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Chemical composition and microbiological quality of Dhanaan: traditional fermented camel milk produced in eastern Ethiopia

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Abstract

The composition and microbiological quality of traditional and laboratory made Dhanaan were investigated in this study. Traditional Dhanaan samples and fresh camel milk used to prepare Dhanaan in the laboratory were collected from pastoralists in Dundumas, Duasho and Gabagabo kebeles in Jijiga district, eastern Ethiopia. The overall mean (±SD) values for pH, titratable acidity, total protein, fat, total solids, solids-not-fat and ash were 4.18 ± 0.29, 1.75 ± 0.34% lactic acid , 4.11± 0.67%, 2.50 ± 0.60%, 11.08 ± 2.47%, 8.64 ± 2.08% 0.96 ± 0.03% and 4.04 ± 0.25, 1.54 ± 0.26% lactic acid, 4.04 ± 0.42%, 2.39 ± 0.56%, 11.83 ± 1.24%, 7.79 ± 1.22%, 0.99 ± 0.09%, respectively for traditional and laboratory made Dhanaan, respectively. No significant difference ($p > 0.05$) was observed between the two Dhanaan types for all the physicochemical parameters considered. The average (±SD) total bacteria, coliform, lactic acid bacteria (grown on MRS and M17 agars), non-lactic acid bacteria and yeast and mould counts were 6.26 ± 1.01l, 5.88 ± 0.84, 5.88 ± 0.97l, 5.98 ± 0.73, 6.25 ± 0.92, 7.05 ± 0.90 $\log_{10}$ cfu/ml and 5.50 ± 0.62, 3.76 ± 0.29, 5.52 ± 0.55, 5.80 ± 0.81, 5.50 ± 0.62, 5.65 ± 0.62 $\log_{10}$ cfu/ml, for traditional and laboratory made Dhanaan samples, respectively. All but lactic acid bacteria counts of traditional Dhanaan were significantly ($p<0.05$) higher than that of laboratory made Dhanaan. The results indicated that the quality of traditional Dhanaan was poor as compared to that made in the laboratory. This calls for the need for hygienic practices during preparation and handling of Dhanaan.

Introduction

Ethiopia has 2.4 million heads of dromedary camels which rank the country third in the world in camel population (FAOSTAT, 2009). The one-humped camel *(Camelus dromedarius)* plays an important role as a primary source of subsistence in the lowlands of Ethiopia. In these areas, camels are mainly kept for milk production and produce milk for a longer period of time even during the dry season when milk from cattle is scarce (Tafesse *et al*., 2002). The annual camel milk production in Ethiopia is estimated to be 165, 117 metric tonnes (FAOSTAT, 2012).

Hadush *et al.* (2008) reported that in most pastoral communities in Ethiopia, camel milk is consumed either fresh or in varying degrees of sourness without heat treatment thus can pose a health hazard to the consumers. The practice of storing milk and milk products in unhygienic containers, pooling milk from different suppliers, prolonged transport time and the high ambient temperature increases the chance of contamination and spoilage of milk (Farah *et al.*, 2007). Milk is an ideal medium for the growth of microbes and loses its quality within a short period of time if not preserved in some way. Poor handling and processing practices of milk usually result in undesirable products. Spontaneous nature of milk fermentation process common in arid areas can result in undesirable products that are sometimes even risky or dangerous for human health (Farah *et al*., 2007).

Traditional fermented camel milk has different names in different countries. In Somalia and Kenya, camel milk is consumed either fresh or in the form of fermented milk, known as Suusac (Farah *et al*., 2007). To prepare Suusac, the milk is left in isolated and warm place, often in a covered container sheltered from dust for 24–48 hours until it becomes sour. The ambient temperature is usually between 25°C and 35°C.

Seifu (2007) reported that pastoralists in Shinile and Jijiga area of eastern Ethiopia produce a traditional fermented camel milk called Dhanaan for its advantages like perceived high nutritional value,

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for it enables collection of milk over a few days and delivery of the milk to the market when surplus milk is produced, its high demand by urban dwellers, it quenches thirsty, preference of consumers for its taste and its long shelf life as compared to raw camel milk. However, this product which is very important in securing food for both pastoralists and urban dwellers even under very harsh seasons of the year didn't get enough attention and hasn't been assessed scientifically for its composition and microbial quality so far. Therefore, this study was conducted with the objectives of determining the physicochemical properties and microbiological quality of laboratory made and traditional Dhanaan produced in eastern Ethiopia.

Materials and Methods

Sampling procedures

Three kebeles (kebele is the lowest administrative unit) namely Dundumas, Duasho and Gabagabo were purposively selected from Jijiga district of Somali Regional State, eastern Ethiopia based on camel possession, accessibility and Dhanaan production. Traditional Dhanaan samples were collected from nine pastoralists (three households each from Dundumas, Duasho and Gabagabo kebeles). Nine Dhanaan samples (two days old) were collected at a time from the selected kebeles three times in July 2011. For preparation of Dhanaan in the laboratory, a total of nine raw camel milk samples were collected from three households (one from each of the above stated kebeles).

Dhanaan (500 ml) and raw camel milk (250 ml) samples were taken aseptically from containers of each producer and placed into separate sterile glass bottles. Then the samples were labeled and transported by placing in icebox to the Dairy Technology Laboratory of Haramaya University. The composition and microbial quality of the Dhanaan samples were analyzed within 24 h of collection.

Physicochemical analysis of Dhanaan

The pH was measured by a digital pH meter (EUTEHC, Model P/N: 54X002606, Malaysia). Acidity (% lactic acid) of the Dhanaan sample was determined by titrating 9 ml of sample with 0.1N NAOH solution, crude protein (%N x 6.38) was determined by the Kjeldahl method, fat by Gerber method, total solids and ash were determined according to Richardson (1985). Solids-not-fat content was determined by subtracting the percentage fat from %total solids (O'Mahony, 1988).

Microbiological analysis of Dhanaan

A Standard Plate Count Agar (Oxoid, CM0325: UK), Violet Red Bile Agar (Oxoid, V37720: UK), MRS (Oxoid, UK) and M17 (Oxoid, UK) agars, Nutrient Agar (Oxoid, UK), and Potato Dextrose Agar (Oxoid, UK) were used for total bacterial count, coliform count, lactic acid bacteria count, non-lactic acid bacteria count and yeast and mould count, respectively. All counts were determined according to Richardson (1985).

Statistical analysis

Microbial count data was first transformed to logarithmic values (log_{10}) before statistical analysis. The physiochemical properties and the transformed microbial count data of traditional and laboratory made Dhanaan samples were compared by the analysis of variance technique using SAS version 9.1 (SAS, 1999).

Results and Discussion

Physical characteristics of Dhanaan

Dhanaan is a traditional fermented dairy product made by pastoralists in eastern Ethiopia from raw milk of camels through spontaneous fermentation. Under normal conditions, Dhanaan has a white opaque color, sour taste and thin consistency. However, it shows syneresis when stored for a longer period. It has similar appearance like cow milk Ergo (Ethiopian traditional fermented milk) but has very thin gel. Farah *et al.* (1990) reported that the consistency of similar fermented camel milk called Suusac produced in Kenya was found to be thin and it precipitates in the form of flakes rather than a coagulum after fermentation. On the other hand, Rahman *et al.* (2009) reported that Shubat (traditional fermented camel milk produced in China) is more or less similar to yoghurt in appearance but unlike yoghurt Shubat is liquid rather than creamy, sparkling due to its CO_2 production and has a high degree of sourness. Hashim *et al.* (2009) reported that camel milk produces thin, flowing and very soft yoghurt based on their study in United Arab Emirates on quality and acceptability of set-type yoghurt made from camel milk.

Proximate composition of traditional and laboratory made Dhanaan

The compositions of traditional and laboratory made Dhanaan samples are indicated in Table 1. No significant difference ($p>0.05$) was observed among the two Dhanaan types for pH, titratable acidity, protein, fat, total solids, solids-not-fat and ash contents. The pH value of Dhanaan observed in

Table 1. Chemical composition and acidity of traditional and laboratory made Dhanaan

Variables	Traditional Dhanaan	Laboratory made Dhanaan
Fat (%)	4.11± 0.67	4.04 ± 0.42
Protein (%)	2.50 ± 0.60	2.39 ± 0.56
Total solids (%)	11.08 ± 2.47	11.83 ± 1.24
Solids-not-fat (%)	8.64 ± 2.08	7.79 ± 1.22
Ash (%)	0.96 ± 0.03	0.99 ± 0.09
pH	4.18 ± 0.29	4.04 ± 0.25
Titratable acidity (% lactic acid)	1.75 ± 0.34	1.54 ± 0.26

No statistically significant difference was observed between mean values of traditional and laboratory made Dhanaan for all parameters (p > 0.05); values in the Table are means ± SD of two replications

the current study is in agreement with the results of Rahman *et al.* (2009) who reported pH values of 4.1 ± 0.07 and 3.7 ± 0.15 for Shubat collected from Kanas and Borjin areas of China, respectively. However, it is relatively higher than the pH value (3.41 ± 1.12 and 3.82 ± 0.49) reported by Hassan *et al.* (2008) for Gariss (fermented camel milk produced in Sudan) samples collected from transhumance and nomadic camel herders in Sudan, respectively. But it is lower than the pH value (5.59) reported for camel milk stored for two days at a temperature of 25-30°C (Omer and Eltinay, 2009).

The titratable acidity observed in this study is slightly lower than that reported for Gariss samples (2.29 ± 1.25% and 2.24 ± 0.68% lactic acid) which were collected from transhumance and nomadic camel herders in Sudan, respectively (Hassan *et al.*, 2008). El-Zubeir *et al.* (2009) reported that contamination increases the acidity of fermented camel milk from comparative study they conducted on Gariss with and without pasteurizing raw camel milk prior to fermentation.

The fat percentage of Dhanaan observed in the present study is similar to the results reported by Farah *et al.* (1990) for three different samples of camel milk Suusac fermented using two different mesophilic lactic cultures and Suusac made without inoculation of mesophilic bacteria which was found to be 4.1%, 4.0% and 4.0% fat, respectively. But it is different from the results of Hassan *et al.* (2008) for Gariss collected from transhumance and nomadic camel herders which was found to be 4.85 ± 0.66% and 3.46 ± 1.18%, respectively. El-Zubeir *et al.* (2009) reported fat percentages of 3.0 ± 0.445% and 3.0 ± 3.076% for Gariss made from pasteurized and non-pasteurized camel milk in Sudan.

The average protein content of Dhanaan observed in the present study is slightly lower than the findings of Hassan *et al.* (2008) who reported protein contents of 2.32 ± 0.59% and 2.58 ± 0.69% for Gariss produced by transhumance and nomadic camel herders, respectively in Sudan. El-Zubeir *et al.* (2009) reported that Gariss prepared from pasteurized and non-pasteurized camel milk in Sudan had protein contents of 3.2 ± 0.311% and 3.1 ± 0.14%, respectively which are higher than that observed in the present study. The average total solids content of Dhanaan obtained in this study is in line with the reports of Hassan *et al.* (2008) for Gariss (11.29 ± 1.40% and 9.81 ± 2.32%) collected from transhumance and nomadic camel herders, respectively in Sudan. Farah *et al.* (1990) also reported total solids content of 12.5 -12.7% for Suusac made in Kenya.

Generally, no significant differences (p > 0.05) were observed in chemical composition between traditional and laboratory made Dhanaan in the present study. The composition of fermented camel milk can be affected by the composition of the milk used for processing rather than the processing condition applied. Hassan *et al.* (2008) reported that the difference in the chemical composition of Gariss made by transhumance and nomadic camel herders of Sudan was due the variation in composition of the milk caused by water availability for the camels, stage of lactation and availability of green fodder as well as the management system under which the herds were kept.

Microbial counts of traditional and laboratory made Dhanaan

The total bacterial count (TBC) of laboratory made Dhanaan was significantly (p<0.05) lower

Table 2. Microbiological counts (log_{10} cfu/ml) of traditionally made and laboratory prepared Dhanaan

Microbial Count	Traditional Dhanaan	Laboratory Made Dhanaan
Total bacteria	6.26 ± 1.01[a]	5.50 ± 0.62[b]
Coliform	5.88 ± 0.84[a]	3.76 ± 0.29[b]
Lactic acid bacteria (MRS)	5.88 ± 0.97	5.52 ± 0.55
Lactic acid bacteria (M17)	5.98 ± 0.73	5.80 ± 0.81
Non lactic acid bacteria	6.25 ± 0.92[a]	5.50 ± 0.62[b]
Yeast and mould	7.05 ± 0.90[a]	5.65 ± 0.62[b]

[*]Superscripts in the same row having different letters indicate significant difference (p < 0.05) among the Dhanaan samples; values in the Table are means ± SD of three replications.

than that of the traditional Dhanaan. Hassan *et al* (2006) reported TBC of 6.16 ± 0.03 log_{10} cfu/ml and 6.45 ± 0.011 log_{10} cfu/ml for Gariss prepared in the laboratory through fermentation for 42 h at 25°C and 37°C, respectively. But the TBC of Gariss collected from transhumant and nomadic pastoralists were 7.26 ± 0.49 and 7.57 ± 0.23 log_{10} cfu/ml, respectively (Hassan *et al*., 2008) which is higher than the TBC observed in the current study.

The lower TBC observed in laboratory made Dhanaan suggests that Dhanaan of good microbial quality can be produced by applying hygienic manufacturing steps. In the study area, pastoralists wash milk vessels using untreated well water and do not wash the udder of camels before and after milking. Moreover, the personal hygiene of the milkers is generally poor which might have contributed to contamination of the milk and caused the high bacteria count observed in the traditional Dhanaan. Higher bacterial counts result in poor quality milk and inferior milk products (Bruhn, 1986). The number of microbes causing spoilage, the level of activity and the microbial environment of a given product give some indication of whether the milk or dairy product can be stored successfully (Kiss, 1984).

The coliform count (CC) of laboratory made Dhanaan was significantly ($p<0.05$) lower than the traditional Dhanaan (Table 2). The CC observed in the present study is much higher than the findings of Hassan *et al.* (2008) who reported that no coliform growth was observed in Gariss. Similarly, no coliform was found in Shubat as reported by Rahman *et al.* (2009). Lore *et al*. (2005) reported a coliform count of < 1 log_{10} cfu/ml for Suusac. The higher coliform count observed in the present study might be attributed to the initial contamination of the milk samples either from the lactating camels, the milkers, milk containers or the milking environment which were of poor hygienic condition. No cleaning of the udder was practiced and milking was done in the open air. Poor personal hygiene and use of untreated well water might have contributed to the contamination of the milk by coliforms and consequently the Dhanaan produced.

No significant difference ($p > 0.05$) was observed between traditional and laboratory made Dhanaan samples for lactic acid bacteria (LAB) that have grown on both MRS and M17 agar (Table 2). Lactic acid bacteria (LAB) play major role in milk fermentation. The LAB count observed in the present study is lower than the findings of Rahman *et al*. (2009) who reported that Shubat collected from different regions in China had LAB count that ranged from 6.8 ± 0.82 - 7.3 ± 0.24 log_{10} cfu/ml. The LAB count of the present study is comparable with the results of Hassan *et al.* (2008) who reported mean counts of LAB grown on MRS agar for Gariss collected from transhumant and nomadic herders of Sudan to be 6.83 ± 0.33 and 6.5 ± 0.32 log_{10} cfu/ml, respectively. The same authors also reported counts of LAB grown on M17 agar to be 6.47 ± 0.35 and 6.85 ± 0.33 log_{10} cfu/ml, respectively for Gariss collected from transhumant and nomadic herders in Sudan which is slightly higher than the result of the current study. Hassan *et al*. (2006) reported *Lactobacillus* counts of 5.44 ± 0.033 and 5.53 ± 0.029 log_{10} cfu/ml for Gariss samples incubated in the laboratory for 42 h at 25°C and 37°C, respectively. Among the lactic acid bacteria which play a major role in fermentation of camel milk, the genus *Lactobacillus* which preferably grows on MRS agar was reported as a dominant flora in fermented camel milk products and it accounted for 74% in Gariss and 8.6 log_{10} cfu/ml in Suusac (Abdel Rahman *et al*., 2009).

Lactic acid bacteria belonging to the genus *Streptococcus* and *Lactococcus* preferably grow on M17 agar medium. The mean counts of LAB grown on M17 agar in the present study were 5.98 ± 0.73 and 5.80 ± 0.81 log_{10} cfu/ml for traditional and laboratory made Dhanaan samples, respectively (Table 2). Hassan *et al.* (2008) reported that the mean LAB grown on M17 agar for Gariss collected from transhumant and nomadic camel herders were found to be 6.47 ± 0.35 and 6.85 ± 0.33 log_{10} cfu/ml, respectively which is higher than the result of the current study. Ashmaig *et al*. (2009) reported that the genus *Lactobacillus* (66.6%) and *Lactococcus* (33.3%) were the main LAB that play a dominant role in the fermentation of Gariss.

Study on the microbiological quality, biochemical changes and sensory evaluation of camel milk fermented by selected starter cultures conducted in Sudan indicated that using mixed starter culture of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* resulted in more LAB counts and produce more acid compared to single starter culture (Osman *et al*., 2010). Abdel Rahman *et al.* (2009) reported that the final fermented product obtained from camel milk was free from pathogenic bacteria, had total coliform, yeast and mould counts of less than 10 cfu/ ml and acceptable sensory quality. Farah *et al.* (1990) also recommended the use of mesophilc bacteria for fermentation of camel milk for it has more fermenting potential and results in a more acceptable aroma of Suusac. Therefore, isolation of the dominant lactic acid bacteria from Dhanaan for use as a starter culture may help to produce Dhanaan with better quality and consumer acceptability.

The non-lactic acid bacteria count of traditional Dhanaan was significantly ($p<0.05$) higher than that of the laboratory made Dhanaan (Table 2). Similar result was reported by Ahmed *et al.* (2010) in which Gariss sample had higher counts ($2.7x10^6$ cfu/ml) of non-lactic acid bacteria count. The mean yeast and mould count (YMC) of traditional and laboratory made Dhanaan were 7.05 ± 0.90 and 5.65 ± 0.62 log_{10} cfu/ml, respectively (Table 2). Similar results (6.99 ± 0.13 and 7.02 ± 0.3 log_{10} cfu/ml) were reported for yeast count by Hassan *et al.* (2008) for Gariss collected from transhumant and nomadic herders in Sudan, respectively. Rahman *et al*. (2009) reported yeast count for Shubat obtained from two different places (Kanas and Borjin areas in China) to be 4.3 ± 0.44 and 4.7 ± 0.14 log_{10} cfu/ml, respectively. Lore *et al*. (2005) reported YMC < 2.1 log_{10} cfu/ml for Suusac which is lower than YMC observed in the present study.

Yeast and mould count of traditional Dhanaan was significantly ($p<0.05$) higher than that of laboratory made Dhanaan (Table 2). The high YMC observed in traditional Dhanaan might be attributed to contamination from the air, the containers or poor hygienic conditions followed by the producers and poor personal hygiene of individuals engaged in the production of traditional Dhanaan. Yeasts and moulds are considered to be spoilage organisms. Some moulds, however, are public health concerns due to their production of mycotoxins, which are not destroyed during food processing or cooking (McLandsborough, 2005).

In general, higher microbial counts were observed in traditionally made Dhanaan samples as compared to laboratory made Dhanaan. This suggests the possibility of improving the microbial quality of Dhanaan by following hygienic processing conditions. In the present study, milk samples used for production of Dhanaan in the laboratory were not pasteurized. Earlier reports (Hassan *et al*., 2006) indicated that the microbiological quality of fermented camel milk (Gariss) was significantly improved by pasteurizing the milk prior to fermentation. Thus, future works aimed at improving the quality of Dhanaan should consider pasteurization of the milk prior to Dhanaan production.

Conclusion

Dhanaan made traditionally by pastoralists was found to be of substandard quality and this was attributed to the poor hygienic conditions followed during handling and preparation of Dhanaan. This calls for the need for teaching pastoralists to follow proper hygienic practices during milking, milk handling and preparation of Dhanaan. In the present study, lactic acid bacteria (LAB) responsible for fermentation of camel milk and production of Dhanaan were not isolated. Thus, isolation of LAB responsible for Dhanaan production deserves detailed study in the future.

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