Microbiological Quality of Raw Cow Milk across the Milk Supply Chain in Eastern Ethiopia

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Abstract: The risk of milk contamination with spoilage and pathogenic microorganisms is high for milk produced in developing countries like Ethiopia especially in lowland region as their milk production practices is traditional type which lack appropriate hygienic control. To protect the raw cow milk from spoilage loss and consumers from milk born public health risk, the availability of documented information on the microbiological quality of raw milk across the milk supply chain is critically important as such information may be important for different organization to undertake relevant development intervention on hygienic practices essential for safe milk production and handling**.** This study was, therefore, conducted to determine the microbiological quality of informally marketed raw cow milk across the milk supply chain in eastern Ethiopia. A total of 360 pooled raw cow milk samples (each with a volume of 450 mL) were collected from udders and milk handling equipment of producers in rural areas of Babile district; from the equipment of collectors/transporters in Harar and Dire Dawa towns as well as from the equipment of vendors and consumer at Babile, Harar and Dire Dawa towns during February 2014 to January 2015. The milk samples were subjected to laboratory analyses to evaluate total aerobic mesophilic bacteria count (TAMBC), total coliform count (TCC), yeast count (YC) and mold count (MC) in the laboratory to determine the microbiological quality of the milk. Mean TAMBC, TCC, YC and MC for raw cow milk samples collected directly from the udders were 6.02±0.14, 4.23±0.12, 2.57±0.10 and 2.67±0.10 $\log_{10}$ cfu mL^{-1}, respectively. The values for the samples collected from the equipment of producers upon arrival at their selling points were 7.17±0.14, 5.86±0.12, 3.46±0.10 and 3.70±0.10 $\log_{10}$ cfu mL^{-1} for TAMBC, TCC, YC and MC, respectively. Mean TAMBC, TCC, YC and MC for samples collected from the equipment of collectors/transporters were 7.96±0.10, 6.49±0.07, 3.99±0.07 and 4.37±0.07 $\log_{10}$ cfu mL^{-1}, respectively. The microbial counts for samples collected from the equipment of vendors were 8.78±0.08, 7.32±0.07, 4.98±0.06 and 5.04±0.07 $\log_{10}$ cfu mL^{-1} for TAMBC, TCC, YC and MC, respectively. The values for samples collected from equipment of consumers were 8.82±0.08, 7.37±0.07, 5.10±0.06 and 5.11±0.07 $\log_{10}$ cfu mL^{-1} for TAMBC, TCC, YC and MC, respectively. It could be concluded that raw cow milk samples collected from all towns and milk source were severely contaminated with aerobic mesophilic and coliform bacteria, yeast and molds, with loads exceeding the respective acceptable limits.

Keywords: Dairy production system; Herd size; Microbiological quality; Milk supply chain, Raw cow milk

1. Introduction

Milk is universally recognized as a complete diet due to its composition of essential nutrients (Pandey and Voskuil, 2011; Melese and Tesfaye, 2015). Cow milk has long been considered a highly nutritious and valuable human food and is consumed by millions daily in a variety of different products in the world (Ali, 2010). It is also an economically important farm commodity and investment option for smallholder farmers in developing countries such as Ethiopia (Haile *et al.,* 2012).

However, milk serves as an excellent growth medium for a wide range of microorganisms because of its high water content, nearly neutral pH, and a variety of available essential nutrients (Ruegg, 2003). Although fresh milk, which is aseptically drawn from clean and healthy cow normally contains low (less than 1000 cfu mL^{-1} of milk) microbial count, it picks many microbes from the time it leaves the teat of the cow until it is used for consumption (Torkar and Teger, 2008). The load of microbes in milk is an indicator of the manner of milk handling from time of milking to consumption (Torkar and Teger, 2008). The microbial load and types found in the milk are influenced by factors such as health and hygiene of milking animal as well as its environment, cleanness of storage and transport

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equipment, milk holding temperature during storage and transport, cleanness of water used for hygienic practices, health and personal hygiene of milkers and milk handlers (Bytyqi *et al.,* 2011).

Mishandling and disregard of hygienic measures by milk handling personnel may enable undesirable microbes to come into contact with milk and in some cases to survive and multiply in sufficient numbers and make the milk unsafe for both direct consumption and further processing (Chatterjee *et al.,* 2006). A high microbial count in milk is an indication of poor hygiene, and reduces the nutritional quality of milk, causes unpleasant effect on the taste and also affect the physical quality of milk. Moreover, it reduces the market value of milk causing income losses to producers and traders. Furthermore, high microbial count in milk threatens the health of consumers due to toxic metabolites produced by different organisms growing in it (Karmen and Slavica, 2008).

The potential risk of milk contamination by spoilage and pathogenic microbes is high for milk produced in traditional system and marketed through the informal channels (Coorevits *et al*., 2008). This is because, in such systems it is a common practice to handle, transport and vend milk in inappropriate equipments and temperature as there is little or no quality control measures. Such practices are very common in developing countries such as Ethiopia and pose a threat to public health as chances of consuming unsafe milk is very high (Kurwijila *et al.,* 2006; Yilma and Faye, 2006).

In Ethiopia, the demand for cow milk is rapidly increasing because of population growth, increase in per capita income and urbanization. Therefore, provision of milk and milk products of good microbiological quality is desirable from consumers' health point of view (Zelalem, 2012). However, there is no well documented information available on the microbiological quality of raw cow milk at different milk source (cow's udder, producers, collectors/tranpsorters, vendors and consumers) across the milk supply chain in the study area. On the other hand, almost all milk produced from pastoral and agro-pastoral areas is supplied to urban centers through informal milk marketing channels. It is, therefore, important to document the microbiological quality of raw milk across the milk supply chain to ensure safety and suitability of raw milk for intended use. The information will be relevant to dairy value chain actors and service providers for introduction of pertinent interventions through the participation of the community. The objective of this study was, therefore, to determine the microbiological quality of informally marketed raw cow milk across the milk supply chain in eastern Ethiopia.

2. Materials and Methods

2.1. Description of the Study Area

The study was conducted in eastern Ethiopia, specifically in Babile district, and Harar & Dire Dawa towns. Babile district is the site of milk production whereas Harar and Dire Dawa towns are the sites of milk distribution (by vendors via informal marketing channel) and consumption.

Babile district is located at $9^{0}08'$ N latitude and $42^{0}21'$E longitude at the distance of about 557 km east of Addis Ababa. The altitude of the district ranges from 950 to 2000 meter above sea level. It has mean annual minimum and maximum temperatures of 18 and 28 ^{o}C, respectively. The mean annual rainfall and humidity of the area ranges from 700 to 900 mm and 33 to 38%, respectively (CSA, 2008). The two prevailing agricultural production systems in the district are pastoral and agro-pastoral production system (CSA 2008). Cattle are the most dominant in population size (56,355 heads) followed by goat (23,020), sheep (12,216) and camel (9,704) (BDLDHA, 2015). The district produced about 12,000 and 6,745 liters of raw cow milk during the wet and dry seasons, respectively (BDLDHA, 2015). Of the total cow milk produced daily in the district, about 50% was used for sale (Bedilu *et al.,* 2015). The total human population of the district is estimated at 115,229, out of which about 21.5% live in Babile town (CSA, 2013).

Harar town is located between 9.11^{0}-9.24^{0} N latitude and 42.03-42.16^{0} E longitude at the distance of about 526 km east of Addis Ababa and 31 km west of Babile district (Abdulwasi, 2009). The altitude of the town is 1850 above sea level and its mean annual rainfall and humidity is 596 mm and 60.3%, respectively (Dinkineh *et al.,* 2014). The town has mean annual maximum and minimum temperatures of 25 and 10^{o}C, respectively (Abdulwasi, 2009). The total human population of the town was estimated at 125,000 with a growth rate of 2.6% (CSA, 2013).

Dire Dawa town is located in the eastern part of Ethiopia at 9°36' N latitude and 41°52' E longitude (Belachew and Zeleke, 2015). The town is situated at the distance of 515 km east of Addis Ababa and 86 km west of Babile district. The altitude of the town is about 1200 meters above sea level. The mean annual rainfall and humidity are 594 mm and 41.82%, respectively. The town has mean annual maximum and minimum temperatures of 31.4 and 18.41 ^{0}C, respectively (Arabali and Amare, 2015). The total human population of the town was estimated at 288,000 with a growth rate of 2.5% (CSA, 2013).

According to Diro *et al.* (2009) and Shimelis *et al.* (2015), the study area has three seasons based on the distribution of rainfall. These are the long rainy season extending from June to September, the short rainy season from February to May, and the dry season that

extends from October to January (NMSA, 1996; Cheung *et al.,* 2008).

2.2. Study Design

A longitudinal study was conducted from February 2014 to January 2015 to determine the microbiological quality of informally marketed raw cow milk across the milk supply chain in the study area. Pooled raw cow milk samples were taken repeatedly from each milk source (from udder, milk handling equipment of producers, collectors/transporters, vendors and consumers) at every month during the study period. Thus, milk samples were collected throughout the year in order to assess the effect of season. The laboratory analysis was done in dairy technology laboratory, Haramaya University, Ethiopia.

2.3. Sampling Targets Across the Supply Chain

Babile district was stratified into pastoral and agro-pastoral production systems. Each production system was further stratified into peasant associations (PAs; the lowest administration unit in Ethiopia). Thus, a total of ten PAs (5 from pastoral and 5 from agro-pastoral systems) with high cow milk production potentail were purposively selected for the study. Each PA was then further stratified into small (1-3 cows) and medium (4-10 cows) herd size groups based on the number of milking cows they possessed (Dayanandan, 2011). Large herd size groups with more than 10 cows were not encountered in the study area. Milk producers households were selected from each herd size group randomly.

Unlike the milk producers in the Babile district, milk traders (collectors/transporters, and vendors), and consumers in Harar and Dire Dawa towns, were selected following a snowball sampling technique. Moreover, the same technique was used to select milk vendors and consumers in Babile town.

2.4. Milk Sampling Across the Supply Chain

A total of 360 raw cow milk (each with 450 mL) samples were collected from the udder, milk handling equipment of producers, collectors/transporters, vendors, and consumers following the sampling stratification described above. The numbers of pooled raw milk samples taken from the cow's udder directly and from the equipment of producers at their selling points (nearby the road side) were 36 each. Similarly, 72 pooled raw milk samples were collected from milk handling equipment of collectors/transporters in Harar (n = 36) and Dire Dawa (n = 36) towns. Moreover, 108 pooled raw milk samples were collected from milk handling equipment of vendors in Babile (n = 36), Harar (n = 36) and Dire Dawa (n = 36) towns. The total number of pooled raw milk samples taken from milk handling equipment of consumers in Babile (n = 36), Harar (n = 36), and Dire Dawa (n = 36) towns was 108. Raw milk samples were collected aseptically using sterile screw-capped sampling bottles. The bottles were then securely capped, labeled with markers and kept in an ice box filled with ice packs and brought to Haramaya University dairy technology laboratory within 3-4 hours of collection. The analysis was carried out within a period of 24 hours after collection. The samples were collected once every month over a period of 12 months (February 2014 to January 2015). In each month, three raw milk samples were collected from the equipment of milk producers, collectors/transporters, vendors, consumers, and from the udders of lactating cows.

2.5. Media Preparation for Microbial Quality Analysis

The total aerobic mesophilic bacterial counts (TAMBC) and total coliform count (TCC) were determined using sterile standard plate count agar and violet red bile agar (VRBA), respectively. Yeast count (YC) and mold count (MC) were also done using sterile Potato Dextrose Agar (PDA) whose pH was adjusted to 3.5 by adding 10 mL of sterile 10% lactic acid to a 1 L volume of the medium. All media except VRBA were sterilized by autoclaving at 121ºC for 15 minutes, while VRBA was sterilized by boiling for two minutes. After sterilization, all media were cooled to 45-47ºC in a water bath before use. The preparation of media was generally done according to the instructions given by the respective manufacturers. Peptone water that was autoclaved at 121ºC for 15 minutes and cooled to 30ºC was used for serial dilution of the milk samples to determine TAMBC, TCC, YC and MC. Each analysis was made in a duplicate.

2.6. Determination Total Aerobic Mesophilic Bacterial Count (TAMBC)

TAMBC was determined using standard plate count agar. One mL of raw milk sample was added into a sterile test tube containing 9 mL of sterile peptone water. After thoroughly mixing, the suspension was serially diluted up to 10^{-11} and duplicate samples from the appropriate dilution (1 mL) was pour-plated using a 15-20 mL of cooled but still molten standard plate count agar (Oxoid, UK) solution and mixed thoroughly. The resulting plates were allowed to solidify and then incubated at 32 ºC for 48 hours (Murphy, 1996). The plates with colonies ranging from 30-300 colony forming units (cfu) mL^{-1} were selected for determination of TAMBC (Kiiyukia, 2003). TAMBC was determined as the total number of cfu per milliliter of milk sample which was calculated using the formula provided by IDF (2004).

$$N = \frac{\sum C}{[(1xn1) + (0.1xn2)]d} \quad (1)$$

Where: N is the number of cfu per milliliter of milk sample; ΣC is the sum of colonies on all plates counted; n_1 is the number of plates in the first dilution counted; n_2 is the number of plates in

the second dilution counted; and d is the dilution from which the first counts were obtained.

2.7. Determination Total Coliform Count (TCC)

TCC was determined using sterile violet red bile agar (VRBA). One mL of raw milk sample was added into a sterile test tube containing 9 mL of sterile peptone water. After thoroughly mixing, the suspension was serially diluted up to 10^{-9} and duplicate samples (1mL) were pour-plated using a sterile 15-20 mL VRBA (Oxoid, UK). After thoroughly mixing, the resulting plates were allowed to solidify and then incubated at 32 ºC for 24 hours (Murphy, 1996). After incubation, typical dark red or purplish-red colonies appearing on the plates were counted as coliforms. For confirmatory test, five to ten typical colonies from each plate were transferred into tubes containing 2% Brilliant Green Lactose Bile Broth (Oxoid, UK) and incubated at 37 °C for 48 hours. Growth and gas production within incubation period was considered as sufficient evidence for the presence of coliforms (Richardson, 1985). Plates with 15 to 150 cfu mL^{-1} were used (Kiiyukia, 2003) for determining total coliform counts using the formula provided by IDF (2004).

2.8. Determination Yeast and Mold Counts

Yeast count (YC) and mold count (MC) were determined using sterile Potato Dextrose Agar (PDA). One mL of raw milk sample was added into a sterile test tube containing 9 mL of sterile peptone water. After thoroughly mixing, the suspension was serially diluted up to 10^{-7} and duplicate samples of 0.1 mL were spread-plated on pre-dried surfaces of media containing PDA (Oxoid, UK). The plates were then incubated at 25°C for 5 days (Andrews, 1992; Roberts and Greenwood, 2003). Creamy to white/gray colonies were counted as yeasts whereas, filamentous (fuzzy) colonies of various colors (yellow, green, light brown) were counted as molds (Yousef and Carlstrom, 2003). When difficulties were faced to differentiate some colonies whether they are yeast or mold, a microscopic examination using the oil immersion objective was carried out to identify whether the cells in the colonies were unicellular or multi-cellular. Plates with 10 - 150 colonies were used for determining yeast and mold counts (IDF, 2004) using the formula provided by IDF (2004).

2.9. Data Analysis

Data on microbial counts, which were expresed as colony forming unit (cfu) per mL were transformed into logarithmic scales (log_{10}) and analyzed using the General Linear Model (GLM) procedure of SAS (SAS, 2008). Mean comparison was made using Tukey's adjustment. The following models were used for the analysis:

Model 1: Effect of production system, herd size group and season on microbial counts of raw cow milk collected directly from the udder of cow at Babile district

$$Y_{ijkl}= \mu+P_i+H_j+S_k+H_jxS_k+E_{ijkl} \quad (2)$$

Where: Y_{ijkl}=Total aerobic mesophilic bacterial count, total coliform count, yeast count and mold count; μ=Population mean (Overall mean); P_i= the effect of i^{th} production system (i=1, 2); H_j= the effect of j^{th} herd size group (j = 1, 2); S_k = the effect of k^{th} seasons (k = 1…3); H_jxS_k = interaction of herd size group with seasons; E_{ijkl} = random error

Since interaction between production system and season (P_i x S_k), production system and herd size group (P_ixH), and production system with herd size group and season (P_i x H_j x S) had no significant effect on microbial load of raw milk samples collected directly from the udder of milking cow, they were excluded from the model.

Model 2: Effect of production system, herd size group and season on microbial counts of raw cow milk collected from milk handling equipment of producers at Babile district

$$Y_{ijkl}= \mu+P_i+H_j+S_k + P_ixS_k+H_jxS_k+ E_{ijkl} \quad (3)$$

Where: Y_{ijkl} = Total aerobic mesophilic bacterial count, total coliform count, yeast count and mold count; μ = Population mean (Overall mean); P_i = the effect of i^{th} production system (i=1, 2); H_j= the effect of j^{th} herd size group (j = 1, 2); S_k = the effect of k^{th} seasons (k=1…3); H_jxS_k = interaction between herd size group and seasons; P_ixS_k= interaction between production system and seasons; E_{ijkl} = random error

Since the interaction between production system and herd size group (P_ixH_j) as well as interaction among production system, herd size group and season (P_i x H_j x S_k) had no significant effect on microbial load of raw milk samples collected from handling equipment of producers, they were excluded from the model.

Model 3: Effect of sources of milk and seasons on microbial count of raw cow milk samples collected in the study area

$$Y_{ijk}= \mu+M_i+S_j+M_ixS_j+ E_{ijk} \quad (4)$$

Where: Y_{ijk} =Total aerobic mesophilic bacterial count, total coliform count, yeast count and mold count; μ=Population mean (Overall mean); M_i=the effect of i^{th} milk source (I = 1…5); S_j = the effect of j^{th} season (j = 1…3); M_ixS_j = interaction between milk source and season; E_{ijk} = Random error

Model 4: Effect of locations and seasons on microbial count of raw cow milk collected from the equipment of

traders (collectors/transporters, and vendors) and consumers in the study areas

$$Y_{ijk} = \mu + L_i + S_j + L_i x S_j + E_{ijk} \quad (5)$$

Where: Y_{ijk} *= Total aerobic mesophilic bacterial count, total coliform count, yeast count and mold count;* μ *= Population mean (Overall mean);* L_i *= the effect of* i^{th} *location (I = 1…3);* S_j *= the effect of* j^{th} *season (j = 1…3);* $L_i x S_j$ *= interaction between location with season;* E_{ijk} *= Random error.*

3. Results and Discussion

3.1. Microbial Count of Raw Cow Milk Samples Collected from the Udder

Mean total aerobic mesophilic bacterial count (TAMBC) and total coliform count (TCC) of raw milk samples were influenced ($P < 0.05$) by herd size group and season differently (Table 1). Thus, milk samples collected from medium size herd during the short rainy season had significantly higher ($P < 0.05$) mean TAMBC than samples collected from small size herd during the dry season. Moreover, among medium size herds, milk samples collected during the short and long rainy seasons had significantly higher TCC than samples collected during the dry season (Table 1). Such differences might be due to the variation in health and hygiene of milking cows between herd size groups as well as between rainy and dry seasons.

Raw cow milk samples collected from medium-sized herds had significantly ($P < 0.05$) higher mean TAMBC and TCC than milk samples collected from small-sized herds (Table 1). This might be due to higher accumulation of effluents in night enclosure areas for cows from medium-sized herds than for small-sized herds. The mean TAMBC, TCC, yeast count (YC) and mold count (MC) of raw cow milk were not influenced ($P > 0.05$) by production system (Table 1).

The mean value of TAMBC and TCC for raw milk samples collected during the short and long rainy seasons were significantly ($P < 0.01$) higher than that for the dry season (Table 1). This might be due to over contamination of teats and udders of milking cows during the rainy seasons compared to the dry season, which might result in milk contamination with bacteria during milking. Moreover, it might be due to higher prevalence of mastitis during the rainy seasons than the dry season (Fox *et al.,* 1995) as the level of udder and teat contamination with mud while lying in night enclosure area is high during the former than the latter season. Moreover, the warm temperature and high humidity during the rainy season favor the growth of organisms, which might result in increased prevalence of mastitis.

The overall mean TAMBC for raw milk samples collected from the udder of milking cows in Babile district was 6.02 ± 0.14 $\log_{10}$ cfu mL^{-1} (Table 1). This was higher than 4.57 ± 0.21 $\log_{10}$ cfu mL^{-1} reported for raw milk samples collected from the udder of milking cows in Hawassa city in Ethiopia (Haile *et al.,* 2012). However, it is lower than 7.18 ± 0.1 $\log_{10}$ cfu mL^{-1} reported for raw milk samples collected from the udder of milking cows in Borana pastoral community in Ethiopia (Tollossa *et al.,* 2012). This difference might be due to the variation in health/hygiene of the milking cows and their environments as well as health care management practices performed by milk producers.

The overall mean TCC of raw milk samples collected from the udder of milking cows in the district was 4.23 ± 0.12 $\log_{10}$ cfu mL^{-1} (Table 1). The finding is in agreement with 4.00 $\log_{10}$ cfu mL^{-1} reported for milk samples collected from the udder of milking cows in Debre Zeit area in Ethiopia (Alehegne, 2004). However, it was lower than 6.88 ± 0.04 $\log_{10}$ cfu mL^{-1} reported for raw milk samples collected directly from the udder of milking cows in Borana pastoral community, Ethiopia (Tollossa *et al.,* 2012). The difference could be attributed to variation in cleanliness of night enclosure area and hygiene of milking cows (e.g. level of soiling of teats, udders, flanks and tails of the milking cows while lying in night enclosure area).

For raw milk samples collected from the udder of milking cows, the overall mean MC was 2.67 ± 0.10 $\log_{10}$ cfu mL^{-1} (Table 1), which was relatively comparable to 3.03 ± 0.26 $\log_{10}$ cfu mL^{-1} reported for raw cow milk samples collected from the udder of milking cows in Hawassa city in Ethiopia (Haile *et al.,* 2012). The overall mean yeast count (YC) for raw milk samples collected from the udder of milking cows was 2.57 ± 0.10 $\log_{10}$ cfu mL^{-1} (Table 1), which was relatively comparable with 2.87 $\log_{10}$ cfu mL^{-1} reported for udder milk samples in the Republic of Benin (Souaibou *et al.,* 2012).

Table 1. Least square mean (± S.E.) microbial counts (log_{10} cfu mL^{-1}) and temperature of raw cow milk samples collected directly from the udder across the different production systems, herd size groups and seasons in Babile district.

	Microbial count				Milk temperature
Variables	TAMBC	TCC	YC	MC	Temperature (°C)
Production system (n=36)	ns	ns	Ns	ns	ns
Pastoral	5.95(0.14)	4.26(0.12)	2.57(0.11)	2.72(0.12)	33.42(0.28)
Agro-pastoral	6.09(0.14)	4.21(0.12)	2.55(0.11)	2.62(0.12)	33.00(0.28)
Herd size group (n=36)	**	*	Ns	ns	ns
Small	5.73(0.14)^{b}	3.99(0.13)b	2.43 (0.11)	2.57(0.21)	33.194(0.28)
Medium	6.31(0.14)^{a}	4.47(0.13)a	2.70 (0.11)	2.78(0.21)	33.22(0.28)
Season (n=36)	**	**	ns	ns	ns
Short rainy season	6.33(0.16)^{a}	4.37 (0.14)^{a}	2.75(0.14)	2.70(0.15)	33.58(0.35)
Long rainy season	6.12(0.16)^{a}	4.57 (0.14)^{a}	2.59(0.14)	2.78(0.15)	33.08(0.35)
Dry season	5.60(0.16)^{b}	3.76(0.14)^{b}	2.36(0.14)	2.54(0.15)	32.96(0.35)
Herd size group X Season	*	*	ns	ns	ns
Medium X Short rainy season	6.66(0.22)^{a}	4.73(0.22)^{a}	2.92(0.20)	2.66(0.21)	33.33(0.49)
Medium X Long rainy season	6.33(0.22)^{ab}	4.84(0.22)^{a}	2.70(0.20)	2.98(0.21)	33.42(0.49)
Medium X Dry season	5.93(0.22)^{ab}	3.85(0.22)^{b}	2.48(0.20)	2.68(0.21)	32.92(0.49)
Small X Short rainy season	6.00(0.22)^{ab}	4.01 (0.22)^{ab}	2.57(0.20)	2.74(0.21)	33.83(0.49)
Small X Long rainy season	5.79(0.22)^{ab}	4.30 (0.22)^{ab}	2.49(0.20)	2.59(0.21)	33.00(0.49)
Small X Dry season	5.39(0.22)^{b}	3.67(0.22)^{b}	2.23(0.20)	2.40(0.21)	32.75(0.49)
Overall mean	6.02(0.14)	4.23(0.12)	2.57(0.10)	2.67(0.10)	33.21(0.24)

*Note: TAMBC = Total aerobic mesophilic bacterial count; TCC = Total coliform count; YC = Yeast count; MC = Mold count; Significance: *=p < 0.05, **= p < 0.01, ns = not significant; S.E. = Standard error; Column mean values with different superscript letters (a, b, ab) are significantly different.*

According to Marshall (1992) and Heeschen (1997), the acceptable limit of TAMBC and TCC for raw milk is 5.30-5.60 and 2.18 log_{10} cfu mL^{-1}, respectively, which was lower than the present findings. Moreover, the mean values of YC and MC for udder milk samples exceeded the upper acceptable limit (2.1 and 1.7 log_{10} cfu mL^{-1} for YC and MC, respectively) (Torkar and Vengust, 2008). This might be due to poor herd/farm hygiene and health care management practices performed by smallholder milk producers.

3.2. Microbial Count of Raw Milk Samples Collected from the Equipments of Producers

The microbial count of raw milk samples collected from the equipment of producers in the Babile district was influenced by the interaction between the production system and season ($P < 0.05$), and herd size group and season ($P < 0.05$) (Table 2). In most cases, milk samples collected during the short rainy season had significantly higher mean TAMBC, TCC, YC and MC than milk samples collected during the dry season within a given production system and herd size group.

Raw milk samples collected from pastoral production system had significantly ($P < 0.05$) higher mean TAMBC, TCC, YC and MC than raw milk samples collected from agro-pastoral production system (Table 2). This might be due to higher milk temperature for the former than the latter production system. Moreover, the quality of water used for hygienic practices in agro-pastoral production system is better than the quality of water used in pastoral production system (Tadele *et al.,* 2016).

The overall mean TAMBC for raw milk samples collected from the equipment of producers upon arrival at their selling points (nearby road side) in Babile district was 7.17±0.14 log_{10} cfu mL^{-1} (Table 2). The finding is in agreement with that of Alehegne (2004), who reported 7.20±0.13 log_{10} cfu mL^{-1} for raw cow milk collected from the equipment of producers in Debre Zeit, Ethiopia. However, it was lower than 7.96±0.07 log_{10} cfu mL^{-1} as reported for Yabello district of Ethiopia by Gurmessa (2015). On the contrary, it is higher than 6.36 log_{10} cfu mL^{-1} as reported by Asrat (2010) for raw cow milk samples collected from the equipment of producers in Wolayta zone in southern Ethiopia. The difference could be attributed to differences in the levels of hygiene of milking equipment, animal, milker wash water and the environment. Moreover, it might be due to the differences in milk holding time and temperature during storage and transportation.

Table 2. Least square mean (± S.E.) microbial load of raw cow milk samples (log_{10} cfu mL^{-1}) collected from milk handling equipment of producers across the different production systems, herd size groups, and seasons in Babile district.

Variables	Microbial count				Milk temperature
	TAMBC	TCC	YC	MC	temperature °(C)
Production system (n=36)	*	**	*	*	*
Pastoral	7.38(0.14)^{a}	6.14(0.12)^{a}	3.66(0.10)^{a}	3.89(0.11)^{a}	27.39(0.20)^{a}
Agro-pastoral	6.95(0.14)^{b}	5.57(0.12)^{b}	3.25(0.10)^{b}	3.51(0.11)^{b}	26.75(0.20)^{b}
Herd size group (n=36)	**	**	ns	ns	ns
Small	6.86(0.14)^{b}	5.58(0.12)^{b}	3.52(0.10)^{a}	3.81(0.11)^{a}	26.92(0.20)a
Medium	7.48(0.14)^{a}	6.13(0.12)^{a}	3.38(0.10)^{a}	3.58(0.11)^{a}	27.19(0.20)a
Season (n=36)	**	***	***	***	**
Short rainy season	7.54(0.17)^{a}	6.09(0.14)^{a}	3.94(0.12)^{a}	4.17(0.13)^{a}	27.96(0.25)^{a}
Long rainy season	7.48(0.17)^{a}	6.31(0.14)^{a}	3.50(0.12)^{a}	3.74(0.13)^{a}	27.26(0.25)^{a}
Dry season	6.48(0.17)^{b}	5.18(0.14)^{b}	2.94(0.12)^{b}	3.18(0.13)^{b}	26.00(0.25)^{b}
Production system X Season	*	*	*	*	**
Pastoral X Short rainy season	7.73(0.23)^{a}	6.37(0.20)^{a}	4.25(0.17)^{a}	4.41(0.19)^{a}	28.42(0.35)^{a}
Pastoral X long rainy season	7.67(0.23)^{a}	6.53(0.20)^{a}	3.62(0.17)^{ab}	3.84(0.19)^{ab}	27.32(0.35)^{ab}
Pastoral X dry season	6.74(0.23)^{ab}	5.53(0.20)^{b}	3.10(0.17)^{bc}	3.40(0.19)^{bc}	26.42(0.35)^{bc}
Agro-pastoral X short rainy season	7.36(0.23)^{a}	5.80(0.20)^{ab}	3.62(0.17)^{ab}	3.85(0.19)^{ab}	27.50(0.35)^{ab}
Agro-pastoral X long rainy season	7.28(0.23)^{ab}	6.10(0.20)^{ab}	3.48(0.17)^{b}	3.71(0.19)^{b}	27.10(0.35)^{b}
Agro-pastoral X dry season	6.22(0.23)^{b}	4.82(0.20)^{c}	2.68(0.17)^{c}	2.96(0.19)^{c}	25.58(0.35)^{c}
Herd size group X Season	**	*	*	*	*
Medium X short rainy season	8.03(0.21)^{a}	6.32(0.20)^{a}	3.90(0.17)^{a}	4.15(0.19)^{a}	27.83(0.35)^{a}
Medium X long rainy season	7.69(0.21)^{ab}	6.54(0.20)^{a}	3.30(0.17)^{ab}	3.77(0.19)^{a}	27.68(0.35)^{a}
Medium X dry season	6.71(0.21)^{bc}	5.54(0.20)^{b}	2.95(0.17)^{b}	3.52(0.19)^{ab}	26.07(0.35)^{b}
Small X short rainy season	7.05(0.21)^{b}	5.85(0.20)^{ab}	3.96(0.17)^{a}	4.20(0.19)^{a}	28.08(0.35)^{a}
Small X long rainy season	7.26(0.21)^{b}	6.09(0.20)^{ab}	3.70(0.17)^{a}	3.70(0.19)^{a}	26.83(0.35)^{ab}
Small X dry season	6.26(0.21)^{c}	4.81(0.20)^{c}	2.92(0.17)^{b}	2.84(0.19)^{b}	25.83(0.35)^{b}
Overall mean	7.17(0.14)	5.86(0.12)	3.46(0.10)	3.70(0.10)	27.06(0.21)

*Note: TAMBC=Total aerobic mesophilic bacterial count; TCC = Total coliform count; YC = Yeast count; MC = Mold count; Significance: *p < 0.05, ** p < 0.01,*** p < 0.001, ns = not significant; S.E.= Standard error*

The overall mean TCC for raw milk samples collected from the equipment of producers was 5.86 ± 0.12 log_{10} cfu mL^{-1} (Table 2), which is relatively comparable with 6.19 ± 0.03 log_{10} cfu mL^{-1} as reported by Gurmessa (2015) for raw cow milk samples collected from the equipment of producers in Yabello district in Ethiopia. However, it is higher than 4.84 log_{10} cfu mL^{-1} as reported by Derese (2008) for raw cow milk samples collected from a milk shed in Bahir Dar in Ethiopia and 4.03±0.09 log_{10} cfu mL^{-1} as reported by Abebe *et al.* (2012) for equipment samples in Ezha district of the Gurage zone in Southern Ethiopia.

The overall mean YC and MC of the present study were 3.46±0.10 and 3.70±0.10 log_{10} cfu mL^{-1}, respectively (Table 2). The result is lower than 4.9±0.6 and 4.61±0.5 log_{10} cfu mL^{-1} for yeast and mold, respectively as reported by Teshome and Ketema (2014) for raw cow milk sample collected from the equipment of producers in Kersa district in Jimma Zone of southwestern Ethiopia. This might be due to differences in hygienic practices during production and postharvest handling as well as due to the variations in milk holding temperatures and time during storage and transportation.

The mean TAMBC and TCC for raw cow milk samples collected from the equipment of producers at Babile district were much higher than the upper acceptable limit given by Marshall (1992) and Heeschen (1997). Moreover, the mean YC and MC of raw milk samples collected from the equipment of producers exceeded the upper acceptable limit as reported earlier (Torkar and Vengust, 2008). This indicates that milk is produced and handled under unhygienic condition at producer's level. Moreover, delayed milk transportation and lack of cooling facilities during milk storage and transportation, which are the common practice in the area might also be another important factor contributing to high microbial loads in the milk. Omore *et al.* (2005) also provided similar suggestion.

3.3. Microbial Count of Raw Milk Samples Collected across the Milk Sources and Seasons

The mean TAMBC, TCC, YC and MC of raw milk samples were significantly ($P < 0.05$) influenced by milk source and season interaction (Table 3). In most cases, raw milk samples collected during the short and long rainy seasons had significantly higher mean TAMBC, TCC, YC and MC than samples collected during the dry season within a given milk source. The mean TAMBC, TCC, YC and MC for raw milk samples collected from the udder were significantly ($P < 0.001$) lower than samples collected from the equipment of producers, which were significantly ($P < 0.001$) lower than samples collected from the equipment of collectors/transporters (Table 3). The mean TAMBC, TCC, YC and MC for raw milk samples collected from the equipment of vendors and consumers were significantly ($P < 0.001$) higher than milk samples collected from udder as well as from the equipment of producers and collectors/transporters (Table 3). This might be due to further contamination of the milk during storage and transportation. The possible sources of contamination might be the use of poorly cleaned equipment, lack of proper protection of milk equipment from risk factors after cleaning, the use of untreated water for hygienic practices, lack of cooling system, poor personal hygiene of milk handlers etc. Moreover, the longer storage time (about 4 hours) elapsed during milk vending by vendors might also contribute to such differences. However, there is there is no difference ($P>0.05$) in mean TAMBC, TCC, YC and MC between milk samples collected from the equipment of vendors and consumers (Table 3).

The mean TAMBC for raw cow milk samples collected from the equipment of collectors/ transporters upon arrival at selling points i.e., 7.96±0.10 log_{10} cfu mL^{-1} (Table 3) is in agreement with 8.0l log_{10} cfu mL^{-1} for raw cow milk samples collected from most dairy cooperatives operating in Ethiopia (Francesconi, 2006), but slightly lower than 8.26±0.31 log_{10} cfu mL^{-1} as reported by Mustefa (2012) for raw cow milk samples collected from the equipment of collectors in Sululta and Welmera districts, Ethiopia. The mean TAMBC for raw milk samples collected from the equipment of vendors in the study areas (8.78±0.08 log_{10} cfu mL^{-1}) (Table 3) was lower than 10.28±0.28 log_{10} cfu mL^{-1} as reported by Haile *et al.* (2012) for raw cow milk samples marketed in Hawassa city, Ethiopia but higher than 7.35±0.18 log_{10} cfu mL^{-1} as reported by Shunda *et al.* (2013) for raw cow milk collected from the equipment of vendors in Mekelle town, Ethiopia. The differences might be attributed to variation in the level of hygiene of cleaning water, milk handling equipment and milk marketing places used by milk vendors.

The mean value of TCC (6.49±0.07 log 10 cfu mL^{-1}) of raw cow milk samples collected from the equipment of collectors/ transporters upon arrival at their selling points (Table 3) is in agreement with 6.46±0.03 log_{10} cfu mL^{-1} reported for raw cow milk samples collected from market in Yabello district, Ethiopia (Gurmessa, 2015), but much higher than 4.11±0.01 log_{10} cfu mL^{-1} reported for raw cow milk samples collected from market at Khartoum in Sudan (Ali and Abdelgadir, 2011). The mean TCC for samples collected from the equipment of vendors was 7.32±0.07 log_{10} cfu mL^{-1} (Table 3). The finding was comparable with Alehegne (2004) who reported mean TCC of 7.32 log_{10} cfu mL^{-1} for raw cow milk samples collected from market upon arrival at processing plant in Addis Ababa, Ethiopia.

Table 3. Least square mean (± S.E.) microbial count (log_{10} cfu mL^{-1}) of raw cow milk samples collected across the milk sources and seasons in the study area.

Variables	Microbial count				Milk temperature
	TAMBC	TCC	YC	MC	temperature(°C)
Source of milk (n=360)	***	***	***	***	**
Udder	6.02(0.14)^{d}	4.23(0.12)^{d}	2.57(0.10)^{d}	2.67(0.10)^{d}	33.21(0.25)^{a}
Producers equipment	7.17(0.14)^{c}	5.86(0.12)^{c}	3.46(0.10)^{c}	3.70(0.10)^{c}	27.06(0.25)^{c}
Collectors equipment	7.96(0.10)^{b}	6.49(0.07)^{b}	3.99(0.07)^{b}	4.37(0.07)^{b}	28.12(0.18)^{b}
Vendors equipment	8.78(0.08)^{a}	7.32(0.07)^{a}	4.98(0.06)^{a}	5.04(0.07)^{a}	28.49(0.18)^{b}
Consumers equipment	8.82(0.08)^{a}	7.37(0.07)^{a}	5.10(0.06)^{a}	5.11(0.07)^{a}	28.37(0.18)^{b}
Season (n=360)	***	***	***	***	**
Short rainy season	8.18(0.09)^{a}	6.47(0.07)^{a}	4.31(0.07)^{a}	4.40(0.07)^{a}	29.85(0.17)^{a}
Long rainy season	8.03(0.09)^{a}	6.64(0.07)^{a}	4.12(0.07)^{a}	4.31(0.07)^{a}	29.09(0.17)^{b}
Dry season	7.04(0.09)^{b}	5.66(0.07)^{b}	3.63(0.07)^{b}	3.82(0.07)^{b}	28.21(0.17)^{c}
Milk Sources X season	*	*	*	*	*
Udder X short rainy season	6.33(0.17)^{d}	4.37(0.16)^{de}	2.75(0.15)^{de}	2.70(0.15)^{de}	33.58(0.41)^{a}
Udder X long rainy season	6.13(0.17)^{de}	4.57(0.16)^{d}	2.59(0.15)^{de}	2.78(0.15)^{de}	33.08(0.41)^{a}
Udder X dry season	5.59(0.17)^{e}	3.76(0.16)^{e}	2.36(0.15)^{e}	2.54(0.15)^{e}	32.96(0.41)^{a}
Producers equipment X short rainy season	7.54(0.24)^{c}	6.09(0.15)^{c}	3.94(0.14)^{bc}	4.17(0.14)^{bc}	27.96(0.41)^{cd}
Producers equipment X long rainy season	7.48(0.24)^{c}	6.31(0.15)^{bc}	3.50(0.14)^{c}	3.74(0.14)^{c}	27.26(0.41)^{d}
Producers equipment X dry season	6.48(0.17)^{d}	5.18(0.15)^{d}	2.94(0.14)^{d}	3.18(0.14)^{d}	26.00(0.41)^{e}
Collectors equipment X short rainy season	8.39(0.17)^{b}	6.68(0.14)^{bc}	4.20(0.13)^{b}	4.54(0.13)^{b}	28.90(0.35)^{bc}
Collectors equipment X long rainy season	8.32(0.17)^{b}	6.81(0.14)^{b}	4.10(0.13)^{bc}	4.51(0.13)^{b}	28.26(0.35)^{c}
Collectors equipment X dry season	7.18(0.17)^{c}	6.00(0.14)^{c}	3.68(0.13)^{c}	4.06(0.13)^{bc}	27.21(0.35)^{d}
Vendors equipment X short rainy season	9.31(0.14)^{a}	7.56(0.11)^{a}	5.25(0.11)^{a}	5.27(0.11)^{a}	29.04(0.28)^{b}
Vendors equipment X long rainy season	9.10(0.14)^{a}	7.76(0.11)^{a}	5.13(0.11)^{a}	5.26(0.11)^{a}	28.68(0.28)^{bc}
Vendors equipment X dry season	7.93(0.14)^{bc}	6.67(0.11)^{bc}	4.55(0.11)^{b}	4.63(0.11)^{b}	27.75(0.28)^{cd}
Consumers equipment X short rainy season	9.33(0.14)^{a}	7.61(0.11)^{a}	5.41(0.11)^{a}	5.33(0.11)^{a}	29.44(0.28)^{b}
Consumers equipment X long rainy season	9.19(0.14)^{a}	7.80(0.11)^{a}	5.26(0.11)^{a}	5.30(0.11)^{a}	28.56(0.28)^{bc}
Consumers equipment X dry season	7.95(0.14)^{bc}	6.71(0.11)^{bc}	4.62(0.11)^{b}	4.70(0.11)^{b}	27.13(0.28)^{d}

*Note: TAMBC = Total aerobic mesophilic bacterial count; TCC = Total coliform count; YC = Yeast count; MC = Mold count; Significance: *p < 0.05, ** p < 0.01, *** p < 0.001, ns = not significant; S.E.= Standard error*

The mean values of TAMBC and TCC for raw milk samples collected from the equipment of traders (collectors/transporter and vendors) and consumers in the study area far exceeded the upper acceptable limit as reported earlier (Marshall, 1992; Heeschen, 1997). Moreover, they were higher than the upper acceptable limit (6.30 and 4.70 $\log_{10}$ cfu mL^{-1} for TAMBC and TCC, respectively) given by East African Community Standard (EACS, 2007). This implies that the sanitary conditions in which milk is being handled in the study area is substandard and leads to high degree of microbial contamination and multiplication. Moreover, unavailability of cooling facilities during milk storage and transportation in the study area could also be another important factor contributing to high TAMBC and TCC in the milk (Omore *et al.,* 2005).

A high TAMBC in the milk may reduce shelf life stability and the nutritional quality of milk (Yousef and Carlstrom, 2003), and also threatens the health of consumers due to toxic metabolites produced by of different organisms growing in it (Karmen and Slavica, 2008). Moreover, the high number of coliform bacteria in raw milk has received considerable attention, partly due to their association with contamination of fecal origin and the consequent risk of more enteric pathogens being present. Apart from safety and public health concerns, occurrence of coliforms in raw milk in high numbers could result in spoilage that makes the milk unsafe for processing (Gamal *et al.,* 2015).

The mean of YC for raw milk samples collected from the equipment of collectors/transporters upon arrival at selling points were 3.99±0.07 $\log_{10}$ cfu mL^{-1} (Table 3), which is relatively similar to 4.15 $\log_{10}$ cfu mL^{-1} reported for raw cow milk marketed in Cairo town, Egypt (Gamal *et al*., 2015). The mean values of YC of raw milk samples collected from the equipment of vendors in the study area was 4.98±0.06 $\log_{10}$ cfu mL^{-1} (Table 3). The finding is far lower than 7.13±0.33 $\log_{10}$ cfu mL^{-1} reported for raw cow milk samples collected from distribution equipment upon arrival at selling points in Hawassa city, Ethiopia (Haile *et al.,* 2012). However, it is higher than 4.11±0.02 $\log_{10}$ cfu mL^{-1} as reported by Gemechu *et al.* (2014) for raw cow milk samples collected from small milk vending shops in Shashemene town, Ethiopia. Such variations might be due to the differences in hygienic handling practices performed by milk vendors and previous actors (like collectors/transporters and producers). Variations in milk holding temperature and time during storage and transportation at each milk source might also contribute to such differences.

The mean MC for raw cow milk samples collected from the equipment of collectors/transporters upon arrival at their selling points was 4.37±0.07 $\log_{10}$ cfu mL^{-1} (Table 3), which was comparable with 4.46±0.04 $\log_{10}$ cfu mL^{-1} as reported by Gurmessa (2015) for raw cow milk samples collected from market in Yabello district in Ethiopia). The mean value of MC (5.04±0.07 $\log_{10}$ cfu mL^{-1}) for raw milk samples collected from the equipment of vendors in the study areas was lower than 5.63±0.24 $\log_{10}$ cfu mL^{-1} reported for raw milk samples collected from El-Beida city in Egypt (El-Diasty and El-Kaseh, 2009). This variation might be due to differences in initial contamination during production, further contamination during post harvest handling as well as due to the differences in milk holding temperatures and time during storage and transportation

The mean YC and MC counts for samples collected from the equipment of traders and consumers in the study area were also much higher than the upper acceptable limit indicated above (Torkar and Vengust, 2008). The presence of high numbers of yeast and mold in milk indicates that the milk has been contaminated with soil, dusts, air and other contaminants due to poor hygienic practices during milk production and postharvest handling. In the study area, delivery of raw milk to the next actors in the study area is carried out at roadsides on the grounds, which are dusty and not protected from wind and road traffic (Tadele *et al.,* 2016). This might be a possible reason for the high yeast and mold counts observed in the present study. Moreover, the absence of milk cooling system at all critical points in the study area might contribute higher YC and MC than the upper acceptable limit indicted above.

High yeast and mold counts in foods including milk cause spoilage (Gamal *et al.,* 2015). Moreover, some molds, however, are public health concerns due to their ability to produce toxic substances (mycotoxins), which may not be easily destroyed during food processing or cooking (Wouters *et al.,* 2002). Therefore, training and guidance should be given to traders and consumers on general hygienic practices to be followed during milk postharvest handling to avoid/minimize the risk of milk contamination with yeasts and molds.

3.4. Microbial Count of Raw Milk Samples Collected from the Equipment of Traders and Consumers across Locations and Seasons

Mean TAMBC, TCC, YC and MC of raw milk samples were significantly ($P < 0.05$) influenced by location and season interaction (Table 4). In most cases, within a season, there is no difference ($P>0.05$) in mean TAMBC, TCC, YC and MC among milk samples collected from Babile, Harar and Dire Dawa towns. However, there is difference ($P<0.05$) in mean TAMBC, TCC, YC and MC among milk samples collected during the short rainy season, the long rainy season and the dry season within a location, except for MC in Harara and YC in Dire Dawa, in that, milk samples collected during short and long rainy seasons had significantly higher microbial load than that for dry season. This might be due to higher milk holding temperature and initial contamination during short and long rainy seasons than during dry season.

The milk samples collected in Dire Dawa and Babile towns had higher ($P<0.01$) mean TAMBC and TCC than milk samples collected in Harar town (Table 4). This might be mainly due to higher milk holding

temperature for Dire Dawa and Babile towns than for Harar town. Although milk holding temperature for Dire Dawa town is significantly higher (P<0.001) than that for Babile town, mean TAMBC and TCC were not significantly (P>0.05) different between milk samples collected in Dire Dawa and Babile towns (Table 4). This might be due to the higher level of contamination of milk with microorganisms for Babile town than for Dire Dawa town.

The mean YC and MC for milk samples collected from Babile town were higher (P<0.05) than for milk samples collected from Harar town (Table 4). This might be mainly due to the higher milk holding temperature of Babile town than Harar town. Moreover, the variation in the level of milk contamination with microorganisms during production, storage and transportation might contribute such differences.

Table 4. Least square mean (± S.E.) microbial count (log_{10} cfu mL^{-1}) of raw cow milk samples collected from the equipment of traders and consumers across location and season.

	Microbial count				Milk temperature
Variables	TAMBC	TCC	YC	MC	temperature(°C)
Location (n=288)	**	**	**	*	***
Babile	8.70(0.09)^{a}	7.26(0.09)^{a}	5.03(0.09)^{a}	5.03(0.08)^{a}	28.67(0.08)^{b}
Harar	8.35(0.11)^{b}	6.95(0.08)^{b}	4.52(0.08)^{b}	4.75(0.07)^{b}	26.40(0.08)^{c}
Dire Dawa	8.76(0.09)^{a}	7.23(0.08)^{a}	4.78(0.08)^{ab}	4.95(0.07)^{ab}	30.10(0.08)^{a}
Season(n=288)	***	***	***	***	***
Short rainy season	9.10(0.10)^{a}	7.37(0.08)^{a}	5.08(0.08)^{a}	5.13(0.07)^{a}	29.31(0.09)^{a}
Long rainy season	8.93(0.10)^{a}	7.54(0.08)^{a}	4.96(0.08)^{a}	5.09(0.07)^{a}	28.45(0.09)^{b}
Dry season	7.79(0.10)^{b}	6.54(0.08)^{b}	4.29(0.08)^{b}	4.51(0.07)^{b}	27.42(0.09)^{c}
Location X season	*	*	**	*	***
Babile X short rainy season	9.15(0.16)^{a}	7.49(0.16)^{a}	5.37(0.16)^{a}	5.32(0.15)^{a}	29.52(0.17)^{bc}
Babile X long rainy season	8.88(0.16)^{a}	7.59(0.16)^{a}	5.22(0.16)^{ab}	5.28(0.15)^{a}	28.81(0.17)^{c}
Babile X dry season	8.08(0.16)^{b}	6.72(0.16)^{b}	4.50(0.16)^{bc}	4.48(0.15)^{b}	27.69(0.17)^{d}
Harar X short rainy season	8.88(0.16)^{a}	7.18(0.13)^{ab}	4.85(0.13)^{ab}	4.96(0.12)^{ab}	27.01(0.14)^{de}
Harar X long rainy season	8.74(0.16)^{ab}	7.41(0.13)^{a}	4.70(0.13)^{b}	4.79(0.12)^{ab}	26.61(0.14)^{e}
Harar X dry season	7.41(0.16)^{c}	6.28(0.13)^{c}	4.00(0.13)^{c}	4.51(0.12)^{b}	25.57(0.14)^{f}
Dire Dawa X short rainy season	9.26(0.16)^{a}	7.45(0.13)^{a}	5.03(0.13)^{ab}	5.13(0.12)^{a}	31.39(0.14)^{a}
Dire Dawa X long rainy season	9.18(0.16)^{a}	7.63(0.13)^{a}	4.94(0.13)^{ab}	5.20(0.12)^{a}	29.92(0.14)^{b}
Dire Dawa X dry season	7.85(0.16)^{bc}	6.61(0.13)^{bc}	4.37(0.13)^{bc}	4.54(0.12)^{b}	28.99(0.14)^{c}

*Note: TAMBC = Total aerobic mesophilic bacterial count; TCC = Total coliform count; YC = Yeast count; MC = Mold count; Significance: *p < 0.05, ** p < 0.01,*** p < 0.001; S.E. = Standard error*

4. Conclusions

The results of the present study revealed that the raw milk samples collected from the equipment of producers in pastoral production system and medium size herds had significantly higher microbial counts than samples collected from the equipment of producers in agro-pastoral production systems and small-sized herds, respectively. Moreover, there were significant microbial quality differences among milk samples collected during the short rainy season, the long rainy season, and the dry seasons at all milk sources except for milk samples collected from the udders (for which the effect of season was not significant for yeast and mold counts). In most cases, milk samples obtained during the short and long rainy seasons had greater microbial loads than those obtained during the dry season. There were significant microbial quality differences among milk samples collected from udder, producers, collectors/transporters, vendors and consumers. Samples collected from the equipment of consumers and vendors had significantly higher microbial loads than samples collected from the equipment of collectors/transporters, which had greater microbial loads than those obtained from the equipment of producers; and also samples collected from the equipment of producers had significantly higher microbial counts than samples collected directly from the udder of milking cows. The mean values of TAMBC, TCC, YC and MC for raw milk samples collected from all milk sources (from udder, producers, collectors/transporters, vendors and consumers) in the study area exceeded the upper acceptable limit. This indicates that the sanitary conditions in which the milk is being produced and handled are substandard. In general, it could be concluded that, the microbial quality of raw cow milk produced and marketed in the study area was poor and a threat to human health. Therefore, improved milk hygienic practices across the milk supply chain is recommended to protect the milk from being unsafe for consumption as well as from being spoiled. Thus, awareness creation and capacity development of producers, collectors/transporters, vendors, consumers and other people involved in the milk supply chain should be done on hygienic practices of producing and handling raw milk. Moreover, introduction of pertinent interventions such as milk cooling facilities, organized and efficient milk storage and transportation systems at all across the supply

chain are highly important. Further investigation is recommended to identify pathogenic strains of mold (like aflatoxins) and coliform (like *Escherichia coli* O155:H7) that cause a potential health risk.

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