes associated with food spoilage and spoilage.

### Solid NonFat

The dry matter without fat or Solid Non-Fat (SNF) can be seen in the table. 1. Based on the statistical analysis results, it was found that the length of the shelf life in frozen conditions significantly affected the SNF levels of Sapera goat milk ( $p < 0,0,5$ ). SNF levels at 0d, 10d, 20d, 30d and 40d were 9.04; 8.97; 8.3; 8.17 and 7.8%. In the 0d, 20d, and 30d treatments, according to Duncan's different test the results were not different, but the 40d sample showed differences in SNF levels compared to the control. This indicates that the chemical quality of milk begins to decrease at 40 days of storage. SNF levels at 0d vs 40d, i.e., 9.04% vs 7.8%, obtained a decrease in milk SNF levels as much as 13.72%. Protein and lactose are parts of SNF, where these two components describe the percentage of SNF in milk. In this study, protein and lactose levels decreased with increasing shelf life (Fig. 3). This is under Zurriyati et al., (2011), stating that SNF is influenced by lactose and milk protein components where feed quality will affect SNF levels related to milk protein levels. The decrease in SNF in Sapera goat's milk stored for up to 40 days resulted in SNF levels that did not meet the TAS standard (minimum SNF 8.25%) but still met the SNI standard; namely, SNF at least 7.8 (Fig. 4).

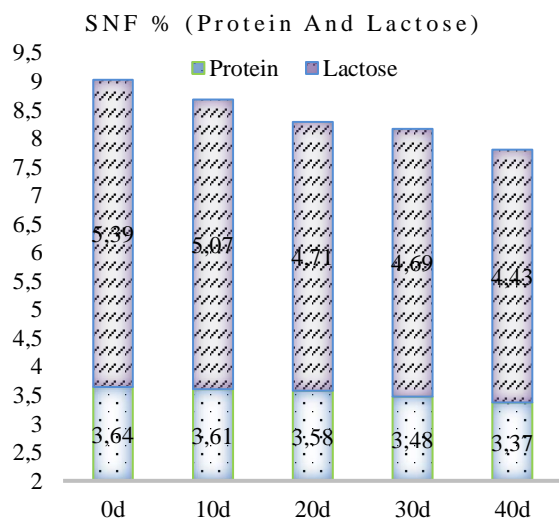


Figure 3. Solid Non-Fat Component (Protein and Lactose)

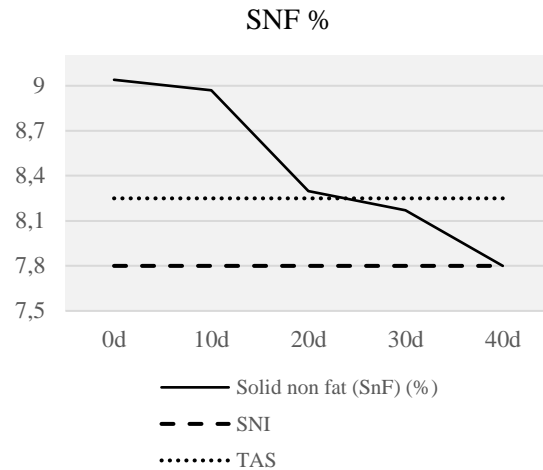


Figure 4. Solid Non-Fat content

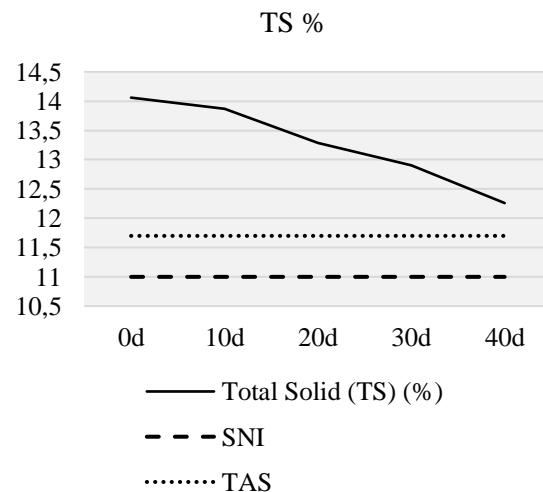


Figure 5. Total Solid content

### Total Solid

Total Solid (TS) is a component of milk other than water, including protein, fat, lactose, and ash. The statistical analysis results on TS levels due to the effect of storage time obtained significantly different results ( $p < 0.05$ ). The TS levels at 0d, 10d, 20d, 30d and 40d were 14.06, respectively; 13.87; 13.29; 12.90 and 12.26%. The TS level decreases with the longer the shelf life. TS values in treatment 0d, 10d, 20d, and 30d were not significantly different. Still, treatment 0h compared to 40h obtained a significant difference of 14.06% vs. 12.26%, where during 40 days of storage there was a decrease in TS levels of 12.8%. At 40 days of storage, it was suspected that microbes, especially lactic acid bacteria, had proliferated, marked by the increasing value of TPC and decreasing pH (Table 2). The decrease in TS levels during the storage process was due to

microbiological activity using milk components such as protein, fat and lactose. This is by Resnawati (2020), which states that milk components such as lactose are a source of energy for the growth of lactic acid bacteria, which in the next process will act as a producer of acid levels in the fermented milk. The TS level in Sapera goat's milk in this study was considered to have met the SNI and TAS standards, namely > 11.7% (Figure 5.)

### Specific gravity

The specific gravity (BJ) of milk in this study can be seen in Table 2. Statistical analysis showed that the length of frozen storage had a significant effect on the BJ value ( $p < 0.05$ ). The BJ value in the 40d treatment showed significantly different results than the other treatments (0d, 10d, 20d, 30d). Specific gravity is influenced by the constituent components of milk such as protein,

lactose and minerals. The difference in milk composition in this study was reflected in the difference in the total solid value between the 40d treatment compared to 0d and 10d. A decrease in total solids decreases the specific gravity of the milk. This is in accordance with the opinion of Eccles et al, (1984) which states, the factors that influence changes in the specific gravity of milk are factors from the milk itself which consists of protein, fat, lactose, gas and minerals in milk. According to Legowo et al. (2009), the specific gravity of milk depends on the fat content and milk solids because the specific gravity of fat is lower than the density of water or milk plasma. The specific gravity of goat's milk is higher than that of cow's milk in the range of 1.0231–1.0398 kg/m<sup>3</sup> but lower than that of sheep's milk in the range of 1.0347–1.0384 kg/m<sup>3</sup> (Park et al., 2007). The value of BJ goat's milk stored for up to 40 days in this study was still considered to meet the TAS and SNI standards.

Table 2. Physical and microbiological qualities of milk

Parameter	Treatment					Standardization	
	0d	10d	20d	30d	40d	SNI <sup>1</sup>	TAS <sup>2</sup>
Specific gravity (g/ml)	1,039 <sup>a</sup>	1,032 <sup>b</sup>	1,031 <sup>b</sup>	1,028 <sup>b</sup>	1,029 <sup>b</sup>	≥1,0270	≥1,0280
pH	6,60 <sup>a</sup>	6,54 <sup>a</sup>	6,58 <sup>a</sup>	6,54 <sup>a</sup>	6,3 <sup>b</sup>	6,3-6,8	6,5-6,8
TPC (cfu/ml)	29.000	69.750	39.500	36.500	91.250	≤ 1.000.000	≤ 200.000
Coliform (cfu/ml)	4,20	4,53	6,78	6,43	9,75	20*	≤ 1.000

Description: - Different superscripts on the same line show significantly different at the 5% level ( $P < 0.05$ ).

- 1: standard of fresh cow's milk (SNI-3141.1:2011)

- 2: Thai Agricultural Standard (TAS) No.6006:2008

- \*: maximum limit of microbial contamination in food (SNI-7388:2009)

### pH

The statistical analysis results of frozen storage have a significant effect ( $p < 0.05$ ) on the pH value of goat's milk. In this study, the pH value decreased with the long shelf life. (Table 2) The difference in pH values in milk may be due to higher microbial activity as the shelf life increases. Storage temperature conditions of 18°C cannot kill bacteria but can slow down their activity. Changes in pH value during storage indicate the presence of bacteria that produce lactic acid fermentation. This agrees with Swadayana et al., (2012) which states that bacterial activity is indicated by a decrease in the pH value below the normal value of 6.5-6.7. The pH value of milk in this study was in the range of 6.3-6.6. This condition is still following the pH standards according to SNI and TAS.

### Total Microbe

The results of the total microbial analysis can be seen in table 2. The difference in frozen storage time, the results were not significantly different from the total microbial content (Total Plate Count) ( $p > 0.05$ ). The total bacterial content at storage time of up to 40 days was statistically not significantly different, but descriptively there was a tendency to increase with increasing shelf life. This is in line with the acquisition of milk's pH value, which decreases with increasing shelf life. It can be assumed that the activity of lactic acid bacteria at 40 days of storage tends to increase; indicated the presence of lactic acid formation. This is following the opinion of Umar et al., (2014), which states that the decrease in pH is caused by lactic acid, which is formed from lactose degradation by lactic acid bacteria, such as

*Streptococcus thermophilus*, *Lactobacillus lactis*, and *Lactobacillus thermophilus*. TPC in Sapera goat's milk in all treatments obtained results that met the standards of SNI ( $\leq 106$  cfu/ml) and TAS ( $\leq 2 \times 10^5$  cfu/ml).

### Coliform

Data on Coliform content in goat's milk due to differences in frozen storage time are shown in Table 2. Statistical analysis showed that the results were not significantly different from coliform content ( $p > 0.05$ ). The storage temperature condition of  $18^\circ\text{C}$  was able to inhibit the growth of coliform bacteria so that the number of coliforms obtained was not significantly different. Coliform bacteria are gram-negative, lactose-degrading bacteria and are singular. Coliform is used as an indicator of the presence of pathogenic bacteria, both in animals and humans, because the number of colonies is positively correlated with the number of pathogenic bacteria (Wiliantari et al., 2018). Coliform content in milk in all treatments showed the results were still within the permissible threshold according to SNI 7388:2009, which was 20 cfu/ml (BSN, 2009).

### CONCLUSION

Based on the study results, it can be concluded that Sapera goat's milk stored in frozen conditions at  $-18^\circ\text{C}$  did not cause differences in the components of fat, protein, lactose, the number of bacteria and coliforms in storage for up to 40 days. Specific gravity, solid non-fat, totally solid, and pH values differed from the control. The research treatment was able to maintain the quality of milk both physically and chemically and inhibit the development of bacteria. In terms of chemical, physical and microbiological quality, Sapera goat's milk stored at  $-18^\circ\text{C}$  for 40 days still complies with SNI and TAS. The process of storing and freezing milk can be an alternative for preservation to ensure the physical and chemical quality of Sapera goat's milk.

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