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Compositional Variation between Cow and Buffalo Milk in the Central and South Region of Punjab with Special Reference to Feed

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Abstract

The increase in demand for nutritionally balanced milk and milk products is due to increasing awareness and scientific evidence of the functional and nutritional properties of milk. The quality and shelf life of dairy products and their yield are directly affected by the composition and properties of raw milk. Variations occur in raw milk quality due to various factors, such as intrinsic factors, which are breed and genotype, lactation stages, age, and the size of the animal, and extrinsic factors, which include season, climatic conditions, change in the feeding system, herd management, and farm management practices. This research aimed to study the effect of different cows and buffalos with feeding patterns and variations on raw milk composition. Quality indicators of raw milk such as pH, acidity, protein, fat, SNF (Solid Not Fat), total solids, BR values, and microbial analysis such as total plate count (TPC) and total coliform count (TCC) were analyzed to explore the variation in milk quality between cows and buffalos. Milk samples were collected in sterilized glass bottles to check variations in milk composition with variations in feed and region of different breeds of cow and buffalo from dairy farms in the southern and central areas of Punjab. Data were analyzed statistically to compare the means of each parameter. The results revealed that the percentage of fat, protein, and total solids of milk varied significantly between cows and buffalos. The result indicated a significant difference in the content of fat. The fodder that was used, such as sorghum millet and maize, and generally dairy cattle were grazed in green pastures, had more fat. Animals that were fed on mustard, rapeseed, berseem, lucern, sugarcane, and oat had more LR, BR, and pH; similarly, those fed on soybean meal had high protein content. The animals that fed on fresh grass had more total solids (TS), solid not fat (SNF), and protein.

Keywords: Raw milk quality; Milk composition; Feeding patterns; Cow and buffalo milk;

Microbial analysis

1. Introduction

Milk is stated as ‘’lacteal secretion, which is obtained from complete milking of one or more healthy dairy animals that are clean and fresh. Lacteal secretion that is obtained 15 days before and 5 days after calving is not included in this category.” Milk is a lacteal secretion that does not consist of colostrum, and one or more healthy milch cows are used to obtain it, consisting of a minimum of 8.25% SNF and a fat content of not less than 3.25%. This definition is mentioned by the Pasteurized Milk Ordinance and Code suggested by the Public Health Service of the United States of America (USA) (Hussain et al., 2010). The use of milk was reported as early as 4000 BC. Milk obtained from different animals like buffalo, ewe, goat, and cattle has been utilized for various purposes due to its nutritional value; as a feed to mammals’ offspring and humans, for the production of different products like yogurt, cheese, butter, and cream. Fortified milk with enhanced nutrition and its products having increased biological value have been in great demand. In general, milk consists of 87% water, 3.5% fat, 3.3% protein, 4.9% lactose, and 0.7% minerals. Milk fat comprises four components: triacyl-glycerides, free fatty acids, diacyl-glycerides, and cholesterol, which make up 98%, 0.1%, 2%, and less than 0.5% of fat, respectively. Apart from these, fat contains vitamins like vitamins A, D, and E. Milk fat has a complex nature because of 400 kinds of different fatty acids present in it, and out of these 400, 70% are saturated, and the rest are unsaturated fatty acids. Casein and whey are two major fractions of milk protein, and 80% of milk protein comprises casein, while whey represents 20% of total milk protein (Gustavsson et al., 2014).

Cow milk is the most commercially acceptable and conventional milk. Holstein-Friesian cows are the main producers of milk. It is the most universal raw material used for the manufacturing of an extensive range of dairy products. In recent years, according to the market requirements, the consumers and dairy industry have shifted the concept of quality: for example, to improve cheese production, milk coagulation characteristics, and fatty acid structure have been studied to enrich the nutritional value of milk for human health. The recent interest in milk composition is likely because of the global demand for nutritionally balanced milk and dairy products, which is due to increasing awareness, changes in eating trends, and scientific evidence of the importance of nutritionally balanced milk in the human diet (Smith et al., 2000). Cow milk is a vital source of minerals. These are found in a very small amount of milk but contribute to various vital physiological processes (Stocco et al., 2019). In milk, minerals are present in various forms. Such as ions, salts, or linked to proteins, fats, carbohydrates, and nucleic acids. The mineral content differs between individual cows due to different feeding practices, breeds, and utilization of different breeding techniques. Lactose is a major carbohydrate in milk and contributes to the energy value of milk. The major quantity in the total solids of milk is lactose. Lactose is the most valuable by-product for the dairy industry. The lactose content differs among individual cows mainly due to feed and different locations (Cashman, 2016).

Currently, the livestock population in Pakistan comprises 38.8 million buffaloes and 46.1 million cattle. Buffalo milk constitutes about 68 percent of the total milk produced in the country, as buffaloes are major milk-producing domesticated animals. Goats and sheep are mainly raised for meat purposes, and there are about 30.5 million sheep and 74.1 million goats (Wing, 2018). According to reports of the FAO, Pakistan stands as the 4th largest country in the world. China and the USA stand second and third, respectively, while India is the largest milk-producing country in the world. The official statistics state that around 6.75 billion liters of milk are lost during the value addition chain due to unclean, contaminated, and inappropriate storage conditions and the lack of technical and educational background of farmers, traders, and milk retailers (King, 2022).

Water buffalo is capable of converting low-grade fodder into valuable products like meat and milk. Water buffalo is found worldwide, but it is widespread in Asia and the Mediterranean region. Buffalo is divided into two breeds: the River which is mainly present in the Mediterranean, and the Swamp buffalo in Asia. 97% of buffalo are scattered in Asian countries, which are fed on low-grade fodder, resulting in less production (Deb et al., 2016).

The FAO estimates that the world's total production of buffalo milk is approximately 75 million metric tons per annum, which is mostly produced in Asian and the Pacific countries (73 million metric tons). Likely, a large portion is not recorded as buffalo milk is mostly produced by small rural families for domestic purposes (Guo and Hendricks, 2010). There are 50 million animals in Pakistan producing 61 million tons of milk with nearly 40 million farming households (Pak economical survey 2019- 20). Punjab province contributes 49 percent of total production of the product (Yasmin et al., 2012). The buffalo and Cattle population of Pakistan is more than 29 and 31.8 million respectively. The buffaloes are the key milk-producing animals and pose a good percentage of 62 percent of all the milk. Buffalo milk comprises more than 12% of the world's milk production, which is second only to cow's milk. However, in tropical and moist countries of the world, the Total milk production of buffalo is more than cows because of their potential to survive under harsher conditions and being disease-resistant. In the Indian subcontinent, buffalo milk production of buffalo is more than cows. Buffalo milk contributes to more than 53% of overall milk production in India, which is growing annually at the rate of 3.6%, and more than 68% in Pakistan. This growth rate will continue to increase in the future. Pakistan and India are producing 90% of the total buffalo milk. Some countries in Asia are also producing buffalo milk. Egypt is the only African country with considerable buffalo milk production. Some countries of Europe & Egypt are producing about two million tons per annum (Medhammaret al., 2011).

The main aim of the research was to estimate the composition of cow and buffalo milk concerning feed and to evaluate milk quality parameters in the central and southern regions of Punjab.

2. Materials and Methods

2.1. Materials

Glass bottles (for milk sample collection), Icebox (for protection of samples from direct sunlight or microbes and samples transport), Gloves (used during samples collection), Volumetric pipets, beakers, flask, thermometer, cylinder, filter paper, conical glass tube with a stopper, centrifugal machine, water bath, pH meter, butyrometer, Kjeldhal's Apparatus, and lactometer. 0.1 N NaOH, phenolphthalein, sulfuric Acid, isoamyl alcohol, 40% sodium hydroxide, boric acid, methylene blue, copper sulfate, sodium tartrate salt, and 5% acetic acid**.**

2.2. Procurement of Raw Material

Sampling was done four times a month from the dairy farm of the central and southern regions of Punjab. Samples from both morning and evening milking were collected. A total of 80 samples were collected in 3 months (from August 2021 to October 2021), including 40 samples from cross crossbreed of cow and 40 samples from the Nilli Ravi buffalo. Glass bottles were sterilized before sample collection in the laboratory. Bottles were labelled with Date, region, time of sample collection, and the temperature of the sample was checked at the time of sampling. Samples were placed in the ice box for transportation to the laboratory for analysis. The analysis was performed at the Dairy Lab of the National Institute of Food science and Technology, University of Agriculture, Faisalabad.

Proximate Analysis of Milk Samples

2.2.1. Crude Protein Determination

Protein of the milk sample was determined by Kjeldahl's method by the standard described by Jeong et al. (2025). The following steps are involved in this method:

I. Digestion

Digestion of milk protein involved taking 10ml of milk into a digestion flask and 5g of digestion mixture (100g K2SO4, 10g CuSO4 and 5g FeSO4) was added into it. The digestion flask was placed in a fume hood where 30 ml sulfuric acid was added to the flask and digestion was carried out until light green color appeared at the end. A mixture of 250 mL after digestion was left to digest after some time and completes this process by bringing the mixture capacity to 250mL by adding distilled water. The material that was digested was pipetted out of the digestion flask into a rinse of distilled water 2-3 times.

II. Distillation

A 10 ml sample was taken for distillation, and 10 ml of sodium hydroxide was added. The distillation occurred in distillation chamber. Ammonia was released during distillation and was captured in separate beaker. Methyl red was used as an indicator, and Boric acid 4 percent was added to the beaker; 10 mL of the same into the beaker as well. Maximum ammonia was trapped 2-3 minutes after bubble formation was stopped, and the color of sample turned from red to yellow, which indicates the completion of the reaction.

III. Titration

After distillation, the titration of the sample was done against 0.1 N H_2SO_4. The titration was discontinued at just before the change to light pink colour. By means of the formula, the percentage of nitrogen was calculated.

$$\% \text{ Nitrogen} = \frac{\text{Volume of 0.1 N } H_2SO_4 \text{ used} \times 250 \times 0.0014}{\text{Sample used for distillation} \times \text{Vol. of milk sample}} \times 100$$

Total protein = nitrogen % x 6.38

One factor is 6.38 and applied to milk.

2.2.2. Crude Fat Determination

The Gerber method by Najafi Seraji et al. (2025) was used to determine the fat content of milk sample. Sample was mixed carefully to avoid air incorporation then milk sample was allowed to stand for some time so that any air bubble trapped gets discharged. 10ml sulfuric acid was added intro butyrometer using pipette. Then 11ml milk was poured gently into butyrometer. After that, 1ml isoamyl alcohol was added into butyrometer and butyrometer neck was cleaned with tissue. Stopper was applied on to butyrometer and it was inverted and shaken gently upside down till acid absorbed all the milk in butyrometer. After that sample was centrifuged by placing it in a centrifugation machine for 5 minutes at 1100 rpm and temperature was set at 65^{o} C. Butyrometer was taken out afterward centrifugation. The level of fat column was regulated by adjusting the stopper position and Fat percentage was read.

2.3. Physio-chemical Analysis of milk samples

2.3.1. Total Soluble Solids

The size of total soluble solids was determined with the help of the methodology outlined by Pasan et al. (2025). Empty weight of China dish was recorded then added 10g of sample then weighed again. Subsequently, the China dish was placed in hot air oven at 105o C to dry the sample. China dish was taken out of the oven after 24 hours and put in to the desiccator. Then weighed again removing water and replaced weighed amounts of Chinese dish after which total soluble solicits were calculated by putting it in the following formula:

Moisture% % = <u>Sample weight after drying </u>x 100

Weight of sample
TSS = 100 - moisture%

2.3.2. Solids not Fat

The milk sample was mixed thoroughly and added to the cylinder. Lactometer was poured into the cylinder, and a reading was taken when it was in equilibrium. The method of solid-not-fat determination was described by Kalogianni & Gelasakis (2025). Lactometer reading was corrected, and SNF was calculated using the formula:
SNF (%) = CLR + (0.22 x Fat %) + 0.72

2.3.3. pH Determination

An electronic pH meter was used for the determination of the pH of the milk sample as described by Tong & Yang (2025). A 10 ml sample was taken in a beaker. Electrodes of the pH meter were rinsed with distilled water, then it was dipped in buffer solution of pH 4. Electrodes were again rinsed with distilled water and dipped in buffer solution of pH 7. After calibration, electrodes were cleaned with distilled water, dried, and dipped in a beaker containing the sample. pH was checked on a meter.

2.3.4. Titratable Acidity

The titration method was used for the determination of titratable acidity as described by Li et al. (2016). Phenolphthalein was used as an indicator as it is colorless with acid but gives a pink color to a basic solution. A burette was filled with N/10 NAOH, and the initial volume of NAOH was noted. A 10 ml sample was added to a flask and 2-3 drops of phenolphthalein were added to the sample. Mixing of the sample was followed by titration of the sample against 0.1N NAOH. Titration was continued till the appearance of a light pink color. Titration was stopped, and the volume of NAOH used was noted. Values were put in the following formula to calculate acidity:

$$\text{Titratable Acidity} = \frac{\text{Volume of 0.1 N NaOH used x 0.009}}{\text{Weight of sample}} \times 100$$

2.3.5. Lactometer reading (LR)

Lactometer reading depends upon the nature and number of solid contents present in it, so that it varies among different animals. The mean LR of cow milk is 1.032, and buffalo milk is 1.033.
LR value of milk determined by lactometer. Every sample was mixed thoroughly and brought to 10 – 20 °C. Then poured into a glass cylinder up to its neck, and slowly inserted a lactometer until it floated. Allowed it to stand for half a minute, when it became stationary, readings were recorded from the stationary that was on level with the milk surface.
The Lactometer reading is determined by the formula as given below:

$$LR = \frac{\text{CLR (corrected lactometer readings)}}{1000} + 1$$

2.3.6. Butyro-refractometer reading

The procedure to determine the BR value of milk is as follows to Ramani et al. (2025):
Milk fat% % was determined through the Gerber centrifuge method. After this, a drop of milk fat was extracted by using a syringe from a fat column of the butyrometer. Temperature of butyro refractometer was maintained at 40ºC by connecting its pipe with water bath set at 40ºC. A drop of fat extracted from the butyrometer was placed on the prism of the BR meter. Reading was noted by looking into the fat column of the BR meter through the lens.

2.4. Microbiological evaluation

The microbiological evaluation of milk method was followed by Amin et al. (2015) with some modification:

2.4.1. Total Plate Count

Total plate count (TPC) is supposed to denote the quantity of micro-organisms in the product.

i. Making of normal saline solution

Normal saline solution was made with 8.9g / L of NaCl, dilution of biological samples and sterilization**.**

ii. Media preparation

The nutrient agar was prepared in autoclave up to 121 C in a period of 15 min.

Samples preparation

Six sterilized test tubes were added and labeled as 10-1, 10-2, 10-3,... and 10-6. 9mL of normal saline was added to each of the test tubes. One ml of the homogenized raw milk sample was transferred to the first test tube and gently shook contents of test tube making it well mixed. In the second test tube, sample was added and it was mixed at the end. Likewise, 5 mL of sample was transferred to a third test tube in the second test tube. Other serial dilutions were also prepared in the above way. The dilutions are depicted in tabe-1.

Table-1. Dilution Table

Test Tube No.	**1**	**2**	**3**	**4**	**5**	**6**
Dilutions	1:10	1:100	1:1000	1:10000	1:100000	1:1000000
Volume of samples per ml	0.1 (10^{-1})	0.01 (10^{-2})	0.001 (10^{-3})	0.0001 (10^{-4})	0.00001 (10^{-5})	0.000001 (10^{-6})

iii. Pouring the plates

The contents of each test tubes containing dilutions were dropped on the surface of Nutrient agar plates to cover the agar plates and well mixed and the agar plates incubated at 37C 24 hrs.

iv. Colony counting

Averages were calculated on the number of colonies after those dilutions that registered the colony size of 30 to 300 colonies using colony counter.

$$\text{Total Plate Count (cfu/ml)} = \frac{\text{Average number of colonies x dilution factor}}{\text{Volume factor}}$$

2.4.2. Coliform count

The coliform number is meant to be a pointer of the degree of microorganisms in the product.

i. Preparation of normal saline solution

Normal saline solution made with 8.5g / L NaCl was used in the normal dilution of samples.

Media preparation

Enumeration of Coliform substances was carried out in raw milk samples by using Eosine Methylene Blue (EMB) Agar that was sterilized at 121 C degree during 15 min.

ii. Sample preparation

Sterilized test tubes were sampled and labeled as 10-1, 10-2, 10-3 ...10^ -6 9ml of normal saline was added to test tube. Each test tube was shaken lightly and 1 ml of the homogenized sample used was transferred to the first test tube and mixing done. The sample was taken out of the first test tube to the second one and was thoroughly mixed. In the same manner I moved around 1ml of sample in the second test tube into a third test tube. Other serial dilutions were also carried out using the above procedure. The above mentioned dilutions were prepared in TPC.

a. Pouring the plates

Each of the test tube dilutions is applied to the surface of the Eosine Methylene Blue Agar (EMB) media, and a spread is made. The plate would be grown at 35-37*C on 24 hrs.

b. Colony counting

Upon incubation, the colonies that turned out dark red in color were counted. Mean number of colonies was observed in the dilutions which gave colony size of 30 to 300 using a colony counter.

$$\text{Total Plate Count (cfu/ml)} = \frac{\text{Average number of colonies x dilution factor}}{\text{Volume factor}}$$

2.5. Statistical analysis

The obtained data was statistically analyzed at a 5% significance level (Montgomery, 2017). LSD with two-way ANOVA was applied by using the software "Statistics'' version 8.1. Data obtained was divided in three groups; highly significant, significant, and non-significant.

3. RESULTS AND DISCUSSION

Milk is an important part of our diet as it is consumed all around the world. It contains macro and micro nutrients which are essential for growth and development of people of all age groups.
Samples were collected four times in a month from the Dairy farm of central and southern region of Punjab. Various physio-chemical analysis, proximate analysis, lactometer reading, butyro-refractometer reading and microbiological analysis were studied in dairy lab at National Institute of food Science and Technology, University of Agriculture, Faisalabad. The results of all the parameters are mentioned below in headings.

3.1. Proximate Analysis

The samples of milk collected from different farms were analyzed for milk major constituents like crude fat and crude protein. Results are presented in tables and discussed as following:

3.1.1. Crude Fat

The fat of any foodstuff is extremely significant with regard to nutritive value. It also composes other functional characteristics like flavor/taste, mouth feel, structure/ texture and palatability but not energy granting. Other nutrients is fatty acids that do not find synthesis in their respective body and thus found in fat food contents and this may as well be envisioned to be one of the main body food of the day.
Analysis of variance indicated that the difference among fat content of milk was highly significant according to specie and according to region it is significant. While specie*region interaction is non-significant. Table 1. showed the mean values for fat of cow and buffalo milk. The highest mean value of buffalo milk was 5.7770±0.3036 in south region while the lowest mean value for buffalo milk samples was5.7750±0.3036 in central region. The highest mean value of cow milk was 3.5330±0.3576 in south region while the lowest mean value for cow milk samples was 3.5310±0.3576 in central region. This is where a representation of how much watering / skimming of milk in various farms selling milk is being engaged in. There is nothing wrong in saying that milk sellers dilute either milk or do skimming and none of them uses a metering or a measuring device.

The current research results are according to that of McCarthy et al. (2017) who showed milk at different shops did not justify the least content of fat.

3.1.2. Crude Protein

In most cases, Milk is a significant protein product in the diet of human beings because it contains almost 32g of protein. Milk is high in protein which contains amino acids, and it is found to stimulate the synthesis of the muscles. It is also a major issue of evaluating the nutritive value of food samples. Milk is the only source that supplies dairy proteins (casein and whey) and thus their profile in milk is significant as regards to nutritive value.
Analysis of variance revealed that there was a highly significant difference of milk protein according to specie and according to region it is significant. While specie*region interaction is non-significant. The highest mean value of buffalo milk was 6.531±0.698 in south region while the lowest mean value for buffalo milk samples was 6.516±0.696 in central region as mentioned in Table 1. The highest mean value of cow milk was 4.2509±0.1640 in south region while the lowest mean value for cow milk samples was 4.2359±0.1592 in central region. It could be revealed from current study data by taking as standard value of cow and buffalo milk mostly milk samples had low protein content than normal. Such low protein milk resulted from highly water adulteration activities. Thus, the final consumer is being destitute of a valued ingredient i.e. milk proteins.
The mean protein content (3.8%) of cow and buffalo milk found high reported by when compared. The current study is in line with Kučević et al. (2016) who showed milk at different farms not justify the least content of protein.

Table 2. Means values for crude fat and crude protein of cow and buffalo milk

	Fat (%)		**Protein (%)**	
Regions	**Cow**	**Buffalo**	**Cow**	**Buffalo**
Central	3.5310±0.3576	5.7750±0.3036	4.2359±0.1592	6.516±0.696
South	3.5330±0.3576	5.7770±0.3036	4.2509±0.1640	6.531±0.698

Values are Mean ± SD for the analyzed sample.

3.2. Physico-chemical Analysis

3.2.1. pH

pH of milk can be said to be the yardstick of acidity yet can be upset by acidity. Analysis of variance revealed that there was a highly significant difference among pH of milk according to region and specie. While specie*region interaction is non-significant. The highest mean value of buffalo milk was 6.7401±0.0540 in south region while the lowest mean value for buffalo milk samples was6.6670±0.0543 in central region. Table 2. shown the highest mean value of cow milk was 6.5282±0.0738 in south region while the lowest mean value for cow milk samples was 6.4542±0.0734 in central region. This may be caused due to addition of foreign additives/ adulterants. Some of literature also tells that pH of milk is normalized by the addition of stabilizers and neutralizers to enhance the pH which wad decreased. So, the normal pH is not necessarily good for consumption but needs more explorations regarding existence of any neutralizer in it.
These values of pH were proved to be very similar to the mean values of the reported pH of Pisano et al., (2016).

3.2.2. Acidity

The reason behind the acidity in milk is the action of lactic acid bacteria which uses the lactose in to develop lactic acid. Total acidity of milk is the basis of quality attributes and subsequent processing of milk. When the acidity is more than the standard one the value of milk will be reduced and through the subsequent procedure it will contract with the heat curdling and the spoilage of milk.

Analysis of variance revealed that there was a highly significant difference among acidity of milk according to region and specie. While specie*region interaction is non-significant. The highest mean value for acidity was 0.13958±0.000873 of buffalo milk in central region are shown in Table 2. The lowest mean value for acidity was 0.13664±0.00090 of cow milk in south region. These values for acidity found to be normal as compared to mean values reported by Bahebecket al. (2024).

The present study results showed that the acidity of all the milk samples was as normal as found in fresh milk. As the acidity of milk is caused due to activity of LAB which are killed by heating. These bacteria need appropriate conditions for their proper growth and activity. While all the milk samples were continuously in contact to heat treatment and the enzymes and LAB are killed by excessive heating, therefore the acidity of milk was not caused and the most of samples had normal range of % acidity.

3.2.3. Total solids (TS)

Milk, free of water and the balance of the constituents, are referred to as total solids. The protein, the fat, the mineral, and the lactose are found in the total solid of milk. Having low amount of total solids could be signified adulteration of milk by some water and also this also skimming of milk. It is no more news that the content of total solids in milk assumes great relevance when it comes to the overall quality properties of the milk, and that its average/normal variations in milk range between 9-16 percentile which is a derivative of the quality properties of end-products as well as the structural/textural properties by times.

Analysis of variance revealed that there was a highly significant difference of milk total solids according to specie and according to region it is significant. While specie*region interaction is non-significant. Table 2. showed the mean values for total solids of cow and buffalo milk. The highest mean value was 14.797±1.173 of buffalo milk in central region. The lowest mean value was 12.550±1.521 of cow milk in south region.

The current study results are in line with Atkinsal, (2016) who disclosed milk at different shop does not justify the minimum total solids content.

3.2.4. Solid not fat (SNF)

Solids not fat contents of milk are the solid proportion without fat or moisture content and so it provides good profile of total solids.

Analysis of variance revealed that according to region there was non-significant difference among SNF of milk and there was highly significant difference among milk SNF according to specie. While the region*specie interaction was non-significant. Table 2. showed the mean values for SNF of cow and buffalo milk. The highest mean value of buffalo milk was 8.8989±0.0630 in south region while the lowest mean value for buffalo milk samples was8.8899±0.0630 in central region. The highest mean value of cow milk was 8.3269±0.0817 in south region while the lowest mean value for cow milk samples was8.3179±0.0817 in central region.

Current study is according to those of Chen et al. (2014); Awan and Naseer. (2014) who revealed, milk marketed was comprehensively prone to malpractice such as adulteration with water and skimming. In addition, it did not have desire minimum level of solid not fat content.

3.2.5. Lactometer Reading

Denatured milk is also the reading of the lactometer the density of milk that is the ratio of the weight of milk sample and the weight of an equal volume of water at a temperature of 15C. All the solids that are present in milk are exactly related to the reason it has a LR value. There is also an alteration in the LR since there are differences in milk constituents of milk. LR value which is calculated with the help of special device known as Lactometer When there is an increase or a decrease in the lactometer reading the specific gravity also increases or decreases correspondingly. Specific gravity of milk is also important since it shows nature of water adulteration.

Analysis of variance revealed that according to region and specie there was a highly significant difference among LR values of milk. While region*specie interaction was non-significant. The highest mean value of buffalo milk was 1.0329±0.0006 in south region while the lowest mean value for buffalo milk samples was1.0323±0.0006 in central region. The highest mean value of cow milk was 1.0307±0.00122 in south region while the lowest mean value for cow milk samples was 1.0301±0.00122 in central region. Collectively the present study's results showed in Table 2. that the lactose content of milks form different localities was very close to normal range and the disturbance relating carbohydrates was comparatively lower expected in the test samples. Thus, further analysis was needed for occurrence of any mismanagement.

The values of LR calculated here appeared to be pretty similar to mean value provided by Chen et al. (2014).

Butyro-Refractometer (BR) reading

Butyro-Refractometer (BR) Reading is the value of purity of foods in form of ghee, sweets and fats as well as oils and it can be read with the assistance of Butyro-Refractometer or the (BR) meter. There was a variation of 37 and 42 BR values in pure cow, buffalo and mixed milk fat. BR values of clear fat of various milk systems falling outside this range may be considered the cutoff points in determination of presence of vegetable oil(s).

Analysis of variance revealed that there was non-significant difference among BR value of milk according to region while according to specie there was a significant difference among BR values of milk. Region*specie interaction was non-significant. The highest mean value of buffalo milk was38.991±0.918 in south region while the lowest mean value for buffalo milk samples was38.952±0.918 in central region. The highest mean value of cow milk was 38.591±0.503 in south region while the lowest mean value for cow milk samples was38.552±0.504 in central region are shown in Table 2.

The present work was in line with the research of Tomar et al. (2019) who received the same values of BR when special kefir grains or starter cultures were added to the various types of milk.

Table 3. Mean values for physicochemical analysis of cow and buffalo milk

	pH	
Regions	**Cow**	**Buffalo**
Central	6.4542±0.0734	6.6670±0.0543
South	6.5282±0.0738	6.7401±0.0540
	Acidity	
Central	0.12726±0.000639	0.12958±0.000873
South	0.13664±0.00090	0.13856±0.0111

	Total solid (TS)	
Central	12.985±0.999	14.797±1.173
South	12.550±1.521	14.169±1.609
	Solid not fat (SNF)	
Central	8.3179±0.0817	8.8899±0.0630
South	8.3269±0.0817	8.8989±0.0630
	Lactometer reading (LR)	
Central	1.0301±0.00122	1.0323±0.0006
South	1.0307±0.00122	1.0329±0.0006
	Butyro-Refractometer reading (BR)	
Central	38.552±0.504	38.952±0.918
South	38.591±0.503	38.991±0.918

Values are Mean ± SD for analyzed sample

3.3. Microbial Analysis of Milk samples

3.3.1. Total plate count (TPC)

Dairy products quality and shelf life is determining through an important criterion of microbiological analysis. Dairy products quality and safety can be evaluated by these microbiological tests. In current study various samples of central and south Punjab were taken for assessment. The results are shown in Table 3. indicated that samples handled with proper hygiene, processing and storing condition have less impact on the overall quality of final product.

Analysis of variance revealed that there was a non-significant difference of milk samples according to region while according to specie it was highly significant. Region*specie interaction was also non-significant. The highest mean value of buffalo milk was 1.9988±0.0484 in south region while the lowest mean value for buffalo milk samples was1.9688±0.0484 in central region. The highest mean value of cow milk was 2.8004±0.3840 in south region while the lowest mean value for cow milk samples was 2.7708±0.3840 in central region. The recent research was by Pisano et al., (2016).

3.3.2. Total coliform count (TCC)

Dairy products quality and shelf life is determining through an important criterion of microbiological analysis. Dairy products quality and safety can be evaluated by these microbiological tests. In current study various samples of central and south Punjab were taken for assessment. Table 3. results indicated that samples handled with proper hygiene, processing and storing condition have less impact on the overall quality of final product.

Analysis of variance revealed that there was a non-significant difference of milk samples according to region while according to specie it was highly significant. Region*specie interaction was also non-significant. The highest mean value of buffalo milk was 2.409±0.458 in south region while the lowest mean value for buffalo milk samples was2.349±0.458 in central region. The highest mean value of cow milk was 3.1249±0.4404 in south region while the lowest mean value for cow milk samples was 3.0649±0.4404 in central region. The study matched with the study of Muhib et al. (2016).

Table 4.Mean values for Microbial Analysis of cow and buffalo milk samples

	Total plate count TPC		**Total coliform count TCC**	
Region	Cow	Buffalo	Cow	Buffalo
Central	2.7708±0.3840	1.9688±0.0484	3.0649±0.4404	2.349±0.458
South	2.8004±0.3840	1.9988±0.0484	3.1249±0.4404	2.409±0.458

Values are Mean ± SD for the analyzed sample

4. Conclusion

It was concluded from the results of the current study there were effects of feeding and regional variation on composition of raw milk that was represented by the total solid, fat and protein contents of milk showed significant results by feeding while pH, acidity and LR contents of milk were highly significantly affected and solid not fat, BR, TPC and TCC were non-significantly affected by feeding practices in both central and southern Punjab. Also, different kind of feeds used in different regions of Punjab shows difference in several compositional parameters of cows and buffalo milk. The findings of current study will assist the researcher and farmer to use nutritious feed for animals to improve the quality of milk.

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