

# Comparative Study of Branded and Unbranded Milk Collected from Different Areas of Kathmandu Valley



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## Abstract

Milk is known as the most complete food for health and promotion of growth as it provides all needed body building proteins, bone-forming minerals and health-giving vitamins. The study aimed at comparing the microbial safety and physicochemical safety of branded and unbranded milk sold in Kathmandu valley. A descriptive study was conducted at Microbiology Laboratory of St. Xavier's college from August 2018 to February 2019, where total of 30 milk samples including 15 branded and 15 unbranded milk from various local dairies and retailer shops of Kathmandu, Lalitpur and Bhaktapur were collected.

In this study, physicochemical parameters include pH, Solid Not Fat (SNF), Fat%, Acidity and Adulteration (Starch, Neutralizer, Glucose, Common salt, Alcohol). The microbial parameters include Total Plate Count and Coliform Count. In case of detection of adulterants, out of 15 branded milk samples, starch was found in 6 milk samples, neutralizer was observed in 4 milk samples. Similarly, in case of 15 unbranded milk samples, starch was found in 3 milk samples, alcohol was observed in 4 milk samples. The average fat percentage of branded milk samples was 2.73% and unbranded milk samples was 3.38%. The percentage of acidity in branded and unbranded milk samples were found to be 0.174% and 0.19% respectively.

The range of Total plate Count (TPC) branded milk samples were  $9.1 \times 10^1$  -  $1.1 \times 10^5$  whereas the range of Total Coliform Count (TCC) was 0.00- $4.5 \times 10^3$ . Similarly, the range of TPC of unbranded milk samples was  $1.8 \times 10^4$  -  $2.1 \times 10^5$  whereas the range of TCC was 0.00- $1.4 \times 10^4$ . The comparative analysis of both milk samples showed the acceptable result of branded milk samples than unbranded milk samples. This comparative study on milk will help consumers to make best choice and it will also help the dairy industries and authorities to take corrective actions for the better quality of milk production.

**Keywords:** Milk; Branded; Unbranded; pH; Acidity; Solid Not Fat; Adulterants

**Abbreviations:** TPC: Total plate Count; TCC: Total Coliform Count; SNF: Solid Not Fat; VRBA: Violet Red Bile Agar

## Introduction

Milk is an inevitable liquid food product produced by the mammary glands of mammals. It is the primary and main source of nutrition for young mammals before they can digest other types of food. Early-lactation milk contains colostrums, which carries the mother's antibodies to newly born babies and can reduce the risk of many diseases [1]. The major components of milk are water, fat, lactose, casein, whey proteins, and minerals in amounts varying with the milk of various species of animals [2,3].

Milk is composed of approximately 3.7% fat, 3.2% protein, 4.8% lactose, 0.19% non-protein nitrogen and 0.7% ash. However, the exact composition of milk varies with individual animals, breed, season, diet and phase of lactation [4]. Milk contains more than 60 different types of known enzymes and during the heat treatment the most of those enzymes will destroy and become inactive. The heat processing at high level of temperatures causes not only digestion enzymes denaturation (amylases, proteinases,

phosphatases, lipases) but also digestion those enzymes having antioxidant and antimicrobial characteristics [5].

Milk is virtually a sterile when discharged into the alveoli of the udder. However, after this stage of milk production, microbial contamination can generally occur from the exterior of the udder and from the surface of milk handling and storage equipment [6]. Milk can get easily contaminated with bacterial contamination and hence it is easily perishable. Common bacteria to be isolated from milk include *Staphylococcus* spp, *Listeria* species, *Salmonella* species, *E. coli*, *Campylobacter* species, *Mycobacterium* species, *Brucella* species, *Coxiella burnetii*, *Yersinia* species, *Pseudomonas aeruginosa* and *Corynebacterium ulcerans*. Others are *Proteus* species, *Leptospira* species, *Clostridium* species, *Streptococcus* species, *Klebsiella* species, *Enterobacter* species and *Bacillus* species [7].

There are many milks borne human diseases which get spread through consumption of contaminated milk and pass from human intestine to blood and from blood to different organs. Few examples of the known milk -borne diseases are bovine tuberculosis, brucellosis, anthrax, listeriosis, salmonellosis, leptospirosis, Q fever, campylobacteriosis and *E. coli* O157:H7 [8]. Spoilage is a word, to describe the deterioration of a food color, texture or flavor which is unsuitable for human consumption. Microbial spoilage involves the degradation of carbohydrates, protein and fats by microbes or their enzymes. Some species of *Clostridium*, *Cornebacterium*, *Bacillus*, *Lactobacillus*, *Arthrobacter*, *Micrococcus*, *Mycobacterium* and *Streptococcus* are heat stable and grow at cooler temperatures which cause spoilage problems [9].

The word "Adulteration" is a legal term meaning that a food produced fails to meet the standards given by the concerned authority. Adulteration of food is the mixing or substitution of impure weaker, cheaper or inferior, possible toxic substances to the food stuff which affect the quality of food. Adulteration food is one that contains low quality substances or the substances which are fatal to our health [10]. Adulterants are mainly added to increase the shelf life of milk. Normally, the adulteration in food is done either for financial gain. To maintain its composition, materials like starch, flour, urea, cane sugar and vegetable oil are added as adulterants.

It also includes preservatives like formalin, hydrogen peroxide, boric acid and various antibiotics. Urea is added for whitening of milk and only few grams of urea are enough to bring milk in its original state. Hydrogen peroxide is used as preservative usually in summer season when environmental temperature is very high [11]. This study aims to analyze and compare the quality of branded and unbranded milk collected from different area of Kathmandu valley by assessing their physiological properties (acidity, fat, pH, and SNF content), detecting common adulterants (starch, glucose, neutralizers, salt, and alcohol), and evaluating their microbiological quality through total bacterial and coliform

counts using the pour plate method.

### Materials and Methods

This was a prospective experimental study conducted in Kathmandu Valley, Nepal. In this study Branded and Unbranded milk samples were purchased from the market and small local dairies of Kathmandu, Lalitpur and Bhaktapur respectively. The laboratory work was carried out in microbiology laboratory of St. Xavier's college. Altogether 30 milk samples (15 branded and 15 unbranded) milk samples were collected from different local dairies and milk vendors, and then triplicate testing was done. All the samples were collected using sample collector ice box at 4°C and transported to the laboratory within 2 hours of collection. In the laboratory, milk was immediately stored in refrigerator at 4°C until the tests were carried out. The study was conducted from the month of August 2018 to February 2019. Physicochemical Analysis of Branded and Unbranded milk.

### Physical Analysis

pH of the samples was determined by using pH paper. The pH paper was dipped in the milk samples, and the color change was observed. The pH was determined by comparing with the standard pH color range.

### Chemical Analysis

#### Fat Percentage Determination

A butyrometer was taken in which 10ml of 80% conc. sulphuric acid and 10.75 ml of milk sample was taken carefully in order not to mix the milk with the sulphuric acid. One ml of amyl alcohol was added and butyrometer was closed with a stopper without mixing the liquids. The content was mixed in an upright position. Centrifugation was then carried out in Gerber's centrifuge for 5 min after which the butyrometer was kept in a warm water bath at 65°C for 5 min. Finally, the scale reading was noted.

#### Acidity Percentage Determination

The burette was filled with 0.1N NaOH solution. Ten ml of milk sample was transferred with the pipette in a conical flask. Then 3-4 drops of Phenolphthalein indicator solution were added and mixed well. The initial reading of the 0.1N NaOH in burette at the lowest point of meniscus was taken. From the burette 0.1N NaOH solution was added drop by drop and continuous mixing until a faint pink color appeared.

#### Solid Not Fat (SNF)

500 ml of milk sample was taken and poured into a measuring cylinder. The temperature of the milk was measured. At an appropriate temperature the lactometer was dipped slowly into the milk sample. The last lactometer reading ( $^{\circ}$  L) just above the surface of the milk was read and recorded. Note: Calibration temperature of the lactometer may be 20°C. Calculate the correction factor for the temperature. For each one  $^{\circ}$ C above the calibration temperature, add 0.2 $^{\circ}$ L; for each  $^{\circ}$ C below calibration

temperature subtract  $0.2 \text{ }^{\circ}\text{L}$  from the recorded lactometer reading.

$$\text{SNF} = 0.25\text{L} + 0.2\text{F} + 0.14$$

### Milk Adulterants Detection Tests

#### Starch

Three ml of milk sample was taken in a test tube. After boiling it thoroughly it was cooled to temperature. One drop of 1% iodine solution was added. The appearance of blue indicated the presence of starch.

#### Glucose

One ml of milk sample was taken in a test tube and 1 ml of modified Barford's reagent was added to it. The mixture was heated in a water bath for 3 min. One ml of phosphomolybdic acid reagent was added to the turbid solution. The immediate change in deep blue color indicated the presence glucose.

#### Preparation for Alcohol Solution

Two ml of milk sample was taken in a test tube. Two ml of 68% of ethanol solution was added. Appearance of clotting or precipitation indicated failure to the quality of the test sample.

#### Common salt

Two ml of milk sample was taken in a test tube, 0.5 ml of 5% potassium chromate and 2ml of 0.1N silver nitrate were added. The appearance of yellow precipitate indicated the presence of sodium chloride.

#### Neutralizer

Firstly, 2 ml of milk samples were taken in a test tube. Then, 0.5 ml rosalic acid solution was added to it. Then, the change in color i.e, pink color was observed.

### Microbial Analysis

#### Total Plate Count

Initially, serial dilution was performed by adding 1 ml of milk samples in 9 ml of distilled water and that will be diluted till 105. Then from dilution 101 to 105, 1 ml of diluents was poured into sterile petriplates. After that 20 ml of PCA was added and mixed gently. The plates were incubated at  $37^{\circ}\text{C}$  for 48 hours. At last, the colonies appeared in the plates were counted and average CFU/ml was calculated.

#### Coliform Count

At first serial dilution was performed by adding 1 ml of milk samples in 9 ml of distilled water and that will be diluted till 104. Then from dilution 101 to 104, 1 ml of dilution was poured in sterile Petriplates. After that 15 ml of Violet Red Bile Agar (VRBA) was added and mixed slowly and allowed to solidify. After that, again 5 ml of VRBA was poured to create anaerobic environment. Then, plates were incubated at  $37^{\circ}\text{C}$  for 24 hours. Finally, the

colonies were counted, and average CFU/ml was calculated.

### Result

#### PH and SNF of Branded Milk

The overall pH values of the 15 branded milk samples ranged from 6-7. Out of this, 20% of the milk samples had pH levels less than 6.3. While 66.7% of milk samples had pH in the range of 6.3-6.8 and 13.3% of the milk samples had pH level more than 6.8. According to DFTQC the standard of pH is 6.3- 6.8. The overall pH values of the 15 unbranded milk samples ranged from 6-7. Out of this, 33.3% of the milk samples had pH level ranged from 6.3-6.8. While 46.7% of the milk samples had pH level less than 6.3 and 20% of the milk samples had pH level more than 6.8. Statistically, there was no difference in the pH of branded milk and unbranded milk samples ( $p > 0.05$ ) (Table 1).

The Solid Not Fat (SNF) test of branded milk showed the range from 6.375- 8.115, the average SNF being 7.01%. Out of 15 branded milk samples SNF of 80% of milk samples were below the standard range 8 whereas 20% of milk samples were found within the standard range. The Solid Not Fat (SNF) test of unbranded milk showed the range from 5.71- 7.63, the average SNF being 6.69%. In case of unbranded milk samples, SNF of all milk samples were found below the standard range. Statistical analysis of branded milk and unbranded milk samples showed that there was no difference in SNF at 5% level of significance ( $p > 0.05$ ).

The percentage of fat in branded milk sample ranges from 1.9-3%, the average fat percent being 2.73%. Out of 15 branded milk samples, fat of 80% milk samples was below the standard range 3 whereas 20% milk samples were in standard range. The percentage of fat in unbranded milk sample ranges from 1.9-4.6%, the average fat percent being 3.38%. Out of 15 unbranded milk samples, fat of 26.6% milk samples was below the standard range and 60% were found above the standard range whereas 13.3% were in range. Statistically, unbranded milk samples have more fat than branded milk samples ( $p < 0.05$ ) (Table 2).

#### Acidity in Branded and Unbranded Milk

The percentage of acidity in branded milk sample ranges from 0.16-0.18%, the average acidity percent being 0.174%. Out of 15 branded milk samples, 60% milk samples were in standard range 0.18%, whereas 33.33% milk samples were below the standard range. The percentage of acidity in unbranded milk sample ranges from 0.15-0.22%, the average acidity percent being 0.19%. Out of 15 unbranded milk samples, 26.6% milk samples were in standard range and 46.66% milk samples were above the standard range, whereas 13.3% milk samples were below the standard range. The statistical analysis showed unbranded milk samples have more acidity than branded milk samples at 5% level of significance ( $p < 0.05$ ). Statistically, branded milk samples have similar acidity at 5% level of significance ( $p > 0.05$ ) while unbranded milk samples have diverse acidity ( $p < 0.05$ ) (Table 3).

**Table 1:** PH in branded and unbranded milk.

SN	Volume(ml)	Branded (pH)	Unbranded (pH)	P-Value
1	500	6.5	6	0.22
2	500	6.5	6	
3	500	6.5	6.5	
4	500	6.5	6.5	
5	500	6	6.5	
6	500	7	7	
7	500	6.5	6	
8	500	6.5	6	
9	500	6.5	6	
10	500	6.5	6.5	
11	500	6.5	7	
12	500	7	7	
13	500	6	6.5	
14	500	6.5	6	
15	500	6	6	

**Table 2:** SNF in branded and unbranded milk.

SN	Volume(ml)	Branded (SNF)	Unbranded (SNF)	P-Value
1	500	8.115	7.63	0.62
2	500	6.375	6.365	
3	500	6.5025	7.1	
4	500	6.7	6.905	
5	500	8.035	7.125	
6	500	8.1125	6.9025	
7	500	6.4825	6.9225	
8	500	7.0375	7.225	
9	500	6.6725	5.71	
10	500	7.23	6.4	
11	500	6.855	6.355	
12	500	6.7625	6.07	
13	500	6.835	6.1325	
14	500	6.675	6.54	
15	500	6.77	6.96	

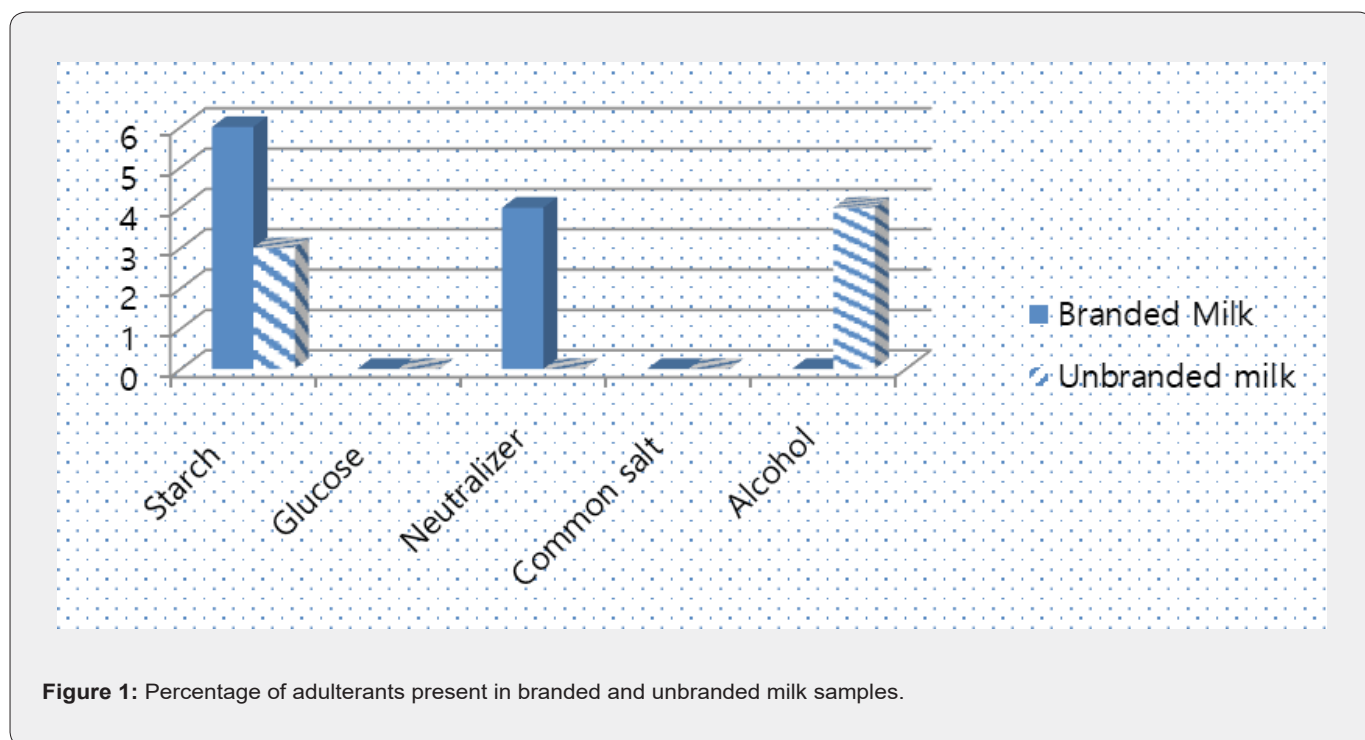
**Table 3:** Fat% in branded and unbranded milk.

SN	Volume(ml)	Branded (Fat%)	Unbranded (Fat %)	P-Value
1	500	3	2.7	0.62
2	500	2.8	3.3	
3	500	2.9	3.8	
4	500	2.8	3.2	
5	500	2.6	4.1	
6	500	3	1.9	
7	500	2.8	4	
8	500	1.9	4.5	

9	500	2.5	4.6	
10	500	2.7	2.8	
11	500	3	3	
12	500	2.7	3.4	
13	500	2.9	2.3	
14	500	2.6	3	
15	500	2.8	4.1	

**Table 4:** Acidity% in branded and unbranded milk.

SN	Volume (ml)	Branded (Acidity%)	Unbranded (Acidity%)	P-value
1	500	0.18	0.22	0.002
2	500	0.18	0.18	
3	500	0.17	0.18	
4	500	0.18	0.19	
5	500	0.18	0.2	
6	500	0.18	0.15	
7	500	0.18	0.16	
8	500	0.16	0.18	
9	500	0.18	0.2	
10	500	0.16	0.21	
11	500	0.17	0.19	
12	500	0.17	0.2	
13	500	0.16	0.21	
14	500	0.18	0.18	
15	500	0.18	0.2	



**Figure 1:** Percentage of adulterants present in branded and unbranded milk samples.

**Test of Adulterants of Branded and Unbranded Milk**

**Comparison Between Branded and Unbranded Milk's Adulteration**

In adulteration test of branded and unbranded milk, there was starch, neutralizer, and alcohol. In branded milk, 40% of starch 26.66% of soda was present whereas in unbranded milk, 26.66% of alcohol and starch were detected on average (Figure 1).

**Microbial Analysis of Branded and Unbranded Milk**

**Total Plate Count of Branded Milk**

The Total Plate Count of 15 different samples of branded milk ranged from  $9.1 \times 10^1$ – $1.1 \times 10^5$ cfu/ml. The statistical analysis of TPC showed that the branded milk samples have different microbial content a( $p < 0.05$ ).

**Coliform Count of Branded Milk**

The Coliform count of branded milk ranged from 0.00- $4.5 \times 10^3$ cfu/ml. The statistical analysis of Coliform count is diverse in branded milk samples ( $p < 0.05$ ).

**Total Plate Count of Unbranded Milk**

The Total Plate Count of 15 different samples of unbranded milk ranged from  $1.8 \times 10^4$  –  $2.1 \times 10^5$ cfu/ml.

The statistical analysis of TPC of unbranded milk samples showed difference in microbial content at 5% level of significance ( $p < 0.05$ ) (Table 4).

**Coliform Count of Unbranded Milk**

The Coliform count of unbranded milk ranged from 0.00- $1.4 \times 10^4$ cfu/m. Statistically, Coliform count are diverse in unbranded milk samples ( $p < 0.05$ ) (Table 5-8).

**Table 5:** Microbial analysis of TPC in branded milk.

S.N	Sample code	Total Plate count	chi2 (X2)
1	BM1	$9.1 \times 10^1$	745652.7
2	BM2	$1.1 \times 10^5$	
3	BM3	$6.1 \times 10^4$	
4	BM4	$3.1 \times 10^4$	
5	BM5	$1.03 \times 10^5$	
6	BM6	$2.8 \times 10^2$	
7	BM7	$1.9 \times 10^4$	
8	BM8	$1.4 \times 10^3$	
9	BM9	$1.06 \times 10^5$	
10	BM10	$3.2 \times 10^4$	
11	BM11	$1.09 \times 10^5$	
12	BM12	$1.4 \times 10^2$	
13	BM13	$3.8 \times 10^3$	
14	BM14	$4.9 \times 10^3$	
15	BM15	$3.8 \times 10^2$	

**Table 6:** Microbial analysis of Coliform count in branded milk.

S.N	Sample code	Coliform count	chi2 (X2)
1	BM1	Nil	32574.923
2	BM2	$6.9 \times 10^2$	
3	BM3	$1.7 \times 10^1$	
4	BM4	$1.02 \times 10^2$	
5	BM5	$2.8 \times 10^3$	
6	BM6	Nil	
7	BM7	$2.3 \times 10^2$	
8	BM8	Nil	
9	BM9	$4.5 \times 10^3$	
10	BM10	$2.3 \times 10^1$	
11	BM11	$1.6 \times 10^3$	
12	BM12	$1.8 \times 10^2$	

13	BM13	Nil	
14	BM14	8.6×10 <sup>2</sup>	
15	BM15	Nil	

**Table 7:** Microbial analysis of TPC in unbranded milk.

S.N	Sample code	Total Plate count	CHI2 (X2)
1	UBM1	2.03×10 <sup>5</sup>	271132.509
2	UBM2	1.4×10 <sup>5</sup>	
3	UBM3	1.8×10 <sup>5</sup>	
4	UBM4	7.1×10 <sup>4</sup>	
5	UBM5	1.6×10 <sup>5</sup>	
6	UBM6	1.25×10 <sup>5</sup>	
7	UBM7	2.1×10 <sup>5</sup>	
8	UBM8	2×10 <sup>5</sup>	
9	UBM9	2.02×10 <sup>5</sup>	
10	UBM10	1.1×10 <sup>5</sup>	
11	UBM11	1.09×10 <sup>5</sup>	
12	UBM12	1.8×10 <sup>4</sup>	
13	UBM13	1.7×10 <sup>5</sup>	
14	UBM14	2.4×10 <sup>4</sup>	
15	UBM15	1.8×10 <sup>5</sup>	

**Table 8:** Microbial analysis of Coliform in unbranded milk.

SN	Sample code	Coliform count	CHI2 (X2)
1	UBM1	3.02×10 <sup>3</sup>	47632.72
2	UBM2	6.8×10 <sup>3</sup>	
3	UBM3	1.4×10 <sup>4</sup>	
4	UBM4	Nil	
5	UBM5	1.8×10 <sup>3</sup>	
6	UBM6	8.7×10 <sup>3</sup>	
7	UBM7	5.9×10 <sup>3</sup>	
8	UBM8	9.1×10 <sup>3</sup>	
9	UBM9	7.2×10 <sup>3</sup>	
10	UBM10	1.006×10 <sup>4</sup>	
11	UBM11	1.5×10 <sup>4</sup>	
12	UBM12	Nil	
13	UBM13	1.02×10 <sup>4</sup>	
14	UBM14	1.1×10 <sup>4</sup>	
15	UBM15	1.3×10 <sup>3</sup>	

## Discussion

This research-based work was carried out to analyze the quality of branded milk and unbranded milk available in packaged and unpackaged form dairy shops situated at various places inside the Kathmandu valley. The milk samples were tested in the laboratory of St. Xavier’s college for six months

study period of August 2018 to February 2019. The milk samples were collected and taken to the laboratory for processing within 120 minutes. The purpose for storing milk samples in ice box was to stop rapid growth of bacteria. The fresh milk samples left under unrefrigerated condition for long period of time allow the multiplication of bacteria to very high concentrations [12]. As

this research is based on comparative study, the pH range of raw milk and pasteurized milk were compared. The maximum value of pH of branded milk was 6.5 and the maximum value of pH of unbranded milk was 6. The pH ranges of both milk samples were 6-7.

The findings of both samples were compared with the standard value given by DFTQC, Nepal. The findings were within the range specified by DFTQC (6.3-6.8). The results of unbranded milk are like the result of the study by [13] (6.4-6.8). The findings of branded milk were like the result of the study carried out by [14] (6.02-6.46). Out of 15 unbranded milk samples, pH of 46.67 percent of milk samples was below the standard range whereas pH of 20 percent of milk samples was above the range. In case of 15 branded milk samples, pH of 13.33 percent of milk samples were above the standard range. There is slight deviation in the pH range of both branded and unbranded milk samples. This might be due to not storing the milk samples in proper temperature as pH of milk samples are directly proportional to the milk temperature milk samples stored in, which means when temperature is increased pH of milk samples decreases and vice versa [15].

The unpaired t-test analysis of data of branded milk and unbranded milk samples showed similar pH at 5% level of significance ( $p < 0.05$ ). The result of this test confirms both milk samples are of good quality. The next parameter to test the quality of branded and unbranded milk was fat percentage. In this research, fat percentage of branded and unbranded milk was compared. The fat percentage of branded milk ranged from 1.9-3%. Out of 15 milk samples, only 3 samples were met the standard of DFTQC, Nepal. The results derived in this study are approximately similar to the study carried out by [16] where the average fat percentage was 3 while the result [17] was 4.4%.

Similarly, the fat percentage of unbranded milk ranged from 1.9-4.6%. Out of 15 milk samples, 4 did not meet the standard range, 9 milk samples are above the standard range and only 3 milk samples are as per the standard range. The findings of this study are like the findings of the study carried out by [18] where the range was 3.5 - 4.2%. This study shows that the unbranded milk had better fat % than branded milk samples. This might be due to lack of addition of water at producers' level and addition of water in milk at collector's level and buying and selling site as well. The fat% of milk also depends upon the animal's breed, stage of lactation, feeding regime, individual conditions of animal and parity [19].

The unpaired t-test analysis of data of unbranded milk samples showed higher fat percentage than branded milk at 5% level of significance ( $p < 0.05$ ). The result of this test confirms both milk samples are of good quality as well as few of bad quality. The Solid Not Fat (SNF) test of the milk was the other parameters to test the quality of milk. According to DFTQC, the standard value

of SNF of milk is 8. In this study, 15 branded milk samples were tested. The range of SNF branded milk samples was 6.375-8.115, the average SNF being 7.01%. Out of 15 milk samples tested only three samples had met the standard. Similarly, 15 unbranded milk samples were 5.71-7.63. The average value of SNF being 6.69%. No milk samples had met the standard.

The results obtained in this study are quite similar to the study by [16] (6.9 -7.3). The SNF test shows that many milk samples had low SNF. Comparatively, branded milk showed better quality than unbranded milk.

The reasons behind low SNF are animals feeding, genetics, season of the year, stage of lactation and disease [16]. The unpaired t-test analysis of data of branded milk and unbranded milk samples showed same SNF of both milk samples at 5% level of significance ( $p > 0.05$ ). The result of this test confirms both milk samples are of good quality. The acidity test is other parameters to compare the quality of branded and unbranded milk samples.

The acidity range in branded milk was found to be 0.16-0.18%. Whereas the acidity range in unbranded milk was found to 0.15-0.22%. Out of 15 branded milk samples, 9 samples only meet the standard of DFTQC whereas only 4 unbranded milk samples meet the acidity standard given by DFTQC. The results of unbranded milk obtained in this research seem to be like the results of the study done by [20] (0.15 -0.2) percentage. Similarly, the result of branded milk in this research was like the study done by [21] (0.16 -0.2%).

The high percentage of acid present in milk samples indicates that the milk is contaminated due to high enzyme and microbial activity in the milk samples, which may result due to lack of inappropriate cold chain in the distribution channels, and the long time taken from the source of market [22]. The result of acidity of both milk samples was compared and branded milk samples showed that there was less presence of acidity in it than in unbranded milk samples. The unpaired t-test analysis of data of branded milk and unbranded milk samples showed different acidity of both milk samples at 5% level of significance ( $p < 0.05$ ).

The result of this test confirms both milk samples are of good quality as well as few of bad quality. The chi-square analysis of acidity and branded milk samples showed similar acidity at 5% level of significance ( $p > 0.05$ ), whereas the analysis of acidity and unbranded milk samples showed diverse acidity at 5% level of significance ( $p < 0.05$ ). Adulteration testing is another step to determine the quality of milk. To analyze the quality of milk, 15 branded and 15 unbranded milk samples were tested. The presence of different adulterants like Starch, Glucose, Neutralizer, Common salt and alcohol were observed. The tests showed there was presence of Starch in 6 branded milk samples Neutralizer in 4 branded milk samples. No presence of Glucose, Common salt and Alcohol in any branded milk samples.

The result of starch in this study on branded milk is slightly above the study carried out by [23] (Out of 10 samples, 40% starch) and the outcome of neutralizer test of branded milk is quite near to the study done by [24] (8 neutralizer). Similarly, the presence of starch was seen in 3 unbranded milk samples, Alcohol in 4 unbranded milk samples and absence of Glucose, Common salt and Neutralizer. The result of unbranded milk samples test is slight above the study carried out by [25] (starch in 2 samples), which showed the presence of Starch and [26] which found the presence of Alcohol in the milk samples.

The Starch is present due to addition of it in milk samples to increase the SNF content of milk. The Starch can be in the form of wheat flour, arrowroot, rice flour etc [11]. The presence of Neutralizer in milk is usually done to minimize the pH and acidity values of badly preserved milk. The presence of Neutralizer in milk may cause disruption of hormones which are important for development and reproduction [11]. Neutralizers are usually added to increase the shelf life of milk. They can be added in the form of caustic soda, sodium bicarbonates, and sodium carbonates etc [27]. In this research the analysis of Total plate Count (TPC) and the Total coliform Count was carried out to analyze the microbial load in branded and unbranded milk samples.

As per the standard of DFTQC the acceptable level of TVC (Total Viable Count) for raw milk is  $<1.5 \times 10^5$  CfU/ml and pasteurized milk is  $<5 \times 10^4$  CfU/ml and TCC must be nil in both cases. The TPC of 15 different branded milk samples ranged from  $9.1 \times 10^1$  -  $1.1 \times 10^5$  and only 10 milk samples met the standard. Similarly, the coliform count of branded milk ranged from  $0.00$  -  $4.5 \times 10^3$  where only 5 milk samples met the standard. The outcome of the study of TPC and TCC of branded milk were contradictory to the outcome of the study done by [28] ( $4.4 \times 10^1$  -  $4.43 \times 10^5$  CFU/ml and  $4.5 \times 10^1$  -  $2.3 \times 10^4$  CFU/ml). In case of unbranded milk, TPC of 15 different unbranded milk samples ranged from  $1.8 \times 10^4$  -  $2.1 \times 10^5$  and only 7 milk samples met the standard.

Similarly, the Coliform count of unbranded milk ranged from  $0.00$  -  $1.4 \times 10^4$  where only 2 milk samples met the standard. The results obtained in case of unbranded milk are lower than the previous study carried out by [29] ( $7.05 \times 10^4$  -  $3.5 \times 10^5$  CFU/ml and  $15$  -  $7.05 \times 10^4$  CFU/ml). After the analysis of both cases, it can be concluded that branded milk showed high quality than unbranded milk though the test showed the presence of coliform in branded and unbranded milk as well which indicates errors in production hygiene, unsatisfactory sanitation, milking environment condition and unsuitable storage temperature [29]. The chi-square analysis of data of branded milk and unbranded milk samples showed diverse TPC and Coliform at 5% level of significance ( $p < 0.05$ ). This shows both milk samples are of low quality.

### Conclusion

The quality analysis of branded and unbranded milk was performed. Different physicochemical and microbial assessments

of 15 branded and 15 unbranded milk samples were carried out. The data and results were compared with the standard values provided by DFTQC, Nepal, to analyze the quality of milk samples. In case of the adulteration test of milk, starch and neutralized was present in branded milk samples whereas starch and alcohol were detected in unbranded milk samples which makes it unfit for consumption.

Furthermore, branded milk samples have met the TPC standard range whereas slight deviation was seen in case of unbranded milk and coliform count was detected high in both milk samples. About statistical analysis of different parameters, similar pH and SNF was observed in both milk samples. There was presence of Fat and avidity more in unbranded milk samples whereas branded milk samples have met the standard range given by DFTQC. It can be stated that branded milk is of higher quality than that of unbranded milk. Though branded milk samples showed high quality, it needs some serious attention by the regulatory body as some deviation than DFTQC standard was observed.

### Conflict of Interest

The author(s) declared that there are no potential conflicts of interest.

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