Journal of Advances in Applied Sciences and Technology
ISSN : 2393-8188 (print)
2393-8196 (online)
www.milliyasrcollege.org.journal.php

MICROBIOLOGICAL AND CHEMICAL ANALYSIS OF BUFFALO MILK SUPPLIES OF BEED DISTRICT

Seema Hashmi [1]*& Quazi Saleem[2]

[1] *Dept of microbiology Milliya College BEED.*

[2] *Dept of Zoology Milliya College BEED.*

***C**orresponding author *quazisaleem@gmail.com*

Abstract : This study was carried out to determine the chemical and microbiological quality of buffalo milk. A total of 120 buffalo raw milk samples were collected monthly from beed district, throughout the year for this study. In the chemical analysis of buffalo milk samples the mean total solid value, non-solid fat, lipid, protein, lactose, ash and pH values were detected as 16.38%, 8.56%, 7.04%, 4.36%, 4.19%, 0.72% and 6.55, respectively. Total bacteria count (TCA), coliform, lactic acid bacteria (LAB), *Escherichia coli*, *Staphylococcus aureus* and yeast-mold (log10 cfu/ml) levels in the milk samples were detected as 6.36, 2.95, 5.74, 1.10, 2.46 and 2.63, respectively.

Key words: Buffalo milk, chemical quality, microbiological quality

INTRODUCTION

Milk is a white liquid produced by the mammary glands of mammals. It is the primary source of nutrition in the world with its high nutritious property. If convenient storage temperatures are not paid attention, milk becomes a suitable propagation medium for microorganisms due to its biochemical composition and high water activity. Milk can be easily contaminated and spoiled when it is produced in unhygienic environment. Milk quality is directly related to its composition and hygiene [14]. Buffalo milk has become a research subject and received increasing attention in many countries due to its rich nutrient content . Compared to cow milk, buffalo milk has a richer taste due to its contents of milk fat, protein, lactose, total dry matter, vitamin and minerals. These properties allow a wider variety for buffalo milk as raw material for milk products like cheese, butter and ice-cream . [11] There has been an increasing demand for cheese made of buffalo milk in many countries throughout the world as it is an organic product . [4]

It is reported that global buffalo milk was 90 million tons in 2009 and accounted for 13% of total milk production. The same report stated that more than 90% of total buffalo milk in the world is produced by India and Pakistan.. [3]Interest and investments in buffalo milk in different countries is increasing each year due to its unique taste and nutritious content .The products made of buffalo milk like mozzarella cheese, cream, ice-cream and yogurt have commercial impor-tance, as well . As in raw milk, microorganisms could rapidly propagate in buffalo milk due to rich nutrient content. In previous studies carried out on microbiological properties of buffalo milk , coliform bacteria, *Escherichia coli*, lactic acid bacteria, yeast-mold and *Staphylococcus* spp., were isolated from buffalo milk. Saprophytic microorganisms in milk could spoil buffalo milk, while the presence of pathogen bacteria could pose a potential health threat [10].

Contamination of milk and milk products mostly results from human factor and unhygienic conditions. Milk is generally contaminated in milk collection places. Coliform bacteria are microorganisms in natural flora of human and animal intestinal tract and they are accepted as indicators for bacteriological quality of milk and milk products . Furthermore, the presence of these microorganisms indica-tes the possible existence of enteric pathogens that could threat public health. [2] The most important indicators for microbiological quality include total bacteria number, coliform, yeast and mold quantity and detection of spe-cific pathogens and their toxins . Among all microorganisms, *E. coli* is an organism that frequently contaminates foods. They generally exist in milk and milk products due to inadequate sanitation. The main thing in micro-biological investigation of milk is the determination of contamination degree and number of indicator micro-organisms. Coliform bacteria are reported to define the suitability of milk for human consumption. [1]

Besides microbiological quality of milk, its physical and chemical properties are also quite important. Changes in milk composition depend on many factors like genetic, lactation time, daily variations, birth, alimentation type, age, udder cleaning and season. These factors greatly affect the quality and processing ability of cheese, butter and other milk products . Geographic region, climate conditions and lactation period are known as seasonal changes and cited among factors affecting milk composition. Especially, there is an inverse proportion between ambient temperature and protein and fat content of milk. Solid fats tend to decrease with increasing air temperature. Fat, protein, casein and all fractions of nitrogen are affected by seasonal change. It is reported that high ambient temperature causes clot hardness to decrease and clot formation rate and clotting time to increase [16]. At high ambient temperatures, fat concentration decreases, while lactation period extends. Some studies reported that significant changes occur in the amounts of short chain fatty acids, and they become minimum in winter and maximum in summer.

The main physical characteristics of milk are defined as pH and electrical conductivity; in addition, fat content of buffalo milk is the most variable milk component, which is caused by genetic and specific factors [13]. Carbohydrates are preliminary form of glucose circulating as lactose in milk. High fat and calorie contents besides dry matter are regarded as the superior and distinctive property of buffalo milk. Buffalo milk is processed into many products including butter, cream, hard and soft cheese, ice-cream and yogurt.

The aim of this study carried out in Beed district an important region for buffalo milk and milk products with dense buffalo population, was to deter-mine the chemical and microbiological qualities of seasonally collected buffalo milk samples and to investigate the effect of seasonal change on milk composition.

MATERIAL AND METHODS

MILK SAMPLES

A total of 120 raw milk samples were collected randomly every month from 10 small sized family enterprises (farms had ≤10 buffaloes) between September 2012 and August 2013. Two hundred and fifty milliliter (250 ml) of milk samples were taken from producer under aseptic conditions and transferred in sterile bottles to laboratory in cold chain 4°C' and then they were analyzed.

CHEMICAL ANALYSES

Fat, protein, lactose, total dry matter (DM) and ash contents of buffalo milk samples were determined by pre-calibrated LactoStar milk analysis device (FUNKE GERBER, Germany). pH values of milk samples were determined by InoLab (pH Level L 01280054) pH meter device.

MICROBIOLOGICAL ANALYSES

In the analysis, 10 ml of milk was taken from each milk sample and homogenized in sterile bags containing 90 ml of sterile buffered peptone water for 1-2 min and then dilutions were prepared & Inoculated on Plate Count Agar (Oxoid CM0325) using spread plate method in order to determine the number of mean total bacteria count (TCA) in the prepared dilutions. Violet Red Bile Agar (Oxoid-CM 107) was used to determine coliform bacteria count and incubated under aerobic conditions at 37°C for 24 h . Inoculation was done on MRS Agar (Oxoid-CM 0361) for lactic acid bacteria (LAB) count and left for 48-72 h of incubation under anaerobic conditions at 30°C .

Inoculations were made on Tryptone Bile X- Glucuronide Medium (TBX) (Oxoid-CM945) for *E. coli* count and incubated at 44°C for 18-24 h (Anonymous, 2001). Egg Yolk Tellurite emulsion (Oxoid-SR54) was included for *S. aureus* count and performed in Baird Parker Agar (Oxoid -CM275) at 37°C for 24-48 h in aerobic conditions. Inoculation was done on Rose Bengal Chlo-ramphenicol Agar (Oxoid-CM549) for yeast and mold count and incubated at 25°C for 72 h under aerobic conditions .

STATISTICAL ANALYSES

The microorganism numbers detected in this study were transferred to base 10 logarithm values and then statistical data was obtained by SPSS statistics software. One way variance analysis was per-formed to determine the differences between microorganism numbers and chemical parameters in terms of seasons, and Duncan test was applied to determine differences among the means.

RESULTS

CHEMICAL ANALYSIS

As a result of the chemical analyses of milk samples col-lected from buffaloes in different seasons, the total solids (TS) (16.38% ± 1.01), non-fat solids (NFS) (8.56 ± 0.32), fat (7.04% ± 0.84), protein (4.36% ± 0.40), lactose (4.19% ± 032), ash (0.72% ± 0.08) and pH (6.55 ± 013) values were determined (Table 1). The data of this study indicated that milk fat and protein contents increase concurrently, while TS, NFS, fat, protein, ash and pH values are highest in winter, while TS, fat and protein levels are lowest in spring.

Table 1. Chemical analysis results of Buffalo milk.

Season	Totalsolids (%w/w)	Non fat solid(%w/w)	Fat(%v/v)	Lactose(%w/w)	Protein(%w/w)	Ash(%w/w)	pH
Rainy	17.41±0.54	8.89±0.28	7.67±0.55	3.99±0.12	4.90±0.26	0.78±0.07	6.68
Winter	15.61±0.74	8.49±0.23	6.38±0.66	4.45±0.20	4.03±0.16	0.69±0.07	6.45
Summer	15.90±1.03	8.49±0.27	6.70±0.89	4.05±0.14	4.43±0.25	0.66±0.06	6.45
Mean	16.38±1.01	8.56±0.32	7.04±0.84	4.36±0.40	4.19±0.32	0.72±0.07	6.55

MICROBIOLOGICAL ANALYSIS

Regardless of seasons, the mean total bacteria count (TBC), coliform bacteria, lactic acid bacteria (LAB), *E. coli*, *S. aureus* and yeast-mold values (log10 cfu/ml) were determined as 6.36 ± 0.28, 2.95 ± 0.21, 5.74 ± 0.31, 1.10 ± 0.17, 2.46 ± 024 and 2.63 ± 0.25, respectively .

Contamination levels of milk with TBC, coliform, *S. aureus* and yeast-mold were found lower in winter than in other seasons,. The highest contamination levels with these bacteria were observed in summer.

DISCUSSION

In this study, the mean total solids, non-fat solids, fat, protein, lactose and pH levels of milk samples collected in different seasons were determined as 16.38%, 8.56%, 7.04%, 4.36%, 4.19%, 0.72% and 6.55, respectively. Fat, protein and ash contents were determined to decrease in hot summer months, while lactose content was reported to increase. It was reported that buffalo milk contain has higher nutritious values with higher protein, fat, lactose and TS than cow milk. Different researchers reported that alimentation, lactation period, milking frequency, milking method and season have important effects on physicochemical parameters of buffalo milk [9].In this study, the mean lactose level of buffalo milks (4.19% ± 032) was found lower On the other hand, milk fat is the most changeable milk component. Fat content is affected by many factors. The most important factors are seasonal change and lactation period. Fat, protein and ash contents tend to increase in winter, and milk yield is reported to increase in the later periods of lactation, while fat and protein contents decrease [6]. It is also reported that habitat and feeding pattern are quite effective on milk fat and protein levels, and milk protein and NFS contents of animals grazing in summer are higher than those of animals closed-fed in winter [7]. In this study, the mean fat (7.04 ± 0.84 %v/v), total solids (16.38 ± 1.01% w/w) and protein (4.36 ± 0.40% w/w) contents are slightly lower . Similar results were reported by [5].

Variance analysis was used to investigate chemical composition. Total solids, protein, non- fat solids and pH values were significantly higher ($p<0.01$) in winter than in other seasons. On the other hand, the lactose content was found higher ($p<0.01$) in summer than in winter and rainy . The mean fat and ash contents were found highest in winter .

Similar to milk components, microbiological quality of milk changes by ambient temperature. In this study, the mean total bacteria count (TBC), coliforms, lactic acid bacteria, *E. coli*, *S. aureus* and yeast-mold (log10 cfu/ml) levels were determined as 6.36±0.28, 2.95±0.21, 5.74 ± 0.31, 1.10 ± 0.17, 2.46 ± 024 and 2.63 ± 0.25, respectively, and microorganism load was determined to increase in warm months.

TBC was determined as 2.30x106 cfu/ml. This level is higher than the standards. The main reason for these relatively higher counts of TBC should be ascribed to poor hygiene conditions during milking, collection and transport. [15]

S. aureus may access bulk milk either by direct excretion from the udder with clinical and subclinical staphylo-coccal mastitis, or by fecal contamination. Interchange of staphylococcal strains and poor microbiological quality of raw milk may be attributed to skin particles in the environment and poor sanitary practice [8]

Chemical composition of buffalo milk provides perfect opportunities for the development of local milk industry and providing nutrient element needs of humans. In addition, the presence of pathogens, indicators and index microorganisms in raw milk and products made of inadequately heat-treated milk could pose a threat for public health [12]. Livestock enterprises making milk production are composed of family enterprises in villages and towns with large numbers. Small sized production units have difficulty in obtaining inputs and services like adequate shelter, feed, technical information, veterinary services for buffalo and cow dairy production. It is possible to precisely organize hygienic and technological stages from production to consumption of milk and milk products only when all potentials are combined. In this regard, the principal that quality milk comes from healthy udder, healthy animal and clean environment gains great importance.

The important rule in food processing is the good quality of raw material. A good quality of end product cannot be obtained from a raw material with poor hygienic quality. Spoilage process of milk starts with milking. Considering the unhealthy conditions in milk production and other contamination sources in milk processes, milk can be a con-veyor of pathogens threatening public health. Microorganisms cause rapid souring, spoilage and undesired color, taste and bitterness in milk and thus resulting in poor quality. It is reported that many epidemic disorders of milk origin are caused by dirty hands of workers in milk production, dirty tools and equipment, insects and dirty water sources. Provision of microbiological quality parameters of raw milk and milk products plays an important role in quality control. It is necessary to minimize technological and economic losses in milk processing and obtain a longer shelf life.

REFERENCES

1. Ali AA, Irshad N, Razaz SA, Manahil AA (2010). Microbiological safety of raw milk in Khartoum state, Sudan: 1-Khartoum and Omdurman cities. Pak. J. Nutri., 9(5): 426-429.
2. Amarjit SN, Toshihiko N. (2003). Role of buffalo in the socioeconomic development of rural Asia: Current status and future prospectus. Anim. Sci. J., (74): 443–445.
3. Ariota B, Campanile G, Potena A, Napolano R, Gasparrini B, Neglia GL, DiPalo R (2007). Ca and P in buffalo milk: curd yield and milk clotting parametters. Ital. Anim. Sci., 6(6): 497-499.

4 Bilal MQ, Suleman M , Raziq A (2006). Buffalo: Black Gold of Pakistan. Livestock Res. Rural Development, 18(9): 128.

5 Chatterjee SN, Bhattacharjee I, Chatterjee SK, Chandra G (2006). Microbiological examination of milk in Tarakeswar, India with special reference to coliforms. Afr. J. Biotechnol., 5: 1383-1385.

6 Coroian A, Coroion CO, Vodnar DC, Trif M (2010). Study on the main microbiological traits in Romanian buffalo milk. Bioflux, 2(2): 92-98.

7 Fernando RS, Spahr SL, Jaster EH (1983). Comparacion of electrical conductivity of milk with other indirecty methods for detection of subclinical mastitis. J. Dairy Sci., 68: 449-456.

8 Kanwal R, Ahmed T, Mirza B (2004). Comparative analsis of quality of milk collected from Buffalo, Cow, Goat and Sheep of Rawalpindi/Islamabad region in Pakistan. Asian J. Plant Sci., 3(3): 300-305.

9 Kilic A, Kilic S (1994). Feeding and milk. Bilgehan Press, Izmir, 1994.

10 Najdenova N, Dimitrov T (2003). Technological qualities of buffalo milk from the Bulgarian Murrah breed for production of Bulgarian yoghurt. J. Anim. Sci., 40(5): 33–35.

11 Ng-Kwai-Hang, KF, Hayes JF, Moxley JE, Monardes HG (1984). Variability of test-day milk production and composition and relation of somatic cell counts with yield and compositional changes of bovine milk. J. Dairy. Sci., 67: 361-366.

12 Parekh TS, Subhash R (2008). Molecular and bacteriological examination of milk from different milch animals with special reference to coliforms. Current Research in Bacteriology, 1: 56-63

13 Sarfarz A, Gaucher I, Rousseau F, Beaucher E, Piot M, Grongnet JF, Gaucheron F (2008). Effect of acidification on physico-chemical charecteristics of buffalo milk: A comparison with cow's milk. Food Chem., 106 (1): 11-17.

14 Waldner DN, Stokes SR, Jordan ER, Looper ML (2002). Managing milk composition: Normal sources of variation.

15 Wells JG, Shipman LD, Greene KD, Sowers EG, Green JH, Cameron DN, Downes FP, Martin ML, Griffin PM, Ostroff SM (1991). Isolation of *Escherichia coli* serotype O157:H7 and other Shiga-like-toxin-producing *E. coli* from dairy cattle. J. Clin. Microbiol., (29): 985–989.

16 Yöney Z (1974). Milk Chemistry. Ankara Univ. Agric. Fac. Publication, No. 530, Textbook: 135, Ankara.