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COMPARATIVE ANALYSIS OF MILK COMPOSITION FROM DIVERSE SOURCES IN SUKKUR: A BOUNTY OF NUTRIENT DIVERSITY

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Abstract

Milk, often hailed as nature's elixir, is a treasure trove of essential nutrients crucial for human health. In this comprehensive study, delve into the fascinating world of milk, exploring its diverse origins and uncovering a tapestry of nutritional richness. Sukkur, a city perched on the banks of the majestic Indus River in Pakistan, serves as a canvas, showcasing the remarkable variations in milk composition obtained from various sources. The findings not only shed light on the distinct nutritional profiles of human and animal milk but also underscore the versatility and safety of these sources for consumption. The analysis unveiled a captivating tableau of milk composition diversity. Protein content reigned highest in human milk samples at 2.343 mg/L, contrasting with rural sheep milk's meagre 0.89 mg/L. Carbohydrates were highest in human milk (1.12 mg/L) but lowest in dairy buffalo milk (0.3 mg/L). TDS levels reached their zenith in rural sheep milk (12%) but ebbed in human milk (7.43%). Fat content triumphed in dairy sheep milk (3.75%) while rural cow milk lagged at 2.75%. Ash content reached its pinnacle in dairy goat milk (0.257%), but rural cow milk displayed a more modest 0.175%. While variations were observed, they generally aligned with global benchmarks. The present study concludes that there is no significant difference among the samples compared to the reported work. Therefore, milk from any of the mentioned sources can be used without any dangerous effects.

Keywords:

analysis of milk, protein, carbohydrate, fat, TDS, ash, spectrophotometer

1. INTRODUCTION

Milk, the life-giving elixir produced by mammalian mothers for their young, is a nutritional powerhouse. This study unveils the captivating diversity of milk sourced from humans and animals, emphasizing its paramount role in nourishing populations across the globe. It contains a complex mixture of carbohydrates, proteins, vitamins, fats, minerals, and other bioactive compounds that are essential for development and growth (USDA 2021). Pakistan, a milk-producing giant, takes centre stage, producing a staggering 48 million tonnes annually. The rich tapestry of milk sources, including buffalo, cow, goat, sheep, and even humans, beckons exploration, with each offering a unique blend of nutrients (Sattar 2020). In terms of goat and buffalo milk, Pakistan ranks 2nd, producing 22.3 and 0.7 billion litres, respectively, after India and Bangladesh. Buffalo milk comprises the highest share (62.8%), followed by Cow (34.9%), Goat (2.0%), and Sheep (0.1%). Regarding the population of animal species, the collected data indicates that Buffaloes, Cows, Sheep, and Goats number at 30.8, 34.3, 27.8, and 1.0 million, respectively. (Bilal M Q 2006).

According to the USDA, cow's milk is the most widely consumed in the US, and it is an important source of nutrition for Americans. It contains high-quality proteins, including all of the essential amino acids. It is also a rich source of calcium, which is essential for bone health, and contains other minerals such as phosphorus, magnesium, and potassium. In addition to its nutritional value, milk has several functional properties that make it useful in food processing. It can be used to create a variety of dairy products like yogurt, cheese, and butter. Milk also contains bioactive compounds such as lactoferrin and immunoglobulins, which have antibacterial and immune-boosting properties. Some people may have lactose intolerance or milk allergies, which make them unable to digest lactose, the primary sugar present in milk. In such cases, alternative milk sources like almond milk, soy milk, and oat milk are available for consumption.

Milk is a chief source to obtain a significant amount of fat, minerals, and protein, which are beneficial for health. Therefore, new-borns and school-age children are prime examples and beneficiaries of milk and its products (Muehlhoff, Bennett et al., 2013). The composition of milk samples varies with the source of animal species, but all sources are excellent for physical appearance and genetics as well (Karmaker, Das et al., 2020). Goat milk differs from human or cow milk in its good buffering capacity, alkalinity, and digestibility (Khan, Choi et al., 2014). Each source of milk has its own characteristics, which change with animal species, but they all share a high priority in human nutrition. Buffalo milk is common worldwide, accounting for about 5% of milk production. Asia yields over 95% of the buffalo milk in the world (Siregar, Susanti et al., 2020). Cow and buffalo milk are extensively used due to their rich nutritious and flavourful properties, as they contain high fat and TDS (Total Dissolved Solids).

The present study was based on correlational ideas, and the composition of milk samples includes protein, carbohydrate, TDS, fat, and ash, as mentioned in the reported work. Milk contains many

nutrients that are important for maintaining good health, such as calcium, vitamin D, and protein. Research into the composition and nutritional value of milk can help us better understand the health benefits of consuming dairy products. Milk is a key ingredient in many food products, such as cheese, yogurt, and ice cream. Researchers in food science might investigate how different processing techniques or ingredients affect the quality and flavour of these products. Some research suggests that milk consumption may be linked to certain health conditions, such as allergies or lactose intolerance. Investigating these associations can help us better understand the role of dairy products in human health.

The aim of the present study was to investigate and explain the biochemical parameters of milk samples and any significant changes found in the Sukkur region of Sindh, Pakistan. The research can help us better understand the nutritional, agricultural, and medical properties of milk, which can have important implications for public health and well-being.

2. MATERIAL AND METHOD

2.1. Area of Study

Sukkur, a city steeped in dairy farming tradition, serves as the study's backdrop. Nestled on the Indus River's western bank, Sukkur is a thriving hub of dairy production, housing a constellation of milk processing plants and dairy cattle farms.

2.2. Chemicals and Reagents

In the quest to unravel the secrets of milk's composition, current work employed a meticulous selection of high-quality chemicals and reagents. These vital components ensured the accuracy and reliability of the analytical methods.

Biuret Reagent was utilized in the determination of protein content. This reagent played a pivotal role in the study. Its formulation consisted of 4 ml of biuret reagent and 1 ml of deionized water, expertly mixed to perfection. Petroleum Ether was used for the precise quantification of fat content; 60 ml of petroleum ether was used in conjunction with milk samples. This solvent allowed for the extraction of fat from the milk matrix, ensuring the accuracy of measurements. Hydrochloric Acid (HCl) used in the carbohydrate determination process; 5 ml of 2.5N HCl was skilfully employed to facilitate the hydrolysis of carbohydrates within milk samples. Sulfuric Acid (H_2SO_4) played a pivotal role in the Phenol-Sulfuric method used for total carbohydrate determination. A mixture consisting of 5% phenol and 96% sulfuric acid was meticulously prepared to ensure precise measurements. Sodium Carbonate (Na_2CO_3) is an essential component for neutralizing the hydrolysed samples during carbohydrate determination. Sodium carbonate was used to maintain the integrity of the results. Bovine Serum Albumin was employed as a standard in protein analysis, and bovine serum albumin was rigorously calibrated to guarantee the accuracy and reliability of protein measurements. Deionized Water, a cornerstone in analytical procedures,

was used for dilution, sample preparation, and as a key component in reagent formulations. Its purity was paramount to success.

These meticulously chosen chemicals and reagents formed the backbone of analytical endeavours, ensuring the robustness of findings and the validity of conclusions. All chemicals were purchased from Merck in Germany.

2.3. Standards and Sample Preparation

To ensure the precision and accuracy of analytical methods, samples were meticulously prepared from standards and reference materials for key parameters. Bovine Serum Albumin (BSA) served as the gold-standard protein reference material. A series of BSA solutions was prepared with known concentrations, i.e., 0.5, 0.04, 0.3, 0.2, and 0.1 mg/mL, allowing us to create a calibration curve for protein determination (Fig. 1).

2.4. Sample Collection

The rigorous sample preparation process was designed to maintain the integrity of the milk samples and guarantee the reliability of the results. Milk Samples were collected using a random sampling method. Samples were collected in triplicate from various sources, including buffalo, cow, goat, sheep, and humans, totaling 90 samples. These samples were promptly chilled at 4°C to preserve their freshness and composition during transportation to the laboratory.

2.5. Analytical Methods

The analytical arsenal included a state-of-the-art UV-Visible Spectrophotometer, a powerful tool for dissecting milk's nutritional content. Assessment of protein, carbohydrate, Total Dissolved Solids (TDS), fat, and ash content following established AOAC methods. For the determination of protein content, 1 ml of fat-free milk sample was precisely measured and combined with 4 ml of biuret reagent and 1 ml of deionized water. This mixture was then cooled and subjected to analysis at 540 nm using a UV-visible spectrophotometer. Before analysing the milk samples, BSA standards of known concentrations were meticulously prepared and analysed to generate a linear calibration graph for accurate protein measurements. To determine ash content, crucibles were heated in an oven at 300°C overnight to ensure they were free of impurities. Five grams of each milk sample were then placed in these pre-prepared crucibles and subjected to heating in the oven. Ash content was calculated by weighing the resulting ash and crucible. For fat determination, 10 ml of each milk sample was combined with 60 ml of petroleum ether. The samples were shaken for 20 minutes, followed by the removal of the aqueous layer. The remaining layer, containing the fat, was gently heated, and its weight was measured to calculate the fat content. For Carbohydrate Analysis (Phenol-Sulfuric Method), fat-free milk samples were taken in triplicate, and a meticulous process ensued. First, 5 ml of each sample was combined with 5 ml of 2.5N HCl and placed in a boiling water bath for three hours. After cooling, the samples were neutralized with

Na_2CO_3, and their volume was adjusted to 100 ml with de-ionized water. These samples were then centrifuged for 5 minutes. Total carbohydrate content was determined using the Phenol-Sulfuric method, with absorbance measured at 490 nm.

During the research process, dedication to precise sample preparation, calibration, and adherence to standard procedures guaranteed the accuracy and reliability of findings, unravelling the captivating diversity of milk composition in the Sukkur region.

3. Instrumentation

3.1. UV -Visible Spectrophotometer

The research harnessed the power of advanced scientific instrumentation, with a primary focus on the UV-Visible Spectrophotometer. This cutting-edge device served as the cornerstone of an analytical arsenal, enabling us to probe the intricate composition of milk samples with precision and accuracy (Sablinskas 2014). The sophisticated UV-Visible Spectrophotometer instrument, depicted in Figure 2, represents a technological marvel in the realm of analytical chemistry. Operating on the fundamental principle of measuring light absorption, it allowed us to examine milk samples in the ultraviolet (UV) and visible (vis) spectrum range.

The spectrophotometer facilitated the quantification of specific substances within the milk samples by precisely measuring the amount of light absorbed at distinct wavelengths (Kakkar 2015). With the ability to measure light in nanometres (nm), the device provided critical insights into the absorption characteristics of various milk components. The UV-Visible Spectrophotometer found versatile applications across the study, from protein analysis using the Biuret reagent method to carbohydrate determination through the Phenol-Sulfuric method. This instrument ensured the generation of highly accurate and reproducible data, essential for establishing robust conclusions. The judicious utilization of the UV-Visible Spectrophotometer underscored commitment to scientific rigor and the quest for a comprehensive understanding of milk composition.

4. Result and Discussion

Table No. 1 shows that the concentration of protein was highest in Human milk (2.343 mg/L) compared to other milk samples analysed during the study. However, the percentage of protein is very low in milk from all sources compared to the available literature (Roy, Ye et al., 2020). The lower value indicates that animals might not be getting the proper nutrients they need, and therefore, farmers are required to play an active role in ensuring adequate nutrition for their animals. The minimum protein was found in rural sheep (0.89 mg/L).

Carbohydrates were also found to be maximum in Human milk samples (1.12 mg/L), while their minimum contents were found in dairy buffalo (0.3 mg/L), likely because dairy buffalo primarily feed on dry foliage most of the time. The carbohydrate contents of the present study were found to be less compared to the available literature for buffalo, cow, goat, and sheep milk samples.

TDS (Total Dissolved Solids) was high in rural sheep (12%) likely due to greater chances of free grazing in the jungle, while less amount of TDS was found in human milk samples (7.43%). TDS levels were within the same range as reported in the literature.

Fat was found to be the highest in dairy sheep milk samples (3.75%), while the minimum percentage was in rural cow milk samples (2.75%). The fat content of buffalo, cow, goat, and sheep milk in the present study was within the same range as reported in the literature.

Ash was found to be the maximum in dairy goat milk (0.257%), while the minimum value was observed in rural cow samples (0.175%). The quantity of ash in the present study was high (Fig. 3: Analysed average results of Protein, Carbohydrate, TDS, Fat, and Ash of different milk samples).

Table No. 2 shows a comparison between the current study and available literature, indicating that protein and carbohydrate levels are the lowest in all categories in the current study. TDS does not show much variation and remains within a similar range as reported in the literature, while fat shows the same pattern, and ash is slightly lower in the current study. Overall, the current study does not show significant differences compared to work reported in different parts of the world.

Table No. 3 shows the percentage (%) contribution of buffalo, cow, goat, and human milk intake with the recommended human daily dietary allowances, RDA (Muehlhoff, Bennett et al., 2013). It clearly indicates that children smaller than 7 months require more protein from milk because they need it for their growth, and they mainly rely on milk for nutrition. As the child grows to 7 months and above, they also start eating other foods, which provide additional protein. After reaching 7 months, they need a slightly greater amount of fat, which is essential for their health and growth.

5. Conclusion

In-depth analysis brings reassuring news that there are no substantial differences among milk samples in the current study compared to worldwide findings. This implies that milk from any of the tested sources can be confidently embraced without concerns of adverse effects on health. These findings not only highlight the nutritional riches of milk but also underscore its safety and versatility.

6. Declarations

6.1 Ethics Approval and Consent to Participate

The research protocol and procedures outlined in this study have been reviewed and approved by Shah Abdul Latif University Ethics Committee (Approval Number: 2018/204). All participants provided informed consent before they participated in the study. They were informed about the study's objectives, procedures, potential risks, and benefits, and were assured that their participation is voluntary. Participants were also informed about their right to withdraw from the

study at any time without consequences. Confidentiality of participants' information and data will be strictly maintained throughout the study and reporting process.

6.2 Consent for Publication

Participants have consented to the publication of anonymized and aggregated data obtained from this study. Any identifiable information will be carefully removed or altered to ensure the privacy and confidentiality of the participants. Images, audio recordings, or other forms of identifying information that could compromise anonymity will not be included unless explicit written consent has been obtained from the participants.

6.3 Availability of Data and Materials

The datasets generated and analyzed during the current study are available from the corresponding author upon reasonable request. All data will be provided in a format that ensures participant anonymity and protects confidential information. Materials used in the study, such as survey questionnaires or interview guides, can also be made available upon request.

6.4 Competing Interests

The authors declare that they have no competing interests related to the research presented in this study. Financial, personal, or professional conflicts of interest that could potentially influence the research, analysis, or reporting process have not influenced this work.

6.5 Funding

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6.6 Authors' Contributions

Dr. Mushtaque Ali Jakhrani contributed to conceptualization and methodology and supervised the whole process. Mr. Safeullah Bullo and Dr. Sanaullah Ansari contributed to data collection, analysis, and writing. Hafeezullah Mazari, Nabidad Bajkani, Abdul Aziz Bakhrani, Laraib Ali Awan, Amjad Hussain Soomro, Qandeel Haider Hundal, Ali Bahar Shahani, Nazia Rind, Pooja Bai, and Nazia Mumtaz Amur helped in review and editing.

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Table 1. Analysed average results of Protein, Carbohydrates, TDS, Fat and Ash, of Buffalo, Cow, Goat, Sheep and Human milk

Animal	Protein mgL^{-1}	Carbohydrate mgL^{-1}	TDS	Fat%	Ash%
Dairy Buffalo	1.37±0.092	0.3±0.023	10.09±0.443	3.16±0.172	0.234±0.375
Rural Buffalo	1.61±0.057	0.38±0.025	10.62±0.472	3.161±0.259	0.201±0.469
Dairy Cow	1.12±0.094	0.39±0.013	10.34±0.485	2.54±0.166	0.252±0.823
Rural Cow	1.66±0.1	0.55±0.015	9.77±0.436	2.75±0.191	0.175±0.441
Dairy Goat	1.35±0.1	0.58±0.01	10.91±0.5	3.27±0.221	0.257±0.495
Rural Goat	0.97±0.068	0.56±0.015	9.77±0.465	3.23±0.232	0.191±0.398
Dairy Sheep	1.23±0.086	0.33±0.015	10.71±0.439	3.75±0.242	0.211±0.475
Rural Sheep	0.89±0.073	0.34±0.019	12±0.444	3.21±0.253	0.238±0.56
Human Milk	2.343±0.13	1.12±0.038	7.43±0.409	3.67±0.299	0.185±0.374

Mean ±Standard Deviation (Triplicates)

Table 2 Comparison between current study with available literature

Animal	Protein mgL^{-1}	Carbohydrate mgL^{-1}	TDS	Fat%	Ash%
Buffalo					
A	2.7-4.7	3.2-4.9	15.7-17.2	5.3-9.0	0.8-0.9
B	N/A	N/A	N/A	N/A	N/A
C	4.6	N/A	18.2	7.26	0.79
D	1.49	0.34	10.355	2.8505	0.2175
Cow					
A	N/A	N/A	N/A	N/A	N/A
B	3.27	4.52	N/A	3.49	N/A

C	3.2	N/A	11.2	3.28	0.74
D	1.39	0.47	10.055	3.01	0.2135
Goat					
A	3.0-5.2	3.2-5.0	11.9-16.3	3.0-7.2	0.7-0.9
B	2.85	4.13	N/A	3.58	N/A
C	3.2	N/A	13.21	4.26	0.83
D	1.16	0.57	10.34	3.49	0.224
Sheep					
A	4.5-7.0	4.2-5.9	18.1-20.0	5.0-9.0	0.8-1.0
B	N/A	N/A	N/A	N/A	N/A
C	4.6	N/A	16.22	4.92	0.94
D	1.06	0.335	11.355	3.44	0.2245
Human Milk					
A	10.7-12.9	5.8-7.4	10.7-12.9	2.1-4.0	0.2-0.3
B	N/A	N/A	N/A	N/A	N/A
C	N/A	N/A	13.53	4.17	0.21
D	2.342	1.12	7.43	3.67	0.185

A = New Zealand (Roy, Ye et al. 2020)
B = UK (Stergiadis, Nørskov et al. 2019)
C = Czech Republic (Borková and Snášelová 2005)
D = Present Study
N/A = not available

Table: 3 Percent (%) contribution of buffalo, cow, goat and human milk intake with the recommended human daily dietary allowances, RDA (2002, FAO USA)

Animal	Protein	Fat	Carbohydrate	Protein	Fat	Carbohydrate
			Children			
		0-6m			7-12m	
Buffalo	110.37	60.67	22.23	91.31	62.69	14.04
Cow	95.55	33.37	20.74	79.05	34.48	13.10
Goat	91.32	32.54	18.49	75.55	33.63	11.68
Human	30.55	33.63	33.49	25.27	34.75	21.15
			Adult			
		Male			Female	
Buffalo	17.94	N/A	10.26	21.83	N/A	10.26
Cow	15.53	N/A	9.57	18.90	N/A	9.57
Goat	14.84	N/A	8.53	18.07	N/A	8.53
Human	--	--	--	--	--	--

N/A = not available

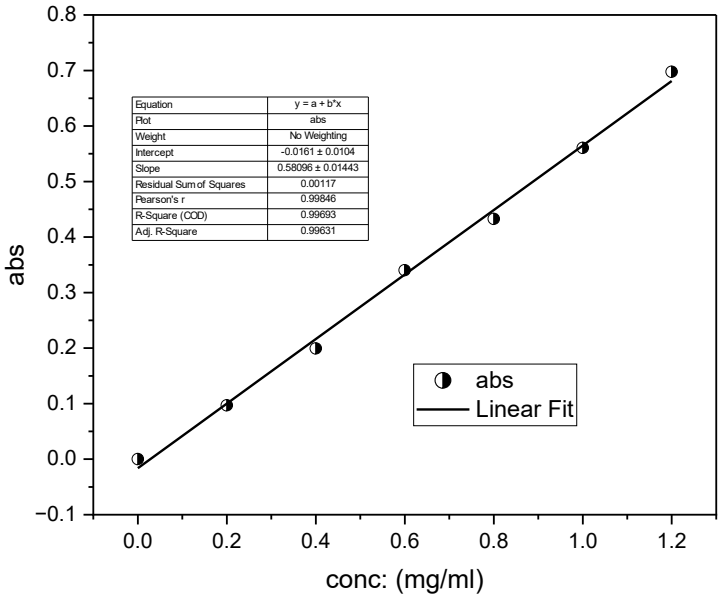

Fig. 1. Calibration graph of standard protein

Fig. 2. UV Visible Spectrophotometer

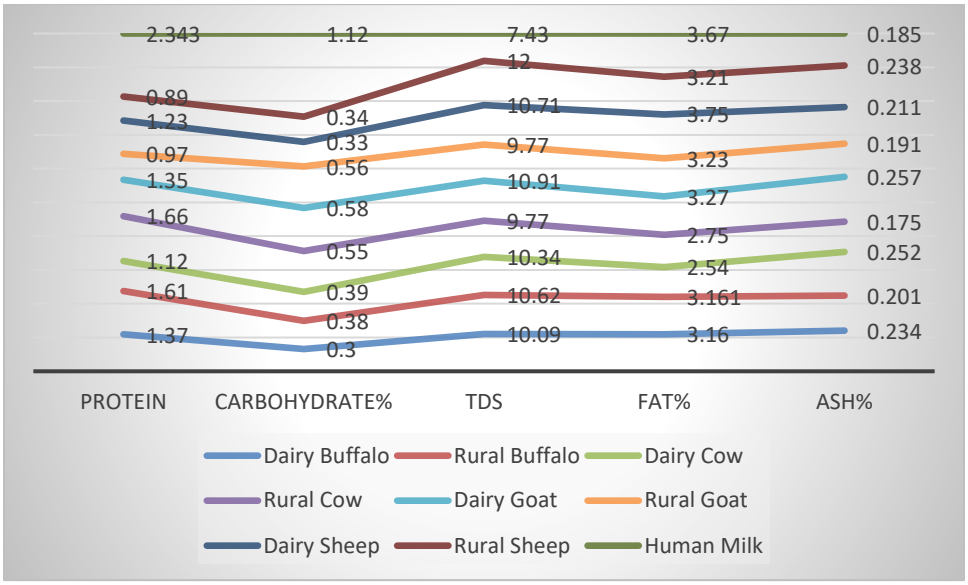

Fig. 3. Analysed average results of Protein, Carbohydrate, TDS, Fat and Ash of different milk samples

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