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Effects of Pasteurized Sheep's Milk Use on Production and Ripening of Siirt Herby Cheese

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Abstract: Siirt herby cheese is a varietty of local cheeses, and raw sheep's and goat's milk are used in its production. The curds obtained by curdling the milk obtained in the highlands is sold in the city centre during the day or the next day. Siirt herby cheese is produced from the retail curds. Three different cheese samples form retail curd, raw sheep's and pasteurized sheep's milks were made. The samples were ripened at 4°C for 120 d. Hygienic quality of raw milk supplied from a farm and raw milk curd bought from local retailers did not have adequate hygiene. Coagulase positive staphylococci were 3.11±0.6 $\log_{10}$ cfu/ml in raw milk, then it increased to 6.6±0.5 $\log_{10}$ cfu/g in the curd before salting. Pasteurization of milk was decreased the total count of the coagulase positive staphylococci below the maximum permissible value (10^3 cfu/g) in the regulation on Microbiological Criteria of the Turkish Food Codex. These samples were only contained >10^3 cfu/g coagulase positive staphylococci at the 120th d of ripening. The pasteurization of milk allowed to obtain cheese that meets the desired hygienic criteria. In addition, sufficient acidity and pH development were observed in pasteurized dairy cheeses even though starter culture was not used. It was found that the traditional cheese production using both retail curd and raw milk may not be suitable for producing cheese in accordance with the official standards and regulations. It was concluded that in the case of the production of these cheeses, it is not appropriate to offer them for sale before the completion of the legal ripening period of 120 d. It was deemed necessary to conduct further research in order to draw up a production diagram including the parameters of the Siirt herby cheese production process.

Keywords: Siirt herby cheese, Hygiene, Pasteurised milk, Quality

Pastörize Koyun Sütü Kullanmanın Siirt Otlu Peynirinin Üretimi ve Olgunlaşması Üzerine Etkileri

Özet: Siirt otlu peyniri yöresel bir peynir çeşidi olup üretiminde çiğ koyun ve keçi sütleri kullanılmaktadır. Yaylalarda elde edilen sütün mayalanması ile elde edilen teleme gün içerisinde veya ertesi gün şehir merkezlerinde satılmaktadır. Siirt otlu peyniri bu telemelerden üretilmektedir. Satın alınan teleme ile çiğ ve pastörize koyun sütü kullanılarak 3 farklı peynir örneği yapıldı. Numuneler 4°C'de 120 gün süreyle olgunlaştırıldı. Bir çiftlikten temin edilen çiğ sütün ve bu sütten yapılan telemenin hijyenik kalitesinin yeterli olmadığı gözlendi. Koagülaz pozitif stafilokoklar çiğ sütte 3.11±0.6 $\log_{10}$ kob/ml iken, tuzlamadan önceki aşamada telemede 6.6±0.5 $\log_{10}$ kob/g'a ulaştığı gözlendi. Sütün pastörizasyonu Türk Gıda Kodeksi Mikrobiyolojik Kriterler Tebliği'nde izin verilen maksimum değerin (10^3 kob/g) altına ancak 120. günde düştüğü gözlendi. Sütün pastörizasyonunun istenen hijyenik kriterlere uygun peynir elde edilmesini sağladı. Ayrıca pastörize süt peynirlerinde de starter kültür kullanılmadığı halde yeterli asitlik ve pH gelişimi gözlendi. Gerek satış yerlerindeki teleme ve gerekse çiğ süt kullanarak geleneksel peynir üretiminin standart ve mevzuata uygun peynir üretmek için uygun olmayabilir. Bu peynirlerin üretilmesi durumunda yasal olgunlaştırma süresi olan 120 günün tamamlanmadan satışa sunulmasının uygun olmadığı sonucuna varıldı. Siirt otlu peyniri üretim prosesine ait parametrleri de içeren bir üretim şemansının çıkarılmas için daha fazla araştırmanın yapılması gerekli görüldü.

Anahtar Kelimeler: Siirt otlu peyniri, Hijyen, Pastörize süt, Kalite

1.Introduction

While there are about 2000 types of cheese in the world, Türkiye, which has 193 of them, has been called "cheese heaven". The journey of each of these cheeses from production to consumption is different (1). It has been reported that the gastronomic value of this rich cheese variety should be brought to the fore (2). It has been stated that the production technology of many cheese varieties in Anatolia should be developed. So, the products should have been made at standard manners to increase export potential of these cheeses should be used (3,4). A total of 753 thousand tons of cheese produced in Türkiye. We could not find out the knowledge about how much traditional cheese, and also how much herby cheese is produced in Türkiye (5). Nevertheless, it has been reported in a study that per capita consumption of herby cheese is 14.74 kg/year in Eastern Anatolia and South-eastern Anatolia regions, and the Türkiye average is 3.2 kg/year (6). The most produced type of herby cheese in Türkiye is Van herby cheese and it has a Geographical Indication Certificate (5). Other herby cheeses are Urfa, Bitlis, Hakkâri, Trabzon, Erzincan Keçene and Siirt herby

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cheeses. Most of the previously made studies on the herby cheeses have been conducted on Van herby cheese (3,7-10).

The minimum technical and hygienic criteria for cheeses have been established with official documents and official inspections are carried out accordingly (11-13). Chemical and microbiological quality characteristics of herby cheeses have been investigated and possible health risks have been emphasized (7,14,15). However, it has been revealed by researchers that there are no product standards in local cheeses produced outside of commercial enterprises, the hygienic quality of these products is low and they do not provide sufficient assurance in terms of public health (16-18). It has been reported that the chemical qualities of herby cheeses on sale have not founded in accordance with related standards (19-23). It has also been reported in the previously made studies that hygienic quality of cheese samples examined have not been determined to be good (14,24-28).

Siirt herby cheese is generally produced from raw sheep's milk or a mixture of sheep's, cow's and goat's milk. While the main herb added to cheese is sirmo (sirik, *Allium* sp.), herbs called heliz (*Ferula orientalis*) and çiriş (*Eremurus spectabilis*) are also used depending on the taste of the household (6).

The unique qualities of cheese are shaped by many factors. Milk constituents, microbiota composition, process parameters used, ripening regimes and many other factors are prevailed effective on the quality characteristics of the cheese (29-34). Developing raw milk cheese technology is very difficult due to the risk of pathogenic bacteria (35). Many scientific researches on the process development for some of the other herby cheese varieties have been conducted previously (36-42). Few studies have been conducted on the Siirt herby cheese (6,23,24,27). Herby Cheese Standard is the reference regulation for making the cheese (43).

In this study, two different cheese samples were prepared; one from raw milk and the other pasteurised milk. Also, as for control samples, one d old curd was purchased from the retail stores in the city centre and used for making herby cheese as for mimicking traditionally produced Siirt herby cheese. So, it is aimed to investigate in this study that differences between raw and pasteurized milk use, and also retailed curd use on the final chemical and microbiological composition of the Siirt herby cheese during a 120 d of ripening period.

2.Materials and Methods

Three different cheese samples were made from retail curd, raw milk and pasteurized milk. So, curd cheese, raw milk cheese and pasteurized milk cheese were made. Experimental cheese samples maintained at 4°C for ripening, and were analysed once a mo. for in a 4 mo. of ripening period.

Raw milk: A 25 l of newly milked sheep's milk was purchased from a sheep farm located in the city centre of Siirt. The milk was filtered through a cloth strainer and brought to the laboratory within 1 hour. Half of the milk was used raw, and the other half was pasteurized and used to make cheese.

Coagulant: A microbial cheese coagulant including rennet at 20.000 MCU/ml activity (Mayasan®, İstanbul Türkiye) was purchased from a local market and stored at 4 °C until use. Herb (Sirmo, Sirik, Allium sp.): Herbs collected from the highlands and sold by the citizens in the market were used. After the herbs were removed and washed with drinking water, they were chopped to a size of approximately 5 mm and added to the curd at a rate of 3%, w/w as recommended by many local Siirt herby chees producers (personal communications).

Salt: Rock salt from the region was used in cheese making was used in cheese making.

Press material: Plastic containers filled with water at a rate of 70 % of total weight of each portion milk used for making each part of the curd samples. Then, the water containers were used as press by putting them on the draining curd portions.

Making herby cheese by using raw milk: Raw sheep's milk was heated up to 35 ˚C. Calcium chloride (200 ppm, w/v) was added to the milk. Coagulant was added to the milk at 35 ˚C to be the same as applied to the pasteurized milk and waited for 40 min for coagulation. The clot was cut into 1x1x1 cm as made by Ocak et al. (38) and waited for 30 min for clot hardening. The clot was added in a press cloth and the press material was put on the curd for 60 min as make it standard for all the samples. So, we could not find a standard whey drainage time period in the literature applied for herby cheese making. The pressed curd was crumbled to the size of chickpeas. Then, a 3% (w/w) chopped Sirmo (Allium sp.) and a 3% salt was added to the crumbled curd and mixed. The same press material was put on the curd in cloth once again for 90 min to make extra drainage. The cheese samples obtained were separately crumbled to the size of chickpeas cheese and also separately filled tightly in 100 ml sample containers (5).

Making herby cheese using pasteurized milk: Raw milk in stainless steel container was heated at 72˚C by using a kitchen type gas stove. The heat temperature was controlled by using a thermometer (Thermopro TP02S, ThermoPro Co., USA) Milk was stirred gently for 3 min after termination of the heating to make heat treatment more uniform. Then, the milk was cooled to 37˚C by soaking the milk container in a cold water bath. All other processes were applied as in the production of raw milk cheese as mentioned above.

Making herby cheese using retail curd: A 2 kg of curd was bought from a local curd retailer in Siirt province and transferred to the laboratory under cold storage (4 °C) in 1 h.

The curd was crumbled to the size of chickpeas. Then it was mixed with 3% (w/w) herb (sirmo) and 3% (w/w) salt, then pressed for 90 min by applying a weight of equal to that of raw milk used. At the end of the pressing, the crumbled curd again in the size of chickpeas, and the plastic sample cups with a volume of 100 ml were filled in such a way that there was no air gap.

Raw milk analysis: pH measurement was made with a handheld pH meter (AZ 8685, Taiwan). Acidity determination was made by titrimetric method and the results were given as % l.a. In addition, milk analysis was performed with a milk analyser (Lactoscan LS, Nova Zagora, Bulgaria).

Determination of coagulant strength: A domestic commercial rennet used for cheese making was diluted 1/10. Raw milk was heated to 35 °C. A 1 ml of reconstituted coagulant was added to 50 ml of heated milk and the clot formation time was determined. Coagulant strength was calculated according to the formula (44).

Analysis of milk, curd and cheese: The pH was measured by using a digital pH meter (Milwaukee AZ8686, USA) (45). Titratable acidity (lactic acid, %) was determined by using 0.1 N NaOH and phenolphthalein indicator as stated to TS 591/ March 2013 (46). The dry matter and ash were determined gravimetrically according to TS EN ISO 5534/AC (47). The fat content was determined by using Van Gulik method (TS ISO 3433) (48). Salt in dry matter was determined by titration according to TS EN ISO 5943; 2007 (49).

For microbiological analysis, reference methods were followed (50). Briefly, 10 g from each sample was used for making ten-fold serial dilutions in 90 ml of sterile physiological saline (PS). For count of total aerobic mesophilic bacteria (TAMB), Plate Count Agar (PCA, Oxoid CM 0463) was used and the petri dishes were incubated at 30±2°C for 72 h for count of *Enterobacteriaceae*, Violet Red Bile Glucose Agar (VRBGA, Oxoid-CM0485) was used and the petri dishes were incubated at 37±2°C for 48 h. For coliforms, Violet Red Bile Lactose Agar (VRBLA, Oxoid CM0107) was used and the petri dishes were incubated at 37 °C for 24 h. The growing pink-red colonies with a pink precipitation ring around were counted. For coagulase positive staphylococci, Baird Parker Agar plates including Baird Parker Agar Base (BPA, Oxoid CM1127) and Egg Yolk Tellurite Emulsion (Oxoid SR0054) were used. The plates were incubated at 37°C for 48 h. Black shiny coagulase positive colonies with a diameter of 1.5- 2.5 mm with a transparent zone around were counted. Then, 5 susceptive colonies selected randomly for each sample were tested for their coagulase reaction before calculation of cfu/g values. For yeasts and molds, Yeast Extract Glucose Chloramphenicol Agar (YGCA, Merck 1.16000) was used and the petri dishes were incubated at 25°C for 5 d. Colonies grown on the medium were counted.

The study was repeated in triplicate and each analysis was made in duplicate. The mean and standard deviation values were calculated by using the Microsoft Excel program. Means with a significant difference were compared by Duncan's multiple range tests by using SPSS v.15.00 program (Chicago, Illinois, USA) and P values <0.05 were considered statistically significant. The standard deviation values (±SD) are given in the Table 2 and Table 3.

3.Results

The mean values from the raw milk used in the study were as follows: pH 6.7, acidity (%, l.a) 0.2, dry matter (%, w/w) 12, protein (%, w/w) 6.1, fat (%, w/w) 7.2, lactose (%, w/w) 4.8, salts (%, w/w) 0.8 and density 1.034.

Raw milk was subjected to microbiological analysis within 1 hour after being taken from the farm. The results are given in Table 1. As seen in Table 1, it was determined that the bacterial and yeasts and molds loads were quite high in curd made with raw milk.

Table 1: Microorganism loads of raw milk and raw milk curd before salting (log10 cfu/g).

	Total mesophilic aerobes	*Enterobacteriaceae*	Coliforms	Yeasts and molds	Coagulase positive staphyllococci	Lactobacilli	Lactic streptococci
Raw milk	6.8±0.6	6.3±03	5.1±0.4	5.1±02	3.1±0.6	3.96±0.2	7.3±0,3
Raw milk curd (before salting)	9.8±0.2	8.2±0,2	8.9±0.5	9.8±0,4	6.6±0.5	9.70±0.3	10.65±0,6

The pH values of the retail curd, raw milk curd and pasteurized milk curd were found to be 6.7, 6.7 and 6.0, respectively, after the curd was filtered and herb was added. Acidity values were found to be 0.3, 0.2 and 0.15 in the same order. The findings are given in Figure 1.

Within the values before salting, the pH of the ready-made curd was found to be higher than the other two samples. Dry matter ratio of pasteurized milk curd was found to be higher than the other two samples (39.4%). There was no statistically significant difference among the samples in terms of fat ratios (Figure 1).

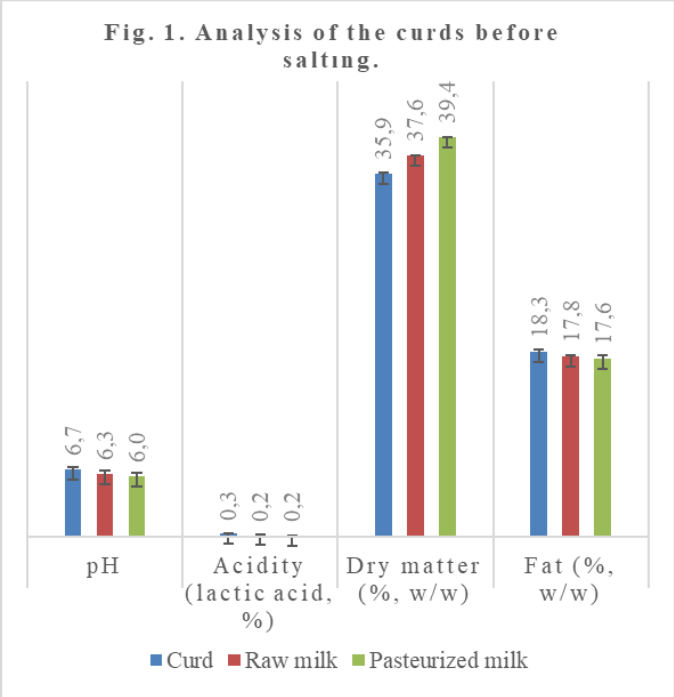

Figure 1: Values of physical and chemical analyzes of curds before salting

Three different cheeses were made by processing three different curds in the same way, and they were packaged in Ø63 x 65 mm polypropylene sample containers (L 102348, Lp Italiana) and kept in the cold. Analyses were performed on the 1st, 30th, 60th, 90th and 120th d of 4°C of cold storage. The pH and chemical analysis results are given in Figure 2.

While the initial pH values were similar in all three samples, the decrease in the following days was more in the retail curd samples ($p<0.05$). After the 90th d, pH decreases followed an approximately horizontal course and decreased to the level of 5 at the 120th d (Figure 2a).

While the initial acidity values were similar in all three samples, the increase in the following days was more in the curd cheeses purchased from the seller. The increase in acidity continued in all three samples until the 120th d and the highest (1.17 %) was observed in cheese made using curd bought from the seller ($p<0.05$). In the other two samples, the acidity reached approximate values on the 90th and 120th d (Figure 2b).

When the values from high to low in terms of dry matter levels were ranked, cheese made from curd were raw milk cheese and pasteurized milk cheese. It was observed that the moisture loss in the samples was not at a significant level (Figure 2c).

While the fat levels were highest in cheese made from curd, this difference disappeared after the 90th d and the fat levels in all three samples were similar. It was observed that the cheese samples completed the ripening period with the fat level between 17.5% and 20.4% (Figure 2d).

In terms of salt levels, it was observed that cheeses made from curd were always high at 120th d, while pasteurized milk cheeses had the lowest salt values. It was observed that the salt added at the rate of 3% of the curd weight remained lower in pasteurized milk samples during ripening compared to the others (Figure 2e) ($p<0.05$).

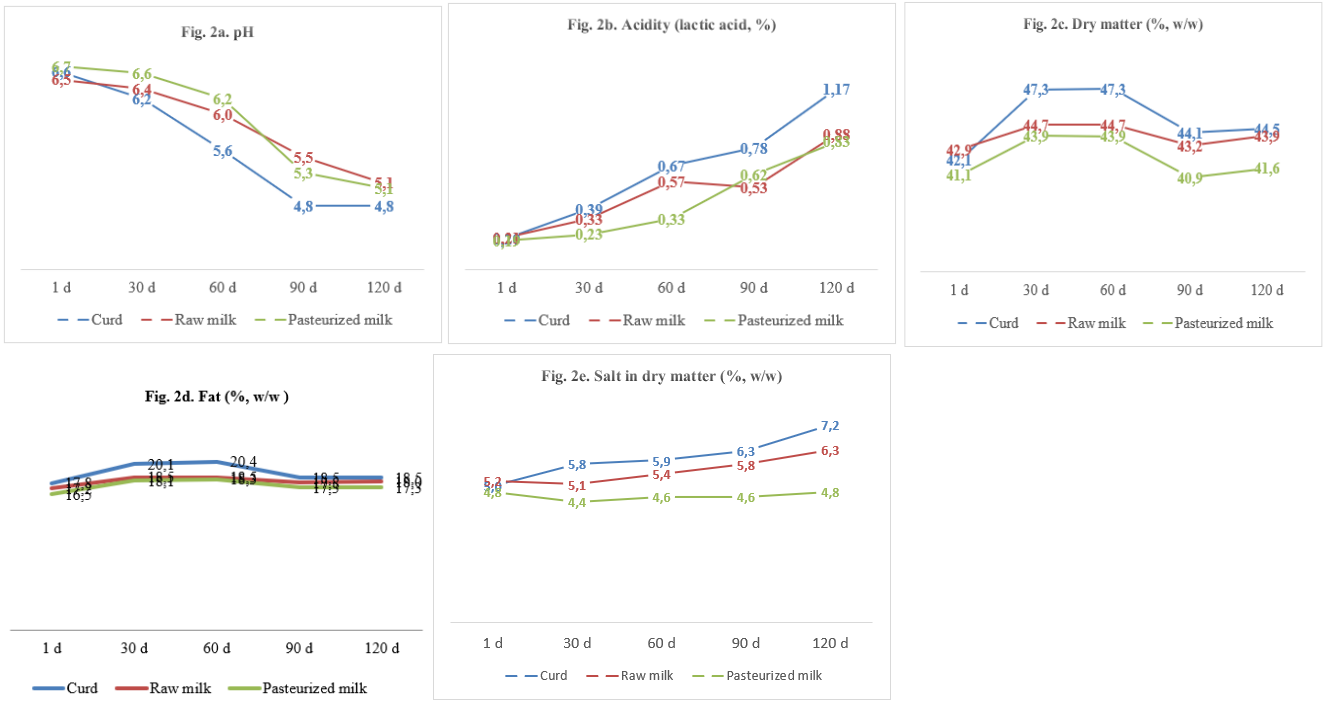

Figure 2 (a, b, c, d, e). Values of physical and chemical analyzes in 120-d cold storage in Siirt herby cheese samples produced by using retail curd, and also raw milk and pasteurized sheep's milk.

Table 2: The standard deviations (±SD) of values given at Figure 2.

Figure no.	Cheese / ripening d	1	30	60	90	120
Fig. 2a. pH	Retail curd cheese	0,2	0,1	0,2	0,3	0,1
	Raw milk cheese	0,2	0,1	0,3	0,3	0,1
	Pasteurized milk cheese	0,3	0,2	0,3	0,2	0,1
Fig. 2b. Acidity, % l.a.	Retail curd cheese	0,02	0,01	0,02	0,02	0,03
	Raw milk cheese	0,01	0,02	0,03	0,02	0,02
	Pasteurized milk cheese	0,01	0,02	0,02	0,01	0,03
Fig. 2c. Dry matter, %	Retail curd cheese	0,1	0,1	0,6	0,6	0,3
	Raw milk cheese	0,4	0,6	0,6	0,5	0,4
	Pasteurized milk cheese	0,8	0,3	0,3	0,5	0,3
Fig. 2d. Fat, %	Retail curd cheese	0,1	0,1	0,2	0,2	0,1
	Raw milk cheese	0,2	0,3	0,3	0,2	0,2
	Pasteurized milk cheese	0,3	0,1	0,1	0,2	0,1
Fig. 2e. Salt in dry matter, %	Retail curd cheese	0,2	0,2	0,4	0,2	0,2
	Raw milk cheese	0,2	0,2	0,2	0,1	0,3
	Pasteurized milk cheese	0,3	0,2	0,3	0,3	0,4

Total bacterial counts were found to be higher in retail curd and raw milk samples on the 1st and 30th d compared to pasteurized milk samples ($p<0.05$). In the following period, the numbers were found to be similar in all three samples (Figure 3a).

Although the numbers of Enterobacteraceae decreased gradually in retail curd and raw milk samples, it was observed that they were in very high numbers than that of the pasteurised milk sample ($p<0.05$). Enterobacteraceae counts were determined to be 3.68 log10 cfu/g at 1st d of cold storage and >1 log10 cfu/g in pasteurized milk cheese samples at the 1st d of ripening (Figure 3b).

Although the numbers of coliforms decreased gradually in the curd and raw milk samples, it was observed that they were quite high. This number was determined as 3.20 log10 cfu/g in pasteurized milk cheeses at the 1st d of ripening. Then, it was determined to decreased to the level of >1 log10 cfu/g in the samples (Figure 3c).

Yeasts and molds counts were found to be considerably higher in retail curd and raw milk samples on the 1st and 30th d compared to pasteurized milk samples ($p<0.05$). In the

following period, the numbers were found to be similar in all three samples. The level in raw milk sample was higher than the other two cheeses ($p<0.05$). The values of the samples decreased by approaching each other at 120th d of ripening (Figure 3d).

While the lactobacilli level was found to be quite low (4.73 log10 cfu/g) in pasteurized milk cheese on the first d compared to the other samples ($p<0.05$), the values for all three samples were at an approximate level on the further analysis d. It was observed that lactobacilli level in pasteurized milk samples was higher than that of other two cheese samples at 60th d (Figure 3f).

While the count of lactotococci were determined to be quite low (5.36 log10 cfu/g) in pasteurized milk samples on the first d ($p<0.05$), as was in lactobacilli counts, on the other analysis days, the values of all three samples were at an approximate level. It was observed that the level in pasteurized milk samples was higher than that of other two cheese samples at 60th d. Although the 1st d level of lactococci was higher than that of lactobacilli, similar levels were observed in the following ripening d (Figure 3g).

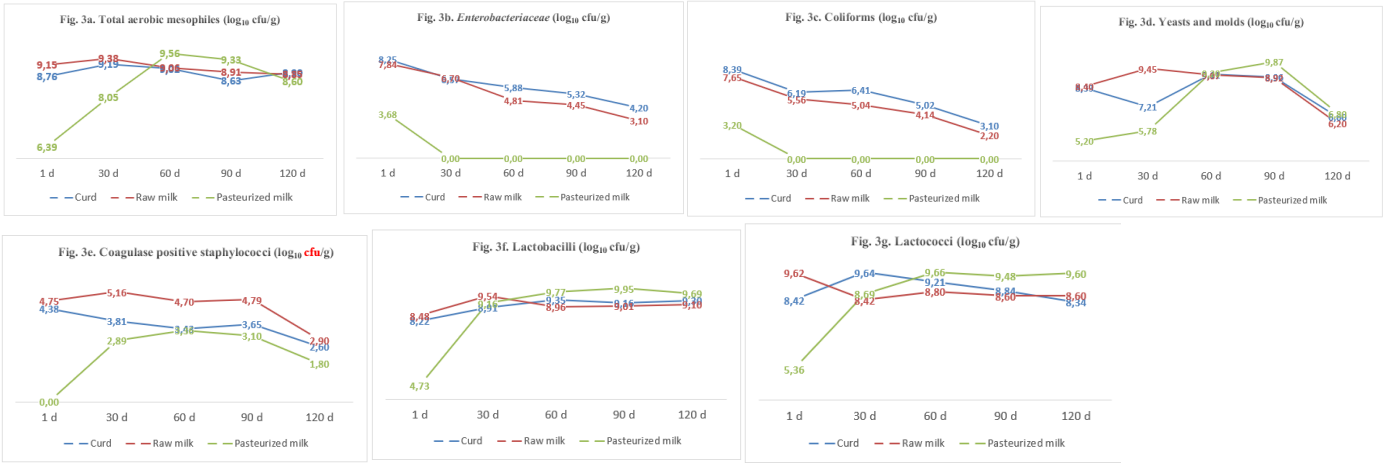

Figure 3 (a, b, c, d, e, f, g): Values of microbiological analyzes in 120-d cold storage in Siirt herby cheese samples produced by using retail curd, and also raw milk and pasteurized sheep's milk.

Table 3: The standard deviations (±SD) of values given at Figure 3.

Figure no.	Cheese / ripening d	1	30	60	90	120
Fig. 3a. Total aerobic mesophiles (log_{10} cfu/g)	Retail curd cheese	0,34	0,31	0,22	0,16	0,34
	Raw milk cheese	0,26	0,14	0,41	0,37	0,27
	Pasteurized milk cheese	0,18	0,33	0,25	0,21	0,23
Fig. 3b. *Enterobacteriaceae* (log_{10} cfu/g)	Retail curd cheese	0,13	0,23	0,41	0,33	0,21
	Raw milk cheese	0,32	0,34	0,21	0,24	0,11
	Pasteurized milk cheese	0,11	0,24	0,18	0,21	0,00
Fig. 3c. Coliforms (log_{10} cfu/g)	Retail curd cheese	0,27	0,31	0,27	0,16	0,21
	Raw milk cheese	0,34	0,12	0,24	0,12	0,19
	Pasteurized milk cheese	0,22	0,30	0,00	0,00	0,00
Fig. 3d. Yeasts and molds (log_{10} cfu/g)	Retail curd cheese	0,21	0,23	0,41	0,29	0,34
	Raw milk cheese	0,32	0,40	0,44	0,39	0,42
	Pasteurized milk cheese	0,16	0,32	0,30	0,33	0,18
Fig. 3e. Coagulase + staphylococci (log_{10} cfu/g)	Retail curd cheese	0,16	0,21	0,23	0,11	0,13
	Raw milk cheese	0,23	0,20	0,20	0,23	0,21
	Pasteurized milk cheese	0,30	0,34	0,17	0,20	0,20
Fig. 3f. *Lactobacilli* (log_{10} cfu/g)	Retail curd cheese	0,34	0,41	0,38	0,70	0,63
	Raw milk cheese	0,30	0,50	0,36	0,47	0,26
	Pasteurized milk cheese	0,22	0,21	0,44	0,44	0,33
Fig. 3g. *Lactococci* (log_{10} cfu/g)	Retail curd cheese	0,43	0,54	0,51	0,29	0,33
	Raw milk cheese	0,40	0,51	0,45	0,37	0,51
	Pasteurized milk cheese	5,36	0,27	0,39	0,40	0,37

4.Discussion and Conclusion

It is well known that factors such as breed, age, race, season, diet of dairy animal are effective on the milk composition. Also, milk composition and all the other applications of cheese making are effective on the cheese characteristics (19). Since the change made in each of these factors causes significant changes in the cheese, so many cheese varieties have been able to be produced. For production of a cheese variety, the effects of many factors separately and/or in combinations on its determinative characteristics have to be taken into consideration during the process. The cheeses traded must be produced in accordance with the standards established by official regulations (5,11-13,43,46).

In the Herby Cheese Standard (43), it is stated that rate of milk fat in dry matter has to be at least 45%, humidity at most 60% and salt at most 7.5%. The minimum, (average) and maximum values of this cheese in the geographical indication document of Van herby cheese are listed as follows: Dry matter (%) 43.81 (46.78) 47.78, protein (%) 20.60 (22.17) 25.52, fat (%) 16.75 (17 .29) 19.21, ash (%) 5.07 (6.85) 7.45, salt (%) 4.60 (5.73) 6.9. When it is classified in terms of % Moisture (in fat free dry matter) in cheese standards, herby cheese has been reported to be from the group of semi-hard cheeses that fall between 57-64% (5). A fully demonstrated process follow-up instruction for making herby cheese including critic control points such as curd break point, press end point, packaging point, and also critic limits such as acidity, pH, dry matter, salt levels at the critic control points has not been documented yet. Van herby cheese is currently being produced and sold by many large companies in Türkiye. However, we could have no information about their production processes. It can be proposed that each company is free for development its own process parameters and the production is made in accordance with the regulations. Although there are many scientific studies conducted on this subject, a complete process definition has not yet been revealed as a result of these studies. Also, this study is a narrowly comprehensive study focused on the pasteurization of sheep's milk only. It may be concluded for this situation that a full scientific study is needed for documentation of a complete herby cheese producing process.

In a study that we compared together with the findings of our own study by previously made studies conducted on herby cheeses, and it was revealed that most of the herby cheeses on sale do not comply with the regulations (51). The production and sale of these cheeses, which are traditionally produced by local people, should be made in accordance with the regulations. In our opinion, the most effective public health measure that can be applied in the current conditions is to prevent the production of raw milk cheese by producers who do not comply with the Special Hygiene Rules Regulation for Animal Food (11) and Microbiological Criteria Regulation (12). It would be a good decision to impose the obligation to pasteurize milk to such enterprises. Because in order to make raw milk cheese, it is necessary to use the milk of animals free from disease and ripening period have to be minimum 4 mo. as mentioned in Cheese Regulation (13). Akyüz and Kurt (52) have reported in 1984 that primitive conditions should be abandoned and production should be carried out in modern factories.

Since we could not come across to a complete process definition, we only followed regulatory parameters for making Siirt herby cheese (5,11-13,43,46). In order for us to take one of the previous studies as a reference, our other parameters should have been similar to those in the reference study. For this reason, we decided to use our own process parameters and our own method with the condition of complying with the regulations. So, pasteurisation of the bulk milk was made at 72˚C for 3 min. Coagulation was made at 35 ˚C for 40 min. The clot was cut into 1x1x1 cm as made by Ocak et al. (38) and waited for 30 min for clot hardening. The clot was added in a press cloth and the press material was put on the curd for 60 min as make it standard for all the samples. The press weight was determined as the rate of 70 % of total weight of each portion milk used for making each part of the curd samples. The press was applied 60 min before salting and 90 min after salting to make a good drainage from the curd samples. We applied dry salting to the crumbled curd and salt was used at a 3% of curd weighed after the first pressing. At the end of many preliminary studies, we realized to follow the parameters of pH between 5.5-6.0 and lactic acid between 0.3-0.5 in the curd before salting, minimum pH of 4.5 and maximum 1% lactic acid, humidity maximum 50%, the protein minimum 20% and the fat minimum 20% in cheese after 120 d of ripening. The parameters followed in this study for making Siirt herby cheese has to be confirmed in comparison with other process parameters that effect the resultant cheese regulatory parameters.

According to the Regulation on supply of raw milk (53), which is required for raw milk trade, animals must be free from disease. Also, milk has to be cooled to 4°C and below immediately after milking and the temperature during transport should not exceed 4°C. The supply of raw milk to the final consumer is carried out within 24 hours after milking. If raw milk obtained from other species other than cow's milk is to be used in the production of raw milk cheese without any heat treatment, the TAMB grown at 30°C in per ml of the milk have to be ≤ 500,000 (11). Most of the herby cheeses are made with the sheep's' milked in the highlands in the form of herby cheese curd in Siirt. Raw milk curd is produced by clotting the milk at milking temperature after draining from strainer cloth. The drained curd is brought to the provincial and district centres in sacks and then sold in the same d or other d. Individuals or small outlet owners buy these retail curds and process it into herby cheese for household use or sale. It was determined in this study that the

number of TAMB in the raw milk was as high as 6.77 log10 cfu/ml (Table 1). It can be difficult to obtain milk with the desired bacterial load in existing sheep barns and by hand milking. For these reasons, it is inevitable that the cheese produced from raw milk or retail curd cheese. It has been pointed out in previously made studies that Siirt herby cheese on sale have not met the regulatory criteria (6,23,24,27,51). We have also determined in this study that both raw milk and retail curd may not ensure the minimum hygiene requirements (Figure 3).

In this study, it was observed that acidity development continued with pH decrease in the samples during ripening (Figure 2a, 2b). This change occurred in the pasteurized milk samples as well as retail curd and raw milk samples. Although the microorganisms causing these changes were not extensively investigated in this study, it was determined that the numbers of lactic acid bacteria and lactic streptococci increased in all three types of cheese during the ripening period (Figure 3f, 3g). However, we did not used any microbial culture for ripening, natural microbiota presented in the samples or secondary contaminated microorganisms could have pH and acidity development in all the samples. Ripening microbiota in the pasteurized milk samples may have came from the herbs used, and/or from raw milk flora that survived pasteurization and/or microorganisms transmitted by secondary contamination during cheese making. In a previous study, the pH of herby cheese has been stated to be decreased from 4.89 to 4.52 and the acidity (%, lactic acid) increased from 0.62 to 1.05 during the 90-d ripening period (54). Emirmustafaoğlu and Coçkun have demonstrated significant differences from goat's cheese than that of cow's and ewes' cheese (55).

Tuncturk et al. (21) have prepared cheese samples by both raw milk and pasteurized milk. According to the changes recorded in the ripening period, it has been reported that difference in bulk milk do not adversely affect the products chemical composition, and pasteurized milk cheeses have been found to have harder structure. We also could not have determined a difference between raw and pasteurized milk samples. More research could be needed for making a net decision on the matter.

Although the production methods of herby tulum cheese is different, its composition and physical appearance are resembling to Tulum (leather bottle) cheese. The humidity rate has been regulated to be maximum 45% for full fat Tulum cheese and maximum 50% for low-fat and fat-free Tulum cheese (13). The minimum, (average) and maximum dry matter (%) values in the Geographical Indication Certificate of Van herby cheese are as follows: 43.81 (46.78) 47.78. In an our previously made study and also in this study we determined dry mater mean values close to 45% (Figure 2c) (51). We experienced in this study that the cheese could be more wet and sticky when the dry matter was as high as 60%. However, in the Herby Cheese Standard, humidity rate has been regulated as maximum 60% (43). This value is the same as the value of white cheese (13). It has been reported that Van herby cheese is produced similarly to white cheese in commercial production enterprises, the moisture rate in the cheese is high, and therefore it would be more appropriate to call these cheeses as herby white cheese instead of Van herb cheese (20). Thus, dry matter or humidity regulations for herby cheese could be reconsidered.

A maximum salt content in the dry matter of Tulum cheese is regulated as 5% (13). It is understood that the salt in Tulum cheese should be maximum 2.25%. Nevertheless, salt value has been regulated as maximum 7.5% in the Herby Cheese Standard (43), and also as 6.9% in the geographical indication document for Van herby cheese. In this study, dry salting was done by adding 3% of salt to the pressed curd as similar manner as the traditional method. So, the salt ratios the cheese samples have been determined to be higher than 2.25%. In most of the previously made studies, the salt rate from the retailed cheeses examined by us and other researchers have been generally higher than 5% (51). Because in our face-to-face interviews, people who produce herby cheese reported that they need more salt to prevent the cheese from spoilage. The salt content of the cheese at retail have to be decreased for public health. The salt content has already been reduced in other cheese varieties (13, 46).

In the Regulation on Turkish Food Codex Microbiological Criteria (12), it has been reported that the number of Enterobacteriaceae in pasteurized drink milk can be maximum 10 cfu/ml as a hygiene criterion. There are no coliform and coagulase positive staphylococcal values for pasteurized milk in the Regulation. In this case, it could be understood that these two microorganisms should not be present in pasteurized milk. As can be seen in Table 1, the numbers detected in all analysed microorganism were quite high. It has been revealed in the studies that the products on sale that are analysed have a high microorganism load and do not comply with the standards (51). The TAMB count in raw milk used in this study have been determined to be 6.77 log10 cfu/ml, and was higher than 500.000 / ml level which is stated in the Regulation (11) and Regulation on Turkish Food Codex Microbiological Criteria (12). Also, coagulase positive staphylococci are allowed to be a maximum 103 cfu/g in in Tulum cheese (12).

The total mesophilic aerobic bacteria, coliforms and yeasts and molds counts determined in this study have not been stated in the Regulation (12). These analyses were made for a possibility of use in next process development studies. When Table 1 and Figure 3 are comparatively examined, it could be understood that the hygienic quality of the samples has been improved by using pasteurized milk, since counts of Enterobacteriaceae, coliforms and coagulase positive staphylococci were lower than that of raw milk and retail curd

cheese samples. The reason for the presence of microorganism load in cheese samples above the standard values reported above may be that the samples were made with traditional methods. Drain cloth, strainer, utensils and other materials used in the study were used without sterilization and only by washing with tap water to mimic the traditional way. Under controlled production conditions, it is possible to reduce these values below the levels allowed in the standards. For this, the milk must be pasteurized and the materials used must be pasteurized or sterilized. The hygiene of the coagulant used should also be checked. Starter culture and/or ripening culture can be used to provide early acidity development and pH reduction in cheese. However, new studies are needed to obtain such microbial cultures.

Tunçtürk et al. (21) have made Van herby cheese by using raw and milk and brine salted the samples. A decrease of 1 log10 cfu/g in the TAMB on the 30th d of the ripening period has been documented. Also, coliforms (log10 cfu/g) has been decreased from 7 to 5.41 on the 30th d, 3.85 on the 60th d, 2.45 on the 90th d and to uncountable level on the 120th d. In this study, the initial bacterial levels were found to be high, similar to the researchers' findings. In parallel to the findings of the researchers, we were also determined high counts of the microorganisms from the 1st d of the ripening. However, unlike the findings of other researchers, it was observed in this study that the numbers of coliforms and coagulase positive staphylococci in the samples decreased to acceptable levels (102-103 cfu/g) after 60th d of the ripening (Fig. 3d, 3f).

Yeasts and molds counts were found to be high in our study and other previously made studies (Fig 3e) (51). When the situation that the number of yeasts and molds gradually increases during the ripening period is accepted as certain, it may be necessary to bring a new interpretation with a new perspective on the acceptance of this parameter as a criterion in hygiene monitoring. More research on this subject should be done to reveal the positive and negative aspects of yeasts and molds growth.

As a result, the positive effect of pasteurization in terms of hygiene assurance was determined. Hygiene may be difficult for producing herby cheese by using retail curd or raw ewe's milk. Regulative standards about moisture and salt content for the herby cheese could be valuable if are revaluated. Since not a fully informative regulatory or another published process schematic has not been documented yet, initial studies such as this study could be more valuable for future studies. This study alone is not enough to propose process parameters, critical control points and critical limits. Nevertheless, accessing the following parameters can be considered as the basic process parameters for further studies. In the process development studies for Siirt herby cheese, following parameters may be valuable to evaluate complex studies including multi-variable parameters. By this way, homogeneity of the future studies may be increased. Then, making new inferences from the results of the studies may be easier. We propose pH between 5.5-6.0 and lactic acid value between 0.3-0.5 in curd before salting, minimum pH of 4.5 and maximum 1% lactic acid in cheese after 120 days of ripening. For full-fat Tulum cheese, it would be appropriate to develop an appropriate process so that the humidity is at most 50%, the protein at least 20% and the fat at least 20%. It may probably be the first time that these values, which are not included in the regulations, are proposed by us.

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Conflict of Interest

The authors declare no conflict of interest.

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