

Changes and interactions of milk components over one year by months

Cambios e interacciones de los componentes de la leche durante periodos mensuales a lo largo de un año

Tahire Darbaz¹, Beyza Ulusoy^{2,3*}, Isfendiyar Darbaz^{2,4}, Feride Zabitler Tepik⁴, Canan Hecer⁵, Selim Aslan⁴

¹Cyprus Health and Social Sciences University, Faculty of Veterinary Medicine, Department of Food Hygiene and Technology. Morphou, Cyprus.

²Near East University, DESAM Institute. North Cyprus Mersin 10, Nicosia, Türkiye.

³Near East University, Faculty of Veterinary Medicine, Department of Food Hygiene and Technology. Nicosia, Cyprus.

⁴Near East University, Faculty of Veterinary Medicine, Department of Obstetrics and Gynecology. Nicosia, Cyprus.

⁵Cyprus West University. Famagusta, Cyprus.

*Corresponding author: beyza.ulusoy@neu.edu.tr

ABSTRACT

This study was carried out by collecting tank milk samples every month, from a total of 240 farms producing cold chain milk for one year. Somatic cell count (SCC), total bacterial count (TBC), some nutritional elements (lactose, protein, casein, fat) and some physicochemical parameters (dry matter, fat-free dry matter, freezing point, density, free fatty acids, Soxhlet Henkel acidity degree) and citric acid were analyzed. According to the results; TBC were at the highest level in March (M), April (A) and June (J) (835.07×10^3 ; 940.25×10^3 and 1007.30×10^3 cfu·mL⁻¹), whereas between July (Ju) and December (De), TBC (446.09×10^3 and 795.15×10^3 cfu·mL⁻¹) were significantly lower ($P < 0.001$). The highest SCC were found in M, A and May (Ma), whereas the lowest SCC were found between September (S) and De. Between S and De, when SCC decreased, (varied between 236.13×10^3 cells·mL⁻¹ and 284.43×10^3 cells·mL⁻¹; $P < 0.05$). Lactose values were found to be significantly higher in spring and summer compared to other months. A significant decrease was determined in protein values in the summer months compared to November (N) and De ($P < 0.05$; $P < 0.01$). It was also revealed that casein values were higher in the summer months Ma–August (Au) compared to the other lower months ($P < 0.01$). For physico-chemical parameters, it was determined that non-fat solids and freezing point (FP) values decreased significantly during the summer months ($P < 0.01$; $P < 0.001$). The results obtained show that the parameters in question are seasonally affected, but these parameters change depending on the changes in both TBC and SCC values (high level of positive or negative correlation).

Key words: Bulk tank milk; somatic cell count; raw milk; total bacteria; milk quality

RESUMEN

Este estudio se llevó a cabo recogiendo muestras de leche en tanque cada mes, de un total de 240 explotaciones que mejoraron la redacción durante un año. Recuento de células somáticas (SCC), recuento bacteriano total (TBC), algunos elementos nutricionales (lactosa, proteínas, caseína, grasa) y algunos parámetros fisicoquímicos (materia seca, materia seca magra, punto de congelación, densidad, ácidos grasos libres, Soxhlet Se analizó el grado de acidez Henkel) y el ácido cítrico. De acuerdo a los resultados; Los TBC alcanzaron su nivel más alto en marzo (M), abril (A) y junio (J) ($835,07 \times 10^3$; $940,25 \times 10^3$ y $1007,30 \times 10^3$ ufc·mL⁻¹), mientras que, entre julio (Ju) y diciembre (De), los TBC ($446,09 \times 10^3$ y $795,15 \times 10^3$ ufc·mL⁻¹) fueron mas bajos nivel significativo ($P < 0,001$). El RCS más alto se encontró en M, A y mayo (Ma), mientras que el RCS más bajo se encontró entre septiembre (S) y De. Entre S y De, cuando el SCC disminuyó (varió entre $236,13 \times 10^3$ células·mL⁻¹ y $284,43 \times 10^3$ células·mL⁻¹; $P < 0,05$). Se encontró que los valores de lactosa eran significativamente más altos en primavera y verano en comparación con otros meses. Se determinó una disminución significativa en los valores de proteína en los meses de verano respecto a noviembre (N) y De ($P < 0,05$; $P < 0,01$). También se reveló que los valores de caseína fueron más altos en los meses de verano de mayo a agosto (Au) en comparación con los otros meses más bajos ($P < 0,01$). Para los parámetros físico-químicos, se determinó que los valores de sólidos no grasos (MS) y punto de congelación (FP) disminuyeron significativamente durante los meses de verano ($P < 0,01$; $P < 0,001$). Los resultados obtenidos muestran que los parámetros en cuestión se ven afectados estacionalmente, pero estos parámetros cambian dependiendo de los cambios en los valores tanto de TBC como de SCC (alto nivel de correlación positiva o negativa).

Palabras Clave: Leche cruda en tanque; recuento de células somáticas; leche cruda; bacterias totales; calidad de la leche

INTRODUCTION

Milk quality and safety carry great importance in terms of milk technology, and also for public health [1]. There is a close relationship between obtaining healthy and high-quality milk, herd health and farm hygiene. For example, an infection due to mastitis pathogens causes the activity of secretory cells to deteriorate, which leads to a decrease in lactose, fat and protein synthesis [2]. In addition to reducing the quality of the product, the permeability of cell membranes increases in subclinical and clinical mastitis cases, and the nutritional properties and chemical structure of milk change as substances from the blood pass into the milk [3]. The transfer of relevant pathogens and possible toxins into milk is also a negative situation in terms of food safety. To ensure milk safety on a farm, rapid cooling and tank storage of milk is important. The mandatory cooling and cold storage of raw milk since the 1950s has had a significant impact on the bacteriological and chemical quality of raw milk [4].

In order to prevent the initial microbial load from increasing until the milk is processed and to prevent the activity of some enzymes that will affect the sensory properties of the milk, it must be cooled to the appropriate storage temperature [5]. The food hygiene legislation package came into force throughout the European Union in 2006. The legislation affects all food chain, including caterers, primary producers (such as farmers), manufacturers, distributors and retailers. For the dairy industry, this package of legislation replaces the requirements of the Dairy Hygiene Directive 92/46/EEC [6]. Samples taken from tank milk and laboratory analysis results are indicators of raw milk quality and reliability as well as parameters that give an idea about herd health. In tank milk analyses, the primary determining factor is the somatic cell count (SCC) and the total bacterial count of the milk [7].

SCC in raw milk is one of the indicators of animal udder health and milk hygiene [8]. Somatic cells are among the cells that form the body's natural defense system, and the SCC count in milk taken from a healthy cow's udder is expected to be below 200,000 cells·mL⁻¹. Acceptable SCC in samples taken from tank milk should be below 400,000 cells·mL⁻¹. There is a positive relationship between increases in the number of somatic cells in milk and the degree of inflammation [6, 9]. It has been reported that in herds where udder health control programs have been implemented for a long time, 80–90% success is achieved in protecting against udder infections, and as a result of these controls, quality and safe raw milk can be obtained [10]. If the microorganism load and SCC in milk are above limits suggests the existence of factors that may threaten human health [11], and is also an indication that it will create quality problems in the processing of dairy products and also cause milk production losses [12].

Since the quality of milk and dairy products depends on the composition of raw milk and the content of nutrients, the factors responsible for changes in the composition and physico-chemical properties of raw milk are of great importance for milk processors [13]. The composition covers the main nutritional elements of milk such as fat, protein, lactose and total dry matter [14, 15]. The effects of seasonal change on milk yield and composition have been investigated by many researchers [16, 17]. It has also been reported that there is a positive correlation between the increase in SCC and changes in milk composition [15].

With this study, it was aimed to reveal the relationship between SCC and TBC depending on seasonal changes and the changes in the nutritional elements and physico-chemical parameters in milk according to months. It is also aimed to reveal how these parameters affect each other.

MATERIAL AND METHODS

This study was carried out by collecting 2,880 milk tank samples from a total of 240 farms producing cold chain milk throughout North Cyprus between March and February. In order for the study to accurately reflect the situation in North Cyprus, all farms producing cold chain milk for at least 1 year were included in the scope of the study. Samples were collected regularly in the first week of each month from 240 farms. Milk samples were filled into sterile sample bottles with a sterile dipping container from the mixed tank within 1 hour of morning milking, in accordance with aseptic rules, and are kept under cold chain (+4°C) without freezing, to be subjected to analysis.

Analyses applied to milk samples

All analyses applied to milk were carried out in the Turkish Cypriot Dairy Industry Institution Quality Control Department Laboratory. BactoScan™ FC, (Foss, Denmark) [18] for total bacterial count, Fossomatic™ FC 5000 (Foss, Denmark) [19] for Somatic Cell Count and MilkoScan™ FT-120 (Foss, Denmark), were used for the detection of Lactose, Total Protein, Casein Fat, Non-Fat Solids, Dry Matter, Freezing Point, Acidity-SH, Density, Free Fatty Acids, Citric Acid (TABLE I) [20].

TABLE I
Devices used in the analysis of milk samples and their relevant references

Device Name	Performed Analyses	Relevant References
BactoScan™ FC	Total Bacterial Count	ISO/IDF and FDA/NCIMS
Fossomatic™ FC 5000	Somatic Cell Count	AOAC ISO 13366-2 / IDF 148-2:2006
MilkoScan™ FT-120	Lactose, Total Protein, Casein Fat, Non-Fat Solids, Dry Matter, Freezing Point, Acidity-SH, Density, Free Fatty Acids, Citric Acid	AOAC and IDF

Statistical analysis

SPSS Statistics 26.0, IBM, USA program was used for statistical evaluation. Descriptive statistics (Mean ± Std. Dev.) were applied for the mean value and standard error. The homogeneity distribution of values was tested with Shapiro-Wilk. For non-homogeneous data, the Kruskal Wallis test was used to test the difference between two groups and the overall difference between all groups was determined, and then the Mann-Whitney U test was used to test the difference between each group. One-Way ANOVA (Tukey's) was used for normally distributed data, and One-Way ANOVA (Tamhane's) was used for the non-homogeneous distribution of values. T-test test was used for homogeneously distributed data. In terms of statistical significance, results with $P < 0.05$ and below were considered significant.

RESULTS AND DISCUSSION

During the data collection of this study, every month, the samples were collected from same farms. Visits were planned in the first week of every month and samples were gathered with same conditions every time. According to the results (TABLE II and FIG. 1); TBC were at their highest level in March (M), April (A) and June (J) (835.07×10^3 ; 940.25×10^3 and 1007.30×10^3 cfu·mL⁻¹), whereas TBC between July (Ju) and December (De) (446.09×10^3 and 795.15×10^3 cfu·mL⁻¹) has been shown to decrease significantly ($P < 0.001$). The reason why it wasn't obtained to be high in Ma is the reflection of weather changes in that period of seasons.

TABLE II
Analysis results of SCC, TBC and some nutrients in tank milk by months¹

n:240	SCC cell·mL ⁻¹	TBC cfu·mL ⁻¹	Lactose %
March 2019	553.94 × 10 ³ ± 417.45 × 10 ³ b*;c*** (33–2121 × 10 ³)	835.07 × 10 ³ ± 1959.43 × 10 ³ a* (10–16933 × 10 ³)	4.50 ± 0.11 b*** (3.94–5.05)
April 2019	526.06 × 10 ³ ± 379.20 × 10 ³ b***;c*** (26–2013 × 10 ³)	940.25 × 10 ³ ± 2247.24 × 10 ³ a* (7–17339 × 10 ³)	4.49 ± 0.11 a***; b***; c*** (3.78–4.73)
May 2019	463.88 × 10 ³ ± 329.94 × 10 ³ b***;c*** (65–1628 × 10 ³)	636.67 × 10 ³ ± 1093.37 × 10 ³ (8–7852 × 10 ³)	4.50 ± 0.10 b**; b***; c***; d*** (4.18–4.71)
June 2019	371.93 × 10 ³ ± 277.57 × 10 ³ b***;c***;c* (35–1564 × 10 ³)	1007.30 × 10 ³ ± 2102.96 × 10 ³ a* (7–13155 × 10 ³)	4.48 ± 0.11 b*** (4.05–5.25)
July 2019	291.27 × 10 ³ ± 235.76 × 10 ³ b**;b***;b* (29–1802 × 10 ³)	522.27 × 10 ³ ± 1101.16 × 10 ³ b* (11–6792 × 10 ³)	4.42 ± 0.10 a***; a**; c*** (3.56–4.65)
August 2019	315.87 × 10 ³ ± 270.71 × 10 ³ b*;b***;d** (34–1802 × 10 ³)	499.40 × 10 ³ ± 1013.08 × 10 ³ (5–9522 × 10 ³)	4.41 ± 0.11 a***; a**; c*** (3.68–4.68)
September 2019	293.06 × 10 ³ ± 254.75 × 10 ³ b*;b*** (36–1662 × 10 ³)	475.95 × 10 ³ ± 1016.20 × 10 ³ b* (6–9913 × 10 ³)	4.40 ± 0.10 a***; a**; d*** (3.91–4.77)
October 2019	236.13 × 10 ³ ± 200.05 × 10 ³ b*;b***;b** (28–1574 × 10 ³)	446.09 × 10 ³ ± 947.12 × 10 ³ b* (6–9604 × 10 ³)	4.39 ± 0.12 a***; a**; d*** (3.93–5.07)
November 2019	278.30 × 10 ³ ± 277.25 × 10 ³ b*;b*** (19–2201 × 10 ³)	791.32 × 10 ³ ± 2130.66 × 10 ³ (6–17339 × 10 ³)	4.40 ± 0.16 a***; a**; d*** (3.46–6.11)
December 2019	284.43 × 10 ³ ± 283.63 × 10 ³ b* (16–2198 × 10 ³)	795.15 × 10 ³ ± 1808.80 × 10 ³ (8–16906 × 10 ³)	4.43 ± 0.10 a***; a**; d*** (3.93–4.70)
January 2020	419.82 × 10 ³ ± 386.08 × 10 ³ a*;a**;a*** (22–2737 × 10 ³)	813.10 × 10 ³ ± 1823.71 × 10 ³ (10–15608 × 10 ³)	4.41 ± 0.10 a***; a** (3.95–4.66)
February 2020	323.69 × 10 ³ ± 281.64 × 10 ³ a*;a**;a***;c*** (25–1397 × 10 ³)	557.46 × 10 ³ ± 1227.65 × 10 ³ (12–12647 × 10 ³)	4.43 ± 0.10 a***; a** (4.03–4.69)

¹The ¹numbers in the table were given as Mean ± Std. Dev. (Min – Max). Values marked with different letters (a;b; a.c, a.d, b;c; c:d) are statistically different from each other. Asterisks indicate *, **, *** $P < 0.05$; $P < 0.01$ and $P < 0.001$ values, respectively. SCC: somatic cell count; TBC: total bacterial

TABLE II cont...
Analysis results of SCC, TBC and some nutrients in tank milk by months¹

n:240	Protein %	Casein %	Fat %
March 2019	3.32 ± 0.14 b*;c*** (2.72–3.92)	2.62 ± 0.13 a***;b***;c** (1.96–3.28)	3.55 ± 0.44 b***;b**;d*** (2.03–4.47)
April 2019	3.30 ± 0.14 b***;c*** (2.72–3.61)	2.60 ± 0.14 a***; b***;c** (1.89–2.93)	3.56 ± 0.37 b***;b** (2.34–4.66)
May 2019	3.26 ± 0.14 b*** (2.89–3.75)	2.57 ± 0.13 b***;b** (2.20–2.95)	3.46 ± 0.35 b*** 2.18–4.36
June 2019	3.22 ± 0.14 b*** (2.76–3.66)	2.54 ± 0.13 b***;c**;c*** (2.05–3.07)	3.45 ± 0.33 b*** (2.43–4.89)
July 2019	3.21 ± 0.13 b*** (2.50–3.49)	2.51 ± 0.12 b***;c*** (1.84–2.78)	3.39 ± 0.29 b***;e*** (2.18–4.06)
August 2019	3.20 ± 0.14 b*** (2.71–3.60)	2.50 ± 0.13 b***;c*** (1.83–2.87)	3.42 ± 0.33 b***;e*** (1.95–4.30)
September 2019	3.31 ± 0.14 c*** (2.90–3.76)	2.60 ± 0.12 b***;b**;c*** (2.17–3.02)	3.50 ± 0.35 b*** (1.75–4.64)
October 2019	3.39 ± 0.15 b***;c*** (3.02–3.74)	2.63 ± 0.13 a***;b***;d*;d*** (2.29–2.95)	3.61 ± 0.36 b***;c*** (2.06–4.36)
November 2019	3.44 ± 0.18 b***;c*** (2.74–4.29)	2.68 ± 0.16 b***;d*** (1.81–3.73)	3.79 ± 0.33 a***;b***;b** (1.95–4.62)
December 2019	3.47 ± 0.15 b***;c*** (3.09–3.94)	2.7 ± 0.13 a***;b***;d*** (2.33–3.04)	3.87 ± 0.34 a***;b***;b**;d*** (2.28–4.84)
January 2020	3.36 ± 0.14 a* (2.98–3.89)	2.64 ± 0.13 a*** (2.21–3.07)	3.80 ± 0.34 a*** (2.42–4.84)
February 2020	3.31 ± 0.13 b**;c*** (2.98–3.77)	2.61 ± 0.12 a***; b*** (2.20–2.93)	3.70 ± 0.33 a***;a**;c*** (2.17–5.12)

¹The ¹numbers in the table were given as Mean ± Std. Dev. (Min – Max). Values marked with different letters (a;b; a.c, a.d, b;c; c:d) are statistically different from each other. Asterisks indicate *, **, *** $P < 0.05$; $P < 0.01$ and $P < 0.001$ values, respectively

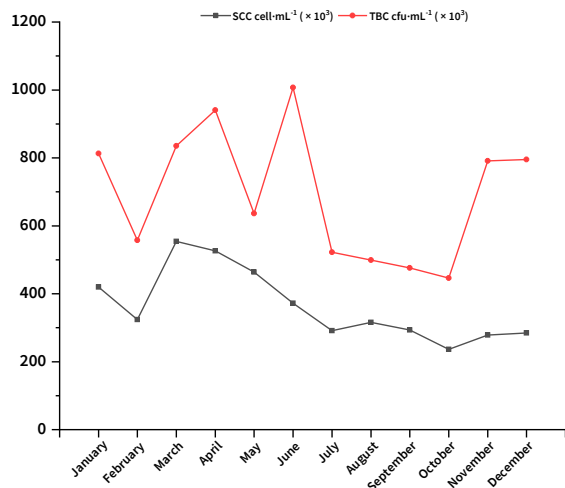


FIGURE 1. Analysis results of SCC and TBC in tank milk by months

In M and A the weather was more rainy but dry in Ma. It is thought that in J warmer climate accelerated the TBC increase. On the other hand, these months are the months when the animals are taken to pasture feeding, the rumen content changes and TBC values are higher due to the stress it creates. The highest SCC were found in M, A and May (Ma), whereas the lowest SCC were found between September (S) and De. Between S and De, when SCC decreased, values varied between 236.13×10^3 cells·mL⁻¹ and 284.43×10^3 cells·mL⁻¹ ($P < 0.05$). While lactose values reached the highest average value of 4.48–4.50% in M, A, Ma and J, it was revealed that significantly different values (between 4.39% and 4.42%) were obtained between these months and other months. ($P < 0.01$; $P < 0.001$).

A high level of positive correlation ($r = 0.979$) was found between both TBC and SCC and lactose values ($P < 0.05$; $P < 0.01$). The acidity may not be directly related with Protein values were found to be significantly lower ($P < 0.0001$), with a range of 3.20% and 3.26% between Ma and Au. On the other hand, between November (N) and De, protein values (FIG. 2) increased to the highest levels with average values of 3.44%

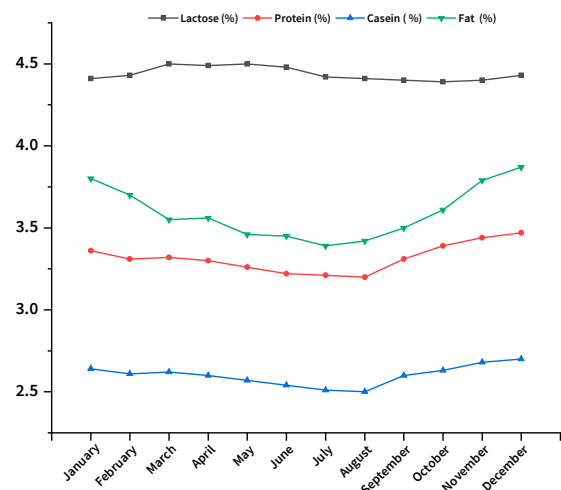


FIGURE 2. Analysis results of Lactose, Protein, Casein and Fat in tank milk by months

and 3.47% ($P < 0.0001$). The lowest casein values were found between Ma and Au (Ma 2.57%; J 2.54%; Ju 2.51%; Au 2.50%), while the data obtained in the other months were between 2.60% and 2.70% and was found to be significantly higher compared to the lowest months ($P < 0.001$; $P < 0.01$). The highest casein values were obtained in N and De with 2.68% and 2.70%. The highest mean fat values were in N (3.79%), De (3.87%), January (Ja) (3.80%), February (F) (3.70%) and decreased to 3.39% and 3.42% in Ju and Au, respectively ($P < 0.001$). It was determined that starting from S, the values increased again ($P < 0.001$; $P < 0.0001$) (TABLE II, FIG. 1).

While individual SCC being higher than 250,000 cells·mL⁻¹ suggests the possibility of intra-mammary infection in the cow, if SCC measured in tank milk is higher than 400,000 cells·mL⁻¹, there are udder health problems, so dairy cows should be checked and the necessary precautions should be taken [9, 21]. According to the results of this study, the mean values were significantly higher ($> 400,000$ cells·mL⁻¹) in M, A and Ma, while they were significantly lower between J and D (236,000–371,000 cells·mL⁻¹; $P < 0.01$; $P < 0.001$). With these results, SCC values were significantly lower in the months when both the environmental temperatures were warmer. According to Northern Cyprus, Ministry of Public Works and Transportation, Meteorology Department; the climatic conditions of Cyprus, the environmental temperature did not fall below 10°C. Tank milk acceptable SCC levels vary between countries from past to present. While maximum limit values of $< 750,000$ cells·mL⁻¹ in the USA, $< 500,000$ cells·mL⁻¹ in Canada, and $< 400,000$ cells·mL⁻¹ in European Union laws are considered ideal levels, in recent years these values have been further reduced to below 200,000 cells·mL⁻¹ [21, 22]. When $< 400,000$ cells·mL⁻¹ in EU legislation is taken as the reference value, it has been observed that there are farms where this value is exceeded even in the months when the average SCC value is lowest.

Under similar conditions, in the north of Cyprus, Darbaz *et al.* [23] reported that the average value of tank milk SCC was 521,583 cells·mL⁻¹ and concluded that SCC remained high. It should be taken into consideration that increase in SCC is the indicator for poor quality and changes the composition of milk and thus will have a negative impact on dairy products technology [23, 24]. The important result of this study is that high SCC values were obtained between M and Ma, and in general, SCC values are significantly below 400,000 mL/cell in other months. Different studies also advocate the view that SCC values may change depending on the season. As Riekerink *et al.* [25] mentioned in their study bulk tank milk SCC in a herd is mainly influenced by the prevalence and incidence of subclinical and clinical mastitis, which depends on variation of factors such as parity, stage of lactation, type of housing, access to pasture, management, and also temperature, humidity, and season.

In New Zealand, SCC achieved the highest levels from Ju to S (around the calving period) and the lowest SCC occurred in S and O, shortly after the calving period, and SCC then slowly increased again towards the end of the season in A to Ma [26]. Morse *et al.* [27] reported that intramammary infection was more common in seasons where temperature and humidity prevailed. The dry and very hot summer climate in Cyprus and the abundant rain in winter and spring explain the high SCC during this period. In the mild and rainy months of the year, the potential for microorganisms to multiply is higher, which creates a hygiene problem in the farm environment. It is thought that there is an increase in SCC average values in these months due to the lack of necessary hygiene, especially in rainy and unsuitable barn conditions. It is noteworthy that SCC values are low

in Ju and Au. As Özlem & Kul [15] concluded in their study, the season had significant effect ($P < 0.01$) on SCC and it was highest in summer and the lowest in winter. Pavel and Gavan [28] and Maciuc et al. [29] concluded the similar seasonal effect on SCC.

The researchers reported that higher mean results of SCC in the summer, agreed on reasons such as high temperatures and humidity which expose animals to a greater number of pathogens and increasing occurrence of mastitis [30]. Considering the climate of the Island of Cyprus, the months when temperatures and humidity increase are generally spring months, and on the contrary, summer months are dry. This situation explains the highest SCC value obtained in M, A and Ma. Although Ju and Au were hot, the environment was dry and the areas where the animals slept and roamed were better cleaned, which also contributed to the result. Apparently, in different countries, depending on different conditions, SCC values may exceed the threshold limit in different months, but may also be lower in other months.

At current study; results of the analyses to determine the total number of bacteria show the total number, not the bacterial profile. Although bacterial identification provides a satisfactory idea to determine the quality status and shelf life of raw milk, critical limits are set up only depending on the total number by international standards. In the European Union and the United States, the legal limit for raw cow's milk TBC is 100,000 cfu·mL⁻¹, in Canada, 50,000 cfu·mL⁻¹, and in Brazil, 100,000 cfu·mL⁻¹ [31]. In this study, the results are given in cfu·mL⁻¹ units (TABLE III). In this study, some samples' results above 100,000 cfu·mL⁻¹, which is specified as critical limit in the European Union Directive (EC) No 853/2004, were observed [6]. TBC mean values were also high, especially between M and J, when SCC values

reached the highest value. This relationship was revealed by the high degree of positive correlation between TBC and SCC values 0.979; $P < 0.005$ (TABLE IV). Berry et al. [32] documented temporal trends in SCC and TBC in Irish dairy herds during the years 1994 to 2004. According to the conclusion of that study, SCC decreased during the years 1994 to 2000, followed by an annual increase thereafter of more than 2,000 cells·mL⁻¹. As concluded by the researchers, the reason of the decline in mean bulk tank SCC may be due in part to a dilution effect of greater yields per cow.

On the other hand; increased awareness of farmers of cows with elevated SCC, and the impact of EU policies were mentioned to be other factors for the decrease. Across all years, bulk tank SCC was the lowest in A and highest in N; TBC were the lowest in May and highest in December. The significant seasonal pattern observed in herd SCC and TBC was an artifact of seasonal calving in Ireland. The climate difference between Ireland and Cyprus can be shown as the reason for the differences in seasonal distribution results between this study and the current study. Apparently, TBC values also vary seasonally in Cyprus. On the other hand, in parallel with results of current study, it was revealed that there was a positive correlation between SCC and TBC. Previous research has also found highly positive correlations between TBC, SCC and milk yield [31]. Berry et al. [32] examined the changes in SCC and TBC according to months and revealed that there was a correlation between increases or decreases according to months.

Another important result obtained in this study was that changes in all parameters generally affected other parameters. TABLE II, FIG.2 and TABLE III, FIG. 3 presents some of the chemical and physico-chemical results, respectively. For example, for casein, it

TABLE III
Analysis results of some physico-chemical parameters by months¹

n:240	FF-DM (%)	DM (%)	FP	SH Acidity (°SH)
March 2019	8.81 ± 0.23 (a;b;d) (7.46–10.13)	12.32 ± 0.52 (b***;#) (10.56–14.27)	0.54 ± 0.01 (a***) (0.48–0.64)	6.89 ± 0.50 (b***) (5.79–10.47)
April 2019	8.75 ± 0.25 (a;b;c;d;e) (7.26–9.31)	12.29 ± 0.48 (b***;#;α;&) (10.05–13.41)	0.54 ± 0.01 (a***) (0.47–0.57)	6.97 ± 0.46 (b***) (5.34–8.03)
May 2019	8.69 ± 0.23 (a;b;c;e) (7.93–9.24)	12.15 ± 0.45 (b***;c***;α;&) (10.41–13.49)	0.54 ± 0.01 (a**;a***) (0.50–0.57)	6.90 ± 0.43 (b**) (5.92–8.12)
June 2019	8.62 ± 0.25 (b;d;c) (7.66–10.02)	12.09 ± 0.44 (b***;c***;α;&*) (10.66–13.31)	0.54 ± 0.02 (a**;a***) (0.49–0.66)	6.97 ± 0.51 (5.24–8.51)
July 2019	8.52 ± 0.22 (b;d;c) (6.94–9.02)	11.95 ± 0.40 (b***;c***;α;μ;*) (10.26–12.84)	0.53 ± 0.01 (b***) (0.45–0.56)	6.70 ± 0.50 (b**;b***) (4.80–8.20)
August 2019	8.48 ± 0.21 (b***;d;e) (7.13–9.04)	11.95 ± 0.45 (b***;c***;α;μ) (9.22–13.12)	0.53 ± 0.01 (b***) (0.45–0.56)	6.65 ± 0.44 (b***) (4.47–8.19)
September 2019	8.62 ± 0.20 (b;d;c) (7.86–9.51)	12.14 ± 0.43 (b***;c***;#; α;μ) (10.73–14.00)	0.54 ± 0.01 (0.49–0.60)	6.99 ± 0.43 (5.59–8.28)
October 2019	8.73 ± 0.23 (a;b;c;e) (7.91–9.70)	12.33 ± 0.47 (b***;b***) (10.40–13.76)	0.53 ± 0.01 (b**;b***) (0.49–0.62)	7.28 ± 0.54 (b**) (5.14–9.38)
November 2019	8.81 ± 0.32 (a*;b;d) (6.94–11.77)	12.56 ± 0.52 (c**;#;α;μ) (9.75–14.25)	0.54 ± 0.02 (0.42–0.74)	7.36 ± 0.63 (b***) (5.15–10.23)
December 2019	8.90 ± 0.22 (b*;b***;d) (8.00–9.41)	12.73 ± 0.43 (b***;c***;α;μ) (11.44–13.81)	0.54 ± 0.013 (0.49–0.57)	7.51 ± 0.73 (b***) (5.91–15.75)
January 2020	8.76 ± 0.22 (a) (7.68–9.44)	12.52 ± 0.44 (a) (11.22–13.99)	0.54 ± 0.01 (a**;a***) (0.51–0.58)	6.88 ± 0.44 (a**;a***) (5.69–8.12)
February 2020	8.71 ± 0.20 (a;b;c) (7.98–9.18)	12.38 ± 0.43 (b**) (10.94–13.96)	0.54 ± 0.01 (a**;a***) (0.51–0.57)	6.77 ± 0.45 (a**;a***) (5.51–8.37)

¹The numbers in the table were given as Mean ± Std. Dev. (Min – Max). Values marked with different letters (a;b; a,c, a,d, b;c; c;d) are statistically different from each other. Asterisks indicate *, **, *** $P < 0.05$; $P < 0.01$ and $P < 0.001$ values, respectively. FF-DM: fat free dry matter, DM: dry matter, FP: freezing point, SH: soxhlet Henkel

TABLE III cont...
Analysis results of some physico-chemical parameters by months¹

n:240	D (gr/cm ³)	FFA (%)	CA (%)
March 2019	1031.63 ± 1.03 (b***;c***) (1026.00-1036.00)	0.66 ± 0.22 (a**;a***) (0.20-1.96)	0.14 ± 0.02 (a**;a***) (0.09-0.18)
April 2019	1031.46 ± 1.02 (b**;c***) (1026.00-1034.00)	0.67 ± 0.20 (a**;a***) (0.31-1.61)	0.13 ± 0.01 (a***) (0.10-0.18)
May 2019	1031.40 ± 0.92 (b*;b**) (1028.00-1034.00)	0.75 ± 0.22 (0.25-1.54)	0.13 ± 0.01 (a**;a***;b**) (0.09-0.18)
June 2019	1031.08 ± 1.01 (a*;a**;a***;d***) (1027.00-1037.00)	0.77 ± 0.24 (b**;b***) (0.11-2.09)	0.13 ± 0.01 (b*;b**) (0.10-0.18)
July 2019	1030.72 ± 0.92 (b**;d***) (1024.00-1033.00)	0.78 ± 0.22 (b**;b***) (0.21-1.86)	0.12 ± 0.01 (a*;a***) (0.10-0.17)
August 2019	1030.56 ± 0.92 (b***;d***) (1026.00-1033.00)	0.80 ± 0.20 (b***) (0.39-1.61)	0.12 ± 0.01 (a***) (0.08-0.18)
September 2019	1030.84 ± 0.84 (b***;d***) (1028.00-1033.00)	0.73 ± 0.18 (b**) (0.28-1.31)	0.12 ± 0.01 (a***) (0.07-0.18)
October 2019	1031.20 ± 1.00 (a**;b***;d***) (1028.00-1037.00)	0.76 ± 0.28 (b***) (0.32-3.38)	0.13 ± 0.01 (a**;a***;c**) (0.10-0.19)
November 2019	1031.36 ± 1.29 (1024.00-1045.00)	0.77 ± 0.20 (a**;b***) (0.18-1.36)	0.13 ± 0.01 (0.10-0.22)
December 2019	1031.60 ± 0.968 (b***) (1026.00-1035.00)	0.78 ± 0.24 (b***) (0.35-2.70)	0.13 ± 0.01 (0.11-0.18)
January 2020	1031.06 ± 0.97 (a**;a***) (1026.00-1035.00)	0.69 ± 0.23 (a**;a***) (0.16-1.82)	0.13 ± 0.01 (a***) (0.10-0.18)
February 2020	1031.07 ± 0.87 (a**;a***) (1028.00-1035.00)	0.69 ± 0.19 (a**;a***) (0.16-1.40)	0.13 ± 0.01 (a***) (0.10-0.17)

¹The numbers in the table were given as Mean ± Std. Dev. (Min - Max). Values marked with different letters (a:b; a:c, a:d, b:c; c:d) are statistically different from each other. Asterisks indicate *, **, *** P<0.05; P<0.01 and P<0.001 values, respectively. D: density, FFA: free fatty acid, CA: citric acid

TABLE IV
Evaluation of the correlation between SHS and total bacterial count and the levels of other parameters

	TBC	Lactose	Protein	Casein	Fat	FF-DM	DM	FP	SH	D	FFA	CA
SCC	0.979** P<0.005 (0.004)	-0.968** P<0.05 (0.007)	0.936** P<0.01	NS	NS	-0.913* P<0.05	NS	-0.910* P<0.05	NS	P>0.05	NS	NS
TBC		-0.978** P<0.01	0.924* P<0.05	-0.876 P<0.05	NS	-0.973** P<0.005	NS	-0.948* P<0.01	NS	-0.879* P<0.05	0.922* P<0.05	NS

** : The correlation is significant at the 0.01 level, * : The correlation is significant at the 0.05 level, NS: Not significant, SCC: somatic cell count, TBC: total bacterial count, FF-DM: fat free dry matter, DM: dry matter, FP: freezing point, SH: Soxhlet Henkel, D: density, FFA: free fatty acid, CA: citric acid

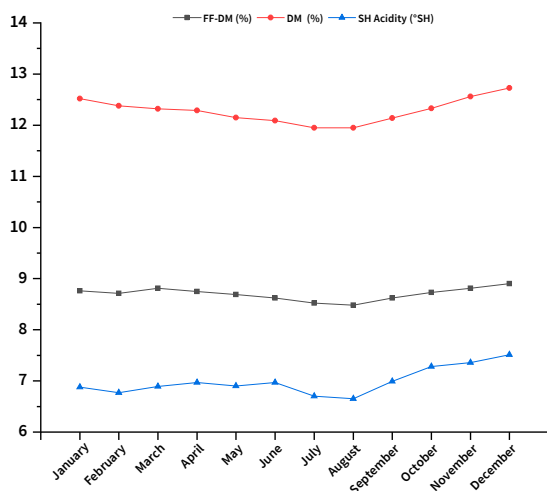


FIGURE 3. Analysis results of some physico-chemical parameters (FF-DM, DM and SH Acidity) by months

was found that there was a positive correlation between 0.702 and 0.975 with protein, fat, FF-DM, SH and DM parameters (P<0.01 and P<0.001). Similarly, FF-DM was significantly positively correlated with protein, casein, fat and DM (r=0.819-0.933; P<0.001), while fat was significantly positively correlated with protein, casein, FF-DM, DM, SH (r=0.661-0.910; P<0.05 and P<0.001). Likewise, there was a high degree of positive correlation between DM, SH, D and CA and these parameters (TABLE IV).

CONCLUSION

According to the results, the increase in TBC with the increase in SCC values showed that there is a possibility of pathogens reaching consumers through raw milk. These two parameters affect each other and show that changes in other milk components also cause changes in other milk components. With this study, we see that this situation also varies depending on climatic conditions through the year. The increase in SCC in the spring months, when the animals are new to pasture and the season in Cyprus is mild and rainy, and even exceeds critical limits in

some months, requires more attention to pasture–farm management, mastitis control programs and farm hygiene during these periods. Because chemical parameters and physico–chemical parameters also change with the changes in SCC and TBC, in which case negative effects on dairy products technology and economic losses are expected to occur. In order to prevent all these negative scenarios, GMP (good manufacturing practice) and GAP (good agricultural practice) are also gaining importance on the basis of farm management.

Conflict of interest

The authors declare there are no conflicts of interest.

BIBLIOGRAPHIC REFERENCES

- [1] Özek K. Süt Sığırlarında Süt Kompozisyonunu Etkileyen Faktörler ve Besleme – Süt Kompozisyonu İlişkisi [Factors affecting composition of milk in dairy cattle and relation between nutrition and milk composition]. Bahri Dağdaş Hayvancılık Araştırma Dergisi [Internet]. 2015 [cited 24 May 2024]; 4(2):37–45. Turkish. Available in: <https://goo.su/u4xo3>
- [2] Ma Y, Ryan C, Barbano DM, Galton DM, Rudan MA, Boor KJ. Effects of somatic cell count on quality and shelf–life of pasteurized fluid milk. J. Dairy Sci. [Internet]. 2000; 83(2):264–274. doi: <https://doi.org/czwd4v>
- [3] Le Roux Y, Laurent F, Moussaoui F. Polymorphonuclear proteolytic activity and milk composition change. Vet. Res. [Internet]. 2003; 34(5):629–645. doi: <https://doi.org/cv38sz>
- [4] Akan E, Yerlikaya O, Kınık Ö. Psikrotrof Bakterilerin Çiğ Süt ve Süt Ürünleri Kalitesine Etkisi [Effect of psychrotrophic bacteria on quality of raw milk and dairy products]. Akademik Gıda. [Internet]. 2014 [cited 15 May 2024]; 12(4):68–78. Turkish. Available in: <https://goo.su/x2s6dz3>
- [5] Günhan T, Demir V, Bilgen H. Çiftlik Tipi Süt Soğutma Tanklarının Performans Değerlerinin Deneysel Olarak Belirlenmesi [Experimental determination of performance of farm type bulk milk coolers]. Tarım Makinaları Bilimi Dergisi [Internet]. 2006 [cited 01 Dec. 2023]; 2(4):369–379. Turkish. Available in: <https://goo.su/HvdIP>
- [6] Komorowski EDS. New dairy hygiene legislation. Int. J. Dairy Technol. [Internet]. 2006; 59(2):97–101. doi: <https://doi.org/cwqqv>
- [7] Diler A, Baran A. Erzurum'un Hınıs İlçesi Çevresindeki Küçük Ölçekli İşletme Tank Sütlerinden Alınan Çiğ Süt Örneklerinin Bazı Kalite Özelliklerinin Belirlenmesi [Determination of some quality characteristics of raw cow milk on smallholder dairy farms in Hınıs district of Erzurum]. Alinteri [Internet]. 2014 [cited 21 May 2024]; 26(1):18–24. Turkish. Available in: <https://goo.su/pzW30P>
- [8] Tsenkova R, Atanassova S, Kawano S, Toyoda K. Somatic cell count determination in cow's milk by near–infrared spectroscopy: A new diagnostic tool. J. Anim. Sci. [Internet]. 2001; 79(10):2550–2557. doi: <https://doi.org/g8v9xx>
- [9] Küplülü Ş, Vural R. Büyük ruminantlarda meme sağlığı kontrol programları [Udder health control programs in large ruminants]. In: Kaymaz M, Fındık M, Rışvanlı A, Köker A, editors. Evcil Hayvanlarda Meme Hastalıkları [Mammary diseases in domestic animals]. Malatya (Türkiye): Medipres Yayıncılık. 2016. p. 261–294. Turkish.
- [10] Petersson–Wolfe CS, Tholen AR, Currin J, Leslie KE. Practical methods for mastitis control. WCDS Adv. Dairy Techn. [Internet]. 2013 [cited 15 Jan. 2023]; 25:341–358. Available in: <https://goo.su/jsqah>
- [11] Manlongat N, Yang TJ, Hinckley LS, Bendel RB, Krider HM. Physiologic–chemoattractant–induced migration of polymorphonuclear leukocytes in milk. Clin. Diagn. Lab. Immunol. [Internet]. 1998; 5(3):375–381. doi: <https://doi.org/g8v9xz>
- [12] Miller RH, Paape MJ, Fulton LA, Schutz MM. The relationship of milk somatic cell count to milk yields for Holstein heifers after first calving. J. Dairy Sci. [Internet]. 1993; 76(3):728–733. doi: <https://doi.org/c76fm6>
- [13] Ulusoy BH, Hecer C, Berkan Ş. Investigation of microbiological hazards in traditional Halloumi/Hellim manufacturing process. Atatürk Üniv. Vet. Bilim. Derg. [Internet]. 2020; 15(3):196–206. doi: <https://doi.org/g8v9x2>
- [14] Guo JZ, Liu XL, Xu AJ, Xia Z. Relationship of somatic cell count with milk yield and composition in chinese Holstein population. Agric. Sci. China [Internet]. 2010; 9(10):1492–1496. doi: <https://doi.org/b967xf>
- [15] Özlem O, Kul E. Effects of some environmental factors on somatic cell count and milk chemical composition in cow bulk tank milk. Akademik Ziraat Derg. [Internet]. 2020; 9(1):163–170. doi: <https://doi.org/g8v9x3>
- [16] Maha EH, Linda Z, Abdellah EH, Mohamed B, Ahmed D, Abdelmajid Z, Khadija O, Mohammed B. Seasonal effect on bovine raw milk composition of Oulmes local race in Morocco. Food Sci. Qual. Manag. [Internet]. 2016 [cited 01 Dec. 2023]; 52:49–55. Available in: <https://goo.su/KiKKQ5>
- [17] Yang L, Yang Q, Yi M, Pang ZH, Xiong BH. Effects of seasonal change and parity on raw milk composition and related indices in chinese Holstein cows in northern China. J. Dairy Sci. [Internet]. 2013; 96(11):6863–6869. doi: <https://doi.org/f5d6vc>
- [18] Wang M, Bai Z, Liu S, Fu B, Liu Y, Wang Z, Zhou G, Gong X, Jiang Y, Sui Z. Rapid and universal quantification of viable bacteria with growth activity in raw milk using a fluorescent d–amino acid–flow cytometry (FDAA–FCM) method. LWT [Internet]. 2023; 184:114998. doi: <https://doi.org/g8v9x4>
- [19] Saeed SI, Mat–Yazid KA, Hashimy HA, Dzulkifli SK, Nordin F, Nik–Him NA, Fahmi–Bin Omar MF, Akilil E, Mohamad M, Salma Zalati CW, Kamaruzzaman NF. Prevalence, antimicrobial resistance, and characterization of *Staphylococcus aureus* isolated from subclinical bovine mastitis in east coast Malaysia. Animals [Internet]. 2022; 12(13):1680. doi: <https://doi.org/g8v9x5>
- [20] Kazou M, Gavriil A, Kalagkatsi O, Paschos T, Tsakalidou E. The Impact of different inoculation schemes on the microbiota, physicochemical and sensory characteristics of Greek Kopanisti cheese throughout production and ripening. Microorganisms [Internet]. 2023; 11(1):66. doi: <https://doi.org/g8v9x6>
- [21] Tosun S, Baki Acar D. Tekirdağ İlinde Bulunan Süt Sığırcılığı İşletmelerinde Sağım Hijyeni Uygulamaları ile Tank Sütü Somatik Hücre ve Toplam Bakteri Sayılarının Karşılaştırılması [The comparison of milking hygiene with bulk tank somatic cell count and total bacterial count in dairy herds in Tekirdag province]. Kocatepe Vet. J. [Internet]. 2019; 12(3):292–299. Turkish. doi: <https://doi.org/g8v9x7>

- [22] Vural R, Ergün Y, Özenç E. Büyük ruminantlarda mastitis [Mastitis in large ruminants]. In: Kaymaz M, Fındık M, Rişvanlı A, Köker A, editors. Evcil Hayvanlarda Meme Hastalıkları [Mammary diseases in domestic animals]. Malatya (Türkiye): Medipres Yayıncılık. 2016. p. 149–259. Turkish.
- [23] Darbaz İ, Baştan A, Salar S. Investigation of udder health and milk quality parameters of dairy farms in Northern Cyprus. Part I: SCC and bacteriologic examination. Ankara Üniv. Vet. Fak. Derg. [Internet]. 2018 [cited 25 May 2023]; 65(2):145–154. Available in: <https://goo.su/fSycx>
- [24] Darbaz I, Ulusoy BH, Darbaz T, Hecer C, Aslan S. The importance of somatic cell count in dairy technology. J. Dairy Prod. Process. Improv. [Internet]. 2023; 73(2):75–84. doi: <https://doi.org/g8v9x9>
- [25] Olde-Riekerink RGM, Barkema HW, Stryhn H. The effect of season on somatic cell count and the incidence of clinical mastitis. J. Dairy Sci. [Internet]. 2007; 90(4):1704–1715. doi: <https://doi.org/d63k8c>
- [26] Clements ACA, Pfeiffer DU, Hayes D. Bayesian spatio-temporal modelling of national milk-recording data of seasonal-calving New Zealand dairy herds. Prev. Vet. Med. [Internet]. 2005; 71(3–4):183–196. doi: <https://doi.org/bp8vqs>
- [27] Morse D, DeLorenzo MA, Wilcox CJ, Collier RJ, Natzke RP, Bray DR. Climatic effects on occurrence of clinical mastitis. J. Dairy Sci. [Internet]. 1988; 71(3):848–853. doi: <https://doi.org/bbs33r>
- [28] Pavel ER, Gavan C. Seasonal and milking-to-milking variations in cow milk fat, protein and somatic cell counts. Not. Sci. Biol. [Internet]. 2011; 3(2):20–23. doi: <https://doi.org/g8v9zb>
- [29] Maciuc V, Radu-Rusu CG, Popescu EC, Radu-Rusu RM, Jurco EC. Influence of season and cows farming system on milk physical, chemical and hygienic traits. Rom. Biotechnol. Lett. [Internet]. 2017 [cited 1 Dec. 2023]; 22(6):13108–13119. Available in: <https://goo.su/oDFOLUW>
- [30] Simioni FJ, Lopes LS, Nespolo CR, Stefani LM, Bordignon R, Bittelbrun MS. Season influence on milk physico-chemical and microbiological aspects in Western Santa Catarina. Semin. Ciênc. Agrar. [Internet]. 2014; 35(4):2033–2046. doi: <https://doi.org/g7x8sw>
- [31] Cassoli LD, Lima WJF, Esguerra JC, Da Silva J, Machado PF, Mourão GB. Do different standard plate counting (IDF/ISSO or AOAC) methods interfere in the conversion of individual bacteria counts to colony forming units in raw milk? J. Appl. Microbiol. [Internet]. 2016; 121(4):1052–1058. doi: <https://doi.org/g8v9zc>
- [32] Berry DP, O'Brien B, O'Callaghan EJ, Sullivan KO, Meaney WJ. Temporal trends in bulk tank somatic cell count and total bacterial count in Irish dairy herds during the past decade. J. Dairy Sci. [Internet]. 2006; 89(10):4083–4093. doi: <https://doi.org/bf8fhx>