



## Nutritional Study of Various Cow Breeds from Bhatta Chowk Lahore (Punjab), Pakistan

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**Abstract:** Present study was conducted to investigate the nutritional study of cow's milk of various breeds from Bhatta Chowk Lahore. Different cow's breeds were found to possess variable amounts of nutritional contents, i.e., highest moisture and ash in Cholistani cow, highest fat in Sahiwal cow, highest calcium and specific gravity in Holstein cow and highest contents of protein, solid-not-fat and total solid in Red Sindhi. Red Sindhi cow's milk was found to be more nutritious in terms of its richness in proteins, solid-not-fat and total solids whereas Holstein cow was rich in calcium. Calcium was found to be in a range of 550 to 630 ppm with the decrease of concentration in the following order: Sahiwal > Holstein Frisian = Red Sindhi > Cholistani. The Cholistani cow milk showed the presence of 3.126 % protein, 3.5 % fat, 88.2 % moisture, 8.3 % solids-not fat, 11.8 % total solids, 0.791 % ash, 30.5° lactometer reading and 1.0305 kg/m<sup>3</sup> specific gravity. Sahiwal cow milk showed 3.318 % of protein, 4.1 % fat, 87.5 % moisture, 30.1° lactometer reading, 1.03 kg/m<sup>3</sup> specific gravity, 8.4 % solids-not-fat, 12.5 % total solids and 0.79 % ash. Holstein Frisian's milk demonstrated the presence of 3.33 % protein, 3.8 % fat, 87.35 % moisture, 31° lactometer reading, 1.03 kg/m<sup>3</sup> specific gravity, 8.85 % solids-not-fat, 12.65 % total solids and 0.77 % ash. Red Sindhi's milk revealed the presence of 3.38 % protein, 3.95 % fat, 85.65 % moisture, 28° Lactometer reading, 1.028 kg/m<sup>3</sup> specific gravity, 10.4 % solids-not-fat, 14.35% total solids and 0.705 % ash.

**Keywords:** Red Sindhi, Sahiwal, Cholistani, Holstein Frisian breeds, Cow Milk, Bhatta Chowk Lahore, Nutritional analysis.

### 1. INTRODUCTION

Dairy products are an important food source throughout the world, for which the milk is mainly produced by four ruminants, i.e., cows, buffaloes, goat and sheep. Milk has special significance due to its nutritional value and its role in growth and resistance to the diseases. It is an important source of magnesium, calcium, phosphorus, potassium and vitamins [1]. Water, proteins, fats, lactose, minerals and other dissolved ingredients (vitamins

and white blood cells) are important components of cow milk. Cow milk is the major source of calcium but the amount of calcium and other constituents varies from a breed to another breed [2]. Milk is a dilute emulsion which is comprised of fat/oil dispersed in aqueous colloidal continuous phase. Physical properties of milk are similar to those of water but the difference lies in the concentration of solutes (salts, lactose and proteins) [3]. The specific gravity of cow's milk is 1.029; its viscosity is significantly lower as compared to the camel

and buffalo's milk [4]. Cow's milk contains fat globules; lipid metabolic differences produce small or large fat globules [5]. It has been reported that the amount of saturated fatty acids, trans fatty acid, linoleic acid and conjugated linolenic acid is lower in cow's milk as compared to that in the buffalo's milk [6]. Milk and dairy products are good sources of fat-soluble vitamins, i.e., A, E, D, and K [7]. Raw milk can be separated into fat-enriched and fat-depleted phases, i.e., cream and skim milk, respectively by gravitational separation. It occurs due to differences in densities of milk serum and emulsified fat globules [8]. Different types of carbohydrates such as lactose, galactose, glucose and other oligosaccharides are found in milk. Cow milk generally contains 4.8 % anhydrous lactose (on average) whereas lactose concentration depends upon the type of milk [9]. Cow's milk is three to four times richer in protein than human milk, [10]. Lingathurai et. al., [11] collected sixty samples of cow's milk in Madurai and found 6.14 % fat, 3.77 % protein, 18.10 % of total solids and 0.08 % ash. Mahdian and Tehrani [12] analyzed total solids in milk by adding bacteria and found the increasing amount of total solids from 14 % to 27 % in milk with the increasing growth of bacteria.

The composition of cow's milk varies with season. The highest level of main components (e.g., solids not fat:  $96.4 \pm 0.04$  g/L; lactose:  $53 \pm 0.02$  g/L; protein:  $35.3 \pm 0.01$  g/L, minerals:  $7.8 \pm 0.04$  g/L) of milk were observed in winter. Moreover, the nutritional components of raw milk were found in higher quantity as compared to the sterilized and pasteurized milk [13]. For macronutrient estimation, 40 milk samples of cow and buffalo were tested and there was a lower concentration of total solids, fat, protein, lactose and ash contents in cow milk than buffalo milk [14]. Dora found the highest significant correlation between total solids, fats and solid-not-fats in cow's milk [15]. Mineral fraction is about 8-9 g/L in cow's milk [16]. The mineral composition varies according the lactation phase, environmental factor, eating and nutritional status of animal and its genes [17]. With the climate changes, it was observed that the Se, Mg and Zn concentrations were fluctuating [18]. A larger fraction of milk contains calcium and phosphorus which take part in bone growth and nurturing of newborns [19]. Calcium is very important as it

involves in many metabolic processes in body, helps in bone growth and prevents from osteoporosis [20]. 0.019 g/liter of calcium, 0.029 g/liter potassium and 0.010 g/liter sodium were estimated through atomic absorption in cow's milk samples in a study [21]. Zinc, magnesium and copper can be determined directly by using atomic absorption spectroscopy according to Association of Official Agricultural Chemists (AOAC) 2000 [22]. For estimating the strength of oxidant or reductant, titration method is used by using a sensitive indicator to analyze the biomolecular interaction specifically calcium milk interaction [23].

With an expected 65.7 million tons of milk produced in 2021–2022, Pakistan is among the top 5 milk-producing nations in the world [24]. Farmers are involved in Milk production in Pakistan. In mixed farming system, farmers keep 1-2 milk animals and are responsible for the production of about 38% of total milk [25].

Sahiwal cattle are considered as one of the best cow breed across the world [26]. Sahiwal cattle are found in parts of districts Sahiwal, Okara, Pakpattan, Multan, and Faisalabad. Sahiwal cow weighs 400–500 kg and is medium-sized with a thick body [27]. The average lactation production of a Sahiwal cow is  $1475 \pm 651$  kg [28]. The Red Sindhi breed emerges from a mountainous region (called Mahal Kohistan) and is extended to Thattha and Dadu districts in Sindh. It is a medium in size and has red colour body [29]. Its milk yield is observed to be the highest (1220 liters) in 3<sup>rd</sup> lactation [30]. Cholistani cattle breed is found in Cholistan tract (a desert area), different areas of Bahawalnagar, Bahawalpur, and Rahimyar Khan districts. In males, it's body weight is 450-500 and in females it is 350-400 kg [31]. Cholistani cattle is as an excellent heat tolerant animal with high milk potential even in desert conditions and is commonly found Cholistan desert in Pakistan [32].

Current studies were performed to investigate the qualitative and quantitative determination of moisture, ash, protein, fat, solids-not-fat, total solids, specific gravity and calcium in milk samples of four cow breeds, i.e., Red Sindhi, Holstein Frisian, Cholistani and Sahiwal breeds.

## 2. MATERIALS AND METHODS

The present work was performed in Food and Biotechnology Research Center (FBRC) at Pakistan Council of Scientific and Industrial Research (PCSIR), Lahore, Punjab Pakistan. The milk samples of four cow breeds (Red Sindhi, Holstein Frisian, Cholistani and Sahiwal breeds) were collected from Bhatta Chowk Lahore which is situated beyond the Cantt area of Lahore, Pakistan. All chemicals were purchased from *BDH*. Distilled water was used for washing all glassware which (after washing) were dried in an oven at 100 °C. Polarized Zeeman Atomic Absorption Spectrophotometer, Hitachi High Technologies America, Inc with model Z-8000 was used. Electronic balance (OHAUS Pioneer Analytical Balance, with Draftshield, 210 g capacity, 0.1 mg Readability, Model Number 80251552) was used in this study.

Gerber's Centrifuge (0-4000 rpm) was used with WTW 1F10-220 Inolab Level 1 Multiparameter Meter without Probe, 110 V. Muffle furnace having temperature range of 0-1000 °C was used. Borosil Gerber milk Pipette (Pyrex) having capacity 10.75 mL and Standard Gerber milk test butyrometers were used.

### 2.1 Determination of calcium contents using permanganate titration method

Calcium was determined by permanganate titration. Solutions containing milk samples were titrated against standard potassium permanganate solution to get persistent pink color (at least 30 sec) with heating.

$$\text{Calcium, percent by weight} = \frac{1.002 \times V}{W}$$

### 2.2 Determination of calcium contents using atomic absorption spectrophotometer

Milk Ash (0.5 g) was mixed with 1 ml concentrated nitric acid and then distilled water was added to make the total volume to 100 mL. Samples were analyzed by Hitachi Polarized Zeeman Atomic Absorption Spectrophotometer, Z-8000. Three readings were taken for precise calculations.

### 2.3 Estimation of Moisture (%)

Moisture was estimated by measuring the difference between weight of dried empty petri dish and sample containing petri dish. The weight of dishes with dried milk samples was noted by analytical balance and % moisture was calculated by following relation:

$$\text{Moisture \%} = \frac{W_1 - W_2}{W}$$

Where,

W = Weight (gram) of milk sample

W<sub>1</sub> = Initial weight (gram) of the dish with sample taken for analysis

W<sub>2</sub> = Final weight (gram) of the dish with sample after drying

### 2.4 Determination of Ash (%)

10 mL milk sample was placed and charred in muffle furnace at 550 °C for 4 hours to get ash. Weight of each crucible was noted to measure the ash percentage.

$$\text{Ash (\%)} = \frac{W_2 - W_1}{W} \times 100$$

Where,

W = Weight (gram) of milk sample

W<sub>1</sub> = Weight (gram) of the empty crucible

W<sub>2</sub> = Weight (gram) of the crucible with sample after ashing.

### 2.5 Evaluation of Fat (%) by Gerber Method

Fat content was determined by Gerber's method using Gerber's Butyrometer as per AOAC methods [33].

### 2.6 Determination of Total Solid (%)

Total solids were determined as per AOAC method.

$$\text{Total solid (\%)} = \frac{W_2 - W}{W_1 - W} \times 100$$

Where,

W = Weight (gram) of dish

W<sub>1</sub> = Weight (gram) of the dish with sample taken for analysis

W<sub>2</sub> = Weight (gram) of the dish with sample after drying

### 2.7 Determination of Solid-not-fat (%)

Total solid and fat were determined by above-described two methods. Then solid-not-fat was determined for each sample by subtracting fat (%) from total solid (%).

$$\text{Solid Not Fat (\%)} = \text{Total solids (\%)} - \text{Fat (\%)}$$

### 2.8 Determination of Nitrogen (%) by Kjeldahl Method

Total nitrogen was determined by standard Kjeldahl's method.

$$\text{Protein (\%)} = \frac{\text{Titre used} \times 0.4 \times 6.38}{\text{Volume of sample}}$$

### 2.9. Evaluation of Specific gravity

Specific gravity was determined by lactometer method.

$$\text{Specific gravity} = \left( \frac{\text{Lactometer reading}}{1000} \right) + 1$$

## 3. RESULTS AND DISCUSSION

The milk samples of four cow breeds (Red Sindhi, Holstein Frisian, Cholistani and Sahiwal breeds) were analyzed for the presence of various parameters (moisture, ash, protein, fat, solids-not-fat, total solids, specific gravity and calcium); the obtained results are summarized in Table 1a whereas the statistical analysis (two-way ANOVA) data has been shown in Table 1b.

### 3.1 Calcium

By potassium permanganate titration method, it was found that Sahiwal, Cholistani, Holstein Frisian and Red Sindhi breeds contain 0.063, 0.055, 0.063 and 0.056 % calcium, respectively. The slight differences between the observed calcium values may be owed to the differences in their habits such as grazing habit, reproductive habit etc.

Atomic absorption spectroscopy was performed to verify the results of calcium concentration as obtained by potassium permanganate method. The Holstein Frisian cow and Sahiwal cow contained 0.06 % and 0.059 % calcium, respectively. The obtained results are thus very close to those obtained by permanganate method.

### 3.2 Moisture percentage

Sahiwal cow, Cholistani cow, Holstein Frisian cow and Red Sindhi cow were found to possess the moisture contents of 87.5, 88.2, 87.35 and 85.65 %, respectively in their milk samples. Red Sindhi cow possessed the lowest moisture content while Cholistani cow demonstrated the highest amount of moisture content in its milk. The moisture content resembles closely with that (82-90%) reported earlier in different milk samples [34].

### 3.3 Ash percentage

Sahiwal cow, Cholistani cow, Holstein Frisian cow and Red Sindhi cow had shown the presence of 0.709, 0.791, 0.77 and 0.705 % ash in their milk. Thus Cholistani cow possessed the highest ash content (0.791 %) as compared to the other three breeds whereas the lowest ash content (0.705 %) was observed in Red Sindhi cow.

### 3.4 Fat percentage

A fat content of 3.8, 4.1, 3.5 and 3.95% was observed in the milk samples of Holstein Frisian cow, Sahiwal cow, Cholistani cow and Red Sindhi cow, respectively. Milk sample of Sahiwal cow has shown the highest fat content (4.1 %) and Cholistani cow possessed the least amount (3.5 %) of fat. However, the obtained fat range 3.5-4.1 % in the investigated samples lies within the range 3.3-4.4 % already reported for the cow's milk [35]. Various factors such as period of lactation, individual traits, nutrition and breed govern the concentration of fat in a milk [35] because various kinds of plant diets govern their nutritional contents [36-38].

### 3.5 Total Solid Percentage

12.5, 11.8, 12.65 and 14.35 % total solids were found in milk samples of Sahiwal cow, Cholistani cow, Holstein Frisian cow and Red Sindhi cow, respectively. Red Sindhi cow possessed the highest percentage (14.35 %) of total solids whereas in Cholistani cow, the lowest percentage (11.8 %) of total solid was observed. The difference in breeds may be due to fodder difference, difference of lactation period, climate and health status of the breed. Actually, different plants have variable amounts of phytochemical and nutritional

ingredients [39-41] and thus affect the nature of milk when they are used as food for cattle.

### 3.6 Solid-not-fat Percentage

It was found that Sahiwal, Cholistani, Holstein Frisian and Red Sindhi breeds have solid-not-fat of 8.4, 8.3, 8.85 and 10.4 %, respectively. Red Sindhi cow contained the highest amount (10.4 %) of Solids-not-fat whereas Cholistani cow demonstrated the lowest value of solids-not-fat (8.3 %) in its milk.

### 3.7 Protein percentage

Sahiwal, Cholistani, Holstein Frisian and Red Sindhi breeds have shown the protein contents of 3.318, 3.126, 3.33 and 3.38 %, respectively in their milk samples. From the above results, it is concluded that Red Sindhi cow has the highest protein content while Cholistani cow shows lowest quantity of protein in its milk sample. Earlier reports verify that beta-lactoglobulin is present in elevated concentrations in cow milk whey protein [42].

### 3.8 Lactometer reading and specific gravity

Lactometer reading and specific gravity were found to be 31 and 1.031 in Holstein Frisian cow, 28 and 1.028 in Red Sindhi cow, 30.5 and 1.0305 in Cholistani cow and 30 and 1.03, in Sahiwal cow as shown in Table 1a. So, it can be concluded that Holstein Frisian possessed the highest value of lactometer reading and specific gravity. Red Sindhi has shown the lowest value of lactometer reading and specific gravity.

## 4. CONCLUSIONS

Nutritional parameters such as calcium, specific gravity, protein, fat, moisture, solid-not-fat, total solids and ash were analyzed in cow milk of four breeds (Red Sindhi, Sahiwal, Cholistani and Holstein Frisian). Calcium in the investigated milks samples was found to be in the range of 0.06-0.05 % as determined by potassium permanganate titration and atomic absorption spectrometry. Protein was observed in the range of 3.312-3.38 % whereas fat contents were found in the range of 3.5-4.1 % in the

**Table 1a.** The values of various parameters in investigated milk samples

Parameters	Breeds			
	Cholistani cow	Sahiwal cow	Holstein Frisian	Red Sindhi
Calcium %	0.055	0.059	0.063	0.056
Protein %	3.126	3.318	3.33	3.38
Fat %	3.5	4.1	3.8	3.95
Moisture%	88.2	87.5	87.35	85.65
Lactometer	30.5	30	31	28
Solid-not-fat	8.3	8.4	8.85	10.4
Total solid	11.8	12.5	12.65	14.35
Ash%	0.791	0.709	0.77	0.705
Specific Gravity	1.030	1.030	1.031	1.028

**Table 1b.** Statistical analysis by using two-way ANOVA was performed which showed significant results in rows ( $P < 0.05$ ) and non-significant in columns  $P > 0.05$

Source of Variation	Rows	Columns	Error	Total
SS	25346.14	0.16	15.12	25361.43
df	8	3	24	35
MS	3168.27	0.05	0.63	
F	5027.89	0.09		
P-value	0.0000	0.9675		
F crit	2.36	3.01		

milk samples. Moreover, moisture (85.65- 88.2 %), solids-not-fat (8.3-10.4 %), total solids (11.8- 14.35 %) and ash (0.705- 0.791 %) were observed in the tested milk of cow breeds. Lactometer reading of cow milk was shown in the range of 28-31°. Specific gravity in cow milk of four breeds was in range of 1.028-1.03 %. A little difference in investigated parameter values is due to the differences in the cow breeds as they vary in their genetics, body and habitat. However, it can be concluded that milk of different cow breeds possesses variable amounts of nutritional contents i.e., highest moisture and ash in Cholistani cow, highest fat in Sahiwal cow, highest calcium and specific gravity in Holstein cow and highest contents of protein, solid-not-fat and total solid in Red Sindhi cow. Red Sindhi cow's milk was found to be more nutritious in terms of its richness in proteins, solid-not-fat and total solids whereas Holstein cow was rich in calcium as compared to other breeds.

## 5. CONFLICT OF INTEREST

The authors declare no conflict of interest.

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