

# Factors affecting global quality of milk produced in a semi-arid Algerian steppe zone

Factores que afectan la calidad global de la leche producida en una zona esteparia semiárida de Argelia

Fatores que afetam a qualidade global do leite produzido numa zona semiárida de estepe argelina

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# **Animal production**

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# Abstract

The purpose of this research was to investigate the effect of the zone, season and collecting point on the quality of milk produced in a semiarid steppe zone of Algeria. Throughout the four seasons of the year and separated into five zones, this study was done on 334 farms and 25 collectors. It involved 1336 milk samples. The obtained results showed that the physico-chemical and microbiological quality of milk produced in semi-arid zones is influenced by the zone, season, and collecting point. The collecting point behaves similarly to the season, except for pH. They showed a highly significant effect ( $p \le 0.01$ ) for Staphylococcus aureus to a very highly significant one ( $p \le 0.001$ ) for all other physico-chemical characteristics (acidity, density, freezing point, wetting, fat, protein and total solids) and microbiological parameters (thermo-tolerant coliforms, aerobic germs at 30°C, Listeria monocytogenes and Salmonella). On the other hand, the effect of the zone was variable. It is significant ( $p \le 0.05$ ) for *Listeria* monocytogenes, highly significant for freezing point and wetting, and very highly significant for the other parameters, except for fat content and Salmonella which were not influenced by the zone. Among other things, pH was not affected by the collection point. This variability in milk's quality is the result of above mentioned factors, either considered independently or in combination. The collection point highlights the mixing effect. The season acts directly through its temperature (condition of transport and storage of milk) or indirectly on the feeding of the animals and the area directly by its climate or indirectly through its plant cover.

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# Resumen

El objetivo de esta investigación es investigar el efecto de la zona, la estación y el punto de recolección en la calidad de la leche producida en una zona de estepa semiárida de Argelia. Durante las cuatro estaciones del año, separadas en cinco zonas, este estudio se realizó en 334 fincas y 25 colectores. Involucró 1336 muestras de leche. Nuestros resultados muestran que la calidad de la leche está influenciada por estos tres factores. El punto de recogida se comporta de forma similar a la estación, excepto pH. Ellos muestran un efecto altamente significativo (p  $\leq$  0,01) para Staphylococcus aureus, muy altamente significativo (p ≤ 0,001) para las características fisicoquímicas (acidez, densidad, punto de congelación, humedad, grasas, proteínas y sólidos totales) y microbiológicos (coliformes termotolerantes, gérmenes aerobios a 30 °C, Listeria monocytogenes y Salmonella). El efecto de la zona es significativo ( $p \le 0.05$ ) para Listeria monocytogenes, altamente significativo para punto de congelación y humedad, y muy significativo para los demás parámetros, excepto el contenido de grasa y Salmonella que no fueron influenciados por la zona. Entre otras, el pH no se vio afectado por el punto de recolección. Esta variabilidad en la calidad de la leche es el resultado de los factores antes mencionados, ya sea considerado de forma independiente o en combinación. El punto de recogida resalta el efecto de mezcla. La estación actúa directamente a través de su temperatura (condición de transporte y almacenamiento de la leche) o indirectamente sobre la alimentación de los animales y la zona directamente por su clima o indirectamente a través de su cobertura vegetal.

Palabras clave: ordeño, lácteos, microbiología, fisicoquímica.

### Resumo

O objetivo desta pesquisa é investigar o efeito da zona, estação do ano e ponto de coleta na qualidade do leite produzido em uma zona de estepe semiárida da Argélia. Durante as quatro estações do ano, separadas em cinco zonas, este estudo foi feito em 334 fazendas e 25 coletores. Envolveu 1336 amostras de leite. Os resultados obtidos mostraram que a qualidade global do leite é influenciada pela zona, estação do ano e ponto de coleta. O ponto de coleta se comporta de maneira semelhante à estação, exceto pH. Eles mostram um efeito altamente significativo ( $p \le 0.01$ ) para *Staphylococcus aureus* a um efeito muito altamente significativo ( $p \le 0.001$ ) para todas as outras características físico-químicas (acidez, densidade, ponto de congelamento, umedecimento, gordura, proteína e sólidos totais) e parâmetros microbiológicos (coliformes termotolerantes, germes aeróbicos a 30 °C, Listeria monocytogenes e Salmonella). Por outro lado, o efeito da zona é variável. É significativo ( $p \le 0.05$ ) para Listeria monocytogenes, altamente significativo para ponto de congelamento e umedecimento e muito significativo para os demais parámetros, exceto para o teor de gordura e Salmonella que não foram influenciados pela zona. Entre outras coisas, o pH não foi afetado pelo ponto de coleta. Esta variabilidade na qualidade do leite é o resultado dos fatores acima mencionados, considerados de forma independente ou em combinação. O ponto de coleta destaca o efeito de mistura. A estação atua diretamente através da sua temperatura (condição de transporte e armazenamento do leite) ou indiretamente na alimentação dos animais e da área diretamente pelo seu clima ou indiretamente através da sua cobertura vegetal.

Palavras-chave: ordenha, laticínio, microbiologia, fisico química.

# Introduction

Milk and milk products play an important role in human nutrition throughout the world. In recent years, the major dairy industry players include dairy farmers declared that milk quality depend largely its safety, hygienic standers intended for consumption and acquisition of technology skills requires dynamic of transformation from farm procedures that distributes milk to retail stores.

Several authors agree that assessment of bacterial levels is a frequently used procedure to measure the microbial quality of milk. Milk can be contaminated at several levels, at milking, on farm storage, during transport and at delivery (Millogo et al., 2010). Furthermore, microbial contamination of milk occurs when bacteria found in the cow's udder mammary, mastitis or from the cow and its environment, milking techniques, methods of disinfection of milking machines, storage equipment, milking utensils and storage conditions (Gebeyehu et al., 2022), non-hygienic handling and handling practices (Islam et al., 2018; Nyokabi et al., 2021). In addition, physicochemical parameters influence the quality of milk in one way or another. Each measurement has its own specificity. For example the pH and acidity of milk are linked to freshness, the density when skimming, the freezing point when wetting, and the dry extract to the richness of the milk. Fat and proteins are involved both in the milk payment system for the quality and the cheese-ability of the latter. Some of these factors are related to the geographical origin (sampling area) (Lingathurai et al., 2009; Mhone et al., 2011; Gemechu and Amene, 2016; Skeie et al., 2019), season of collection (Celano et al., 2022; Dolango et al., 2021) and at different collection points from the farm to the processing unit (Millogo et al., 2010; Islam et al., 2018; Tobar-Delgado et al., 2020; Dolango et al., 2021; Nyokabi et al., 2021).

In Algeria, milk production is no exception to this rule, in most subjects, variations across the area and sampling point was documented (Kaouche, 2018; Meklati *et al.*, 2023). In addition, avoiding or limiting the presence and subsequent growth of microorganisms in milk is an ongoing obstacle for those involved in milk production (Tobar-Delgado *et al.*, 2020). In this order of idea and, to verify or disprove this hypothesis, our study fits. Therefore, physicochemical properties (pH, density, freezing point, added water, fat, protein, total dry solids, and titratable acidity) and microbiological quality (aerobic germs, thermotolerant coliform, *Staphylococcus aureus, Salmonella*, and *Listeria monocytogenes*) of milk produced in a semi-arid Algerian steppe zone were described with emphasis on some variation's factors (zone, season and collecting point).

# Materials and methods

#### Study area and feeding system

This study was conducted in Sétif area located in high plains of eastern Algeria (latitude: 35.0 - 36.5 °N and longitude: 5 - 6 °E) at 1300 m above sea level. This region is characterized by a semiarid continental climate, with dry and hot summer and cold and wet winter. Three agro-ecological zones characterize the region: North with black and deep vertic soils and an annual rainfall of 600 mm, Center with brown calcareous soils and annual rainfall which does not exceed 300 mm, and South with common stony soils and some saline soils in depression and annual rainfall less than 200 mm. The main annual temperature is 14.3 °C with significant variation's seasonal (with a maximum in July 25.5 °C, a minimum in January of 5 °C) (Rouabhi *et al.*, 2019). Cows fed benefiting mixed ration ad libitum based on hay, straw during all the year plus supplementation with concentrate and for all animals, traditional management in the area consists of a winter housing period and access to pastures during spring and summer seasons.

#### Sampling proceedings

Table 1 summarizes the distribution of sampling protocol. A total of 1336 raw milk were collected from dairy producers from different collectors and geographical area. Each farm was visited once time in different seasons (autumn, winter, spring and summer). Four levels of sampling were carried out: at the farm, at the collector tank (before and after refrigeration), and at the dairy. All farms have mechanical milking equipment (milking trolley).

Samples were collected in sterilized bottles and immediately refrigerated in a portable isothermal glacier and transferred to the laboratory of the Dairy unit of Sétif for analysis. The analysis was carried out in less than 3h.

## Physicochemical and microbiological analyses

Analyses were carried out according Beerns and Luquet (1987) protocols. Physicochemical analysis of milk samples was carried using the Lactoscan Milk Analyzer of the Alpes Industries Services brand (Serial 24936; Supply 12-14V DC 50W). It included pH, density (DEN) (kg.cm<sup>-3</sup>), freezing point (FP) (°C), added water rate (AW) (%), fat content (FC) (g.L<sup>-1</sup>), protein content (PC) (g.L<sup>-1</sup>) and total dry solids (TDS) (g.L<sup>-1</sup>). Titratable acidity (TA) (°D) was evaluated in presence of 0.5 % phenolphthalein indicator and sample was titrated with Dornic sodium (N/9).

Microbiological determination included aerobic germs count (FAMT), Count of Thermotolerant Coliform (CTT), *Staphylococcus aureus* (SA), *Salmonella*, and *Listeria monocytogenes* (LM). A serially diluted from 10<sup>-1</sup> to 10<sup>-6</sup> from the mother solution, were prepared with a tryptone salt solution (TSE) and employed to determine the quantity of microbiota in milk. Colony forming units per mL (UFC.mL<sup>-1</sup>) were used to express count for each germ.

#### Statistical analysis

The data collected was entered in Excel version 2010 and were analyzed using IBM SPSS (Statistical software Package for Analysis for window 7, Chicago, version 21). Descriptive statistics were established to report the variability of the different parameters involved in the evaluation of the milk quality. Results of the analysis were expressed as means  $\pm$  SD (standard deviation). The significant differences between means were evaluated by one-way ANOVA using Turkey range test, where sampling point, zone and season are the factors of variation. Statistical significance was determined at the 95 % confidence interval and p<0.05.

## **Result and discussion**

#### General quality

The present work revealed milk fat content of approximately  $32.72 \pm 3.38$  g.L<sup>-1</sup>; protein level around  $30.90 \pm 1.91$  g.L<sup>-1</sup>; and total dray solid  $104.67 \pm 34.52$  g.L<sup>-1</sup>.

These values were slightly lower than those reported for common milk. It is well recognized that that fat level of milk is unquestionably the most valuable constituent of milk; although current consumers prefer skimmed milk. In Algeria, fat content is an incentive criterion for quality payment of the milk.

As regards the physical characteristics of milk, expressed by pH, acidity, density, freezing point and added water, these have the following respective mean values  $6.67 \pm 0.09$ ;  $16.67 \pm 0.34$  °D;  $1.0309 \pm 0.003$  at 20 °C;  $-0.51 \pm 0.16$  °C and  $3.13 \pm 1.81$  %. These values generally agree with Algerian and European standards. The values observed for the freezing point may suggest an addition of water, which could be due to residual water after washing milk recipients (Nyokabi *et al.*, 2021).

It is well admitted that the count total mesophilic aerobic flora is carried out for the payment of the milk to the quality, among other things, this flora is seen as a general indicator of overall quality related to the conditions of hygiene, collection and keeping of the product (Dolango et al., 2021). The data showed in table 2 indicate an average microbial load of  $6.25 \pm 3.04.10^6$  UFC.mL<sup>-1</sup> which was higher than standard permissible limits of Algeria fixed at 3.105 (JORADP N°39, 2017) and the European standards established at 10<sup>5</sup> UFC.mL<sup>-1</sup> for raw cow's milk (Anonyme, 1992). The presence of coliforms in milk indicates a recent fecal contamination, as these bacteria cannot survive outside the gut for a long time (Beerns and Luquet, 1987). In current study, the average contamination of milk samples by coliforms was  $5.48 \pm 0.37.10^3$  UFC.mL<sup>-1</sup> which exceeding the Algerian standards (JORADP N°39, 2017). These results may reflect a poor state of freshness of raw milk, and an indicator of poor hygienic and sanitary practices during milking and further handling, as transport and storage conditions (Islam et al., 2018). S. aureus was detected with an average rate of  $0.83 \pm 1.38.10^2$  UFC.mL<sup>-1</sup> witch is not conform to the national standard (JORADP N°39, 2017) fixed at 10<sup>2</sup> of the minimum threshold value, is commonly associated with intoxications of food through its capacity to produce different kinds of potent enterotoxins (Islam et al., 2018). Contamination of milk by Salmonella at the average rate equal to  $0.21 \pm 0.69$  UFC. mL<sup>-1</sup> not meets the Algerian standards which require total absence of Salmonella in milk (JORADP N°39, 2017). This contamination may be the origin of infected cattle feces, infected udders, contaminated milking equipment, air, feed and water, and milkers (Gebeyehu et al., 2022). The presence of L. monocytogenes at the level of 0.35  $\pm$ 0.98.10<sup>1</sup> UFC.mL<sup>-1</sup> is lower than the Algerian standard. This species excreted by livestock can contaminate milk and food production chain and among pathogen in human health (Steingolde et al., 2021).

# Factors affecting milk quality

Several factors have been described to assess variation in milk composition. Some are linked to the surrounding environment, others linked to the animal (Nyokabi *et al.*, 2021). Among these factors, area, season and collection points are reviewed in this study.

#### Table 1. Distribution of number cow's milk samples per farms, area and collector.

| Zone                  | Northen            | Central            | Estern             | Western             | Southern            | Total |
|-----------------------|--------------------|--------------------|--------------------|---------------------|---------------------|-------|
| Number of Collectors  | 5                  | 5                  | 5                  | 5                   | 5                   | 25    |
| Number of farms*      | 67(15-10-10-16-16) | 52 (12-10-9-11-10) | 62 (14-8-13-14-13) | 73 (13-18-12-14-16) | 80 (15-13-11-18-23) | 334   |
| Number of sample milk | 268                | 208                | 248                | 292                 | 320                 | 1336  |

\*In parentheses: number of farms per collector

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#### Effect of zone on milk quality

The physicochemical and microbiological variables were affected by sampling area, except for the fat level and Salmonella count (table 2). Milk obtained from dairy farms in northern was more acidic, with greater water added and a higher protein concentration compared with all other regions. In fact, the milk samples collected from eastern province was lower added water. Samples taken from central zone was more dense and richer in TDS content. Similar level in density, Dornic acidity and added water was recorded in Southern and Western zone. The effect of zone (region) on milk quality varies according literature. Lingathurai et al. (2009) states that total solids, fat, and crude protein of southern cow milk sample were significantly influenced by regions; pH was slightly influenced. For Gemechu and Amene (2016), there was no significant difference in fat and protein content observed among the study areas; however titratable acidity, specific gravity and pH value were significantly influenced by collection area. According Meklati et al. (2022), titratable acidity, density, fat level, total dry extract were significantly affected by region but not pH.

Table 2. Effect of zone on milk quality.

in milk fat acid composition according to different geographical region. Many authors use milk fatty acids composition to discriminate and authenticate the area origin of bulk milk. The results of these predicting models remains uncertain whether the results were due to the direct effect of altitude or more likely to the cow-feeding system (Coppa *et al.*, 2014).

The effects of photoperiod on milk composition were highlighted but studies about it are fuzzy. Dahl *et al.* (2000) consider that milk composition is generally unaffected by photoperiod while other authors plead in favor of its act, positively by increasing milk fat content as well as milk yield (Espinoza and Oba, 2017) or negatively by reducing milk fat percentage for animals exposed to long period compared to short one (Phillips and Schofield, 1989). But the act of photoperiod can be masked when changing management practices (Espinoza and Oba, 2017).

Likewise, several studies discussed the effect of altitude on milk quality, and results are disparate and sometimes contradictory. Leiber *et al.* (2006) found a clear influence of high altitude grazing on major milk constituents and on SCC; but according to Correddu *et al.* 

|                             | Northern                 | Central                  | Eastern                   | Western                  | Southern                  | Mean±SD            | р   |
|-----------------------------|--------------------------|--------------------------|---------------------------|--------------------------|---------------------------|--------------------|-----|
| pН                          | 6.65±0.09 <sup>b</sup>   | 6.66±0.13 <sup>b</sup>   | 6.68±0.10ª                | 6.68±0.06ª               | 6.68±0.06ª                | 6.67±0.09          | *** |
| Acidity                     | 16.86±0.49 <sup>b</sup>  | 16.96±0.41 ª             | 16.99±0.38ª               | $17.00{\pm}0.38^{a}$     | 17.00±0.38ª               | 16.67±0.34         | *** |
| Density                     | $1.0312{\pm}0.002^{ab}$  | $1.0314{\pm}0.003^{a}$   | $1.0312{\pm}0.003^{ab}$   | $1.0308 \pm 0,003^{bc}$  | $1.0308 \pm 0,003^{bc}$   | $1.0309 \pm 0.003$ | *** |
| FP                          | $0.51{\pm}0.01^{ab}$     | $0.51 \pm 0.02^{\circ}$  | $0.51{\pm}0.01^{ab}$      | $0.51{\pm}0.14^{\rm bc}$ | $0.51{\pm}0.14^{\rm bc}$  | 0.51±0.16          | **  |
| % AW                        | $3.41{\pm}1.68^{a}$      | $3.20{\pm}1.87^{ab}$     | 2.96±1.64 <sup>b</sup>    | 3.36±1.99ª               | 3.36±1.99ª                | 3.13±1.81          | **  |
| FC                          | 32.72±3.10 <sup>a</sup>  | $32.99{\pm}3.55^{a}$     | $32.68{\pm}3.56^{a}$      | 32.69±3.30ª              | 32.62±3.45ª               | 32.72±3.38         | NS  |
| PC                          | 31.59±2.50ª              | $30.61 \pm 1.52^{bC}$    | 30,90±1.92 <sup>b</sup>   | $30.56 \pm 1.60^{\circ}$ | $30,81{\pm}1.68^{bc}$     | 30,90±1.91         | *** |
| TDS                         | $107.23{\pm}30.76^{ab}$  | $110.87{\pm}26.20^{a}$   | $106.72{\pm}32.82^{ab}$   | $104.17 \pm 34.45^{b}$   | 96.77±41.56°              | $104.67 \pm 34.52$ | *** |
| CTT<br>(x10 <sup>2</sup> )  | 37.72±26.29 <sup>c</sup> | 53.94±31.46 <sup>b</sup> | 54.49±36.38 <sup>ab</sup> | 62.11±33.15ª             | 59.56±34.12 <sup>ab</sup> | 54.67±3.65         | *** |
| FAMT<br>(x10 <sup>5</sup> ) | 52.85±30.55°             | 59.89±28.63 <sup>b</sup> | 72.09±28.01ª              | 64.06±28.68 <sup>b</sup> | 63.25±32.59 <sup>b</sup>  | 62.46±30.49        | *** |
| SA<br>(x10 <sup>2</sup> )   | 0.51±0.92°               | $0.75 \pm 0.93^{bc}$     | 1.05±1.61ª                | $0.91{\pm}1.68^{ab}$     | $0.89{\pm}1.40^{ab}$      | 0.83±1.38          | *** |
| LM<br>(x10 <sup>1</sup> )   | 0.22±0.60 <sup>b</sup>   | $0.37{\pm}0.67^{ab}$     | $0.35{\pm}0.95^{ab}$      | 0.49±1.29ª               | 0.29±1.09 <sup>b</sup>    | 0.35±0.98          | *   |
| Salmonella                  | $0.17{\pm}0.45^{a}$      | $0.24{\pm}0.67^{a}$      | $0.25{\pm}0.90^{a}$       | $0.20{\pm}0.70^{a}$      | $0.21{\pm}0.67^{a}$       | 0.21±0.69          | NS  |

 $^{abc}$  : from same line, the Value from different letters are significantly different at p<0.05

Concerning microbiological quality, a higher count of CTT was observed in Western zone, FAMT in Estern region. *S. aureus* was most present in Estern zone and *L. monocytogene* in Western area. However, area sampling did not affect *Salmonella* count. The effect of area in TBC, CTT was reported (Mhone *et al.*, 2011). A significant effect of area in total count of *S. aureus* is similar to the finding reported by Lingathurai *et al.* (2009), Mhone *et al.* (2011) and Skeie *et al.* (2019). No signification variation in *Salmonella* charge between the farms was obtained by Gebeyehu *et al.* (2022).

It is necessary to remember that weather and climate are directly related to geographic zone. According to Leiber *et al.* (2006), variations in milk quality are the result of complex interactions of many environmental factors, as well as geographic origin, photoperiod, and altitude which comprises hypoxia, climatic conditions including ambient temperature and humidity, solar radiation and topographic challenges along with grazing. Coppa *et al.* (2014) showed differences (2021), the contents of protein and lactose and the pH value of the milk were not affected by altitude, except fat concentration but not FA profile. Alrhmoun *et al.* (2023) found two type of relation between altitude and milk quality; positive relationship for milk fat, protein, free fatty acid, and somatic cell count and negative one for lactose content, milk urea nitrogen, and pH-value, and attribute the impact of altitude on milk composition to its effect on dairy cow physiology by direct parameters (atmospheric pressure, temperature, and turbidity) or indirect factors (solar radiation, moisture, wind, season length, feed quantity and quality, and geology). For example, at high altitudes, a higher body fat mobilization was induced when feed intake is limited, then somatic cell count tends to increase due to management and housing factors, also it cause pulmonary hypertension, which results in an increased susceptibility against some pathogen related diseases like mammary infections (Alrhmoun *et al.*, 2023).

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## Effect of season on milk quality

The season has a significant effect on all physico-chemical and microbiological parameters. This variation differs in each parameter by season sampling (table 3).

Milk from cold periods (autumn and winter) was lower acid and had higher pH values than milk collected in hot period (spring and summer). Only, the milk collected in winter was showed a higher milk density. The highest fat level were recorded in winter, spring and summer, the lowest fat means was obtained in autumn; and the higher in protein content was noted in winter/spring while the poorest were found in summer and autumn. Milk collected in winter was richer in dry matter compared to other season sampling. Autumn milk was higher added water followed by spring and summer. Winter milk was less wet. animals are less frequently transferred to outside because of feeding on dry forage so contamination is developed in closed farms affecting milk microbial load, in winter season. Gebeyehu *et al.* (2022) in southern of Ethiopia were demonstrated that, though the overall isolation rate of *Salmonella* was higher during the wet season than the dry season; the difference was not statistically significant.

# Effect of sampling point on milk quality

Milk samples were collected from farm, collector container before cooling (tank end collection), after cooling (cold tank) and upon arrival at the processing dairy unit (delivery tank). Results of variance analysis indicated a significant difference on physicochemical and microbiological parameters between sampling point, except for pH (table 4).

|            | Autumn                   | Winter                    | Spring                   | Summer                  | р   |
|------------|--------------------------|---------------------------|--------------------------|-------------------------|-----|
| pН         | 6.72±0.09ª               | 6.71±0.05ª                | 6.63±0.11 <sup>b</sup>   | 6.63±0.05 <sup>b</sup>  | *** |
| Acidity    | 16.96±0.39ª              | 16.97±0.29 ª              | 16.89±0.39 <sup>b</sup>  | $17.01{\pm}0.28^{a}$    | *** |
| Density    | 1.0305±0.003ª            | $1.0319{\pm}0.003^{b}$    | 1.0308±0.002ª            | 1.0306±0.002ª           | *** |
| FP         | 0.50±0.12°               | $0.51{\pm}0.02^{a}$       | $0.51{\pm}0.01^{ab}$     | 0.51±0,14 <sup>b</sup>  | *** |
| % AW       | 3.41±1.76ª               | 2.75±1.86°                | 3.25±1.94 <sup>ab</sup>  | 3.04±1.63 <sup>bc</sup> | *** |
| FC         | 31.33±2.93ª              | 33.94±3.57 <sup>b</sup>   | 32.86±3.36°              | 32.72±3.14°             | *** |
| PC         | 30.12±1.16 <sup>a</sup>  | 32.37±2.25 <sup>b</sup>   | 31.16±1.67 <sup>b</sup>  | 29.89±1.24ª             | *** |
| TDS        | 96.50±40.10 <sup>a</sup> | 112.58±27.33 <sup>b</sup> | 104.82±34.73°            | 104.43±32.99°           | *** |
| CTT        | 51.60±43,55 <sup>b</sup> | 40.85±30.11°              | 54.54±27.25 <sup>b</sup> | 71.66±22.46ª            | *** |
| FAMT       | 51.42±43.80°             | 61.08±21.53 <sup>b</sup>  | 61.07±24.32 <sup>b</sup> | 72.26±20.86ª            | *** |
| SA         | $0.95{\pm}2.06^{a}$      | $0.63{\pm}1.00^{\rm b}$   | 0.99±1.21ª               | 0.73±0.91 <sup>b</sup>  | **  |
| LM         | $0.58{\pm}1.64^{a}$      | $0.25 \pm 0.69^{bc}$      | $0.36{\pm}0.64^{b}$      | 0.19±0.45°              | *** |
| Salmonella | 0.33±1.10ª               | $0.14{\pm}0.42^{b}$       | 0.26±0.59ª               | 0.11±0.36 <sup>b</sup>  | *** |

#### Table 3. Effect of season on milk quality.

<sup>abc</sup>: from same line, the Value from different letters are significantly different at p<0.05

Millogo *et al.* (2010) did not show any difference between pH, FC and PC taken during the rainy and dry seasons. Seasonality did not affect protein and total solids at the two sampling periods (winter vs summer). In contrast, fat and pH were higher in summer than in winter (Celano *et al.*, 2022). Differences in the mean milk composition were observed seasonally in total solids levels but not in density, protein and fat content (Nateghi *et al.*, 2014). These variations can be attributed mainly to the feeding (Larsen *et al.*, 2014). In summer, animals are grazing on natural pastures, and in winter, animals feed on dry forage; so summer milk is higher quality when compared to winter milk (Nateghi *et al.*, 2014).

Concerning microbiological quality, milk collected in summer reported a highest count for FAMT and CTT, the lowest one were recorded in cold period: autumn and winter respectively. A contamination by *S. aureus* and *Salmonella* was a similar profile in autumn and spring with lowest level recorded in winter and summer. Total number of colonies from LM is the most variable among seasons, and decreases from autumn to summer. It was found to be higher in autumn and lower in summer. These trends were observed by Petróczki *et al.* (2020) who assigned the highest values to heat stress of cows during the summer because of higher temperatures and humidity and lowest values to the inhibition of growth of mesophilic microorganisms at low temperature. On contrary, Celano *et al.* (2022) consider that water and medium-low temperatures could favor the growth of these microorganisms. Also, Nateghi *et al.* (2014) state that There is no effect of sampling point of milk in pH value, which agrees with Millogo *et al.* (2010). Findings of the present study for density agree with Nyokabi *et al.* (2021), and disagree with previously research when concerning FP. The present work revealed that fat, protein and total dray sec were affected by collection point; while Millogo *et al.* (2010), Tobar-Delgado *et al.* (2020) and Nyokabi *et al.* (2021) didn't find any. PC varied between farm level/delivery unit, while the fat level and TDS revealed significant variation between farm, tank before and after cooling and tank dairy unit. It seems that milk fat level highly susceptible to time of transport and storage conditions. According to Nyokabi *et al.* (2021), the absence of significant differences in milk composition (protein and TDS levels) may be due to similarities in agricultural practices, the use of a similar breed of cattle, and similar food management strategies.

In this research, the type and bacterial load has been used as a determinant of the acceptance of the microbiological quality of milk provides information on the sanitary and hygienic designed for milk handling technique associated with storage condition and transport. Bacterial enumeration at the final stage of the chain can reveal the hygienic conditions of previous milk handling (cleanliness of equipment and storage and transport conditions "refrigeration". Bacteria numbers will decrease with cleaner equipment and faster milk cooling. In this regard, our findings on the microbiological raw milk quality should be interpreted from the levels of primary producers (farms) and development in later stage of point dairy value chain.

Table 4. Effect of collection point on milk quality.

|            | Farms                    | Tank collection           | Cold Tank                | Delivery tank                | р   |
|------------|--------------------------|---------------------------|--------------------------|------------------------------|-----|
| pН         | $6.67{\pm}0.09^{a}$      | 6.67±0,05 <sup>a b</sup>  | 6.66±0,11 <sup>b</sup>   | 6.67±0.05ª                   | NS  |
| Acidity    | 16.67±0.34°              | 16.99±0,09 <sup>b</sup>   | 16.99±0,09 <sup>b</sup>  | $17.02{\pm}0.18^{a}$         | *** |
| Density    | $1.0317{\pm}0.003^{a}$   | $1.0313{\pm}0.009^{a}$    | $1.0307{\pm}0.001^{ab}$  | $1.0298{\pm}0.001^{b}$       | *** |
| FP         | 0.51±0.01°               | $0.51{\pm}0.009^{b}$      | 0.52±0.01ª               | 0.52±0.01ª                   | *** |
| % AW       | 3.16±1.84 <sup>a</sup>   | 2.02±1.37 <sup>b</sup>    | 2,02±1.37 <sup>b</sup>   | 2.02±1.37 <sup>b</sup>       | *** |
| FC         | 32.70±3.84ª              | 32.79±1.53ª               | 31.49±1.45 <sup>b</sup>  | 30.37±1.10°                  | *** |
| PC         | 30.85±2.19ª              | 30,87±1,26ª               | 30.78±1,23ª              | 30.65±1.22 <sup>b</sup>      | *** |
| TDS        | $116.60{\pm}40.10^{a}$   | 109.56±22.24 <sup>b</sup> | 107.90±21.61°            | 107.89±21.61°                | *** |
| CTT        | 54.67±33.65 <sup>d</sup> | 57.11±21.65°              | 60.42±21,09 <sup>b</sup> | 65.35±19.99ª                 | *** |
| FAMT       | $62.46{\pm}30.49^{d}$    | 66,99±23,56°              | 68.63±21,33 <sup>b</sup> | $74.34\pm21.21^{\mathtt{a}}$ | *** |
| SA         | $0.83 \pm 1.38^{b}$      | 1.06±0,97ª                | $1.10{\pm}0.94^{a}$      | $1.10\pm0.95^{\rm a}$        | **  |
| LM         | $0.35{\pm}0.98^{b}$      | $0.75{\pm}0,88^{a}$       | $0.73{\pm}0.85^{a}$      | $0.74{\pm}0.89^{a}$          | *** |
| Salmonella | 0.21±0.69 <sup>b</sup>   | 0.48±0,63ª                | 0.47±0.63ª               | 0.46±0.62ª                   | *** |

<sup>abc</sup> : from same line, the Value from different letters are significantly different at p<0.05

Our data showed that the profile level of CTT and FAMT increase by sampling levels. Similarly data was found by Islam *et al.* (2018) and Tobar-Delgado *et al.* (2020). Likewise, a significant interaction for this germ was observed between the area and sample collection chain in raw and pasteurized milk samples (Mengstu *et al.*, 2023). Previous studies revealed that the microbial contamination of raw milk increase between farms and sampling times (Skeie *et al.*, 2019). Moreover, Mengstu *et al.* (2023) showed that differences in estimated levels of thermotolerant bacteria at different points in the milk chain were specific to each region and differed significantly across the daily value.

*S. aureus* is commonly associated with intoxications of food through its capacity to produce different kinds of potent enterotoxins (Islam *et al.*, 2018). The evolution of this germ between the collection points follows the same pace as that found by another authors (Kaouche, 2018; Skeie *et al.*, 2019; Nyokabi *et al.*, 2021). Data profile variation of *L. monocytogenes* showed highly significant difference among farm and other collect points. This result was consistent with finding report by Kaouche (2018) and Šteingolde *et al.* (2021). These latter authors reported that highlighted feeding of silage and indoor keeping as the main factors which could promote the overall mean count of Listeriosis onset during winter and spring.

# Conclusion

The findings of this paper state that all parameters (both physicochemical and microbiological) were affected by the zone, season, and collecting point; except that fat content and *Salmonella* were not influenced by the zone and pH by the collection point. These variations could be the direct or indirect effect of the above factors. Season act directly on climate (winter milk is more dense than summer milk/ condition of transport and storage of milk) or indirectly through the vegetation (pastures during spring and summer seasons/ winter housing); collection point by mixing effect and more milk is handled, more it is contaminated; and area by its relief and climate (north rainier than the south). Also, hygienic practices during milk collection (from farm to dairy) and animal health must be taken in consideration.

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