Full Length Research Paper

Microbial quality and chemical composition of raw milk in the Mid-Rift Valley of Ethiopia

Fikrineh Negash¹*, Estefanos Tadesse¹ and Tatek Woldu²

¹Adami Tulu Agricultural Research Center, P. O. Box 35, Ziway, Ethiopia. ²Jimma University, College of Agriculture and Veterinary Medicine, P. O. Box 307, Jimma, Ethiopia.

Accepted 15 June, 2012

The study was carried out to assess the microbial quality and chemical composition of raw milk collected from selected districts of East Shoa and West Arsi zones of Oromia Regional State in Mid-Rift Valley of Ethiopia. A total of 60 and 48 milk samples collected from individual households were analyzed for microbial quality and chemical composition of milk, respectively. Means of total bacterial counts were significantly different (P<0.05), but coliform counts were not significantly different (P>0.05) among districts. The overall mean total bacterial counts and coliform counts in the study area were 7.08 \pm 0.07 and 4.35 \pm 0.06 ml⁻¹, respectively. The result indicated that the milk samples collected from all districts were subjected to microbial contamination. Therefore, adequate sanitary measures should be taken at all stages from production to consumption. The overall mean for fat, protein and solid-not-fat percents were 5.48 \pm 0.19, 3.46 \pm 0.04 and 9.10 \pm 0.09, respectively and fat percent was significantly different (P<0.05) between breeds and among districts. The results of chemical composition were found to be adequate as compared to the standard level.

Key words: Mid Rift Valley, microbial quality, chemical composition, raw milk.

INTRODUCTION

Milk is one of the most precious natural materials and has been a basic component of human food for a long time (Edgar, 1998). It also provides an excellent medium for the growth of bacteria which may spoil the milk or render it unsafe for human consumption or unfit for further processing (O'Connor, 1994). The initial microbiological quality of milk can vary substantially based on factors such as the health of the animal, the sanitary condition of the milking environment and the milker (Biruk et al., 2009). Microbial contamination of milk can therefore originate from within the udder; the exterior of the teats and udder; and from the milk handling and storage equipment (Chambers, 2002; Biruk et al., 2009).

The safety of dairy products with respect to food-borne diseases is a great concern around the world. This is especially true in developing countries where production of milk and various dairy products take place under rather unsanitary conditions and poor production practices (Mogessie, 1990; Zelalem and Faye, 2006; Alganesh et al., 2007; Asaminew and Eyassu, 2011). A commonly used procedure to measure the sanitary quality of milk is to estimate its bacterial content. Different quality tests can be performed through quantifying bacterial population and other microorganisms present in milk and milk products, the major ones being aerobic mesophilic bacterial count (total bacterial count) and coliform count (Biruk et al., 2009).

On the other hand, the solid components of milk mainly fat and protein make milk an economically and nutritionally important asset (Zelalem et al., 2004). Similarly according to O'Connor (1994), the compositional quality may be ascertained by measuring the fat, protein and total solids content. Milk composition is affected by a number of factors including genetic and environmental factors (O'Connor, 1994). The factors responsible for variations in milk composition include breed and individuality of the cow, strain, interval between milking,

^{*}Corresponding author. E-mail: fiker_2mesi@yahoo.com. Tel: +251464419003. Fax: +251464419108.

stage of lactation, age and health of the cow, feeding regime and completeness of milking (O'Connor, 1994; McDonald et al., 1995; Zelalem et al., 2004). There is scanty information on the microbial properties and chemical composition of raw milk in the country (Ethiopia) in general and in the study area in particular. The objective of this study was to assess microbial quality and chemical composition of raw milk collected from East Shoa and West Arsi zones of Oromia Regional State, Central Rift Valley of Ethiopia.

MATERIALS AND METHODS

Study area

The study was conducted in three districts of East Shoa zone (Adama, Lume and Adami Tulu Jido Kombolcha) and two districts of West Arsi zone (Arsi Negele and Shashemene) of Oromia Regional State, Ethiopia. These five districts were purposively selected since they are potential areas for milk production. The districts are found in the Mid-Rift Valley of Ethiopia. The altitudes of these areas range from 1500 to 2300 m above sea level and have a semi-arid type of climate. The Mid-Rift Valley has an erratic, unreliable and low rainfall averaging between 500 and 900 mm per annum. The rainfall is bimodal with the short rains from February to May and long rains from June to September. The predominant production system in these areas is mixed crop-livestock farming. Cattle are the most important livestock species in the areas (Lemma, 2004).

Milk sampling procedure

Milk samples were collected in June 2010, from households producing milk from local and crossbred cows. A total of 60 samples (12 from each district) for microbiological analysis and 48 samples (12 from each district excluding Shashemene district) for chemical analysis were collected aseptically in sterile bottles, kept in an ice box (at < 5°C) and delivered to laboratory for analysis within 8 to 22 h of sampling.

Microbial analysis

A total of 60 milk samples were collected from individual households. The microbial analysis considered were total bacterial counts (TBC) and coliform counts (CC). For the determination of both counts, peptone water was sterilized by autoclaving at 121℃ for 15 min. Similarly the Standard Plate Count Agar (SPCA) (Oxoid) used for determination of total bacterial counts was sterilized by autoclaving at 121℃ for 15 min, while the Violet Red Bile Agar (VRBA) (Oxoid) used for determination of coliform counts was sterilized by boiling (Richardson, 1985). The media used were prepared according to the guidelines given by the manufacturers.

Total bacterial count

Briefly, 1 ml of milk sample was added into sterile test tube having 9 ml peptone water. Appropriate decimal dilution of milk samples were pour-plated on 15-20 ml SPCA solution and mixed thoroughly. The plated sample was allowed to solidify and then incubated at 30° C for 48 h. Colony counts were made using colony counter (Marth, 1978).

Coliform count

In brief, 1 ml of milk sample was added into sterile test tube having 9 ml peptone water. Appropriate decimal dilutions of milk samples were pour-plated on 15-20 ml Violet Red Bile Agar solution (VRBA). After thoroughly mixing, the plated sample was allowed to solidify and then incubated at 30°C for 24 h. Finally, colony counts were made using colony counter (Marth, 1978). Typical dark red colonies were considered as coliform colonies.

Chemical analysis

Fat, protein and solid-not-fat (SNF) percents were determined with calibrated milk analyzer (EKOMILK, Milkana KAM 98-2A). A total of 48 (26 from indigenous Arsi zebu and 22 from crossbred cows) samples were analyzed for chemical composition.

Statistical analysis

The number of microorganisms (colony forming unit) per milliliter of milk was calculated using the following mathematical formula (APHA, 1992):

 $N = \sum c / (1 \times n_1 + 0.1 \times n_2) d$

Where N is the number of colonies per milliliter of milk, Σc is the sum of colonies on plates counted, n_i is the number of plates on the lower dilution counted, n₂ is the number of plates in next higher dilution counted and d is the dilution from which the first counts are obtained. Total bacterial and coliform counts were log transformed before statistical analysis in order to make the frequency distribution more symmetrical. The transformed value and the chemical composition values were analyzed using the General Linear Model (GLM) for least square mean in Statistical Analysis Software (SAS) version 9.0 (SAS, 2002). A fixed effect model was used to estimate the effects of location on the microbial quality and chemical composition of milk. The Least Significant Difference (LSD) test was used to separate the means and differences were considered significant at P < 0.05.

RESULTS AND DISCUSSION

Microbial quality of milk

Results of coliform and total bacterial counts of the study area are presented in Table 1. According to the result obtained, total bacterial counts were significantly different (P<0.05) by locations and it was generally high as compared to the acceptable level of 1×10^5 bacteria per ml of raw milk (O'Connor, 1994). This implies that the sanitary conditions in which milk has been produced and handled are substandard, subjecting the product to microbial contamination and multiplication (Biruk et al., 2009). Total bacterial count in the present study (7.08 ± 0.07 ml⁻¹) is comparable with the result of Alganesh et al. (2007) which indicated 7.60 ml⁻¹ in milk sampled from a small scale producer in East Wollega. The present result is also comparable with the finding of Asaminew and Eyassu (2011) who reported 7.58 ml⁻¹ in cow milk sampled from around Bahir Dar and Mecha district. The result of current

Variables	Districts					Meen	C)/0/		<u></u>
	Shashemene	Arsi Negele	ATJK	Adama	Lume	Mean	60%	L3D	эL
N	12	12	12	12	12	60			
CC (ml ⁻¹)	4.41 ± 0.02^{a}	4.44 ± 0.13^{a}	4.16 ± 0.10^{a}	4.46 ± 0.12^{a}	4.26 ± 0.08^{a}	4.35 ± 0.06	10.61	0.38	NS
TBC (ml ⁻¹)	6.88 ± 0.20^{b}	7.18 ± 0.13^{ab}	7.16 ± 0.13^{ab}	6.80 ± 0.16^{b}	7.41 ± 0.12^{a}	7.08 ± 0.07	7.35	0.43	*

Table 1. Microbial quality of raw milk (LSM \pm SE) collected from five districts.

Means with different superscripts within the same raw are significantly (P<0.05) different. N = Number of observation; ATJK = Adami Tulu Jido Kombolcha; CC = coliform counts; TBC = total bacterial counts; CV = coefficient of variation; LSD = least significant difference; SL = significant level; NS = not significant; *= Significant (P<0.05).

Table 2. Chemical composition of raw milk (LSM ± SE) collected from four districts.

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variables	Arsi Negele	ATJK	Adama	Lume	wean	CV%	L9D	SL
Ν	12	12	12	12	48			
Fat (%)	6.21 ± 0.39^{a}	5.45 ± 0.33^{ab}	5.24 ± 0.34^{ab}	5.03 ± 0.39^{b}	5.48 ± 0.19	23.00	1.08	*
Protein (%)	3.50 ± 0.05^{a}	3.48 ± 0.05^{a}	3.43 ± 0.06^{a}	3.43 ± 0.11 ^a	3.46 ± 0.04	7.47	0.21	NS
Solid-not-fat (%)	9.22 ± 0.13 ^a	9.08 ± 0.10^{a}	9.05 ± 0.16^{a}	9.05 ± 0.31 ^a	9.10 ± 0.09	7.38	0.55	NS

Means with different superscripts within the same raw are significantly (P<0.05) different. N = Number of observation; ATJK = Adami Tulu Jido Kombolcha; CV = coefficient of variation; LSD = least significant difference; SL = significant Level, NS=Not Significant, *= Significant (P<0.05).

study, on the other hand, was slightly lower than the result of Zelalem and Faye (2006) who reported total bacterial count of 8.34, 8.63 and 8.18 ml⁻¹, respectively on milk samples collected from small scale, large scale producers and research centers.

Generally, the higher total bacterial counts are the indication of a diseased udder, unsanitary handling of milk, or unfavorable storage temperatures (Benson, 2001; Biruk et al., 2009). Coliform counts did not show significant difference (P>0.05) among districts (Table.1). The average coliform counts in the present study (4.35±0.06) is comparable with the finding of Alganesh et al. (2007) which reported average coliform counts of 4.46 m⁻¹ in milk samples collected from small holder producers in East Wollega. It is also comparable with the result of Asaminew and Eyassu (2011) which reported mean coliform count of 4.49 ml⁻¹ in milk sampled from around Bahir Dar and Mecha district. On the other hand, the result of this study was lower than the report of Zelalem and Faye (2006) obtaining higher coliform count of 6.57ml⁻¹ for cows' milk collected from different producers in the central highland of Ethiopia.

Coliform counts can indicate fecal contamination or contamination from equipment that has not been properly cleaned and sanitized (Schmidt, 2008; Bintsis et al., 2008; Biruk et al., 2009). Therefore, the higher coliform count observed in general, could be due to the initial contamination of the raw milk samples either from the cows, the milkers, milk containers and the milking environment.

Chemical composition of milk

Table 2 shows the result of fat, protein and solid-not-fat (SNF) percents in the study area. Protein and SNF percents of milk did not show significant differences (P>0.05) among districts. The current result for fat, SNF and protein were 5.48 \pm 0.19, 9.10 \pm 0.09 and 3.46 \pm 0.04, respectively (Table 2). The current result have similarity with the results obtained by Rehrahie and Yohannes (2000) which reported 5.88% fat and 9.27% SNF, but a little higher than 2.67% protein. The result of fat, protein and solid-not-fat (SNF) percents in the current study was respectively lower than, comparable and higher than the finding of Alganesh et al. (2007) obtaining 6.05% fat, 3.31% protein and 8.22% SNF. The result of present finding was also comparable with the result of Zelalem et al. (2004) in terms of fat and protein percents, indicating 5.43 and 3.17%, respectively, but to some extent higher in terms of SNF (8.43%). Fat and protein present in the current study fall within the acceptable range which is between 2.5 to 6.0% and between 2.9 to 5.0% for fat and protein respectively (O'Connor, 1994).

Moreover, there was significance difference (P<0.05) between breeds and among districts for fat percent. Accordingly, local Arsi zebu cows (5.87 \pm 0.25%) were observed to have high fat percent than crossbred cows (5.02 \pm 0.25%). This could be due to the exotic blood level of the crosses (they contain 50 to 75% Holstein Friesians blood level). The genetic factor of both breeds may contribute to the higher percent of fat for crossbred

cows. Friesians contains about 3.5% fat and zebu cows can give milk containing up to 7% fat (O'Connor, 1994). As far as districts are concerned, significant differences might be due to the difference in terms of available feed resources in the respective districts.

Conclusions

The higher coliform counts and total bacterial counts observed in the present study could be due to contamination of raw milk samples either from the cow, the milker, milk container and the milking environment. Therefore, sanitary measures should be installed at all stages starting from production to consumption in the study area since milk is a known vehicle for a number of human pathogens. However, the chemical compositions of the milk collected from all districts met the acceptable standards; this requires further investigation with different level and type of feeds across the season.

REFERENCES

- Alganesh T, Ofodile LN, Fikadu B (2007). Microbial quality and chemical composition of raw whole milk from Horro cattle in East Wollega, Ethiopia. Available on: www.ajol.info/index.php/ejesc/article/View/41995
- American Public Health Association (APHA) (1992). Standard Method
- for the Examination of Dairy Products. 16th ed., APHA, Washington, pp. 213-223.
- Asaminew T, Eyassu S (2011). Microbial quality of raw cow's milk collected from farmers and dairy cooperatives in Bahir Dar Zuria and Mecha district, Ethiopia. Agric. Biol. J. N. Am. 2(1):29-33
- Benson HJ (2001) Microbiological Applications Laboratory Manual. Laboratory Manual for General Microbiology. Eighth Edition. The McGraw-Hill Companies, New York.
- Bintsis T, Angelidis AS, Psoni L (2008). Modern Laboratory Practices: Analysis of Dairy Products. In: Advanced dairy science and technology. Britz, T. J. and Robinson R. K. (eds). Blackwell Publishing Ltd, Oxford, UK. DOI: 10.1002/9780470697634.ch6.
- Biruk H, Samson W, Zelalem Y (2009). Microbial properties of milk and traditionally produced Ethiopian fermented milk products: a review. Proceedings of the 17th Annual conference of the Ethiopian Society of Animal Production (ESAP) held in Addis Ababa, Ethiopia, September 24 to 26. pp. 137-148

- Chambers JV (2002). The Microbiology of Raw Milk. Dairy Microbiology Handbook. Third Edition. Edited by Richard K. Robinson. John Wiley and Sons, Inc., New York. pp. 39-90.
- Edgar S (1998). Milk and Dairy Product Technology. Marcel Dekker, inc, New York. pp. 22-23.
- Lemma F (2004). Assessment of butter quality and butter making efficiency of new churns compared to smallholders' butter making techniques in East Shoa Zone of Oromia. MSc thesis, Alemaya University, Alemaya, Ethiopia, 129 pp.
- Marth EH (1978). Standard Methods for the Examinations of Dairy Products. American Public Health Association, Washington, DC, 416 pp.
- McDonald P, Edwards RA, Greenhalgh JFD, Morgan CA (1995). Animal Nutrition. 5th edition. Longman Scientific and Technical. Copublished in the United States with John wiley and Sons, Inc., New York, 607 pp.
- Mogessie A (1990). Microbiological quality of Ayib, a traditional Ethiopian cottage cheese. Int. J. Food Microbiol. 10(1990):263-268.
- O'Connor CB (1994). Rural dairy technology. ILRI training manual No.1. International Livestock Center for Africa (ILCA), Addis Ababa, Ethiopia, 133 pp.
- Rehrahie M, Yohannes G (2000). Effect of supplementary concentrate feeds with different ingredients for local and crossbred cows on major milk constituents. Proceedings of 7th annual conference of Ethiopian Society of Animal Production (ESAP) held in Addis Ababa, Ethiopia, May 26-27 1999. pp. 263-267.
- Richardson GH (1985). Standard Methods for the Examination of Dairy Products. 15th ed. American Public Health Association. Washington, D. C, pp. 168-196.
- SAS (2002). SAS Users' guide, Statistical Analysis System (SAS) Institute, Inc, Cary, NC.
- Schmidt RH (2008). Microbiological considerations related to dairy processing. In: Dairy processing and quality assurance. Chandan, R., Kilara, A. and Shah, N (eds). A John Wiley and Sons, Ltd., Publication, New Delhi.
- Zelalem Y, Faye B (2006). Handling and microbial load of cow's milk and *Irgo*- fermented milk collected from different shops and producers in central highlands of Ethiopia. Eth. J. Anim. Prod. 6(2):67–82.
- Zelalem Y, Yohannes G, Gizachew B, Alemu GW, Sendros D (2004). Major milk chemical composition and feed dry matter intake of multiparous Boran cows as affected by level of milk production under bucket feeding and partial suckling calf-rearing methods. Proceedings of the 11th annual conference of the Ethiopian Society of Animal Production (ESAP) held in Addis Ababa, Ethiopia, August 28-30, 2003, pp. 397-402.